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**INTERPLAY BETWEEN COMPETITION AND EVOLUTION IN INVADED AND  
NATIVE PLANT COMMUNITIES**

BY

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B. S., UNIVERSITY OF KANSAS, 2004

Dissertation

presented in partial fulfillment of the requirements  
for the degree of

Doctor of Philosophy

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May 2012

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## ABSTRACT

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Organismal Biology and Ecology

INTERPLAY BETWEEN COMPETITION AND EVOLUTION IN INVADED PLANT COMMUNITIES

Chairperson: Dr. Ragan M. Callaway

The distributions and abundances of organisms are affected by ecological processes, such as competition, predation, and abiotic stress, and these processes can also produce rapid evolutionary change in plant communities. Although our understanding of ecological and evolutionary interactions is growing, so far little is known about how competition among plants interacts with evolution to shape communities. In my dissertation, I use species invasions to investigate the evolutionary and ecological consequences of plant interactions and their effects on plant community assembly.

In my first chapter, I investigated complex ecological interactions between *Euphorbia esula*, an invasive plant, and *Balsamorhiza sagittata*, a native plant. I found that direct negative effects of *Euphorbia* on *Balsamorhiza*, due to competition, were greatly reduced by indirect positive effects. In the second chapter, I investigated whether selection favored competitive “suppression” or “tolerance” strategies in the native *Pseudoroegneria spicata* when competing with the invasive *Centaurea stoebe*. I found that tolerance had far greater fitness benefits than suppression. This observation has important consequences for understanding the outcome of evolution in plant communities. In my third chapter, I investigated the ecological consequences of intraspecific diversity in *Pseudoroegneria*. I found that functional diversity within the species increased ecosystem productivity, and that this pattern was strongest for ecotypes from mesic environments, suggesting that adaptive variation influences emergent consequences of intraspecific interactions. Finally, in my fourth chapter I found evidence that selection by *Euphorbia* on native and invasive grasses influenced how those grasses responded to other competitors, herbivory, and changes in resource availability. I also found evidence that this selection varied among sites in a manner akin to a geographic mosaic. Together, these chapters demonstrate how plant invasions can inform our understanding of interactions between ecological and evolutionary processes that affect plant community assembly.

## ACKNOWLEDGEMENTS

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To my coworkers, you not only helped me keep my projects in motion, but you also made them better. Although I thank everybody who worked in the Callaway Lab, I worked most closely with Carolyn Bauer, Annalisa Ingegno, Becky Fletcher, and Emmanuel Matos-Negrón, who worked hard in the field and in the lab and who truly made it fun to show up for work every day. In particular, however, I must thank David Hooper, who worked tirelessly to help me make my deadlines in the chaotic final weeks before this dissertation was due.

To all of my teachers and former advisors, I learned everything I know about science from you, and I am grateful. To my Ph. D. advisor, Ray Callaway, I thank you for providing me with the most fertile possible environment for my research. You were always available when I needed to discuss my ideas, and you provided me with encouragement and the freedom to follow my interests and choose my own path through my dissertation. I learned from you about how to be a scientist, but most importantly you taught me what it means to be a mentor.

Lastly, I dedicate this dissertation to my family and friends, who kept me going when times were difficult and my confidence, wavered. In particular I thank Nora Lahr, who, in addition to her friendship and support, helped with my research and applied her considerable writing skills to improving everything written in this thesis.

## PREFACE

Plant competition is one of the best studied ecological interactions, and its effects on the ecology of natural systems are well understood at almost all levels of organization (Hairston et al. 1960; Tilman 1982; Grace & Tilman 1990; Weiner 1990). However, with the exception of a handful of studies demonstrating that invasive plants cause selection on neighboring native plants (Callaway et al. 2005; Lau 2006; Meador and Hild 2006; Meador and Hild 2007; Cipollini and Hurley 2008; Leger 2008; Ferrero-Serrano et al. 2010; Rowe and Leger 2011; Goergen et al. 2011), little is known about the evolutionary consequences of plant competition (Dayan & Simberloff 2005; Strauss *et al.* 2006) or the ecological consequences of rapid evolution in plant communities. Because rapid evolution may have important ecological consequences (Thompson 1998; Carroll et al. 2007; Fussmann et al. 2007; Kinnison and Hairston 2007), this may be an important gap in our understanding of the ecological consequences of plant competition (Fig. 1). In this dissertation, I explore the ecological and evolutionary consequences of plant interactions from multiple perspectives, in the context of plant invasions. Exotic plant invasions provide a useful context for this research because of their intense effects on the ecology of plant communities (Braithwaite et al. 1989; Memmott et al. 2000; Grigulis et al. 2001; Ridenour and Callaway 2001; Maron and Marler 2008a; Hejda et al. 2009; Vilá et al. 2011) and native plant demography (Gordon 1998; Maron and Marler 2008b, Atwater et al. 2011), and because

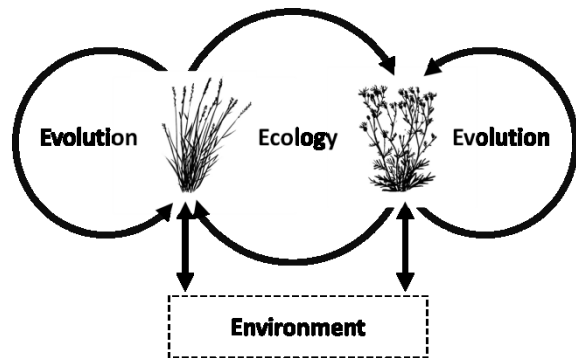


FIG. 1: Both ecological and evolutionary processes influence interactions between plants and the environment. In this dissertation I test multiple links between ecology, evolution, and environment in plant communities, and their influence on community and ecosystem processes.

plant invasions may lead to strong evolutionary forces (Strauss et al. 2006).

In Chapter 1 of this dissertation, I present novel evidence that direct negative effects of invasive *Euphorbia esula* (leafy spurge) on native *Balsamorhiza sagittata* (arrowleaf balsamroot) are reduced roughly 75% due to indirect interactions with native plants and herbivores (Atwater et al. 2011). While I do not explicitly discuss these results in the context of evolution, they suggest that indirect interactions with invasive plants are an important and complex component of the overall interaction between native and invasive plants. The evolutionary consequences of such interactions for plant communities may also be complex.

In Chapter 2, I use computational models to simulate invasion of *Pseudoroegneria spicata* (bluebunch wheatgrass, a native bunchgrass) populations by *Centaurea stoebe* (spotted knapweed, an invasive forb). Contrary to current theoretical predictions (Miller and Werner 1987; Aarssen 1989; Goldberg 1990), my results indicate that tolerance ability – i.e. the ability to endure competition from *Centaurea* – is a far more important component of fitness than ability to competitively suppress *Centaurea*. This occurs because the benefits of neighbor suppression are shared among all members of a community, while the benefits of tolerance are exclusive to each individual. Thus, I propose that competition is less like a boxing match, in which both suppression and tolerance determine success (Aarssen 1989; Goldberg 1990), and more like a demolition derby, in which tolerating competition is paramount. These results have important and wide ranging implications for the ecological and evolutionary consequences of plant competition. One intriguing possibility is that coevolution between plants could counteract competitive exclusion and stabilize plant communities.

In Chapter 3, I use ecotypes from throughout the native range of *Pseudoroegneria spicata* to demonstrate that *Pseudoroegneria* populations with greater ecotypic richness have greater

yield than populations with low ecotypic richness. I also find that this richness-dependent “overyielding” is strongest in ecotypes adapted to more mesic climates. Although species richness is known to influence productivity (Hooper et al. 2005; Balvanera et al. 2006; Cardinale et al. 2007), the effects of intraspecific diversity on species richness are poorly understood (but see Crutsinger et al. 2006, 2008, Cook-Patton et al. 2011). This is only the second study to show that the relationship between intraspecific diversity and productivity is driven by complementary interactions and not sampling effects (*sensu* Loreau and Hector 2001), and it is the first study to show that local adaptation predictably influences diversity-productivity relationships. This finding is important both because it suggests that species have a tremendous capacity to evolve cryptic but ecologically relevant functional diversity and because it suggests that local adaptation can predictably affect emergent properties of ecosystems.

In Chapter 4, I investigate selection caused by *Euphorbia esula* on *Pseudoroegneria spicata*, *Poa secunda* (two native bunchgrasses), and *Bromus japonicus* (an exotic annual grass). I find support for the hypothesis that invasive plants cause selection on native plants (Callaway et al. 2005; Lau 2006; Meador and Hild 2006), and I also show evidence that invaders cause selection on other invasive species. Furthermore, I provide novel evidence that selection by invasive plants influences how species compete with other native species, respond to abiotic stress, and tolerate simulated herbivory. My results do not indicate that these “extended” consequences of selection follow patterns predicted by ecological theory (Grime 1977, Herms and Mattson 1992), however. These findings have implications for understanding the evolutionary consequences of plant invasion and the nature of eco-evolutionary feedbacks in plant communities.

Taken together, my dissertation chapters provide important insights into the ecology and evolution of plant interactions, particularly in the context of plant invasion. They illustrate multiple links between ecological and evolutionary processes in plant communities, and suggest ways in which these links could influence the distribution and abundance of plant species, and the structuring of plant communities. Lastly, my dissertation highlights the importance of spatial heterogeneity and fine-scale local adaptation and illustrates ways that local adaptation can predictably influence community structure and ecosystem processes.

*Note to Readers:* I have formatted these chapters for submission to various journals. For this reason, formatting of text, references, and section titles vary somewhat throughout this thesis.

#### LITERATURE CITED

- Aarssen L. W. 1989. Competitive ability and species coexistence: A “plant’s eye” view. *Oikos* 56:386-401.
- Atwater, D. Z., C. M. Bauer and R. M. Callaway. 2011. Indirect positive effects ameliorate strong negative effects of *Euphorbia esula* on a native plant. *Plant Ecology* 212:1655-1662.
- Braithwaite, R. W., W. M. Lonsdale and J. A. Estbergs. 1989. Alien vegetation and native biota in tropical Australia: The spread and impact of *Mimosa pigra*. *Biological Conservation* 48:189-201.
- Callaway, R. M., W. M. Ridenour, T. Laboski, T. Weir and J. M. Vivanco JM. 2005. Natural selection for resistance to the allelopathic effects of invasive plants. *Journal of Ecology* 93:576-583.



- Carroll, S. P., A. P. Hendry, D. N. Reznick, C. W. Fox. 2007. Evolution on ecological time-scales. *Functional Ecology* 21:387-393.
- Cipollini, D., and K. A. Hurley. 2008. Variation in resistance of experienced and naïve seedlings of jewelweed (*Impatiens capensis*) to invasive garlic mustard (*Alliaria petiolata*). *Ohio Journal of Science* 108:47-49.
- Cook-Patton S. C., S. H. McArt, A. L. Parachnowitch, J. S. Thaler, A. A. Agrawal. 2011. A direct comparison of the consequences of plant genotypic and species diversity on communities and ecosystem function. *Ecology* 92:915-923.
- Crutsinger G. M., M. D. Collins, J. A. Fordyce, Z. Gompert, C. C. Nice, et al. 2006. Plant genotypic diversity predicts community structure and governs an ecosystem process. *Science* 313:966-968.
- Crutsinger G. M., L. Souza, N. J. Sanders. 2008. Intraspecific diversity and dominant genotypes resist plant invasions. *Ecology Letters* 11:16-23.
- Dayan T., D. Simberloff. 2005. Ecological and community-wide character displacement: The next generation. *Ecology Letters* 8:875-894.
- Ferrero-Serrano A, A. Hild and B. Meador. 2010. Can invasive species enhance competitive ability and restoration potential in native grass populations? *Restoration Ecology* 19:545-551.
- Fussmann G. F., M. Loreau, P. A. Abrams. 2007. Eco-evolutionary dynamics of communities and ecosystems. *Functional Ecology* 21:465-477.
- Goergen, E. M., E. A. Leger and E. K. Espeland. 2011. Native perennial grasses show evolutionary response to *Bromus tectorum* (cheatgrass) invasion. *PLoS ONE* 6:e18145.

- Goldberg D. E. 1990. Components of resource competition in plant communities. Pages 27-49 in J. B. Grace, and D. Tilman, ed. Perspectives on Plant Competition. Blackburn Press, Caldwell, NJ.
- Grace J. B., D. Tilman, Eds. 1990. Perspectives on Plant Competition. Blackburn Press, USA.
- Grigulis, K., A. W. Sheppard, J. E. Ash and R. H. Groves. 2001. The comparative demography of the pasture weed *Echium plantagineum* between its native and invaded ranges. Journal of Applied Ecology 38:281-290.
- Grime, J. P. 1977. Evidence for the existence of three primary strategies in plants and its relevance for ecological and evolutionary theory. American Naturalist 111:1169-1194.
- Hairton N. G., F. E. Smith, L. B. Slobodkin. 1960. Community structure, population control, and competition. American Naturalist 94:421-425.
- Heijda M., P. Pyšek and V. Jarošík. 2009. Impact of invasive plants on the species richness, diversity and composition of invaded communities. Journal of Ecology 97:393-403.
- Herms, D. A. and W. J. Mattson. 1992. The dilemma of plants: to grow or defend. The Quarterly Review of Biology 67:283-335.
- Kinnison M. T., N. G. Hairton Jr. 2007. Eco-evolutionary conservation biology: Contemporary evolution and the dynamics of persistence. Functional Ecology 21:444-454.
- Lau, J.A. 2006. Evolutionary responses of native plants to novel community members. Evolution 60:56-63.
- Leger, E. A. 2008. The adaptive value of remnant native plants in invaded communities: an example from the great basin. Ecological Applications 18:1226-1235.
- Loreau M., A. Hector. 2001. Partitioning selection and complementarity in biodiversity experiments. Nature 412:72-76.

- Maron J. L. and M. Marler. 2008a. Effects of native species diversity and resource additions on invader impact. *American Naturalist* 172:S18-S33.
- Maron J. L. and M. Marler. 2008b. Field-based competitive impacts between invaders and natives at varying resource supply. *Journal of Ecology* 96:1187-1197.
- Mealor BA and Hild AL. 2006. Potential selection in native grass populations by exotic invasion. *Molecular Ecology* 15:2291-2300.
- Mealor, B. A. and A. L. Hild. 2007. Post-invasion evolution of native plant populations: a test of biological resilience. *Oikos* 116:1493-1500.
- Memmott, J., S. V. Simon, Q. Paynter, A. W. Sheppard and P Syrett. 2000. The invertebrate fauna on broom, *Cystus scoparius*, in two native and two exotic habitats. *Acta Oecologia* 21:213-222.
- Miller T. E., and Werner P. A. 1987. Competitive effects and responses between plant species in a first-year old-field community. *Ecology* 68:1201-1210.
- Ridenour, W. M. and R. M. Callaway. 2001. The relative importance of allelopathy in interference: the effects of an invasive weed on a native bunchgrass. *Oecologia* 126:444-450.
- Rowe, C. L. J. and E. A. Leger. 2011. Competitive seedlings and inherited traits: a test of rapid evolution of *Elymus multisetus* (big squirreltail) in response to cheatgrass invasion. *Evolutionary Applications* 4:485-498.
- Strauss S. Y., J. A. Lau, S. P. Carroll. 2006. Evolutionary responses of native to introduced species: What do introductions tell us about natural communities? *Ecology Letters* 9:357-374.
- Thompson J. N. 1998. Rapid evolution as an ecological process. *TREE*. 13:329-332.

Thompson J. N. 2005. *The Geographic Mosaic of Coevolution*. University of Chicago, USA.

Tilman D. 1982. *Resource Competition and Community Structure*. Princeton UP, USA.

Vilá, M., J. L. Espinar, M. Hejda, P. E. Hulme, V. Jaršík, J. L. Maron, J. Pergl, U. Schaffner, Y.

Sun and P. Pyšek. 2011. Ecological impacts of invasive alien plants: a meta-analysis of their effects on species, communities and ecosystems. *Ecology Letters* 14:702-708.

Weiner J. 1990. Asymmetric competition in plant populations. *TREE* 5:360-364.

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### CHAPTER 1

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## CHAPTER 1

### INDIRECT POSITIVE EFFECTS AMELIORATE STRONG NEGATIVE EFFECTS OF *EUPHORBIA ESULA* ON A NATIVE PLANT.

*Abstract:* Invasive plant species can have strong direct negative effects on native plants. Depending on the nature of interactions among competitors and consumers within a community, strong indirect interactions may either augment or offset direct effects. We used path analysis to estimate the relative importance of direct and indirect effects of *Euphorbia esula*, an unpalatable invasive plant, on *Balsamorhiza sagittata*, a native forb, through ‘shared defense’ and by suppression of native competitors. Our results indicate that *E. esula* had strong direct negative effects on *B. sagittata*, but also that its net effect was reduced by 75% due to indirect positive effects. This reduction was due in equal parts to lessened competition from other native plants eliminated from *E. esula* stands and to lower levels of herbivory inside *E. esula* stands, apparently caused by indirect defense of *B. sagittata* by *E. esula*. To our knowledge, this is the first evidence that invaders may indirectly reduce herbivory on native plants, a phenomenon that may commonly occur with unpalatable invaders. Furthermore, our results highlight the potential complexity of interactions between native and invasive plants.

