Top-down and bottom-up controls on Dalmatian toadflax (*Linaria dalmatica*) performance along the Colorado Front Range, USA

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Abstract Plant performance is influenced by both top-down (e.g., herbivores) and bottom-up (e.g., soil nutrients) controls. Research investigating the collective effects of such factors may provide important insight into the success and management of invasive plants. Through a combination of observational and experimental field studies, we examined top-down and bottom-up effects on the growth and reproduction of an invasive plant, *Linaria dalmatica*. First, we assessed attack levels and impacts of an introduced biocontrol agent, the stem-mining weevil *Mecinus janthinus*, on *L. dalmatica* plants across multiple years and sites. Then, we conducted a manipulative experiment to examine the effects of weevil attack, soil nitrogen availability, and interspecific competition on *L. dalmatica*. We found substantial variations in weevil attack within populations as well as across sites and years. Observational and experimental data showed that increased weevil attack was associated with a reduction in plant biomass and seed production, but only at the highest levels of attack. Nitrogen addition had a strong positive effect on plant performance, with a two-fold increase in biomass and seed production. Clipping neighboring vegetation resulted in no significant effects on *L. dalmatica* performance, suggesting that plants remained resource limited or continued to experienced belowground competitive effects. Overall, our research indicates that *M. janthinus* can exert top-down effects on *L. dalmatica*; however, weevil densities and attack rates observed in this study have not reached sufficient levels to yield effective control. Moreover, bottom-up controls, in particular, soil nitrogen availability, may have a large influence on the success and spread of this invasive plant.

Keywords *Linaria dalmatica* · Biological control · *Mecinus janthinus* · Invasive plant · Herbivory · Soil nitrogen availability · Plant competition

Introduction

Non-native invasive plants are responsible for significant environmental damage and economic costs worldwide (reviewed in Vitousek et al. 1996; Mack et al. 2000). A multitude of hypotheses (e.g., fluctuating resource availability, biotic resistance, enemy release, etc.) have been proposed to explain plant invasions (reviewed in Inderjit et al. 2005; Richardson and Pyšek 2006). Recent reviews highlight the overlap and theoretical redundancy in many of these hypotheses and recommend a more synthetic conceptual

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framework employing basic ecological concepts for understanding invasions (Catford et al. 2009; Gurevitch et al. 2011; Seastedt and Pyšek 2011). When considering an invasive or native species, plant performance (i.e., growth and reproduction) is likely to be influenced by the collective effects of interacting abiotic and biotic factors; moreover, the success of a species in a particular habitat may be context-dependent due to spatial and temporal heterogeneities in these controlling factors.

Understanding the importance of top-down consumer and bottom-up resource controls on plant productivity and community structure is a central and longstanding theme in ecology (Hairston 1960; Gruner et al. 2008). Over the years, research has shown that the relative strengths of these factors vary across plant communities and ecosystems (Borer et al. 2006; Hillebrand et al. 2007; Gruner et al. 2008). Further, top-down and bottom-up forces are often interdependent, with interactions between consumers and resources yielding synergistic or reduced effects on producer biomass in some cases (Gruner et al. 2008). Thus, the research examining the combined effects of both biotic and abiotic factors on plant growth and reproduction may be especially important for understanding the success of invasive plants and for informing management decisions related to these species.

Insect herbivores play an important role in reducing plant performance, regulating plant populations, and shaping the composition of plant communities (Bigger and Marvier 1998). Two prominent hypotheses in invasion biology (enemy release and evolution of increased competitive ability) propose that escape from natural enemies (e.g., herbivores) contributes to the invasion success of introduced plants (Blossey and Notzold 1995; Keane and Crawley 2002). Based on the same rationale, insect biological control programs reintroduce invasive plants with co-evolved specialist herbivores from their native range in an attempt to reduce the abundance and spread of these species. For a number of invasive plants, insect biocontrol agents demonstrate great potential as top-down controls (e.g., McEvoy et al. 1993; Seastedt et al. 2003), and understanding the influence of resource conditions on the success of such controls presents an ongoing challenge (Lejeune et al. 2005; Blumenthal 2006; Knochel and Seastedt 2010). Myers and Bazely (2003) indicate that insect biocontrol efforts have led to regional control of approximately 40 invasive plant

species. However, those authors also note that little is known about the effects of biocontrol efforts for many other invasive plants and suggest that the greatest weakness of biocontrol programs is the failure to monitor and evaluate the efficacy of released insect biocontrol agents.

Other prominent hypotheses in invasion biology, such as the fluctuating resource hypothesis, stress the importance of bottom-up controls (Davis et al. 2000). In particular, soil nitrogen availability may be an important factor influencing the colonization, establishment, and spread of non-native plants. Numerous studies indicate that increased nitrogen availability can contribute to plant invasions (e.g., Huenneke et al. 1990; Suding et al. 2004). However, the effects of nitrogen enrichment on plant growth and reproduction may depend on biotic interactions (e.g., competition and herbivory) in the invaded habitat (e.g., Seastedt and Suding 2007; Knochel et al. 2010). In this study, we investigated top-down herbivore and bottom-up resource effects on the invasive plant, Dalmatian toadflax, *Linaria dalmatica* (L.) P. Mill. (Plantaginaceae). Specifically, we conducted observational and experimental field studies to examine the influence of an introduced biological control agent, soil nitrogen availability, and interspecific competition on *L. dalmatica* performance.

Linaria dalmatica is an invasive species found throughout the United States and Canada (Wilson et al. 2005). In a review by Duncan et al. (2004) summarizing the environmental, economic, and societal losses caused by invasive plants, this species was identified as one of the 16 key invasive plants on rangelands and wildlands in the United States. Although the extent of *L. dalmatica* infestation was lower than other important invaders, the estimated spread rate (8–29%) of this species was among the highest of the key invaders. Chemical and mechanical controls of *L. dalmatica* have been largely unsuccessful except in small infestations, and biological control is thought to be the most promising management strategy for this invasive plant (Wilson et al. 2005). In particular, the stem-mining weevil, *Mecinus janthinus* Germar (Coleoptera: Curculionidae), has demonstrated great potential as a biological control agent of *L. dalmatica* (Saner et al. 1994; Peterson et al. 2005; Van Hezewijk et al. 2010; Schat et al. 2011).

In this study, we collected field data from multiple sites across multiple years to evaluate the effectiveness of this species as a biocontrol agent of *L. dalmatica*

along the Colorado Front Range. Using these data, we characterized weevil attack levels among sites and years and examined the consequence of weevil attack on plant biomass and reproduction potential (number of flowers and seed capsules produced). In addition, we conducted a manipulative field experiment to investigate the influence of soil nitrogen availability and interspecific competition on plant growth, reproduction, and weevil attack. Through this experiment, we addressed the following questions: (1) What is the relative strength of top-down herbivore controls (weevil attack) and bottom-up resource constraints (soil nitrogen availability and interspecific competition) on plant growth and reproduction? (2) Does plant response to nitrogen availability depend on competitive relationships with neighboring plants? and (3) Does nitrogen availability and/or interspecific competition influence weevil attack levels?

Methods

Study organisms

Linaria dalmatica is an invasive species in the United States and Canada, which was originally introduced to North America in the early 1900s as an ornamental plant from its native range in Eurasia. The biology and biological control of this species are described in detail by Wilson et al. (2005). *L. dalmatica* produces two groups of chemical defenses, iridoid glycosides (Jamieson and Bowers 2010), and alkaloids (Gröger and Johne 1965), and these compounds have been implicated in its impacts and invasion success. It is a perennial plant, which reproduces both sexually and clonally. *L. dalmatica* is a self-incompatible, insect pollinated species, and plants can produce high seed yields (estimates ranging from 60 to 300 seeds per capsule and up to 500,000 seeds per plant). The relative importance of seedling recruitment versus vegetative propagation for population growth and spread is not known.

Mecinus janthinus is also native to Eurasia and was released in North America (Canada and the US) during the early 1990s as a biocontrol agent for invasive *Linaria* species (Wilson et al. 2005). Adults of *M. janthinus* emerge from natal stems in late spring and early summer, feed on leaf and stem tissues, reproduce, and lay eggs in stems (Jeanneret and Schroeder

1992). Female weevils chew a hole into the stem, insert a single egg, and then cover the hole with plant tissue, leaving a distinctive oviposition scar (see Wilson et al. 2005). Complete metamorphosis occurs within stems during spring–summer months. The negative effects of adult and larval feeding include reduce photosynthetic rates, growth, and flowering (Jeanneret and Schroeder 1992; Saner et al. 1994; Peterson et al. 2005; Schat et al. 2011). In addition, plant injury by *M. janthinus* may decrease *L. dalmatica* iridoid glycosides concentrations (Jamieson and Bowers in press). Moreover, Van Hezewijk et al. (2010) showed that *M. janthinus* reduced *L. dalmatica* plant density and distribution at a regional scale in British Columbia, Canada. However, the efficacy of this biocontrol agent at a regional scale in the United States remains uncertain.

Observational study

Field site descriptions

Field surveys and plant collections were conducted from late June to mid-July of 2007, 2009, and 2010 at five field sites located along the Colorado Front Range, USA: Rabbit Mountain (40°14′13″N; 105°12′53″W), Superior-Kelsall (39°55′5″N; 105°11′56″W), Marshall Mesa (39°57′11″N; 105°13′35″W), Johnson (40°8′38″N; 105°15′55″W), and Lefthand Canyon (40°07′38″N; 105°18′37″W). Field sites ranged from 1,650 to 2,000 m in elevation and are generally characterized as dry, rocky foothills grassland habitat with vegetation dominated by mixed grasses, forbs, and shrubs. *M. janthinus* was released at two of our five study sites—Rabbit Mountain and Lefthand Canyon between 2003 and 2005. The other field sites had no documented releases of *M. janthinus*; however, this species was released at nearby locations (approximately 1–3 km from each of these study sites) during that timeframe (personal communication with local weed managers).

Data collection

We examined live and dead *L. dalmatica* stems to assess the distribution, abundance, and efficacy of *M. janthinus*. Collection methods and sample sizes varied during survey years, and Table 1 summarizes data