*Key words:* associational resistance, indirect interactions, leafy spurge, plant invasion, path analysis, shared defense

## INTRODUCTION

Invaders often dominate communities in their introduced ranges, causing losses in native species abundance and diversity (Braithwaite et al. 1989; Memmott et al. 2000; Grigulis et al. 2001) and reducing growth and reproduction of surviving native species. Direct, negative interactions between native and invasive plants are recognized as important drivers of these community changes (Levine et al. 2002; Hierro and Callaway 2003; Maron and Marler 2008). However, indirect interactions between native and invasive plants may also play an important and potentially complex role in the ecology of an invaded community, particularly because indirect interactions with invasive plants may have strong inhibitory and facilitative effects.

Indirect interactions can have substantial effects on the organization of uninvaded plant communities (Miller 1987; Levine 1999; Callaway 2007), but have only recently attracted the attention of researchers studying plant invasions. Indirect interactions between native and invasive plants can be mediated by native plants (Saccone et al. 2010), soil microbes (Callaway et al. 2003; Klironomos 2002; Inderjit et al. 2007) or by pollinators (Munoz and Cavieres 2008), but perhaps most importantly by herbivores (Parker et al. 2006; White et al. 2006; Meiners 2007; Orrock et al. 2008; Pearson and Callaway 2008). Because herbivores can have such powerful indirect effects on community composition in uninvaded systems, understanding their indirect effects in invaded systems is also likely to be important. To our knowledge previous studies on the indirect effects of native herbivores in plant invasions have focused on their role as causes of invader success, but two other potential *positive* indirect interactions between native and invasive plants are “shared defense” and “associational resistance.” Defenses are considered to be “shared” when a palatable beneficiary is protected by a nearby unpalatable species. The positive effect of growing with many other species that limit herbivory by other mechanisms, such as



visual or olfactory masking, is called “associational resistance” (Tahvanainen and Root 1972, Atsatt and O’Dowd 1976, Feeny 1976, Callaway 2007).

Shared defense and associational resistance may be important processes in some plant invasions. Although some exotic plant species evolve lower levels of defense when released from herbivore pressure (Siemann and Rogers 2003), other invasive plant species decrease investment in defense against specialist herbivores but increase investment in defense against generalists (Müller-Schärer et al. 2004; Joshi and Vrieling 2005), or increase overall investment in defense (Ridenour et al. 2008). Also, some plants may possess defensive compounds that are novel to generalist herbivores in the introduced range and thus experience lower rates of herbivory (Callaway and Ridenour 2004; Branson and Sword 2008; Jogesh et al. 2008; Tallamy et al., in press; but see Lind and Parker 2010). Regardless of the underlying mechanisms, herbivores avoid some invasive species, resulting in dramatically lower rates of herbivory on exotic plants than native plants in some systems (Cappuccino and Carpenter 2005; Cappuccino and Arnason 2006). If invasive plant species are avoided by native herbivores in the invaded range, palatable native plant species may benefit indirectly by growing closely to these invaders. This might also happen any time an invasive species forms a dense thicket that obscures plants from visual herbivores. In such a case invaders might release native plants from predation by visual but not olfactory herbivores (as in Hambäck *et al.* 2003).

*Euphorbia esula* (leafy spurge) is one of North America’s most problematic invasive plants due to its competitive ability and vigorous vegetative habit, but also to its toxicity and unpalatability to grazers (Dittberner et al. 1983; Cyr and Bewley 1989; Trammell and Butler 1995). *Euphorbia esula* reduces native diversity in many community types (Belcher and Wilson 1989); nonetheless, some native species persist during the early stages of *E. esula* invasion. We

observed that the North American native *Balsamorhiza sagittata* (arrowleaf balsamroot) maintains substantial populations in low to moderate density *E. esula* stands. *Balsamorhiza sagittata* is a large, dominant, native forb whose shoots, flowers and seeds are commonly eaten by native generalist herbivores such as pronghorn, bighorn sheep, mule deer and deer mice (Wikeem and Pitt 1979, 1991; Burell 1982; USDA 1988). Seeds of *B. sagittata* are also consumed by specialist insects (Amsberry and Maron 2006) that live in the seed head (capitulum) and feed as larvae.

We hypothesized that *B. sagittata* growing within *E. esula* patches would experience strong direct negative effects from *E. esula*, but that *E. esula* would also have indirect effects in the form of shared defense due to its unpalatability, as well as through reduction in the density of native competitors. We used a path analysis to estimate the relative strength of these effects using observational data.

## MATERIALS AND METHODS

We chose five sites in montane grasslands around the Missoula Valley in Western Montana, and at each of the five sites we established a 10 m x 10 m plot in both an *E. esula* invaded area and in a nearby uninvaded area. The uninvaded plot was always within 20 m of the *E. esula* plot and did not differ noticeably in elevation, slope, or aspect. We chose sites where the general plant community resembled the remnants in the *E. esula* patch. All of these sites were historically used for cattle grazing, but had been set aside for conservation and were only experiencing natural grazing by the time *E. esula* invasion occurred. Intense *E. esula* invasion occurred relatively recently in the Missoula Valley and we estimate that dense patches became common only within the past 15 years. *Euphorbia esula* is clonal and expands vegetatively from

the center of the colony. We located plots within several meters of the edge of active expansion fronts, thus our plots were probably situated where *E. esula* had been present for only a few years. We also chose plots that contained relatively uniform, but moderate densities of *E. esula*.

Our sites were located at the Cox Property (46°50'12''N;113°57'59''W, elev. 1265 m, slope 13°, aspect S), the Rocky Mountain Elk Foundation reserve (46°56'17''N;114°01'01''W, 1130 m, 21°, W), Waterworks Hill (46°52'53''N;113°59'08''W, 1090 m, 18°, ENE), the North Hills (46°53'34''N;113°58'45''W, 1085 m, 16°, E), and Mt. Jumbo (46°51'55'';113°58'03'', 1030 m, 39°, SSW). All sites had grassland plant communities dominated by *B. sagittata*, *Pseudoroegneria spicata* (bluebunch wheatgrass), *Bromus tectorum* (cheatgrass) and *Bromus japonicus* (Japanese brome).

From 27 June, 2008 through 7 July, 2008, we measured 1041 individual *B. sagittata* plants at the five sites. We counted each *B. sagittata* individual within our plots and measured canopy height, the longest diameter of the leafy canopy (plant width), number of leaves, number of flower heads, number of damaged leaves, and gave each plant a subjective leaf damage rating based on a scale from 0-10. We also measured the distance to the nearest *B. sagittata* neighbor, distance to nearest *Lupinus sericeus* (if within 2 m), and distance to nearest *Pseudoroegneria spicata* (if within 2 m). *Lupinus sericeus* and *P. spicata* are prominent native plants that, together with *B. sagittata*, covered a majority of the landscape at the study sites (D. Atwater, *pers. obs.*). We also recorded the presence of any other plant species within a 50 cm radius of the target plant. Local *E. esula* density was measured as the number of living stems within a 1 m x 1 m square centered on each *B. sagittata* plant.

We collected one capitulum, or seed head, from each flowering *B. sagittata* plant (total n= 357) and dissected it to look for signs of herbivory. Capitula were scored positively for

herbivory if they contained an intact seed herbivore, boring holes, or frass (insect excreta) produced by insect seed predators. We performed a chi-squared test of homogeneity to determine whether herbivory was contingent on the presence of *E. esula*.

To generate integrative variables for *B. sagittata* performance we performed a principal components analysis (PCA) on plant height, plant width, and number of leaves to create a single score for plant size ( $S_{PCA}$ ). These variables were log-transformed to improve normality and because herbivory is hypothesized to affect plant performance on a logarithmic scale (Rees and Brown 1992, Hambäck & Beckerman 2003). Likewise, we performed a PCA to generate a single, integrative variable for plant damage ( $D_{PCA}$ ) because neither the proportion of leaves damaged per plant nor the subjective damage rating alone was a satisfactory indicator of overall leaf damage. Factors were generated using a correlation matrix. We used a variance explained criterion (retain enough factors to explain 70% of variance) to determine whether the data reduction was appropriate and how many factors to retain. For both  $S_{PCA}$  and  $D_{PCA}$  this resulted in a single factor being retained for further analysis. This analysis and all other analyses reported here were performed with SPSS (SPSS, Inc., 2008-2010), unless otherwise indicated.

To determine the importance of direct and indirect effects, we conducted a path analysis (model illustrated in Fig. 1). For this analysis, we used only data from flowering plants because young, non-flowering plants were almost entirely absent inside of *E. esula* patches and would have caused inappropriate reduction in our estimate of plant size outside of *E. esula* patches (Fig. 2). In the analysis, we modeled the effects of *E. esula* density on the performance of *B. sagittata* ( $S_{PCA}$ ), allowing it to have both a direct effect and indirect effects via changes in leaf herbivory ( $D_{PCA}$ ) and proximity of the target *B. sagittata* to other native plants ( $-1 * \text{nearest-neighbor distance}$ ). In our initial model, we allowed proximity of neighbors to have an effect on herbivory

but removed these effects from the final model because they did not contribute significantly to the model. We used logical inference rather than model comparison to construct the final model, and we used random site effects for each of the endogenous variables.

In our path analysis, we conducted a series of multiple linear regressions of effect variables against response variables. The arrows in the diagram represent effect paths from one variable to another (effect variable  $\rightarrow$  response variable). The path coefficients on the diagram are regression coefficients ( $\beta$ ). A large value for the path coefficient indicates that changes in the effect variable cause large changes in the response variable. To estimate the indirect effect of A on C via B ( $A \rightarrow B \rightarrow C$ ), we take the product of the path coefficients of A on B and of B on C ( $A \rightarrow B * B \rightarrow C$ ). The total effect of A on C is the sum of all direct and indirect effects of A on C. For example, the indirect effect of *E. esula* density on *B. sagittata* size via herbivory is the product of the path coefficient from *E. esula* to herbivory and from herbivory to size. To calculate the total effect of *E. esula* on size, we added the coefficient of the direct path from *E. esula* to size to the products of the coefficients along each of the indirect paths.

## RESULTS

One factor ( $S_{PCA}$ ) was extracted from the PCA for plant size and 85.5% of variance in the initial variables was explained by that single factor. Component loadings were: leaves = 0.903, height = 0.885, and width = 0.935; plants with high  $S_{PCA}$  scores were wider, taller, and had more leaves than plants with low  $S_{PCA}$  scores. We also extracted one factor from the PCA for leaf damage, and 72.7% of variance in the initial variables was explained by that factor. Component loadings were: subjective damage = 0.853 and proportion of leaves damaged = 0.853; plants with

higher  $D_{PCA}$  scores had higher subjective damage ratings and more leaves damaged than plants with lower scores.

Relatively few small, immature plants were present inside of *E. esula* patches, therefore we only considered data from flowering adult plants in further analysis (Fig. 2). Among mature plants, those inside *E. esula* patches were smaller but also experienced about a 25% reduction in herbivory compared to mature plants outside of *E. esula* patches (Table 1).

The path analysis (Fig. 1) suggested that *B. sagittata* adult size was negatively affected by the degree of leaf herbivory ( $\beta = -0.175$ ,  $p = 0.001$ ), proximity to neighboring *P. spicata* ( $\beta = -0.174$ ,  $p = 0.002$ ) and conspecific *B. sagittata* plants ( $\beta = -0.104$ ,  $p = 0.004$ ), and importantly, *E. esula* density ( $\beta = -0.168$ ,  $p = 0.002$ ). However, because *E. esula* density also had strong direct negative effects on leaf herbivory ( $\beta = -0.291$ ,  $p < 0.001$ ), and on proximity to *P. spicata* ( $\beta = -0.266$ ,  $p < 0.001$ ) and conspecific *B. sagittata* plants ( $\beta = -0.145$ ,  $p = 0.004$ ), the *indirect* effects of *E. esula* density on *B. sagittata* size were positive. As a result, the overall negative effect of *E. esula* on *B. sagittata* size was reduced by  $\approx 75\%$  (from  $-0.145$  to  $-0.036$ ). Our model indicated that indirect effects caused by changes in herbivory and in proximity to other competitors contributed equally to the 75% amelioration of direct effects caused by *E. esula* (Table 2). Plants in *E. esula* patches did not have reduced seed herbivory ( $X^2 = 0.106$ , d.f. = 1,  $n = 1041$ ,  $p = 0.744$ ).

## DISCUSSION

In this study we estimated the relative strengths of direct and indirect effects of invasive *Euphorbia esula* on Montana native *Balsamorhiza sagittata* plants. We hypothesized that negative effects of association with *E. esula* would be reduced via protection from herbivores

and through reduction in the density of native competitors. To estimate the importance of these effects, we used observational data to perform a path analysis to quantify the interactions between these plant species. Our model required the following assumptions: 1) *E. esula* density affected native plant abundance, 2) *E. esula* density influenced herbivory on natives and not the opposite, and 3) herbivory negatively influenced the size of natives and not the opposite.

Our model indicated that strong indirect benefits of association with *E. esula* reduced the cost of association with *E. esula* by about 75%. Roughly half of the indirect positive effect was due to relief from competition with native plants inside of *E. esula* patches, and half was due to reduced herbivory inside of *E. esula* patches, probably because native ungulates avoided grazing in *E. esula* patches. Although the positive effects we observed were strong, we do not believe that they will ultimately allow *B. sagittata* to persist within *E. esula* patches, as the overall effect of *E. esula* on *B. sagittata* was still negative despite the strength of the indirect positive effects.

Instead, our results suggest that, for the adult plants that survived initial *E. esula* invasion, the indirect effects of *E. esula* provided substantial benefits and may allow them to persist for longer than might be expected if only direct costs of association with *E. esula* were measured. This may be a common feature of many species invasions; such indirect benefits could occur any time an invader was less palatable than neighbors, any time an invader reduced the abundance of native competitors, or any time an invader obscured native plants from herbivores.

According to our model, half of the indirect benefits of association with *E. esula* resulted from reduced competition from native plants. Because invasive plants often reduce the abundance of native competitors, remaining native plants may experience relief even while experiencing intense competition from the invader. These indirect interactions are important to acknowledge because native plants species may respond differently to invasive species, and may

have different competitive effects on other natives. For example, two of the three species we identified as native competitors in this experiment appeared to have similar negative effects on the performance of target *B. sagittata* plants. In contrast, the nitrogen fixing *Lupinus sericeus* trended towards a weak positive effect. Therefore, the overall cost of association with *E. esula* could be expected to differ depending on the initial abundance of *L. sericeus*, and depending on the degree to which these three species were affected by *E. esula*.

Our model also indicated that *E. esula* indirectly facilitated *B. sagittata* due to reduced rates of leaf – but not seed – herbivory on plants inside of *E. esula* patches. Because it is hypothesized that invasive plants often evolve increased defense against generalist herbivores (Müller-Schärer et al. 2004; Joshi and Vrieling 2005), and that native generalists sometimes cannot tolerate the chemical defenses of novel species, associational resistance could be an important but overlooked aspect of the interactions between many native and invasive plants.

To properly test for causal relationships among the variables we measured, experimental manipulation of native plant competition, herbivory, and *E. esula* density over multiple seasons is necessary. However, in this study path analysis allowed us to explore interactions between *B. sagittata* size, herbivory, native plant abundance, and *E. esula* density using purely observational data. Such analyses may prove powerful in systems where logistical constraints or conservation concerns prohibit experimental manipulation. Our results indicated that indirect interactions among native and invasive plants can be strong, and can be an important component of the net effects of invasive plants. A complete understanding of the ecology of species invasions, and prediction of how native plants will respond to invasion, may therefore require consideration of the strong indirect effects that invaders can have on native species.