Table 1 Summary of observational field study data

Year	Collected	n	Data	Estimate
2007	Dead stems	290	Proportion of stems attacked	2006 Weevil attack
			Number of oviposition scars (per cm stem)	
2009	Dead stems	725	Proportion of stems attacked	2008 Weevil attack
			Number of oviposition scars (per cm stem)	
2009	Live stems	75	Proportion of stems attacked	2009 Weevil attack
			Number of oviposition scars (per cm stem)	
			Number of flowers/seed capsules (per ramet)	2009 Plant performance
			Biomass (per ramet)	
2010	Dead stems	210	Proportion of stems attacked	2009 Weevil attack
			Number of oviposition scars (per cm stem)	
2010	Live stems	337	Proportion of stems attacked	2010 Weevil attack
			Number of oviposition scars (per cm stem)	
			Number of flowers/seed capsules (per ramet)	2010 Plant performance
			Biomass (per ramet)	

collected across years and sites. In general, individual ramets were collected haphazardly from distinct *L. dalmatica* plants and/or patches located throughout each field site (areas ranging from approximately 500–1,500 m²). Because *L. dalmatica* is clonal, it is impossible to determine whether distinct patches represent unique individuals without genetic analyses. Samples were taken back to the lab and were inspected for *M. janthinus* oviposition scars. For each site, we calculated the proportion of stems attacked by weevils (number of stems with oviposition scars divided by total number of stems collected). For attacked stems, we measured lengths, counted scars, and calculated a standardized measure of weevil attack (number of oviposition scars per cm stem).

Statistical analysis

Oviposition scars from dead stems reflect the previous year's weevil activity (Table 1). To determine whether oviposition scar data collected from dead stems approximate data from live stems collected during the previous summer, we performed an analysis of variance (ANOVA) comparing mean scar densities for dead-collected stems in 2010 and live-collected stems in 2009, collected from Lefthand Canyon, and found no significant difference between the two methods of estimation ($F_{1,58} = 2.86$, $P = 0.10$). Thus, in the following analyses, we combined dead and live stem scar data (Table 1). We used a χ^2

analysis to examine variation in the proportion of stems attacked by weevils across sites and years. For stems with oviposition scars, we performed an ANOVA to compare weevil attack levels across sites and years.

To evaluate the effects of *M. janthinus* on plant performance, we used ANOVA to examine variation in (1) ramet biomass, and (2) the number of flowers/fruits produced per ramet. In these analyses, we treated the number of oviposition scars as a categorical variable because the large number of stems with no oviposition scars created a highly skewed data distribution, which could not be transformed to meet ANOVA assumptions. Moreover, we presumed that plant response would not necessarily demonstrate a linear response to oviposition scar density, and that examination of categorical weevil attack levels (e.g., low, moderate, and high) would allow us to better detect plant responses. Data for 2009 and 2010 were analyzed separately because the distribution of attack levels differed in these 2 years; however, categories were created so that we could compare relative values for 2009 and 2010 and also to optimize the number of data points at each categorical level.

Experimental study

Site description

In 2010, we conducted a manipulative experiment at the Lefthand Canyon field site, approximately 0.5 km

west of the area sampled for the observational study. The background plant community was dominated by grasses including *Sporobolis cryptandrus*, *Bouteloua gracilis*, *Bromus tectorum*, and *Poa compressa*, as well as common forb species, such as *Erigeron flagellaris*, *Potentilla recta*, *Ambrosia psilostachya*, and *Alyssum parviflorum*. *L. dalmatica* exhibited a patchy distribution, representing less then 10% of the vegetation cover, which consisted of approximately 60% grasses, 35% forbs, 5% woody plants, and <1% bare ground (unpublished data).

Data collection

In order to evaluate the influence of resource availability on *L. dalmatica* performance, we conducted a full-factorial experiment manipulating nitrogen (N) availability (control and fertilized) and competition (neighbors present and neighbors clipped). In April 2010, we established 60 experimental plots within an area measuring approximately 25×70 m. Each 0.5×0.5 m plot included one haphazardly selected *L. dalmatica* plant that was randomly assigned to one of the four treatment groups (15 plots per treatment). To statistically control for any effects of initial plant size, we estimated plant size before treatments by measuring the total distance across the widest portion of the prostrate basal ramets.

In fertilized plots, we elevated soil nitrogen availability by adding granular calcium ammonium nitrate $(5Ca(NO_3)_2 \cdot NH_4NO_3 \cdot 10H_2O$; Nordst-hydro brand) in two separate additions (March 30, and May 25, 2010) for a total increase approximating 15 g N m^{-2}. This rate was chosen because it just exceeds estimates of N mineralization in our study area—1.5 to 10 g N m^{-2} year^{-1} (Burke et al. 1997). Control plots had no added N. To reduce the competitive effects of neighboring vegetation, we clipped neighboring vegetation surrounding target *L. dalmatica* plants to ground level in half of the plots ("neighbors clipped" treatment group). During the growing season, we implemented the clipping treatment three times, using the same approach as described in Knochel and Seastedt (2010). In the remaining plots, all vegetation was left intact ("neighbors present" treatment group).

Throughout this experiment, *M. janthinus* had access to all experimental plots. Because we were interested in potential interactive effects between weevil attack and plant resource availability, we allowed insects to select plants and inflict a natural gradient of plant damage rather than manipulating insect densities. This approach resulted in a broad range of weevil attack levels against which to examine plant performance and the opportunity to evaluate weevil attack levels across experimental treatments. Examination of natural variation in plant damage by insect biocontrol agents under altered abiotic and biotic conditions has proven informative in prior research (e.g., Knochel and Seastedt 2010).

In October, we harvested the aboveground biomass of target *L. dalmatica* plants (minus prostrate basal ramets which persist year round and do not produce flowers). In the laboratory, we weighed plant biomass, counted seed capsules, measured stem lengths, and calculated the number of oviposition scars per cm stem to make estimates of plant performance and weevil attack at the whole plant level. We also harvested and weighed the aboveground biomass of all neighboring plants in plots with vegetation left intact ("neighbors present" treatment group) to estimate the collective growth response of these plants to nitrogen addition relative to the response of *L. dalmatica*.

Statistical analysis

We used ANCOVA to evaluate the influence of fertilization, neighbor clipping, and weevil attack on estimates of plant performance (aboveground biomass and number of seed capsules), controlling for initial plant size. As in the observational study, weevil attack level was treated as a categorical variable. We examined three levels of weevil attack, which were selected so that sample sizes were approximately equal among groups ($n = 17$ plants per group).

To evaluate the use of oviposition scar density as a measure of plant injury, we conducted a correlation analysis examining the relationship between the number of oviposition scars and number of adult weevils completing development in stems. Finally, to determine whether fertilization and/or clipping treatments influenced weevil attack levels, we conducted an ANCOVA examining variation in oviposition scar density among treatment groups, with experimental treatments (fertilization and clipping) as main effects and initial plant and final plant size as covariates.
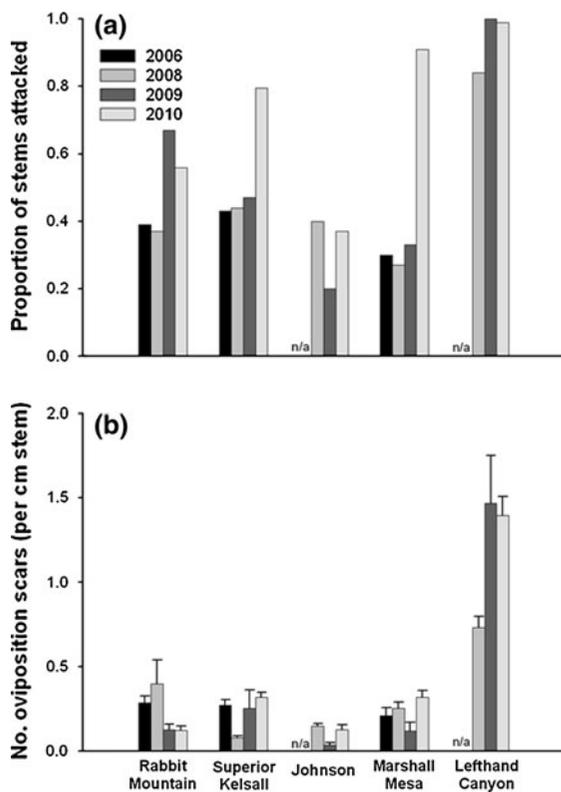
In the above analyses (observational and experimental studies), data were transformed to meet assumptions of statistical models. Specifically, plant

size estimates were log transformed, and flower/seed capsule counts were square-root transformed. Statistical analyses were performed in SYSTAT (version 11) and SAS (version 9.1).

Results

Observational study

The proportion of *L. dalmatica* stems attacked by *M. janthinus* demonstrated significant variation across sites and years, with four of the five sites showing significant overall increases from 2006 to 2010 (Fig. 1a; Table 2). By 2010, the proportion of stems attacked ranged from 40 to 100% across sites (overall

mean $= 55\% \pm 9$ SE; Fig. 1a). Oviposition scar densities varied widely within and among field sites (Fig. 1b), with the number of oviposition scars ranging from 0 to 231 per stem. ANOVA results indicated that mean differences were significant among sites ($F_{4,740} = 53.93$, $P < 0.0001$; Fig. 1b) and over time ($F_{3,740} = 3.22$, $P = 0.022$; Fig. 1b). However, there was a significant site by time interaction ($F_{10,740} = 3.84$, $P < 0.0001$; Fig. 1b). While the overall level of weevil attack increased over time, the only significant within-site increase in oviposition scar density was at Lefthand Canyon (Fig. 1b).

In general, we found mixed results in relation to the effects of weevil attack (measured as oviposition scar density) on plant performance (Fig. 2). In general, there was a significant negative relationship between weevil attack levels and aboveground biomass in 2010 ($F_{5,331} = 11.89$, $P < 0.001$; Fig. 2b), but not in 2009 ($F_{3,70} = 0.96$, $P = 0.42$; Fig. 2a). However, in 2010, there was also a positive relationship between weevil attack and biomass between the two lowest levels of attack (Fig. 2a), and this trend was similar for flower/seed capsule data as well (Fig. 2b). For the moderate-to-high attack categories, increased levels of weevil attack were associated with decreased plant reproductive output in both 2009 ($F_{3,70} = 7.02$, $P < 0.001$; Fig. 2b) and 2010 ($F_{5,331} = 15.15$, $P < 0.001$; Fig. 2b).

Fig. 1 Comparison of *M. janthinus* attack across multiple years and field sites along the Colorado Front Range, USA. Estimates of weevil attack levels across sites and years include **a** proportions of collected stems with oviposition scars and **b** mean (±SE) number of oviposition scars per cm stem (oviposition scar density). Table 1 summarizes data collected across years and includes sample sizes. Data were not collected from Johnson and Lefthand Canyon during the first study year, thus estimates of 2006 weevil attack data are missing (n/a) for those sites

Table 2 Results of χ^2 analyses examining variation in proportions of *L. dalmatica* stems attacked by *M. janthinus* from observational data across five sites and four years

Variable	χ^2	df
Differences within sites across years		
Rabbit Mountain ($n = 302$)	7.77	3
Superior-Kelsall ($n = 361$)	**38.19**	3
Johnson ($n = 214$)	3.72	3
Marshall Mesa ($n = 269$)	**73.54**	3
Lefthand ($n = 284$)	**135.71**	3
Differences within years across sites		
2006 ($n = 290$)	3.95	4
2008 ($n = 729$)	**123.48**	4
2009 ($n = 74$)	**22.44**	4
2010 ($n = 337$)	**70.57**	4