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## REFERENCES

- Amsberry LK, Maron JL (2006) Effects of herbivore identity on plant fecundity. *Plant Ecol* 187:39-48
- Atsatt PR, O'Dowd DJ (1976) Plant defense guilds. *Science* 193:24-29
- Belcher JW, Wilson SD (1989) Leafy spurge and the species composition of a mixed-grass prairie. *J Range Manag* 42:172-175
- Braithwaite RW, Lonsdale WM, Estbergs JA (1989) Alien vegetation and native biota in tropical Australia: The spread and impact of *Mimosa pigra*. *Biol Conserv* 48:189-201
- Branson DH, Sword GA (2008) Grasshopper herbivory affects native plant diversity and abundance in a grassland dominated by the exotic grass *Agropyron cristatum*. *Rest Ecol* 17:89-96
- Burell GC (1982) Winter diets of mule deer in relation to bitterbrush abundance. *J Range Manag* 35:505-510
- Callaway RM, Mahall BE, Wicks C, Pankey J, Zabinski C (2003) Soil fungi and the effects of an invasive forb on grasses: Neighbor identity matters. *Ecology* 84:129-135

- Callaway RM, Ridenour WM (2004) Novel weapons: Invasive success and the evolution of increased competitive ability. *Front Ecol Env* 2:436-443
- Callaway RM (2007) *Positive Interactions and Interdependence in Plant Communities*. Springer, NY
- Cappuccino N, Carpenter D (2005) Invasive exotic plants suffer less herbivory than non-invasive exotic plants. *Biol Lett* 1:435-438
- Cappuccino N, Arnason JT (2006) Novel chemistry of invasive exotic plants. *Biol Letts* 2:189-193
- Cyr DR, Bewley JD (1989) Carbon and nitrogen reserves of leafy spurge (*Euphorbia esula*) roots as related to overwintering strategy. *Physiol Plant* 77:67-72
- Dittberner PL, Olson MR (1983) *The plant information network (PIN) data base: Colorado, Montana, North Dakota, Utah, and Wyoming*. FWS/OBS-83/86. Washington, DC: U.S. Department of the Interior, Fish and Wildlife Service
- Feeny P (1976) Plant apparency and chemical defense. *Rec Adv Phytochem* 10:1-40
- Grigulis KA, Sheppard W, Ash JE, Groves RH (2001) The comparative demography of the pasture weed *Echium plantagineum* between its native and invaded ranges. *J App Ecol* 38:281-290
- Hambäck PA, Pettersson J, Ericson L (2003) Are associational refuges species-specific? *Funct Ecol* 17:87-93
- Hierro JL, Callaway RM (2003) Allelopathy and exotic plant invasion. *Plant and Soil* 256:29-39
- Inderjit, Callaway RM, Vivanco JM (2007) Can plant biochemistry contribute to understanding of invasion ecology? *Trends in Plant Sci* 11:574-580

- Jogesh T, Carpenter D, Cappuccino N (2008) Herbivory on invasive exotic plants and their non-invasive relatives. *Biol Invasions* 10:797-804
- Joshi J, Vrieling K (2005) The enemy release and EICA hypothesis revisited: Incorporating the fundamental difference between specialist and generalist herbivores. *Ecol Lett* 8:704-714
- Hambäck PA, Beckerman AP (2003) Herbivory and plant resource competition: A review of two interacting interactions. *Oikos* 101:26-37
- Klironomos J N (2002) Feedback with soil biota contributes to plant rarity and invasiveness in communities. *Nature* 410:651-652
- Levine JM (1999) Indirect facilitation: Evidence and predictions from a riparian community. *Ecology* 80:1762-1769
- Levine J M, Vilà M, D'Antonio CM, Dukes JS, Grigulis K, Lavorel S (2002) Mechanisms underlying the impacts of exotic plant invasions. *Proc Royal Soc Biol Sci* 270:775-881
- Lind EM, Parker JD (2010) Novel weapons testing: are invasive plants more chemically defended than native plants? *PLoS ONE* doi:10.1371/journal.pone.0010429
- Maron J L, Marler M (2008) Field-based competitive impacts between invaders and natives at varying resource supply. *J Ecol* 96:1187-1197
- Meiners SJ (2007) Apparent competition: an impact of exotic shrub invasion on tree regeneration. *Biol Invasions* 9:849-855
- Memmott J, Simon SV, Paynter Q, Sheppard AW, Syrett P (2000) The invertebrate fauna on broom, *Cystus scoparius*, in two native and two exotic habitats. *Acta Oecologia* 21:213–222
- Miller TE (1987) Effects of emergence time on survival and growth in an early old-field community. *Oecologia* 72:272-278

- Müller-Schärer H, Schaffner U, Steinger T (2004) Evolution in invasive plants: Implications for biological control. *TREE* 19:418-422
- Munoz A A, Cavieres LA (2008) The presence of a showy invasive plant disrupts pollinator service and reproductive output in native alpine species only at high densities. *J Ecol* 96:459-467
- Orrock JL, Witter MS, Reichman OJ (2008) Apparent competition with an exotic plant reduces native plant establishment. *Ecology* 89:1168-1174
- Parker JD, Burkepille DE, Hay ME (2006) Opposing effects of native and exotic herbivores on plant invasions. *Science* 311:1459-1461
- Pearson DE, Callaway RM (2008) Herbicide reveals weed biocontrol insects reduce native plant recruitment through second-order apparent competition. *Ecol App* 18:1489-1500
- Rees M, Brown VK (1992) Interactions between invertebrate herbivores and plant competition. *J Ecol* 80:353-360
- Ridenour WM, Vivanco JM, Feng Y, Horiuchi J, Callaway RM (2008) No evidence for tradeoffs: *Centaurea* plants from America are better competitors and defenders than plants from the native range. *Ecol Monogr* 78:369-386
- Saccone P, Pagés J, Girel J, Brun J, Michalet R (2010) *Acer negundo* invasion along a successional gradient: early direct facilitation by native pioneers and late indirect facilitation by conspecifics. *New Phytologist* 187:831-842
- Siemann E, Rogers WE (2003) Reduced resistance of invasive varieties of the alien tree *Sapium sebiferum* to a generalist herbivore. *Oecologia* 135:451-457

- Tahvanainen JO, Root RB (1972) The influence of vegetational diversity of the population ecology of a specialized herbivore, *Phyllotreta cruciferaea* (Coleoptera: Chrysomelidae). *Oecologia* 10:321-346
- Tallamy DW, Ballard M, D'Amico V. In press. Can alien plants support generalist insect herbivores? *Biol Invasions* 00:000-000
- Trammell MA, Butler JL (1995) Effects of exotic plants on native ungulate use of habitat. *J Wildlife Manag* 59:808-816
- U.S. Department of Agriculture, Forest Service (1988) Range plant handbook. 2nd ed. Dover Publications Inc. Mineola, NY
- White EM, Wilson JC, Clarke AR (2006) Biotic indirect effects: a neglected concept in invasion biology. *Diversity and Distributions* 12:443-455
- Wikeem BM, Pitt MD (1979) Interpreting diet preference of California bighorn sheep on native rangeland in south-central BC. *Rangelands* 1:200-202
- Wikeem BM, Pitt MD (1991) Grazing effects and range trend assessment on California bighorn sheep range. *J Range Manag* 44:466-470

TABLE 1: Effect of *E. esula* on *B. sagittata* size and herbivory. Data (mean +/- s.d.) are only shown for flowering *B. sagittata* plants. P-values for differences in trait means, with respect to *E. esula* presence, were calculated using T-tests (n = 357). These P-values are provided for general information only, as the path analysis presented elsewhere in this manuscript provides a more rigorous test of the effects of *E. esula* on *B. sagittata*.

	Outside <i>E. esula</i>	Inside <i>E. esula</i>	P-value
Num. leaves per plant	21.96 (+/- 12.33)	22.09 (+/- 11.75)	0.690
Num. flowers per plant	9.92 (+/- 11.41)	7.76 (+/- 7.09)	0.029
Canopy height (cm)	41.7 (+/- 11.56)	39.06 (+/- 10.43)	0.034
Canopy width (cm)	70.75 (+/- 16.1)	70.11 (+/- 14.35)	0.862
Plant Size (S <sub>PCA</sub> )	0.96 (+/- 0.42)	0.93 (+/- 0.39)	0.408
Num. damaged leaves	8.01 (+/- 4.25)	6.04 (+/- 3.6)	<0.001
Prop. leaves damaged (%)	41.31 (+/- 19.00)	30.32 (+/- 16.54)	<0.001
Subj. damage rating	2.42 (+/- 1.07)	1.63 (+/- 0.72)	<0.001
Leaf Damage (D <sub>PCA</sub> )	-0.06 (+/- 0.71)	-0.64 (+/- 0.55)	<0.001
Dist. to nearest <i>P. spicata</i> (cm)	64.2 (+/- 59.21)	98.68 (+/- 72.89)	<0.001
Dist. to nearest <i>L. sericeus</i> (cm)	103.18 (+/- 80.56)	139.91 (+/- 75.68)	<0.001
Dist. to nearest conspecific (cm)	53.35 (+/- 27.49)	58.8 (+/- 31.69)	0.087

TABLE 2: Direct and indirect effects of *E. esula* on *B. sagittata* size (S<sub>PCA</sub>). Standardized coefficients are shown with un-standardized coefficients in parentheses.

Direct effect	-0.145 (-0.004)
+ Total indirect effect	0.109 (0.003)
(Indirect effect via plant community)	0.057 (0.002)
(Indirect effect via herbivory)	0.051 (0.001)
= Total effect	-0.036 (-0.001)

FIG. 1. Path analysis diagram. Arrows indicate effects of effect variables on response variables from effect → response variable. The size of the arrow indicates the strength of the effect. Standardized path coefficients are shown on each arrow, with unstandardized coefficients in parentheses. All paths are significant ( $p < 0.05$ ) except ‘Proximity to *L. sericeus*’ →  $D_{PCA}$  (dotted line). Direct random effects of site were also modeled for each endogenous variable (paths not shown).

FIG 2. Frequency distribution of *Balsamorhiza* size PCA score. The upper panel shows the frequency distribution for immature plants, and the lower panel shows the distribution for flowering plants. Color indicates whether plant was located outside (solid grey) or inside (stippled) of a *Euphorbia* patch. The distributions are similar, except that very small, young plants (one or two leaves) are conspicuously absent from *Euphorbia* patches.

FIG. 1

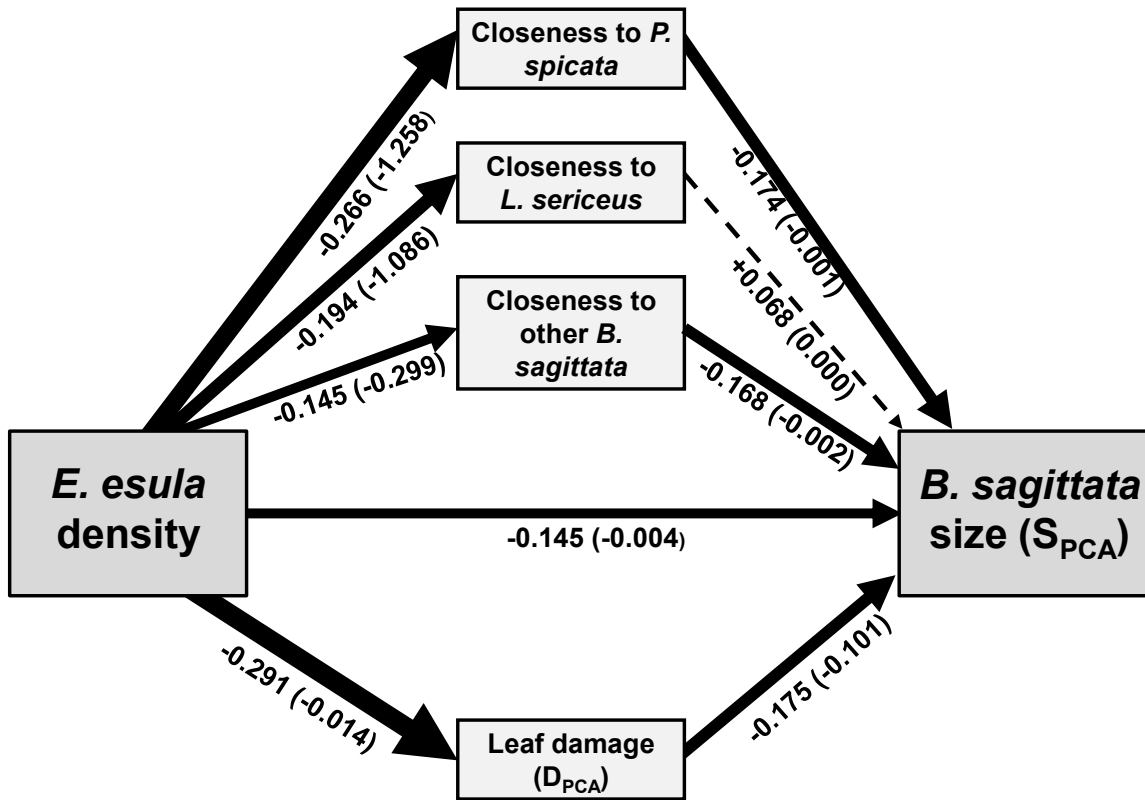
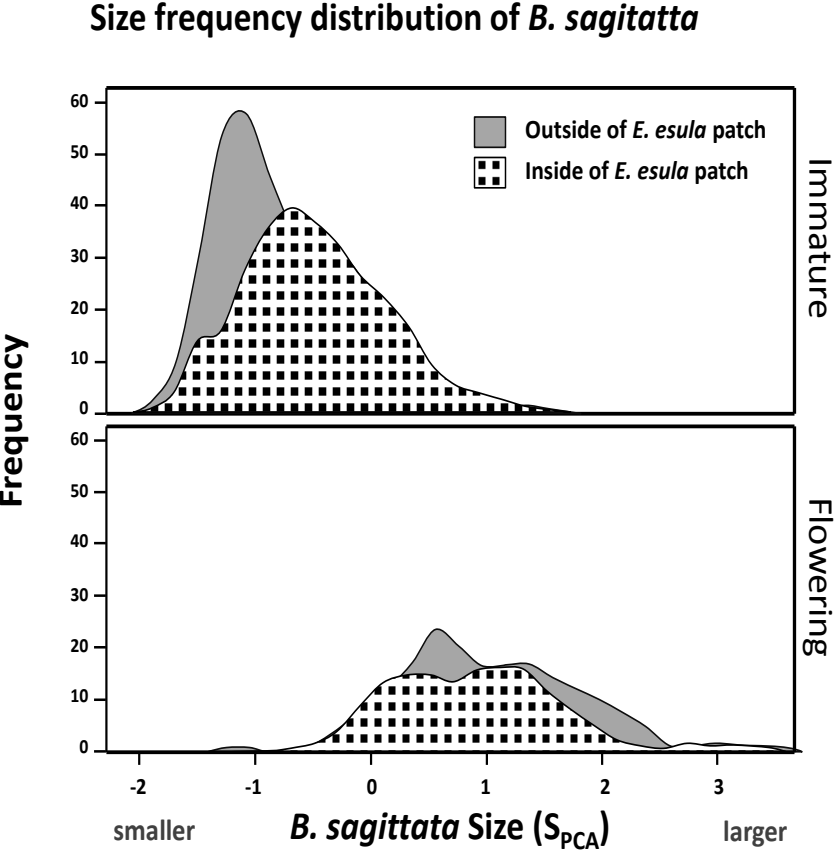




FIG. 2



## CHAPTER 2

### IS COMPETITION AMONG PLANTS MORE LIKE A BOXING MATCH OR A DEMOLITION DERBY?

*Abstract:* We used simulation models informed by experimental competition trials to ask whether tolerance of the competitive effects of an invader or the ability to competitively suppress an invader is more important for the survival of native plant ecotypes following exotic invasion. We calculated tolerance and suppression abilities of 23 ecotypes of the native grass *Pseudoroegneria spicata* competing against the invasive forb *Centaurea stoebe*, and modeled *C. stoebe* invasion into *P. spicata* populations varying in ecotypic diversity. The ability to tolerate competition from *C. stoebe* was far more important for the success of *P. spicata* than the ability to suppress *C. stoebe*. This is because the benefits of suppressing the invader were shared with other *P. spicata* ecotypes, whereas the benefits of tolerance were exclusive to individual ecotypes. Thus competition in communities may be more analogous to a demolition derby than a boxing match, in which plants benefit much more by enduring damage than by causing it, because the benefits of damaging an opponent are shared. This finding has implications for our interpretation of competitive hierarchies formed using pair-wise competition trials and for our understanding of selective forces caused by competition, the structuring of plant communities, the coexistence of plant species, and the outcome of invasions.

*Key Words:* boxing match, competitive effect, competitive response, demolition derby, suppression, tolerance

## INTRODUCTION

Ecologists generally evaluate the competitive ability of plant species in terms of the overall outcome of competition, measured as the size, survival, or fitness of a target individual growing with neighbors versus without neighbors. But there are two distinct components of competitive ability that influence this overall outcome – the suppression of neighbors and the tolerance to the competitive effects of neighbors (Fig. 1A). A species' or genotype's competitive ability in a given situation has been thought to be the product of how good it is at a particular combination of suppression and tolerance (Miller & Werner 1987; Aarssen 1989; Goldberg 1990). From this perspective, competition is analogous to a boxing match in which success is determined both by knocking down one's opponent and by not being knocked down (Aarssen 1989). We do not know what specific traits influence the ability to tolerate or suppress neighbors (but see Goldberg and Landa 1991, Goldberg 1996; Olesen et al. 2004; Cahill et al. 2005; Wang et al. 2010), but because these two components of competition are often not correlated (Peart 1989; Goldberg and Landa 1991; Keddy et al. 1994; Cahill et al. 2005; Fraser and Miletti 2008; Wang et al. 2010; but see Wilson and Keddy 1986; Goldberg and Fleetwood 1987; Miller and Werner 1987; Gurevitch et al. 1990; Novoplansky & Goldberg 2001; Thomsen et al. 2006), competitive tolerance and suppression are thought to be determined by partially independent underlying traits (Goldberg and Landa 1991; Goldberg 1996; Wang et al. 2010). If tolerance and suppression are independent they may respond differently to selective forces and may have independent ecological consequences. Explicitly recognizing and testing this two-part conceptual model of competition therefore has substantial value for understanding the ecological and evolutionary consequences of competition.

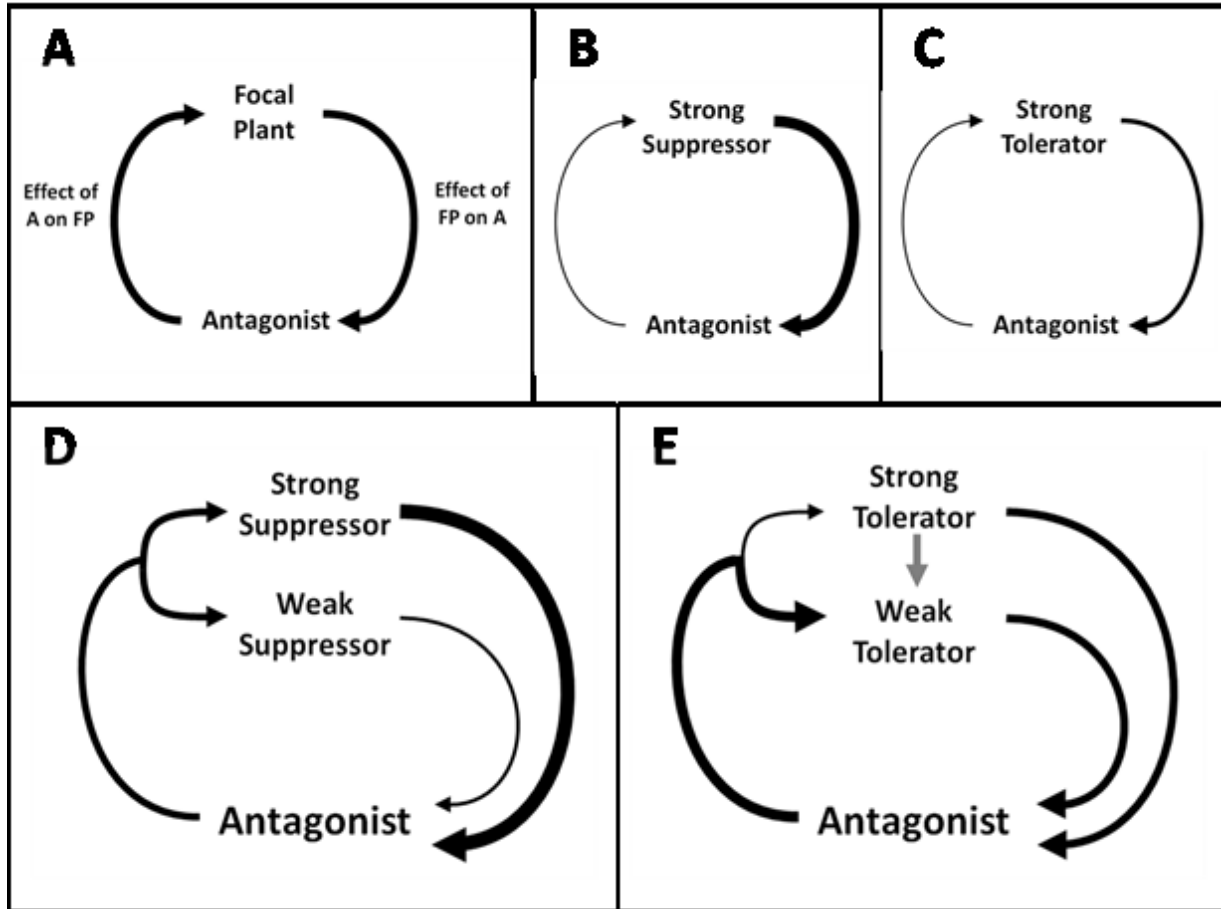


FIG. 1: The boxing match and demolition derby models of plant competition. Panel 1A depicts a “boxing match” between two competitors. In our models *P. spicata* is considered to be the focal plant, and *C. stoebe* the antagonist, and the left and right arrows indicate tolerance and suppression ability of *P. spicata*, respectively. The focal plant can reduce the suppression it experiences from the antagonist either indirectly, by suppressing it and thereby weakening its effects (Panel 1B), or directly by tolerating its effects (Panel 1C). When two or more focal plants compete with an antagonist, in the “demolition derby” scenario, suppression ability does not provide an exclusive advantage because the advantages of reduced competition with the antagonist are shared by both focal plants (Panel 1D). However the benefits of tolerance are not shared; if focal plants vary in their tolerance ability, the more tolerant plant will experience reduced competition from the antagonist and will have an advantage (grey arrow) whenever it is present (Panel 1E).

For similarly sized species competing in a “boxing match” scenario, the current paradigm is that tolerance and suppression either are equal in relative importance or suppression is more important, depending upon which resources are limiting and whether competitors are similarly sized (Goldberg 1990). This is because suppression is thought to have important indirect benefits via the exclusion of competitors. However, we propose that this paradigm may be likely to hold only in the context of pair-wise competition; i.e. just two individuals or genotypes competing rather than many at the same time. In multi-individual or multispecies communities we hypothesize that the benefits of having a greater ability to suppress other species will be low.

For just two competing individuals, the boxing match model applies because each individual competitor is the sole beneficiary of both suppressing and tolerating its competitor. If a focal plant suppresses its competitor, it will also experience reduced competition from its weakened competitor (Fig. 1B). Alternatively, it can achieve the same result by simply being more tolerant of its competitor’s effects (Fig. 1C). But in complex communities consisting of more than two individuals (or genotypes), the suppression of a competitor will benefit all other individuals that interact with that competitor (Fig. 1D). Importantly these “undeserving” beneficiaries (those that are poor suppressors but gain from the suppression done by others) still have competitive effects on other species in the system, including the plants that have indirectly benefitted them. Thus, strong suppressors might not only have to share the benefits of their suppression but they might even sabotage themselves by indirectly making some of their opponents stronger. In contrast, tolerating a competitor will only benefit the individual experiencing competition (Fig. 1E). Thus we predict that in complex communities the species or genotypes that demonstrate disproportionately high abilities to tolerate competition will survive invasion or the presence of strong competitors in general, and become more abundant over time

than those that demonstrate disproportionately high abilities to competitively suppress their neighbors. If so, the multiplayer chaos of a demolition derby may provide a better analogy than a boxing match for competition in plant communities. In a demolition derby, the benefits of dealing damage are limited by strong indirect benefits to other competitors, while the benefits of enduring or avoiding damage are not (Fig. 2). As a result, in a demolition derby the best strategy is simply to avoid collisions, and this is true even if there is no inherent cost to offensive driving. Because driving in such a way is counter to the spirit of the sport, in demolition derbies excessively defensive driving is called “sandbagging” and is outlawed. Plants, however, have no such restriction.



FIG. 2: A demolition derby. The winner is the last car that can still move. Offensive drivers indirectly benefit all other competitors every time they damage or eliminate an opponent. Because damaging opponents helps other cars on the track the soundest strategy is to “sandbag” – i.e. to completely avoid contact with other cars. This is true even if there are no risks to offensive driving. Because it is counter to the spirit of the sport and unfair to more aggressive competitors, in demolition derbies – unlike plant communities – sandbagging is not allowed.

Although competitive suppression and tolerance, considered separately, have each been shown to affect species abundances in the field (e.g. Grubb 1982; Roush and Radosevich 1985; Mitchley and Grubb 1986; Keddy 1990; Howard 2001; Howard & Goldberg 2001), to our knowledge, only a few studies have explicitly compared the relative ecological importance of tolerance versus suppression. Miller and Warner (1987) found that suppression and tolerance abilities were tightly correlated among five old-field species, which produced a strong competitive hierarchy that predicted the abundances of species in the field. But because suppression and tolerance were tightly correlated they could not tease apart the relative contribution of each competitive strategy. MacDougall and Turkington (2004) found that competitive tolerance and not suppression in competition trials predicted the relative abundance of two exotic invaders in the field. In a complex model intended to analyze tradeoffs for selection on herbivore defense versus competitive ability in *Solidago altissima*, Uriarte et al. (2002) found that selection favored only competitive tolerance traits, and not suppression traits, in populations experiencing intraspecific competition. More recently Willis et al. (2010) showed that both competitive suppression and tolerance abilities had important effects on the fitness of *Arabidopsis thaliana* competitors in two-genotype populations.

Here we link competitive suppression and tolerance abilities to potential field performance through simulation models informed by experimental competition trials between 23 different ecotypes of the native grass *Pseudoroegneria spicata* (Pursh) Á. Löve (bluebunch wheatgrass) and the North American invader *Centaurea stoebe* L. ssp. *micranthos* (Gugler) Hayek (spotted knapweed). We used the results of these trials to construct single-ecotype models in which we simulated invasion of populations containing single ecotypes of *P. spicata*, and multi-ecotype models in which we invaded mixed populations of *P. spicata* consisting of

multiple ecotypes. We performed our study in the context of exotic invasion because competition is often thought to play an important role in invasions in general (Vila and Weiner 2004) and for *C. stoebe* specifically (Maron and Marler 2008; Callaway et al. 2011). We tested the hypotheses that 1) in single ecotype models, the “boxing match” scenario will play out and suppression and tolerance will have similar effects on the ability of ecotypes to survive and resist invasion, and 2) in the multiple ecotype models, the “demolition derby” scenario will play out, and tolerance will be a more important determinant of abundance than suppression.

## METHODS

*Pseudoroegneria spicata* is a large and long-lived cool-season, drought-tolerant bunchgrass and a dominant member of native grasslands throughout the western United States. *Centaurea stoebe* is an invasive short-lived perennial that is native to Eurasia and has become highly invasive in the USA since its introduction at the beginning of the 20<sup>th</sup> century (Roché and Talbott 1986). These two species are often found together in the grasslands of the Mountain West (Watson and Renney 1974; Strang et al. 1979; Chicoine et al. 1985; Ridenour and Callaway 2001), but *C. stoebe* can also form mono-dominant stands that exclude many natives and that strikingly alter rangeland and grassland habitat throughout the Northern Great Plains and Intermountain West (Harris and Cranston 1979; Maddox 1979; Tyser and Key 1988; Ridenour and Callaway 2001). The competitive ability of *C. stoebe* has been attributed to many factors, including its ability to capitalize on disturbance (Tyser and Key 1988), strong competition in general or for resources (Herron et al. 2001; Ridenour and Callaway 2001; Maron and Marler 2008; Callaway et al. 2011; Aschehoug et al. 2012), allelopathy (Ridenour and



Callaway 2001; He et al. 2009), and release from inhibition by herbivores and soil biota (Story et al. 2006; Callaway et al. 2004; Ridenour et al. 2008; Schaffner et al. 2011).

We acquired seeds of 23 *Pseudoroegneria spicata* ecotypes from the Plant Germplasm Introduction and Testing Research Station in Pullman, WA, USA. These seeds were from true-bred lines collected from various populations in nine states throughout the USA and in British Columbia. *Centaurea stoebe* seeds were field-collected from a single site near Missoula, MT, USA to minimize variation in *C. stoebe* competitive ability. We germinated seeds of each *P. spicata* ecotype in 500 mL pots containing a 50/50 mix of sand and native soil from intermountain grassland near Missoula. After four weeks we thinned seedlings to one per pot, and added seeds of *C. stoebe* to half of the pots for each ecotype. The remaining half of the *P. spicata* plants were grown without competition. We also planted *C. stoebe* alone. Growing each species both alone and in competition allowed us to calculate the average tolerance of each *P. spicata* ecotype to *C. stoebe* and the average suppression of *C. stoebe* by each *P. spicata* ecotype.

We measured tolerance and suppression ability using the relative interaction intensity (RII) index (Armas et al. 2003), which is calculated as follows:

$$\text{RII} = [\text{biomass in competition} - \text{biomass grown alone}] / [\text{biomass in competition} + \text{biomass grown alone}]$$

Values of this metric can range from 1 to -1. A negative value indicates a negative competitive interaction and a positive value indicates facilitation. The RII for *P. spicata* represents the cost of competition with *C. stoebe* for *P. spicata*. It can be interpreted either as the tolerance of *P. spicata* to competition with *C. stoebe*, or as the suppression ability of *C.*

*stoebe*. Likewise, the RII for *C. stoebe* represents both the tolerance of *C. stoebe* and the suppression ability of *P. spicata*. The RII indices are net metrics of competition, and the values we measure are particular to the interaction between each ecotype and its competitor. A change in that competitor would produce different RIIs, and therefore different estimates of the tolerance and suppression ability of each *P. spicata* ecotype. But because we held the competitor constant, we can interpret differences in the RII indices of each ecotype as the result of differences in competitive abilities of just those ecotypes. And because each RII index describes the net outcome of competition between a particular ecotype of *P. spicata* and *C. stoebe*, we do not need to know the gross competitive traits of each ecotype in order to model its invasion by *C. stoebe*. We therefore define tolerance and suppression ability as follows:

$$\text{Tolerance} = RII_{C \text{ on } P}$$

$$\text{Suppression} = RII_{P \text{ on } C}$$

More negative values for tolerance indicate poorer performance of a particular *P. spicata* ecotype when competing with *C. stoebe*. For suppression, all values were also negative, and a more negative value of suppression indicated a *stronger* suppressive effect of *P. spicata* on *C. stoebe*. The terms “tolerance” and “suppression” are often used interchangeably with “competitive response” and “competitive effect” in the literature; however, for clarity we use the former throughout this paper.

To simulate the long term effects of competition between *P. spicata* and *C. stoebe*, we used the RII indices described above in an individual-based spatially-explicit dual-lattice simulation model (Travis et al. 2005; 2006; Michalet et al. 2011). Simulations were performed

in NetLogo (Wilensky 1999), a multi-agent modelling language particularly well suited for modelling complex systems that develop over time. In this model, *C. stoebe* and *P. spicata* occupied two overlapping two-dimensional lattices of equal sizes ( $100 \times 100$  cells). Each cell on the lattice could be occupied by no more than one individual of the appropriate species. Only directly overlapping *P. spicata* and *C. stoebe* plants interacted in our model; there were no direct interactions among adjacent cells in a lattice, although within each lattice all plants competed neutrally for space during reproduction. We assumed that instantaneous survival rate of *P. spicata* increased linearly with increased tolerance ability of *P. spicata* (less negative  $RII_{C \text{ on } P}$ ) and that survival of *C. stoebe* decreased linearly with increased suppression ability of *P. spicata* (decreased  $RII_{P \text{ on } C}$ ). Therefore, the instantaneous survival rate of each ecotype of *P. spicata* was modeled as:

$$S_P = S_{P_{max}} + RII_{C \text{ on } P} \quad \text{when it overlaps with } C. \text{ stoebe}$$

$$S_P = S_{P_{max}} \quad \text{when it overlaps with empty cell}$$

where  $S_P$  is the maximum survival rate of *P. spicata* (between 0 and 1) and where we assumed  $S_{P_{max}}$  was the same for all *P. spicata* ecotypes. The instantaneous survival rate of *C. stoebe* was modeled as:

$$S_C = S_{C_{max}} + RII_{P \text{ on } C} \quad \text{when it overlaps with } P. \text{ spicata genotype } i$$

$$S_C = S_{C_{max}} \quad \text{when it overlaps with empty cell}$$

where  $S_{Cmax}$  is the maximum survival rate of *C. stoebe*. These equations result in a linear decrease in instantaneous survival probability with increased competitor suppression. Linear survival functions were used for parsimony but we experimented with other functions and found that they did not qualitatively affect model outcomes. Thus, tolerance ability directly improved survival in a linear fashion, and suppression indirectly improved survival by reducing the survival probability of the competitor species in a linear fashion (Fig. 1A).

We used asynchronous updating in the model in the following way: First a single individual of *C. stoebe* or *P. spicata* was selected at random. Next, we determined whether the individual survived. If the individual survived it was allowed to reproduce and disperse propagules. Each time step was made up of  $N_C + N_P$  of such updates, where  $N_C$  and  $N_P$  refer to the number of all individuals of *C. stoebe* and all individuals of *P. spicata*, respectively. During reproduction, each individual produced propagules with competitive traits that were identical to those of that individual. To simplify our assumptions, all individuals of each species produced propagules at the same reproductive rate,  $r$  ( $r_C$  for *C. stoebe* and  $r_P$  for *P. spicata*). This rate was independent of the age of each plant or whether it had a competitor. Propagules from each species were dispersed sequentially to cells that were randomly selected within that species' own lattice. The propagules were only allowed to establish in empty cells and the one arriving first occupied the cell. Thus, there was competition among individuals within the same lattice for recruitment into empty cells. We used a "wraparound" (torus) approach to avoid edge effects (Yamamura *et al.* 2004). Parameters used in the final, reported simulations were:  $r_C = 1$ ,  $r_P = 1$ ,  $S_{Cmax} = 0.8$ ,  $S_{Pmax} = 0.8$ . The robustness of the model was tested with many different combinations of parameters and results were qualitatively the same as for the combinations reported here (data not shown).