Significant differences in weevil attack are highlighted in bold ($P < 0.05$)

Fig. 2 Observational relationship between *M. janthinus* attack (oviposition scar density) and *L. dalmatica* **a** biomass and **b** reproductive potential. Individual ramets from distinct plants/patches were collected from five Colorado field sites in 2009 ($n = 75$) and 2010 ($n = 337$). Data are means ± SE
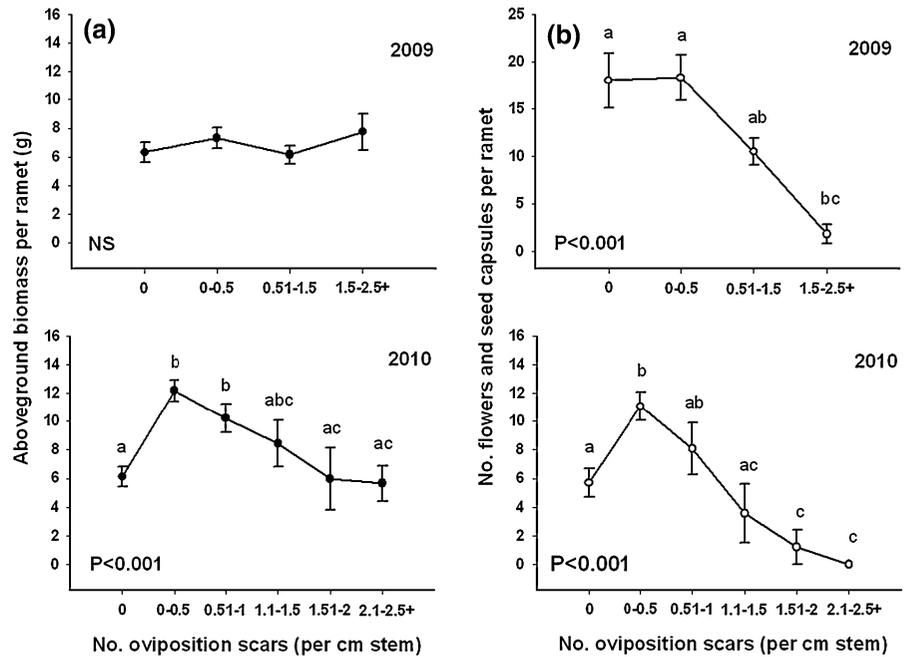
Table 3 Summary of ANCOVA examining the effects of nitrogen fertilization (control, N addition), vegetation clipping (control, neighbors clipped), and weevil attack (low, medium, high) on *L. dalmatica* performance traits, controlling for initial plant size

| Source of variation	Aboveground biomass			Number of seed capsules		
	df	F	P	df	F	P
Fertilization	**1**	**6.82**	**0.013**	**1**	**5.47**	**0.025**
Clipping	1	0.73	0.399	1	1.27	0.266
Weevil	**2**	**5.23**	**0.008**	**2**	**9.14**	**0.001**
Fertilization × clipping	1	0.64	0.430	1	0.81	0.374
Fertilization × weevil	2	1.01	0.373	2	3.03	0.060
Clipping × weevil	2	0.41	0.665	2	0.29	0.753
Fertilization × clipping × weevil	2	0.35	0.709	2	0.32	0.729
Initial plant size	1	0.05	0.822	1	0.06	0.802
Error	39			39		

Significant effects are highlighted in bold ($P < 0.05$)

Experimental study

ANCOVA results indicated a significant positive effect of nitrogen fertilization (Table 3; Fig. 3) and significant negative effect of weevil attack (Table 3; Fig. 4) on plant biomass and number of seed capsules. We found no significant effects of clipping treatments on either estimate of performance (Table 3; Fig. 3). In addition, there were no significant interaction effects or effects of initial plant size (Table 3). Overall, fertilized *L. dalmatica* plants had approximately double the biomass and number of seed capsules of plants in control plots. In comparison, the collective

aboveground biomass of neighboring plants increased by only 20% (200 ± 24.4 g m^{-2}) in response to fertilization, relative to neighboring plants in control plots (166.7 ± 16.1 g m^{-2}).

In experimental plots, the number of oviposition scars per plant ranged from 5 to 339 and the number of adult weevils from 0 to 225. There was a significant positive correlation between densities of oviposition scar and adult weevil (Pearson's $R = 0.66$, $P < 0.0001$), indicating that oviposition scars served as an adequate estimate of larvae developing to the adult stage. We found no significant effects of treatment, either fertilization or clipping, on weevil