We used these models to simulate the invasion of both single-ecotype and multi-ecotype populations of *P. spicata* by *C. stoebe*. In the single-ecotype simulations we separately invaded lattices composed of individual populations of each of the 23 *P. spicata* ecotypes with *C. stoebe*. Since the RIIs we used were averages for each ecotype, in the single ecotype models all *P. spicata* plants were identical. In the multi-ecotype models, we pooled all or some combination (see below) of the 23 separate ecotypes into a single, genetically diverse population containing each ecotype in equal abundance. Per-capita reproduction was assumed to be the same for each population of *P. spicata*.

All simulations were run for 10000 time steps in order to allow the system to reach equilibrium. All measurements were determined as the mean values of 100 independent replicate runs for each time step. Since the initial population sizes of invaders are likely to be small at the beginning of invasions, all simulations were started with 100 individuals of *C. stoebe* (1% of maximum population size). The *P. spicata* lattice was initially saturated, and in the models containing multiple genotypes, all genotypes started with equal abundance. All individuals of *C. stoebe* and *P. spicata* were randomly dispersed across their own lattices.

In the single-ecotype models, we calculated the competitive rank of *P. spicata* ecotypes by ranking those ecotypes that stably coexisted with *C. stoebe* according to their equilibrium abundance. Ecotypes that caused extinction of *C. stoebe* were ranked first, in order of the speed at which they drove *C. stoebe* extinct. Ecotypes that failed to persist were ranked last, and in order of the speed at which they went extinct. In the multi-ecotype models, when 23 *P. spicata* ecotypes were assembled into a single community, only one ecotype usually survived invasion. We assigned this ecotype a rank of “one” and then assembled the remaining 22 ecotypes into a community and again simulated the invasion of that community, now lacking the one ecotype

that survived in the previous simulation. The surviving ecotype in the second simulations was given a rank of “two” and was excluded from a subsequent simulation intended to identify the third-ranked ecotype. This process continued until only one ecotype remained, and that ecotype was given rank “23.”

To assess the relative importance of tolerance and suppression for the performance of *P. spicata* we used multiple linear regressions with tolerance and suppression ability as predictors of competitive rank in both the single- and multi-ecotype models. We calculated the individual contribution of both tolerance and suppression to variance explained in the regression as:

$$\% \text{ Variance explained by tolerance} = \beta_t r_{rt} / R^2$$

$$\% \text{ Variance explained by suppression} = \beta_s r_{rs} / R^2$$

Where  $\beta_t$  and  $\beta_s$  are the standardized regression coefficients for tolerance and suppression in the regression of rank against tolerance and suppression, with  $R^2$  being the coefficient of determination of that regression, and where  $r_{rt}$  and  $r_{rs}$  are the Pearson correlations between rank and tolerance or suppression.

The generality of our results from the empirically informed models was tested with theoretical models simulating 289 ecotypes with different combinations of competitive suppression and tolerance abilities and modelling the outcome of *C. stoebe* invasion on these artificial communities. These ecotypes represent a wider range of combinations of tolerance and suppression abilities, in which we set a minimum value of 0 and a maximum value of 0.8, with an interval of 0.05 between possible values of effect and response. Thus, there were 17 regularly spaced values of effect and response with 289 unique combinations. For the simulated ecotypes,

models were constructed identically to the models used for the real ecotypes, and the same procedures were used to determine competitive ranks.

## RESULTS

The ability to tolerate competition from *C. stoebe* and the ability to suppress *C. stoebe* varied substantially among the 23 ecotypes of *P. spicata*. RIIs for suppression ranged from -0.54 to -0.17 and RIIs for tolerance ranged from -0.54 to -0.14, and suppression and tolerance were not correlated ( $r = -0.281$ ,  $p = 0.195$ ,  $n = 23$ , Table 1, Fig. 3). The values for tolerance and suppression for the two species had similar distributions; *C. stoebe* and *P. spicata* were roughly equivalent competitors in measured conditions and our results were not skewed by one species being a vastly superior competitor than the other.

In the single-ecotype models based on the 23 natural *P. spicata* ecotypes, three ecotypes (2, 4, and 7) competitively excluded *C. stoebe*, and four ecotypes (5, 11, 12, and 19) did not survive invasion. The remaining 16 ecotypes coexisted stably with *C. stoebe* in the models but varied substantially in final abundance. Competitive hierarchies of *P. spicata* ecotypes (Table 1) were determined both by the tolerance and suppression abilities of *P. spicata* in single-ecotype models (Fig. 3, Fig. 4A). Linear regression analyses indicated that ability to tolerate competition accounted for 74% of the explained variance in final rank of natural *P. spicata* ecotypes, whereas ability to suppress *C. stoebe* contributed 26% to the variance in final rank of *P. spicata*. In the single-ecotype models based on 289 simulated ecotypes, 52% of the explained variance in rank was determined by tolerance and 48% by suppression (Table 2). This discrepancy occurred because the relative importance of tolerance versus suppression was not uniform throughout the parameter space. In some regions of parameter space tolerance played more of a role in

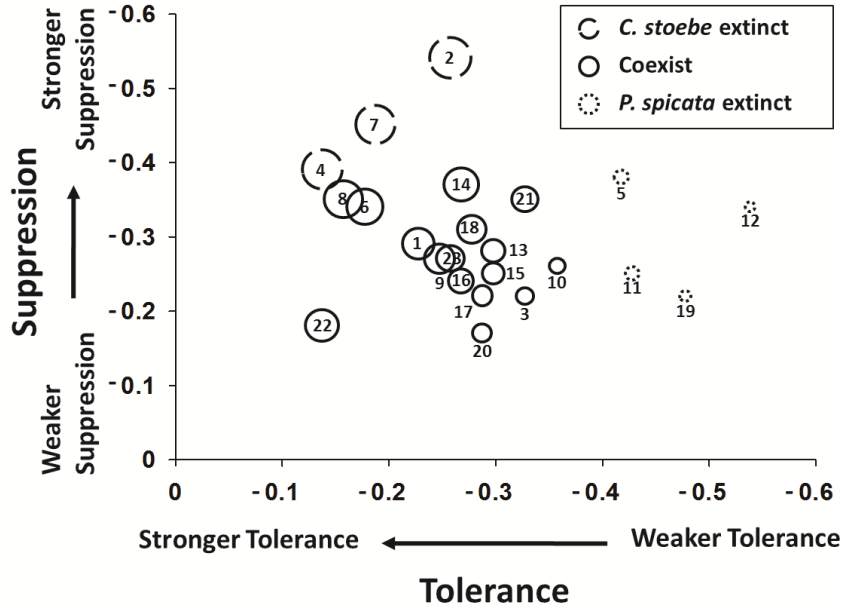


FIG. 3: Tolerance and suppression abilities of the 23 ecotypes of *P. spicata*. Number indicates the ID number assigned to each ecotype. The size of the circle corresponds to the competitive rank of each ecotype in the single-ecotype simulations, with better competitors having larger circles (see Table 1 for values). Line style indicates whether that ecotype competitively excluded *C. stoebe* (dashed), coexisted with *C. stoebe* (solid), or went extinct (dotted).

determining fitness and in others suppression played a stronger role. The natural ecotypes happened to occupy a part of the parameter space where tolerance was more important (Fig. 4A). The simulated ecotypes, however, were evenly distributed across the entire possible parameter space. When the entire parameter space was considered, tolerance and suppression played an equal role in determining competitive rank. Therefore, in the broadest consideration, tolerance and suppression were roughly equal in determining rank in the single-ecotype models.

The performance of *P. spicata* ecotypes in single-ecotype models was a poor predictor of their success in the multi-ecotype models (Table 1). When all 23 *P. spicata* ecotypes were mixed together for the complete multi-ecotype model, only one ecotype survived (ecotype 4),



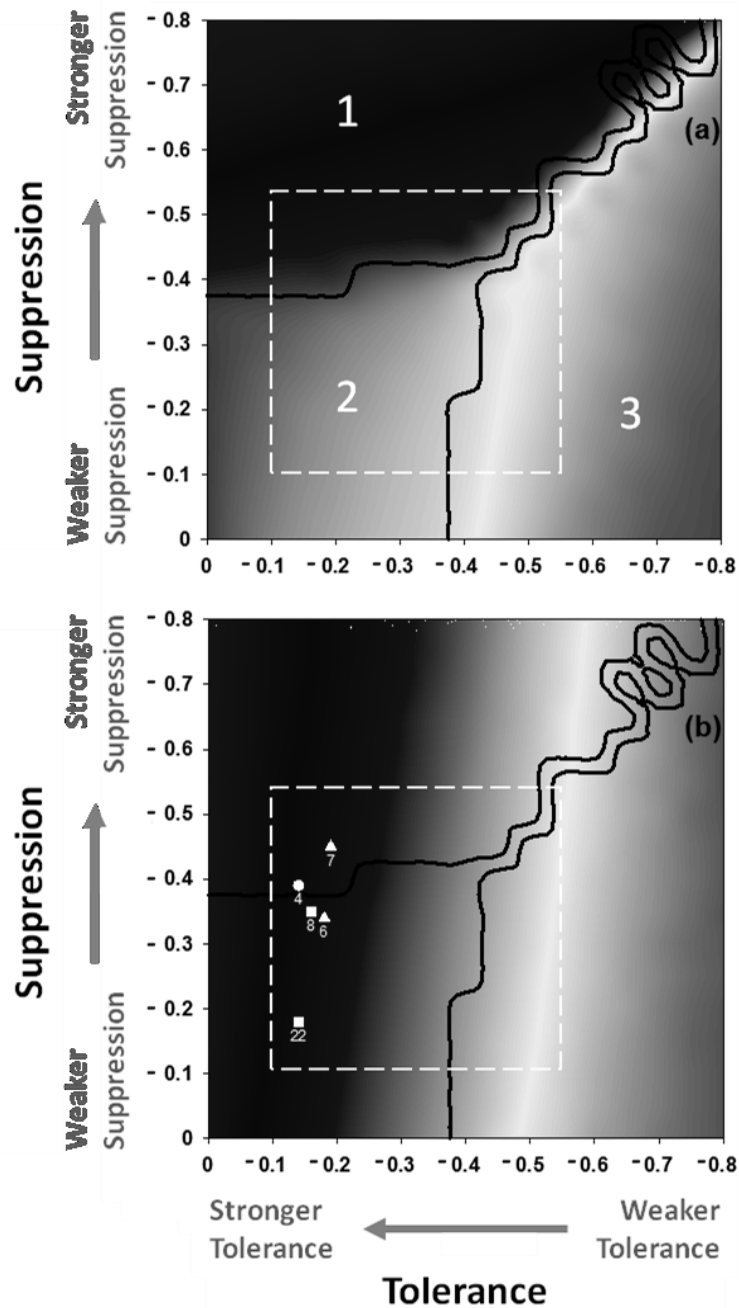


FIG. 4: Outcome of the simulations involving 289 artificial ecotypes. Panel 4A shows the outcome of the single-ecotype models and Panel 4B shows the outcome of the multi-ecotype models. Color indicates the competitive rank of the ecotype with the indicated combination of tolerance and suppression ability. Blue (upper left) indicates higher rank and red (lower right) indicates lower rank. Panel 4A is divided into three regions by black lines. Region 1 indicates ecotypes that could competitively exclude *C. stoebe*, Region 2 indicates ecotypes that could coexist with *C. stoebe*, and Region 3 indicates ecotypes that were driven extinct. These same regions are superimposed over Panel 4B. On each graph the approximate parameter space occupied by the real ecotypes is pictured as a box with a dotted white line. Also on Panel 4B are marks indicating the position of five of the actual ecotypes

(4: circle, 8 and 22: square, 6 and 7: triangle). These are the survivors of the 23-, 22-, and 20-ecotype models, respectively. Although these succeeded primarily due to their competitive response ability, the final outcome for *C. stoebe* is determined by whether at least one survivor was able to competitively exclude *C. stoebe* in the single-ecotype model (*i.e.* it whether it falls in Region 1 or 2).

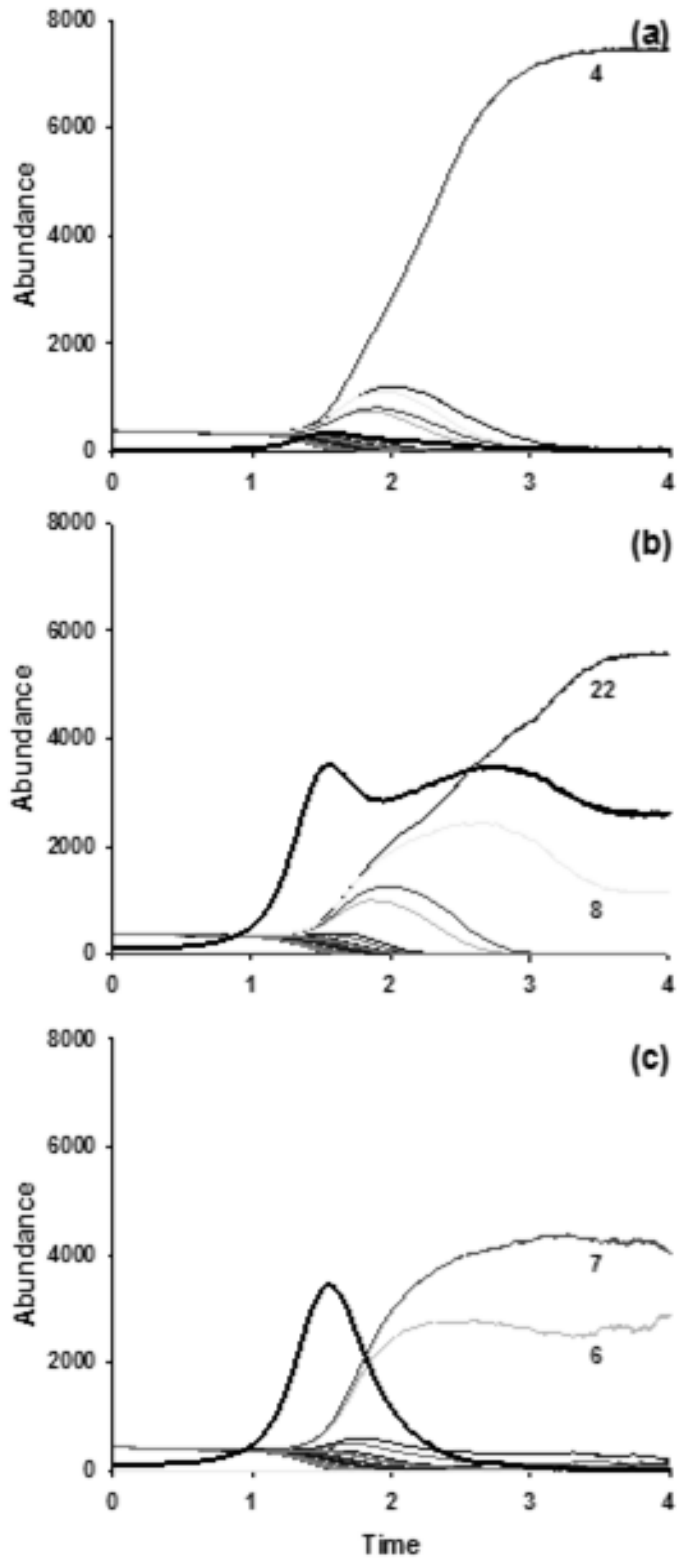


FIG. 5: Change in abundance of each ecotype over time in the multi-ecotype models. The outcomes of the 23-ecotype (Panel 5A), 22-ecotype (Panel 5B), and 20-ecotype (Panel 5C) models are shown. Each line indicates the abundance of the corresponding ecotype as time passed in our models. The bolded, black line indicates *C. stoebe* abundance. Surviving *P. spicata* ecotypes are labeled on each figure.

and the 18 ecotypes that coexisted with *C. stoebe* in the single-ecotype models went extinct (Fig. 5A). In fact, two *P. spicata* ecotypes that went extinct in the 23-ecotype model competitively excluded *C. stoebe* in the single ecotype models (ecotypes 2 and 7). After all *P. spicata* ecotypes other than ecotype 4 were eliminated from the model, *C. stoebe* also went extinct. To determine whether one of these ecotypes would rise to dominance if ecotype 4 was absent, we ran a 22-ecotype model with ecotype 4 excluded. In this model, ecotypes 8 and 22 persisted but ecotypes 2 and 7 again were eliminated. Furthermore, *C. stoebe* persisted at a high final abundance in this 22-ecotype model (Fig. 5B). Finally, we removed ecotypes 4, 8 and 22 from our simulation. In this 20-ecotype model, *C. stoebe* went extinct and ecotypes 6 and 7 became dominant (Fig. 5C).

Ecotypes went extinct from the multi-ecotype models roughly in order of their ability to tolerate competition from *C. stoebe*. Thus, rank in the multi-ecotype models was mostly, but not entirely, determined by tolerance ability (Table 1). Ninety three percent of the explained variance in overall competitive rank of natural ecotypes was determined by tolerance, with just 7% determined by the ability to suppress *C. stoebe* (Table 2). Results of the models using 289 simulated ecotypes were similar, with tolerance ability accounting for 99% of the explained variance in competitive rank (Table 2, Fig. 4B).

## DISCUSSION

In this study we observed a surprisingly large degree of natural variation in the ability of native *P. spicata* ecotypes from different parts of their natural North American range to competitively suppress *C. stoebe* and to tolerate its competitive effects. Moreover, suppression ability and tolerance ability were not correlated for *P. spicata*, suggesting that natural populations of this species have strikingly different abilities to respond to or to attenuate invasion by *C. stoebe*.

To determine whether tolerance or suppression ability is more important for how native *P. spicata* ecotypes might respond to invasion, we used individual-based simulation models. Specifically, we used two types of models: single-ecotype models, in which each ecotype competed one-on-one with *C. stoebe*, and multi-ecotype models in which the ecotypes all interacted with *C. stoebe* at the same time. As predicted, in the single-ecotype models tolerance and suppression played approximately equal roles in establishing competitive hierarchies (Table 2, Fig. 4A). This result is important because it shows that, despite the fact that the fitness benefits of tolerance were direct and the benefits of suppression were indirect, overall benefits of both strategies had the opportunity to be equivalent in these models. Suppression was important in the single-ecotype models for two reasons: First, *P. spicata* plants with better suppression ability were able to more quickly remove their competitor from the overlapping cell in the *C. stoebe* lattice. Second, suppression of *C. stoebe* by the entire *P. spicata* population enabled *P. spicata* to control *C. stoebe* abundance, thereby generating a feedback process that reduced the strength of competition from *C. stoebe* globally.

Despite tolerance and suppression being equally important in determining competitive rank in the single-ecotype models (in which each ecotype competed alone with *C. stoebe*), in the multi-ecotype models, in which multiple coexisting ecotypes competed with *C. stoebe*, tolerance eclipsed suppression for establishing competitive hierarchies and for determining the relative abundance of the different ecotypes (Tables 1 and 2, Fig. 4B). The reasons for this are illustrated in Figure 1. When multiple ecotypes (or genotypes) compete with a single competitor, the benefits of suppression are shared (Fig. 1D) but the benefits of tolerance are not (Fig. 1E). In the simplified case presented in Figure 1, the benefits of suppression are shared completely between the strong and weak suppressor. In such a scenario suppression of neighbors would not confer

any fitness advantage at all. In our simulations, however, suppression provided a small benefit because strong suppressors gained a transient exclusive (i.e. un-shared) benefit from clearing their overlapping cell of *C. stoebe*. But since strong suppressors were constantly inundated with *C. stoebe* propagules, even after successfully outcompeting their neighbors, this exclusive benefit of suppression was temporary and roughly 100 times less important than tolerance in our theoretical models (Table 2). This finding is surprising because it suggests that even though neighbor competition caused a many-fold reduction in the survival probability of individual plants in our models, there was virtually no fitness benefit of neighbor exclusion, which is consistent with Uriarte et al. (2002).

The exclusive benefits of suppression were weak in our models because individuals were not able to engineer a favorable environment either for themselves or for their offspring by suppressing their neighbors. However, is it possible that under different conditions suppression has stronger benefits than we observed in this study? We can hypothesize about how our model assumptions influenced our results by considering the degree to which benefits of suppression are shared versus exclusive under different conditions. For example, we assumed that dispersal was global, so our model lacked spatial structure. In our models, the exclusive advantage of strong suppressors was their ability to temporarily remove *C. stoebe* from an overlapping cell. So if dispersal was limited, strong suppressors could more strongly engineer their local environment by removing *C. stoebe* over a larger area, and therefore potentially enjoy a longer-lasting exclusive benefit of suppression. But still, any *P. spicata* propagule entering the cleared area, regardless of its competitive strength, would benefit from the absence of *C. stoebe*. Moreover, strong suppressors would have the same difficulty in controlling the global abundance of *C. stoebe* as they do in our models, because weak competitors would form patches that would

act as refuges and propagule sources for *C. stoebe*. As a result, suppression might not actually improve the average fitness of a genotype over an entire landscape. Thus we predict that dispersal limitation should not strongly affect the importance of tolerance versus suppression. Uriarte *et al.* (2002) obtained a very similar result to the one reported here, using a model that had strong dispersal limitation, but that differed from our model in many other ways. For example, Uriarte *et al.* modeled direct competition between adjacent plants, while we restricted direct competition only between exactly overlapping intraspecific competitors. And in their model, competitive interactions reciprocally affected plant size, which in turn affected reproductive rate, while we did not model plant size at all. The fact that two independent models with very different structure reached the same result increases our confidence in our theoretical model (Fig. 1D & E) and its assumptions. Nonetheless, more research is needed to determine whether and under what conditions neighbor suppression conveys a fitness advantage in nature.

Our results suggest that competition in a community may not be played out as a conglomeration of pair-wise boxing matches in which species or genotypes attain success in equal measure by suppressing and by tolerating their neighbors. Instead, our results suggest that competition in plant communities more closely resembles the multiplayer chaos of a demolition derby, where damaging an opponent helps everyone else on the track to the same degree. Because sandbaggers – the drivers who intentionally avoid causing damage to other cars – also benefit from the destruction caused by offensive drivers, the benefits of such offensive driving are unfairly shared in a demolition derby. If there is no personal advantage to damaging your opponents, there is no reason to risk your car by doing so. In the same way, in plant communities, tolerating competition appears to be of paramount importance, and even successful

elimination of neighbors by competitive suppression has very limited benefits, even if those neighbors are strong suppressors themselves.

This theoretical model of plant competition has important implications for our understanding of the ecology of plant systems, and for our interpretation of data derived from pair-wise competition trials or competition trials involving two populations consisting of only one genotype each. This is because pair-wise competition may unrealistically favor strong suppressors – particularly if the general competitive interaction is relatively strong (Fig. 4A) – and thus poorly predict competitive outcome in more diverse natural conditions in which suppression plays a lessened role in determining fitness. Thus competitive hierarchies established using pair-wise competition trials may be misleading even when not confounded by other factors such as herbivory or environmental heterogeneity.

The “demolition derby” model of plant competition also has particular importance for understanding selective pressures imposed by invasive species as they re-shape native plant communities. For example, if invaders select for native genotypes with superior ability to tolerate competition (see Aarssen 1983; Strauss et al. 2006; Lau 2008; Thorpe et al. 2011), and if these traits are heritable, invasions may lead to evolutionary increases in the competitive tolerance of native plant species and shifts in the composition of the plant communities in ways related to these traits. Eventually, this could lead to a more stable coexistence of native and invasive species and attenuated invasion intensity over time. Evolutionary responses of native plants have been hypothesized to weaken invasion over time (Simberloff and Gibbons 2004; Callaway et al 2005; Lau 2008), but understanding the relative importance of competitive tolerance and its potential fitness benefits sheds new light how this process may occur. Extending this argument, we also propose that coevolution between competing plants in general

might attenuate the strength of competitive interactions over time. However, plants certainly express traits that strongly suppress neighbors, and this can correspond with variation in abundance among species in the field (e.g. Grubb 1982; Roush and Radosevich 1985; Mitchley and Grubb 1986; Keddy 1990).

How can we reconcile the co-existence of both strong suppression ability and strong tolerance in natural communities with our demolition derby model of competition? We propose that selection for many traits associated with increased suppression ability, particularly those related to size and resource uptake, may be incidental and not directly related to suppression ability *per se*, but instead is related to other inherent advantages of such traits. For example, plants with wide canopies might suppress their neighbors by shading them. However, the selective forces that promote canopy width might relate only to the direct benefits of increased light uptake – counter-intuitively, suppression of neighbors might be an ancillary consequence of these traits that is not beneficial in *any* context, even if it improves survival by excluding neighboring competitors. In sum our results suggest that a demolition derby model of plant competition, with its surprising prediction that suppression strategies provide few advantages in multi-species plant communities, has important implications for the design of studies investigating the importance of competitive hierarchies, our understanding of the selective forces that competing plants experience and how ecological conditions alter those forces, and our knowledge of how competitive interactions between plants affect the distribution and abundance of plants in nature.



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#### LITERATURE CITED

- Aarssen L. W. 1983. Ecological combining ability and competitive combining ability in plants: Toward a general evolutionary theory of coexistence in systems of competition. *American Naturalist* 122:707-731.
- Aarssen L. W. 1989. Competitive ability and species coexistence: A “plant’s eye” view. *Oikos* 56:386-401.
- Armas C., Ordiales R., and Pugnaire F. I. 2004. Measuring plant interactions: A new comparative index. *Ecology* 85:2682-2686.
- Aschehoug, E.T., Metlen, K.L., Callaway, R.M. and Newcombe, G. 2012. Fungal endophytes directly increase the competitive effects of an invasive forb. *Ecology* 93:3-8.
- Cahill J. F., Kembel S. W., and Gustafson D. J. 2005. Differential genetic influences on competitive effect and response in *Arabidopsis thaliana*. *Journal of Ecology* 93:958-967.
- Callaway R. M., Ridenour W. M., Laboski T., Weir T., and Vivanco J. M. 2005. Natural selection for resistance to the allelopathic effects of invasive plants. *Journal of Ecology* 93:5-15.

- Callaway R. M., Thelen G., Barth S., Ramsey P. W., and Gannon J.E. 2004. Soil fungi alter interactions between the invader *Centaurea maculosa* and North American natives. *Ecology* 85:1062-1071.
- Callaway R. M., Waller L. P., Diaconu A., Pal R., Collins A. R., Müller-Schärer H., and Maron J. L. 2011. Escape from competition: Neighbors reduce *Centaurea stoebe* performance at home but not away. *Ecology* 92:2208-2213.
- Chicoine T. K., Fay P. K., and Nielsen G. A. 1985. Predicting weed migration from soil and climate maps. *Weed Science* 41:57-61.
- Fraser L. H., and Miletti T. E. 2008. Effect of minor water depth treatments on competitive effect and response of eight wetland plants. *Plant Ecology* 195:33-43.
- Goldberg D. E. 1990. Components of resource competition in plant communities. Pages 27-49 in J. B. Grace, and D. Tilman, ed. *Perspectives on Plant Competition*. Blackburn Press, Caldwell, NJ.
- Goldberg, D. E. 1996. Competitive ability: Definitions, contingency and correlated traits. *Philosophical Transactions of the Royal Society of London: Biology* 351:1377-1385.
- Goldberg D. E., and Fleetwood L. 1987. Competitive effect and response in four annual plants. *Journal of Ecology* 75:1131-1143.
- Goldberg D. E., and Landa K. 1991. Competitive effect and response: Hierarchies and correlated traits in the early stages of competition. *Journal of Ecology* 79:1013-1030.
- Grubb P. J. 1982. Control of relative abundance in roadside *Arrhenatheretum*: Results of a long-term garden experiment. *Journal of Ecology* 70:845-861.

- Gurevitch J., Wilson P., Stone J. L., Teese P., and Stoutenburgh R. J. 1990. Competition among old-field perennials at different levels of soil fertility and available space. *Journal of Ecology* 78:727-744.
- He W., Feng Y., Ridenour W. M., Thelen G. C., Pollock J. L., Diaconu A., and Callaway R. M. 2009. Novel weapons and invasion: Biogeographic differences in the competitive effects of *Centaurea maculosa* and its root exudate ( $\pm$ )-catechin. *Oecologia* 159:803-815.
- Harris P., and Cranston R. 1979. An economic evaluation of control methods for diffuse and spotted knapweed in Western Canada. *Canadian Journal of Plant Science* 59:375-382.
- Herron G. J., Sheley R. L., Maxwell B. D., and Jacobsen J. S. 2001. Influence of nutrient availability on the interaction between spotted knapweed and bluebunch wheatgrass. *Restoration Ecology* 9:326-331.
- Howard T. G. 2001. The relationship of total and per-gram rankings in competitive effect to the natural abundance of herbaceous perennials. *Journal of Ecology* 89:110-117.
- Howard T. G., and Goldberg D. E. 2001. Competitive response hierarchies for germination, growth, and survival and their influence on abundance. 83:979-990.
- Keddy P. A. 1990. Competitive hierarchies and centrifugal organization in plant communities. Pages 266-290 in J. B. Grace, and D. Tilman, ed. *Perspectives on Plant Competition*. Blackburn Press, Caldwell, NJ.
- Keddy P. A., Twolan-Strutt L., and Wisheu I. C. 1994. Competitive effect and response rankings in 20 wetland plants: Are they consistent across three environments? *Journal of Ecology* 82:635-643.
- Lau J. A. 2008. Beyond the ecological: Biological invasions alter natural selection on a native plant. *Ecology* 89:1023-1031.

- MacDougal A. S., and Turkington R. 2004. Relative importance of suppression-based and tolerance-based competition in an invaded oak savanna. *Journal of Ecology* 92:422-434.
- Maddox D. M. 1979. The knapweeds: Their economics and biological control in the Western States, U.S.A. *Rangelands* 1:139-141.
- Maron, J. L., and Marler M. 2008. Field-based competitive impacts between invaders and natives at varying resource supply. *Journal of Ecology* 96:1187-1197.
- Michalet, R., Xiao S., Touzard B., Smith D. S., Cavieres L. A., Callaway R. M., and Whitham T. G. 2011. Phenotypic variation in nurse traits and community feedbacks define an alpine community. *Ecology Letters* 14:433-443.
- Miller T. E., and Werner P. A. 1987. Competitive effects and responses between plant species in a first-year old-field community. *Ecology* 68:1201-1210.
- Mitchley J., and Grubb P. J. 1986. Control of relative abundance of perennials in chalk grassland in Southern England: I. Constancy of rank order and results of pot- and field-experiments on the role of interference. *Journal of Ecology* 74:1139-1166.
- Novoplansky A., and Goldberg D. E. 2001. Effects of water pulsing on individual performance and competitive hierarchies in plants. *Journal of Vegetation Science* 12:199-208.
- Olesen J. E., Hansen P. K., Berntsen J., and Christensen S. 2004. Simulation of above-ground suppression of competing species and competition tolerance in winter wheat varieties. *Field Crops Research* 89:263-280.
- Peart D. R. 1989. Species interactions in a successional grassland. II. Colonization of vegetated sites. *Journal of Ecology* 77:252-266.
- Ridenour W. M., and Callaway R. M. 2001. The relative importance of allelopathy in interference: The effects of an invasive weed on a native bunchgrass. *Oecologia* 126:444-

450.

- Ridenour W. M., Vivanco J. M., Feng Y., Horiuchi J., and Callaway R. M. 2008. No evidence for tradeoffs: *Centaurea* plants from America are better competitors and defenders than plants from the native range. *Ecological Monographs* 78:369-386.
- Roché B. F. Jr., and Talbott C. J. 1986. The collection history of *Centaureas* found in Washington State. Agricultural Research Center of Washington State University, Research Bulletin XB 0978.
- Roush M. L., and Radosevich S. R. 1985. Relationships between growth and competitiveness of four annual weeds. *Journal of Applied Ecology* 22:895-905.
- Schaffner U., Ridenour W. M., Wolf V.C., Bassett T., Müller C., Müller-Schärer H., Sutherland S., Lortie C. J., and Callaway R. M. 2011. Plant invasions, generalist herbivores, and novel defense weapons. *Ecology* 92:829-835.
- Siemens D. H., Garner S. H., Mitchell-Olds T., and Callaway R. M. 2002. Cost of defense in the context of plant competition: *Brassica rapa* may grow *and* defend. *Ecology* 83:505-517.
- Simberloff D., and Gibbons L. 2004. Now you see them, now you don't – population crashes of established introduced species. *Biological Invasions* 6:161-172.
- Story J. M., Callan, N. W., Corn, J. G., and White, L. J. 2006. Decline of spotted knapweed density at two sites in western Montana with large populations of the introduced root weevil, *Cyphocleonus achates* (Fahraeus). *Biological Control* 38: 227-232.
- Strang R. M., Lindsay K. M., and Price R. S. 1979. Knapweeds: British Columbia's undesirable aliens. *Rangelands* 1:141-143.
- Strauss S. Y., Lau J. A., and Carroll S. P. 2006. Evolutionary responses of natives to introduced species: What do introductions tell us about natural communities? *Ecology Letters* 9:357-

374.