Fig. 3 Effects of experimental treatments on *L. dalmatica* performance (*n* = 52). Fertilization increased plant **a** biomass and **b** seed production. Clipping neighboring vegetation had no significant effect on either estimate of performance. Data are mean ± SE
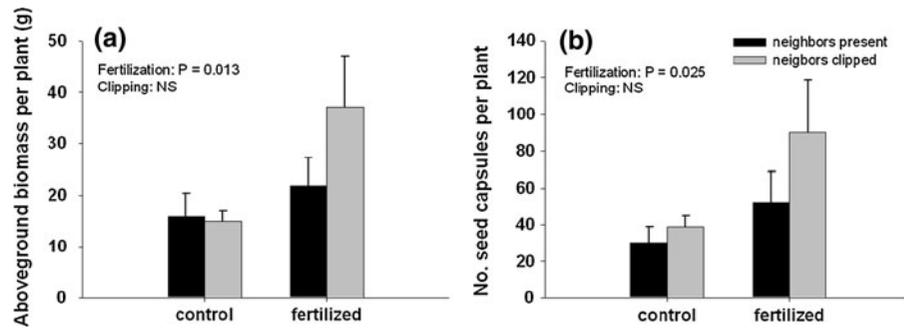
Fig. 4 Relationship between *M. janthinus* attack (oviposition scar density) and *L. dalmatica* **a** biomass and **b** seed production for plants growing in the experimental study (*n* = 52). Weevil attack categories were selected so that each of the three groups is approximately equal. Data are means ± SE
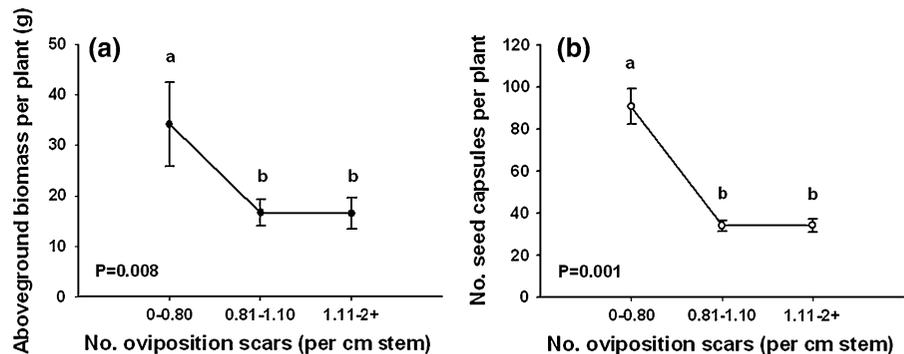
Table 4 Summary of ANCOVA examining the effect of fertilization and clipping neighboring vegetation on weevil attack levels (oviposition scar densities) with initial and final size of target *L. dalmatica* plants as covariates

Source of variation	df	*F*	*P*
Fertilization	1	1.89	0.176
Clipping	1	0.32	0.579
Fertilization × clipping	1	0.21	0.649
Initial plant size	1	0.2	0.659
Final plant size	1	1.5	0.227
Error	46		

attack levels (Table 4). Moreover, there was no significant effect of initial plant or final plant size on oviposition scar density (Table 4).

Discussion

Our observational study demonstrated that attack levels by *M. janthinus* generally increased over the 5-year study period. In particular, we found significant increases in the proportion of stems attacked across the majority of field sites (four of five). However, when examining stems that showed evidence of weevil attack, we found a significant increase in attack level (estimated by oviposition scar density) at only one site (i.e., Lefthand Canyon).

Results from the observational and experimental studies indicate that *M. janthinus* densities and attack levels may need to be above a certain threshold to have a significant negative effect on *L. dalmatica* performance. We found that high levels of weevil attack were associated with diminished reproductive output. In the observational study, we saw negative effects on individual ramets when oviposition scar densities were above 1.5 scars per cm stem. On the other hand, in the experimental study, negative effects at the whole plant level were observed when oviposition scar densities were above 0.8 scars per cm stem. At the majority of our study sites, with the exception of Lefthand Canyon, weevil attack estimates are below this hypothesized threshold level. Schat et al. (2011) manipulated adult *M. janthinus* densities and found similar threshold effects; however, that study indicated that densities above five larvae per stem were adequate to compromise plant growth.

In the observational study, we found a negative association between weevil attack and plant biomass

only during 2010. Moreover, we observed an initial positive association between oviposition scar density and plant performance estimates in 2010. This positive relationship may be explained by *M. janthinus* feeding and oviposition preferences for larger plants. Egan and Irwin (2008) found a similar positive association between *Linaria vulgaris* performance metrics and damage by an accidently introduced specialist beetle in a 2-year observational study, but a negative association in an experimental study and attributed the discrepancy to selective feeding on larger ramets. Herbivores commonly demonstrate oviposition and feeding preferences for larger plants (Price 1991). As Egan and Irwin (2008) note, such selective feeding makes it difficult to evaluate the efficacy of biocontrol agents in observational and correlational studies, and may lead to equivocal findings in regard to the effects of natural enemies on target weeds.

In the experimental study, we found strong negative effects of weevil attack on plant growth and reproduction. In the middle and high weevil attack categories, plants had more than a 50% reduction in biomass and seed production compared to plants in the low attack category. Resource availability can increase plant tolerance to herbivory (Wise and Abrahamson 2007). Although we did not directly examine tolerance, we found that unfertilized plants had about a quarter of the biomass and number of seeds as fertilized plants under low levels of weevil attack. However, it is unclear whether unfertilized plants were less tolerant to weevil attack or if fertilized plants demonstrated an especially strong positive response to increased N under low levels of weevil attack. Surprisingly, we did not find any significant effects of fertilization, neighbor clipping, or plant size on oviposition scar density. Although insects generally prefer to oviposit on larger plants growing in resource rich environments (Price 1991), the influence of plant traits on herbivore attack is complex. For example, in addition to showing preferences for higher plant nitrogen content, specialist insects may demonstrate preferences for higher levels of plant allelochemicals (Prudic et al. 2005), as these compounds can act as oviposition and feeding stimulants (Bernays and Chapman 1994; Bowers 1991). In a greenhouse study examining *L. dalmatica*, Jamieson and Bowers (in press) found that soil nitrogen enrichment decreased iridoid glycoside concentrations. However, the influence of these compounds on the behavior of *M. janthinus* has not been investigated.