- Thomsen M. A., Corbin J. D., and D'Antonio C. M. 2006. The effect of soil nitrogen on competition between native and exotic perennial grasses from northern coastal California. *Plant Ecology* 186:23-35.
- Thorpe A. S., Aschehoug E. T., Atwater D. Z., and Callaway R. M. 2011. Interactions among plants and evolution. *Journal of Ecology* 99:729-740.
- Travis, J. M. J., Brooker R. W., Clark E. J, and Dytham C. 2006. The distribution of positive and negative species interactions across environmental gradients on a dual-lattice model. *Journal of Theoretical Biology* 241:896-902.
- Travis, J.M. J., Brooker R. W., and Dytham C. 2005. The interplay of positive and negative species interactions across an environmental gradient: insights from an individual-based simulation model. *Biology Letters* 1:5-8.
- Tyser R. W., and Key C. H. 1988. Spotted knapweed in natural area fescue grasslands: An ecological assessment. *Northwest Science* 62:151-160.
- Uriarte M., Canham C. D., and Root R. B. 2002. A model of simultaneous evolution of competitive ability and herbivore resistance in a perennial plant. *Ecology* 83: 2649-2663.
- Vilá, M., and Weiner J. 2004. Are invasive plant species better competitors than native plant species? - evidence from pair-wise experiments. *Oikos* 105:229-238.
- Wang P., Stieglitz T., Zhou D. W., and Cahill J. F. Jr. 2010. Are competitive effect and response two sides of the same coin, or fundamentally different? *Functional Ecology* 24:196-207.
- Wardle D. A., Nilsson M., Gallet C., and Zackrisson O. 1998. An ecosystem-level perspective of allelopathy. *Biological Reviews* 73:305-319.
- Watson A. K., and Renney A. J. 1974. The biology of Canadian weeds, *Centaurea diffusa* and *C.*

- maculosa. *Canadian Journal of Plant Science* 54:687-701.
- Wilensky, U. 1999. NetLogo.<http://ccl.northwestern.edu/netlogo/>. Center for Connected Learning and Computer-Based Modeling, Northwestern University, Evanston, IL.
- Willis C. G., Brock M. T., and Weinig C. 2010. Genetic variation in tolerance of competition and neighbour suppression in *Arabidopsis thaliana*. *Journal of Evolutionary Biology* 23:1412-1424.
- Wilson S. D., and Keddy P. A. 1986. Species competitive ability and position along a natural stress disturbance gradient. *Ecology* 67:1236-1242.
- Yamamura, N., Higashi M., Behera N., and Wakano J. Y. 2004. Evolution of mutualism through spatial effects. *Journal of Theoretical Biology* 226:421-428.

TABLES

TABLE 1: Competitive ranks of ecotypes in the single-ecotype and multi-ecotype models.

Pop.	Rank Single	Rank Multi	RII <i>P. spicata</i> (Tolerance)	RII <i>C. stoebe</i> (Suppression)
2	1	6	-0.255	-0.536
7	2	4	-0.187	-0.453
4	3	1	-0.142	-0.392
8	4	2	-0.162	-0.346
6	5	5	-0.179	-0.343
14	6	9	-0.272	-0.370
22	7	3	-0.144	-0.183
1	8	7	-0.230	-0.291
9	9	8	-0.248	-0.270
18	10	11	-0.284	-0.310
23	11	10	-0.260	-0.268
21	12	17	-0.331	-0.349
16	13	12	-0.274	-0.239
13	14	16	-0.304	-0.276
15	15	15	-0.300	-0.254
17	16	13	-0.292	-0.219
20	17	14	-0.289	-0.175
3	18	18	-0.334	-0.224
10	19	19	-0.364	-0.257
5	20	20	-0.424	-0.378
11	21	21	-0.433	-0.248
19	22	22	-0.484	-0.218
12	23	23	-0.540	-0.344

Competitive ranks of *P. spicata* ecotypes from the single-ecotype models “Rank Single” and multi-ecotype models “Rank Multi” are shown, along with the RII values for each ecotype. Data are sorted in terms of single-ecotype model rank.



TABLE 2: Results of linear regressions of ecotype competitive rank against tolerance and suppression.

Type of model		R <sup>2</sup>	n	p	β	Tolerance % Exp p		Suppression % Exp p		
Single	Empirical	0.952	23	0.001	-0.803	74.2%	< 0.001	0.429	25.8%	< 0.001
	Simulated	0.861	289	0.001	0.666	51.6%	< 0.001	-0.646	48.4%	< 0.001
Multi	Empirical	0.936	23	0.001	-0.915	92.8%	< 0.001	0.19	7.2%	0.004
	Simulated	0.997	289	0.001	0.993	98.9%	< 0.001	-0.106	1.1%	< 0.001

Regressions were performed on both the single-ecotype (Single) and multi-ecotype (Multi) models, using RIIs gathered both from 23 natural ecotypes (Empirical) and from 289 simulated ecotypes (Simulated). For each model, the determination coefficient (R<sup>2</sup>), sample size (n), and p value (p) are shown. The separate individual contribution of both tolerance and suppression traits on rank for each regression is indicated by the standardized regression coefficient of that trait (β), percent of total explained variance accounted for by that trait (% Exp), and p value (p).

## CHAPTER 3

### ECOTYPIC DIVERSITY OF A DOMINANT GRASSLAND SPECIES INCREASES PRODUCTIVITY THROUGH COMPLEMENTARITY

*Abstract:* Plant diversity enhances ecosystem processes, including productivity, but these effects have been studied almost exclusively at the taxonomic scale of species. Here we find that intraspecific diversity within one of North America's most widespread and dominant grassland species, *Pseudoroegneria spicata*, increases net annual productivity to a degree similar to that reported for different species. Importantly, this effect of ecotypic diversity is shown to be due to complementarity, or diversity *per se*, and not to the sampling of larger, stronger competitors in diverse assemblages. These results suggest that functional diversity within a species can be very high, and that cryptic biological diversity below the species level has the potential to strongly affect the functioning of ecological systems.

*One Sentence Summary:* Increasing ecotype richness causes a 40% increase in productivity through complementary, indicating very high functional diversity within a species.

#### MAIN TEXT

Concern over the ecological consequences of anthropogenic species loss has led to the discovery of profound effects of species richness on ecosystem function (Tilman et al. 1996, Knops et al.

1999, Hooper et al. 2005, Balvanera et al. 2006). One common finding is that species and functional group richness increases the productivity of plant communities, with diverse communities yielding on average 70% more than monocultures (Cardinale et al. 2007). Studies of diversity-dependentoveryielding have focused almost exclusively on diversity among species (Hooper et al. 2005, Balvanera et al. 2006) or functional groups (Díaz and Cabido 2001), as these are considered to define the key operational units in ecosystems (McGill et al. 2006).

However, a great deal of Earth's biological diversity also resides *within* species, and intraspecific diversity can strongly influence ecological processes (Hughes et al. 2008). We now know that functional diversity within a single plant population can be very high (Garnier et al. 2004, Albert et al. 2010a). Species also vary across their local and regional distributions, with many natural populations consisting of locally adapted "ecotypes" (Clausen et al. 1941, Cordell et al. 1998, Albert et al. 2010b).

The effects of intraspecific diversity on productivity previously have been explored for two species; *Festuca ovina* and *Solidago altissima*. For *F. ovina*, genotypic richness does not influence productivity (Fridley and Grime 2010), but for *S. altissima*, genotypic richness increases productivity to the same extent as interspecific diversity influences productivity (Crutsinger et al. 2006, 2008). However, it is not known whether this diversity-productivity relationship for *S. altissima* is due to ecological complementarity among ecotypes (Hector 1998), or to the selection effect – an increased probability of high-diversity populations containing larger, stronger competitors (Aarssen 1997, Huston 1997, Loreau 1998, Wardle 2001). Thus a crucial question remains; can intraspecific diversity, *per se*, affect ecosystem function (Loreau and Hector 2001)?

Here we find that experimentally constructed assemblages containing diverse ecotypic variants of *Pseudoroegneria spicata* (bluebunch wheatgrass), a widely distributed and dominant species in western North America, are substantially more productive than synthetic assemblages composed of low ecotypic diversity (Figure 1A). Importantly, these effects are almost exclusively due to diversity *per se*, caused by functional complementarity among ecotypes (Table 1, Figure 1B).

*Pseudoroegneria spicata* ecotypes used in this study were collected from 12 sites spanning their North American distribution, and were grown at four levels of ecotypic richness (1-, 3-, 8-, and 12-ecotypes). We estimated overyielding for each plot and partitioned the contributions of selection and complementarity effects after Loreau and Hector (2001). The 8- and 12-ecotype plots were 20-25% more productive than the monocultures, and were 40% more productive than 3-ecotype plots, which underyielded by 20% compared to the monocultures. Individual ecotypes varied in their responses to overall ecotypic diversity; some overyielded at high richness and others had constant yield at all levels of richness (Table S1). Overyielding at high diversity was due to positive complementarity effects, which were five times stronger than selection effects, and opposite in sign (Fig. 1B). This result is consistent with many other studies investigating overyielding produced by species diversity (Cardinale et al. 2007).

Complementary effects result from many processes, but resource partitioning is the most commonly cited cause of complementary overyielding (Tilman et al. 2001, Mulder et al. 2001, Ruijven and Berendse 2005, Cardinale et al. 2007, Fargione et al. 2007). Because negative correlations between available nitrogen and species richness have been taken as evidence for resource partitioning (Tilman et al. 2001) we measured plant -available nitrate and ammonium in each plot. We did not find that nitrogen concentrations varied with ecotypic richness or plot

productivity. Instead, plots that overyielded produced more biomass than expected for a given soil concentration of available nitrogen, meaning that *P. spicata* assemblages that were rich in ecotypes used soil resources more efficiently than single-ecotype plots. Increased nutrient use efficiency is also important in systems with complementary overyielding driven by species richness (Ruijven and Berendse 2005, Fargione et al. 2007).

Soil fungal pathogens have also been shown to determine species diversity – productivity relationships (Maron et al 2011, Schnitzer et al. 2011). Therefore we applied fungicide to half of our plots throughout the experiment. We did not find significant effects of fungicide on yield, complementarity, or selection effects (Table 1). In light of the strong effect of fungi on the relationship between species richness and productivity in other Montana grassland species grown the same experimental area (Maron et al. 2011), this result raises the possibility that fungi might function as broad generalist consumers among ecotypes, but as specialists among species (Schnitzer et al. 2011). However, it is also possible that our treatment was simply not efficacious or that *P. spicata* is less susceptible to fungi than the species used by Maron et al.

In contrast to complementarity effects, which increased with diversity, weak but statistically significant selection effects decreased with diversity (Fig. 1B, Table 1). Selection effects were positive in the 3-ecotype plots, but negative in the 12-ecotype plots, suggesting that when diversity was low, large ecotypes had a competitive advantage in a manner consistent with theory (Weiner 1990). However, in more diverse plots, the competitive advantage of large ecotypes waned and the relative competitive ability of smaller ecotypes improved. This novel evidence of intraspecific diversity leading to more “equitable” intraspecific competitive interactions (Fridley and Grime 2010) parallels the effects of increasing species richness on competition among species (Cardinale et al. 2007). It not clear how ecotypic diversity might

even the playing field among otherwise asymmetrical competitors, but our results suggest that genotypic diversity may result in unusual indirect interactions among competing ecotypes.

We focused on among-population rather than within-population diversity in order to capture as much functional diversity as possible, and to relate performance differences among ecotypes to differences in their local climate. We observed that at all levels of plot diversity, ecotypes from cooler, wetter environments overyielded more than ecotypes from hotter, drier environments (Fig. 2,  $R^2 = 0.020$ ,  $p = 0.023$ ), and that the ability of home climate to predict changes in complementarity effect increased when variance caused by diversity and monoculture yield was removed ( $R^2 = 0.026$ ,  $p = 0.009$ ). Thus, overyielding of mesic ecotypes occurred despite, and not because of, the fact that mesic ecotypes tended to have higher monoculture yields than xeric ecotypes. This is consistent with the theory that diversity-productivity relationships should be stronger for species adapted to more productive environments (Fridley & Grime 2010, but see Mulder et al. 2001).

Our use of ecotypes from a wide geographic range demonstrates that throughout its range *P. spicata* has undergone tremendous yet cryptic functional diversification. In fact, the importance of within-species functional diversity of the selected ecotypes matches that of functional variation among species from similar habitats (Maron and Marler 2007, Maron et al. 2011). It is unknown whether such differentiation is likely to occur within a single population, but other studies have found important functional variation within populations of *P. spicata* (Ridenour et al. 2008), as well as other species (Crutsinger 2006, Crutsinger 2008, Albert et al. 2010a). Nevertheless we caution that our results should not be interpreted as direct evidence of the importance of within-population functional diversity.

In sum, our results demonstrate strong overyielding caused by complementary interactions among ecotypes, substantial functional variation within a species, and that intraspecific diversity *per se* has important and independent effects on ecosystem processes. Expanding our knowledge of how ecotypic variation can affect ecosystem functioning may be crucial for predicting the effects of anthropogenic species losses and gains (Wardle et al. 2011) and for improving our understanding of how assemblages of organisms function in nature (Hughes et al. 2008, Brooker et al. 2009, Cianciaruso et al. 2009).

#### REFERENCES AND NOTES

- D. Tilman, D. Wedin, J. Knops, Productivity and sustainability influenced by biodiversity in grassland ecosystems. *Nature* **379**, 718-720 (1996).
- J. M. H. Knops, D. Tilman, N. M. Haddad, S. Naeem, C. E. Mitchell, *et al.*, Effects of plant species richness on invasions dynamics, disease outbreaks, insect abundances, and diversity. *Ecol. Lett.* **2**, 286–293 (1999).
- D. U. Hooper, F. S. Chapin III, J. J. Ewel, A. Hector, P. Inchausti, *et al.*, Effects of biodiversity on ecosystem functioning: a consensus of current knowledge and needs for future research. *Ecol. Monogr.* **75**, 3–35 (2005).
- P. Balvanera, A. B. Pfisterer, N. Buchmann, J. He, T. Nakashizuka, D. Raffaelli, *et al.*, Quantifying the evidence for biodiversity effects on ecosystem functioning and services. *Ecol. Lett.* **9**, 1146-1156 (2006).
- B. J. Cardinale, J. P. Wright, M. W. Cadotte, I. T. Carroll, A. Hector, *et al.*, Impacts of plant diversity on biomass production increase through time because of species complementarity. *Proc. Natl. Acad. Sci., U.S.A.* **104** 18123-18128 (2007).

- S. Díaz, M. Cabido, Vive la différence: Plant functional diversity matters to ecosystem processes. *TREE* **16**, 646-655 (2001).
- B. J. McGill, B. J. Enquist, E. Weiher, M. Westoby, Rebuilding community ecology from functional traits. *TREE* **21**, 178-185 (2006).
- A. R. Hughes, B. D. Inouye, M. T. J. Johnson, N. Underwood, M. Vellend, Ecological consequences of genetic diversity. *Ecol. Lett.* **11**, 609-623.
- E. Garnier, J. Cortez, G. Billès, M. Navas, C. Roumet, *et al.*, Plant functional markers capture ecosystem properties during secondary succession. *Ecology* **85**, 2630-2637.
- C. H. Albert, W. Thuiller, N. G. Yoccoz, R. Douzet, S. Aubert, *et al.*, A multi-trait approach reveals the structure of intra- vs. interspecific variability in plant traits. *Funct. Ecol.* **24**, 1192-1201 (2010).
- J. Clausen, D. D. Keck, W. M. Hiesey, Regional differentiation in plant species. *Am. Nat.* **75**, 231-250 (1941).
- S. Cordell, G. Goldstein, D. Mueller-Dombois, D. Webb, P. M. Vitousek, Physiological and morphological variation in *Metrosideros polymorpha*, a dominant Hawaiian tree species, along an altitudinal gradient: The role of phenotypic plasticity. *Oecologia* **113**, 188-196 (1998).
- C. H. Albert, W. Thuiller, N. G. Yoccoz, A. Soudant, F. Boucher, *et al.*, Intraspecific functional variability: Extent, structure and sources of variation. *J. Ecol.* **98**, 604–613 (2010).
- J. D. Fridley, J. P. Grime, Community and ecosystem effects of intraspecific genetic diversity in grassland microcosms of varying species diversity. *Ecology* **91**, 2272-2283 (2010).