Our experimental study revealed great phenotypic plasticity in *L. dalmatica* performance traits, which is typical of many invasive weeds (Daehler 2003). In particular, we saw a two-fold increase in plant biomass and seed production with nitrogen fertilization. Overall, this was the strongest effect observed in our study. The opportunistic response of *L. dalmatica* was especially striking, relative to the modest response of the resident plant community to N addition, which as a group increased by only about 20%. These findings (both for *L. dalmatica* and neighboring plants) parallel results reported in a study examining *L. dalmatica* seedling establishment (Blumenthal 2009). Although we did not find an effect of the clipping treatment on *L. dalmatica* performance, we suspect that this treatment did not entirely remove competitive effects of surrounding vegetation, as we did not eliminate belowground biomass.

Linaria dalmatica control remains a high priority for land managers in North America. Through a combination of observational and experimental research, we show that *L. dalmatica* plants are vulnerable to top-down control from the introduced stem-mining weevil, *M. janthinus*. Depending on the initial densities of the plant and success of agent release, it may take several years for insect populations to reach high enough densities to see reductions in target weeds (e.g., De Clerck-Floate and Harris 2002; Van Hezewijk et al. 2010). Our research suggests that the populations surveyed in this study have not reached those levels. Moreover, our research indicates that *L. dalmatica* is able to greatly exploit resource opportunities, possibly hindering effective top-down control, especially in areas with low weevil densities or high resource levels. Effective management of this species will likely require an integrated approach that combines top-down and bottom-up controls (e.g., as recommended in Blumenthal et al. 2010)—one that maintains adequately high densities of biological control agents over several years and increases plant competition or employs methods to minimize resource availability.

References

Bernays EA, Chapman RF (1994) Host-plant selection by phytophagous insects. Chapman and Hall, New York

Bigger DS, Marvier MA (1998) How different would a world without herbivory be? A search for generality in ecology. Integr Biol 1:60–66

Blossey B, Notzold R (1995) Evolution of increased competitive ability in invasive nonindigenous plants—a hypothesis. J Ecol 83:887–889

Blumenthal DM (2006) Interactions between resource availability and enemy release in plant invasion. Ecol Lett 9:887–895

Blumenthal DM (2009) Carbon addition interacts with water availability to reduce invasive forb establishment in a semi-arid grassland. Biol Invasions 11:1281–1290

Blumenthal DM, Norton AP, Seastedt TR (2010) Restoring competitors and natural enemies for long-term control of plant invaders. Rangelands 32:16–20

Borer ET, Halpern BS, Seabloom EW (2006) Asymmetry in community regulation: effects of predators and productivity. Ecology 87:2813–2820

Bowers MD (1991) Iridoid glycosides. In: Rosenthal GA, Berenbaum MR (eds) Herbivores: their interactions with secondary plant metabolites. Academic Press, New York, pp 297–326

Burke IC, Lauenroth WK, Parton WJ (1997) Regional and temporal variation in net primary production and nitrogen mineralization in grasslands. Ecology 78(5):1330–1340

Catford JA, Roland J, Christer N (2009) Reducing redundancy in invasion ecology by integrating hypotheses into a single theoretical framework. Divers Distrib 15(1):22–40

Daehler CC (2003) Performance comparisons of co-occurring native and alien plants: implications for conservation and restoration. Annu Rev Ecol Syst 34:183–211

Davis MA, Grime JP, Thompson K (2000) Fluctuating resources in plant communities: a general theory of invasibility. J Ecol 88:528–534

De Clerck-Floate R, Harris P (2002) *Linaria dalmatica* (L.) Miller, Dalmatian toadflax (Scrophulariaceae). In: Mason PG, Huber JT (eds) Biological control programmes in Canada 1981–2000. CABI Publishing, New York, pp 368–374

Duncan CA, Jachetta JJ, Brown ML, Carrithers VF, Clark JK, Ditomaso JM, Lym RG, McDaniel KC, Renz MJ, Rice PM (2004) Assessing the economic, environmental, and societal losses from invasive plants on rangeland and wildlands. Weed Technol 18:1411–1416

Egan FJ, Irwin RE (2008) Evaluation of the field impact of an adventitious herbivore on an invasive plant, yellow toadflax, in Colorado, USA. Plant Ecol 199(1):99–114

Gröger VD, Johne S (1965) Über das vorkommen von peganin in *Linaria*-Arten. Planta Med 13(2):182–188

Gruner DS, Smith JE, Seabloom EW, Sandin SA, Ngai JT, Hillebrand H, Harpole WS, Elser JJ, Cleland EE, Bracken MES, Borer ET, Bolker BM (2008) A cross-system synthesis of consumer and nutrient resource control on producer biomass. Ecol Lett 11:740–755

Gurevitch J, Fox GA, Wardle GM, Inderjit, Taub D (2011) Emergent insights from the synthesis of conceptual frameworks for biological invasions. Ecol Lett 14(4):407–418

Hairston NG (1960) Community structure, population control, and competition. Am Nat 94:421–425

Hillebrand H, Gruner DS, Borer ET, Bracken MES, Cleland EE, Elser JJ et al (2007) Consumer vs. resource control of producer diversity depends on ecosystem type and producer community structure. Proc Natl Acad Sci USA 104:10904–10909

Huenneke LF, Hamburg SP, Koide R, Mooney HA, Vitousek PM (1990) Effects of soil resources on plant invasions and community structure in Californian serpentine grasslands. Ecology 71:478–491

Inderjit, Cadotte MW, Colautti RI (2005) The ecology of biological invasions: past, present and future. In: Inderjit (ed) Invasive plants: ecological and agricultural aspects. Birkhäuser Verlag, Basel, pp 19–44

Jamieson MA, Bowers MD (2010) Iridoid glycoside variation in the invasive plant Dalmatian toadflax, *Linaria dalmatica* (Plantaginaceae), and sequestration by the biological control agent, *Calophasia lunula*. J Chem Ecol 36:70–79

Jamieson MA, Bowers MD (in press) Soil nitrogen availability and herbivore attack influence the chemical defenses of an invasive plant (*Linaria dalmatica*; Plantaginaceae). Chemoecology. doi:10.1007/s00049-011-0087-1

Jeanneret P, Schroeder C (1992) Biology and host specificity of *Mecinus janthinus* Germar (Col: Curculionidae), a candidate for the biological control of yellow and Dalmatian toadflax, *Linaria vulgaris* (L.) Mill. and *Linaria dalmatica* (L.) Mill. (Scrophulariaceae) in North America. Biocontrol Sci Technol 2:25–34

Keane RM, Crawley MJ (2002) Exotic plant invasions and the enemy release hypothesis. Trends Ecol Evol 117:164–169

Knochel DG, Seastedt TR (2010) Reconciling contradictory findings of herbivore impacts on the growth and reproduction of spotted knapweed (*Centaurea stoebe*). Ecol Appl 20:1903–1912

Knochel DG, Monson ND, Seastedt TR (2010) Additive effects of above- and belowground herbivores on the dominance of spotted knapweed (*Centaurea stoebe*). Oecologia 164:701–712

Lejeune KD, Suding KN, Sturgis S, Scott A, Seastedt TR (2005) Biological control insect use of fertilized and unfertilized diffuse knapweed in a Colorado grassland. Environ Entomol 34:225–234

Mack RN, Simberloff D, Lonsdale WM, Evans H, Clout M, Bazzaz FA (2000) Biotic invasions: causes, epidemiology, global consequences and control. Ecol Appl 10:689–710

McEvoy PB, Rudd NT, Cox CS, Huso M (1993) Disturbance, competition, and herbivory effects on ragwort *Senecio jacobaea* populations. Ecol Monogr 63:55–75

Myers JH, Bazely D (2003) Ecology and control of introduced plants. Cambridge University Press, Cambridge

Peterson RKD, Sing SE, Weaver DK (2005) Differential physiological responses of Dalmatian toadflax, *Linaria dalmatica* L. Miller, to injury from two insect biological control agents: implications for decision-making in biological control. Environ Entomol 34:899–905

Price PW (1991) The plant vigor hypothesis and herbivore attack. Oikos 62:244–251

Prudic KL, Oliver JC, Bowers MD (2005) Soil nutrient effects on oviposition preference, larval performance, and chemical defense of a specialist insect herbivore. Oecologia 143:578–587

Richardson DM, Pyšek P (2006) Plant invasions: merging the concepts of species invasiveness and community invasibility. Prog Phys Geogr 30(3):409–431

Saner MA, Jeanneret P, Mullerscharer H (1994) Interaction among 2 biological-control agents and the developmental stage of their target weed, Dalmatian toadflax, *Linaria dalmatica* (L) Mill (Scrophulariaceae). Biocontrol Sci Technol 4(2):215–222

Schat M, Sing SE, Peterson RKD, Menalled FD, Weaver DK (2011) Growth inhibition of Dalmatian toadflax, *Linaria dalmatica* (L.) Miller, in response to herbivory by the biological control agent *Mecinus janthinus* Germar. J Entomol Sci 46(3):232–246

Seastedt TR, Pyšek P (2011) Mechanisms of invasions in North American and European grasslands. Annu Rev Ecol Evol Syst 42:133–153

Seastedt TR, Suding KN (2007) Biotic constraints on the invasion of diffuse knapweed (*Centaurea diffusai*) in North American grasslands. Oecologia 151(4):626–636

Seastedt TR, Gregory N, Buckner D (2003) Reduction of diffuse knapweed by biocontrol insects in a Colorado grassland. Weed Sci 51:237–245

Suding KN, LeJeune KD, Seastedt TR (2004) Competitive impacts and responses of an invasive weed: dependencies on nitrogen and phosphorus availability. Oecologia 141:526–535

Van Hezewijk BH, Bourchier RS, De Clerck-Floate RA (2010) Regional-scale impact of the weed biocontrol agent *Mecinus janthinus* on Dalmatian toadflax (*Linaria dalmatica*). Biol Control 55(3):197–202

Vitousek PM, D'Antonio CM, Loope LL, Westbrooks R (1996) Biological invasions as global environmental change. Am Sci 84:468–478

Wilson LM, Sing SE, Piper GL, Hansen RW, De Clerck-Floate R, MacKinnon DK, Randall C (2005) Biology and biological control of Dalmatian and yellow toadflax. USDA Forest Service, Morgantown, FHTET-05-13

Wise MJ, Abrahamson WG (2007) Effects of resource availability on tolerance of herbivory: a review and assessment of three opposing models. Am Nat 169:443–454