- G. M. Crutsinger, M. D. Collins, J. A. Fordyce, Z. Gompert, C. C. Nice, *et al.*, Plant genotypic diversity predicts community structure and governs an ecosystem process. *Science* **313**, 966-968 (2006).
- G. M. Crutsinger, L. Souza, N. J. Sanders, Intraspecific diversity and dominant genotypes resist plant invasions. *Ecol. Lett.* **11**, 16-23 (2008).
- A. Hector, The effect of diversity on productivity: Detecting the role of species complementarity. *Oikos* **82**, 597-599 (1998).
- L. W. Aarssen, High productivity in grassland ecosystems: Effected by species diversity or productive species? *Oikos* **80**, 193-184 (1997).
- M. A. Huston, Hidden treatments in ecological experiments: Re-evaluating the ecosystem function of biodiversity. *Oecologia* **110**, 449-460 (1997).
- M. Loreau, Separating sampling and other effects in biodiversity experiments. *Oikos* **82**, 600-602 (1998).
- D. A. Wardle, Experimental demonstration that plant diversity reduces invasibility – evidence of a biological mechanism or a consequence of sampling effect? *Oikos* **95**, 161-170 (2001).
- M. Loreau, A. Hector, Partitioning selection and complementarity in biodiversity experiments. *Nature* **412**, 72-76 (2001).
- D. Tilman, P. B. Reich, J. Knops, D. Wedin, T. Mielke, *et al.*, Diversity and productivity in a long-term grassland experiment. *Science* **294**, 843–845 (2001).
- C. P. H. Mulder, D. D. Uliassi, D. F. Doak, Physical stress and diversity-productivity relationships: The role of positive interactions. *Proc. Natl. Acad. Sci., U.S.A.* **98**, 6704-6708 (2001).

- J. van Ruijven, F. Berendse, Diversity-productivity relationships: Initial effects, long-term patterns, and underlying mechanisms. *Proc. Natl. Acad. Sci., U.S.A.* **102**, 695-700 (2005).
- J. Fargione, D. Tilman, R. Dybzinski, J. HilleRisLambers, C. Clark, *et al.*, From selection to complementarity: Shifts in the causes of biodiversity-productivity relationships in a long-term biodiversity experiment. *Proc. R. Soc. Lond. Ser. B* **274**, 871-876 (2007).
- J. L. Maron, M. Marler, J. N. Klironomos, C. C. Cleveland, Soil fungal pathogens and the relationship between plant diversity and productivity. *Ecol. Lett.* **14**, 36-41 (2011).
- S. A. Schnitzer, J. N. Klironomos., J. HilleRisLambers, L. L. Kinkel, P. B. Reich, *et al.*, Soil microbes drive the classic plant diversity-productivity pattern. *Ecology* **92**, 296-303 (2011).
- J. Weiner, Asymmetric competition in plant populations. *TREE* **5**, 360-364.
- J. L. Maron, M. Marler, Native plant diversity resists invasion at both low and high resource levels. *Ecology* **88**, 2651-2661.
- W. M. Ridenour, J. M. Vivanco, Y. Feng, J. Horiuchi, R. M. Callaway, No evidence for tradeoffs: *Centaurea* plants from America are better competitors and defenders than plants from the native range. *Ecol. Monogr.* **78**, 369-386.
- D. A. Wardle, R. D. Bardgett, R. M. Callaway, Wm. H. Van der Putten. Terrestrial ecosystem responses to species gains and losses. *Science* **332**, 1273-1277.
- R. W. Brooker, R. M. Callaway, L. A. Cavieres, Z. Kividze, C. J. Lortie, *et al.*, Don't diss integration: A comment on Ricklefs's disintegrating communities. *Am. Nat.* **174**, 919-927 (2009).
- M. V. Cianciaruso, M. A. Batalha, K. J. Gaston, O. L. Petchey, Including intraspecific variability in functional diversity. *Ecology* **90**, 81-89 (2009).

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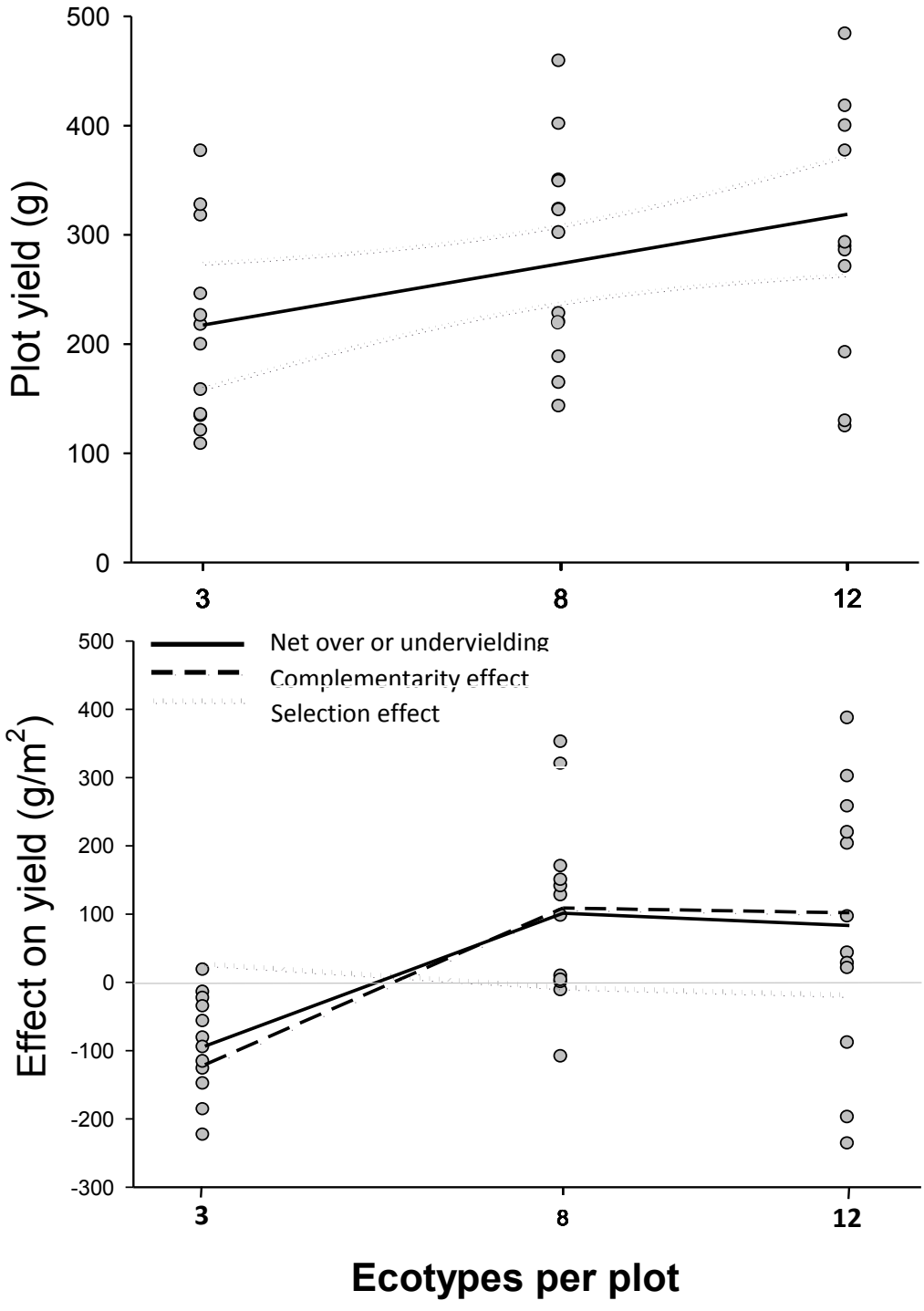


FIG. 1. Effect of ecotypic richness on total plot yield (A:  $R^2 = 0.129$ ,  $p = 0.031$ ), and on total overyielding (B: solid), complementarity (B: dashed), and selection effects (C: stippled).

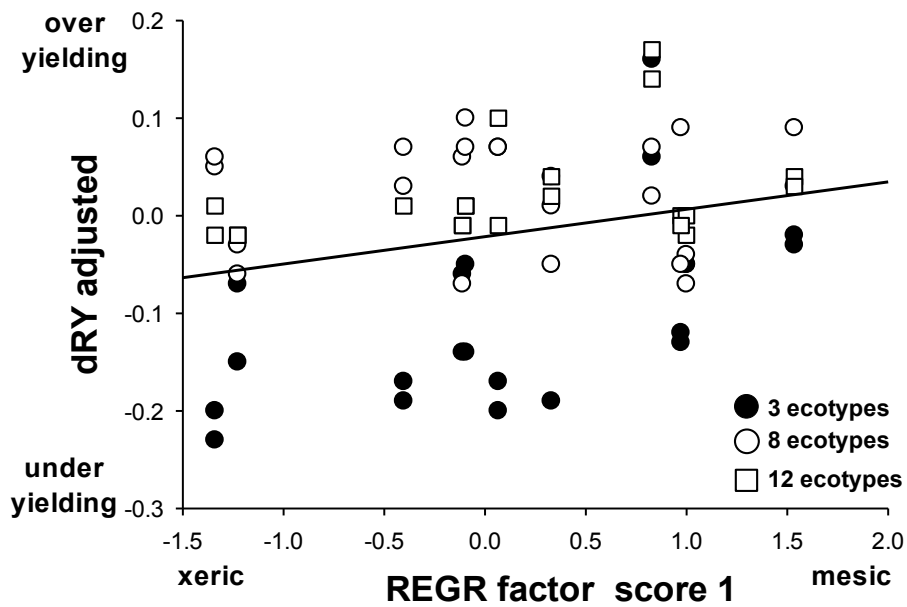


FIG. 2. Relationship between ecotype home climate and relative yield. The climate factor score CPCA1 is shown on the x-axis. Greater values indicate cooler, moister conditions. Relative yield represents the difference in performance of each ecotype relative to its monoculture yield. Symbols correspond to plots containing 3- (closed circles), 8- (open circles), and 12-ecotypes (open squares).

TABLE 1.

Term	df	Total Overyielding			Complementarity Effect			Selection Effect		
		MS	F	p	MS	F	p	MS	F	P
Intercept	1	107769	4.596	0.040	186029	6.626	0.015	10616	3.956	0.055
Richness	1	203850	8.693	0.006	318975	11.361	0.002	12833	4.782	0.036
Fungicide	1	9205	0.393	0.535	17252	0.614	0.439	1253	0.467	0.499
Rich. × Fung.	1	5338	0.228	0.637	19204	0.684	0.414	4291	1.599	0.215
Error	32	23451			28077			2684		
Total	36									

Results of linear models of total overyielding, complementarity, and selection effect ( $\text{g/m}^2$ ) against main effects and interaction effects of ecotypic richness and fungicide treatment.

#### SUPPLEMENTARY MATERIALS AND METHODS

We used ecotypes collected from 12 sites throughout the North American range of *P. spicata* (Figure S1). Seeds were field collected or acquired from true-bred lines collected and managed by the USDA Plant Germplasm Introduction and Testing Research Station in Pullman, WA, USA. The one exception was the purchase from a commercial vendor of seeds of a high-yielding wild-selected cultivar from southeastern Washington, “Goldar.” We determined latitude-longitude coordinates for each seed source and identified the nearest National Weather Service Cooperative Observer Program (COOP) climate station with similar topography to the source locality. Using information provided by the Western Regional Climate Center (<http://www.wrcc.dri.edu>), we collected data on average July maximum temperature; number of days over 90 °F per year; total spring, summer, and yearly precipitation; heating degree days

below 65 °F; and cooling degree days above 55 °F. All data were averages over the entire operation time of each station. Climate data were reduced into two factors using a principle components analysis (Table S2). Scores were not rotated and were extracted using a correlation method. Two scores were extracted according to the Kaiser criterion (retain factors with Eigenvalues > 1) but only the first factor (CPCA1) was used for analysis because the meaning of the second component axis was difficult to interpret. Increased values for CPCA1 corresponded to wetter, cooler conditions (Table S3).

Seeds from each ecotype were planted into conical starter pots and grown for two months in a greenhouse at the University of Montana. Transplants were placed into 96 cm × 64 cm common garden plots located at Fort Missoula, Missoula, MT, USA, early in the spring of 2010. Each plot contained 24 plants set in rows 16 cm apart. Adjacent plots were separated by 50 cm of bare ground. We planted plots at four levels of ecotypic richness (1, 3, 8, and 12 ecotypes per plot). Ecotypes were randomly selected for each 3- and 8-ecotype plot and planting position was randomized. We treated half of these plots with fungicide consisting of 1 g \* m<sup>-2</sup> of Cleary 3336 WP (active ingredient: thiophanate-methyl, a systematic broad-spectrum fungicide) and 117.3 µl Ridomil Gold EC (active ingredient: metalzoxyl, a soil drench preventative fungicide). Fungicide was applied as a soil drench from a pressurized sprayer. Fungicide treatment followed Maron et al. (2011), and fungicide was applied to plots after they had been wetted by rainfall or watering. Each fungicide-treated plot was sprayed in the spring during initial leaf flush and again in midsummer, in both years of the study.

For the 3-, 8-, and 12-ecotype plots, there were six replicates of the control and six of the fungicide treatment. These replicates were paired such that each fungicide treated plot corresponded to a matching control plot with the same ecotypic makeup. Because of poor

germination rates and problems with seed availability, we did not have enough seeds to plant monoculture plots for each ecotype, and we could not establish six replicates for any single ecotype. We planted monoculture plots for six of the twelve ecotypes, and had 1-2 replicates for each of these in the control and in the fungicide treatments.

Plants were grown for two full seasons (in 2010 and 2011). In the first season we watered as necessary to reduce transplant stress. In the second season, however, plots only received natural precipitation. We estimate that common garden conditions in 2011, when most growth occurred, represented a climate intermediate in conditions relative the climatic ranges of the ecotypes, but slightly more mesic than average (estimated factor score = 0.403, Table S2).

In the fall of 2011 we harvested, dried, and weighed the aboveground biomass from each plot. Immediately after harvest three soil cores were taken from each plot at a depth of 8 cm, and pooled. Cores were sieved to 2 mm and extracted overnight in 2 M KCl buffer to remove free nitrate and ammonium. Samples were analyzed for ammonium and nitrate concentration colorimetrically at the University of Montana Soil Biogeochemistry Lab. We obtained wet and dry mass of the soil to determine water content and calculate nitrogen concentrations on a dry weight basis.

We estimated diversity-dependent overyielding and partitioned the influences of selection and complementarity effects on net overyielding, after Loreau and Hector (2001). This method required us to measure monoculture yields for all ecotypes. For reasons described above, we only measured monoculture yields for 6 of the 12 the ecotypes used in this study. To address this limitation we followed two different approaches. In the first approach, we restricted initial analyses to the six ecotypes for which we had monoculture yields. In the second approach, we estimated monoculture yields for 11 ecotypes using projected linear estimates of yield as a



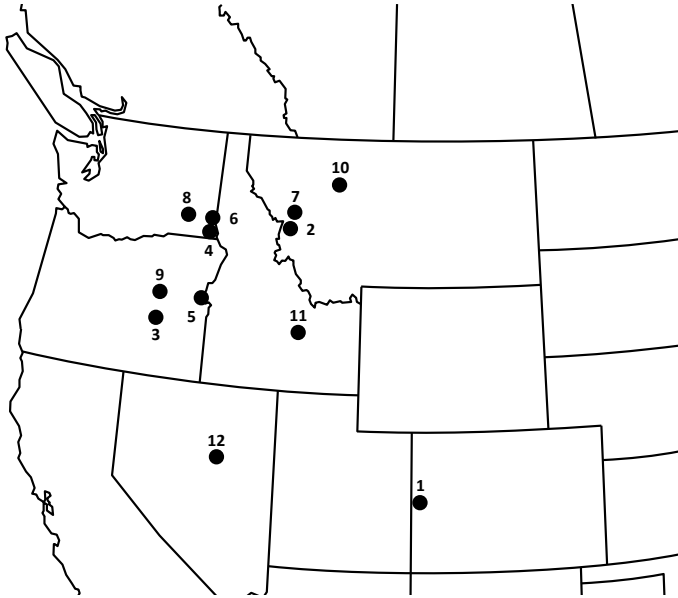
function of richness. Briefly, we calculated least-squares best fit lines of yield against richness using data from the 3-, 8-, and 12-ecotype plots for each ecotype. We then extrapolated an estimate of the yield at richness = 1 for each ecotype other than ecotype 5, which we did not attempt to estimate monoculture yield for because it was not present in any 3-ecotype plots. Because all six ecotypes that were grown in monoculture under-yielded in the 3-ecotype treatment, this approach consistently underestimated their monoculture yields for those six ecotypes. To account for this, we adjusted our extrapolated estimate so that the average projected yield for these six ecotypes matched their average actual yield. We applied this correction to all ecotypes by adding 70 g to the monoculture yield estimate for each ecotype. This approach was conservative, because it had the effect of making positive complementarity and selection effects harder to detect. Both approaches yielded quantitatively and qualitatively similar results. Results based on estimated monoculture yield are reported in the manuscript, and results of analyses restricted to ecotypes with measured monoculture yields are presented in the supplementary materials (Table S4).

In order to determine whether ecotypes responded consistently to increased richness, we used a general linear model with natural log-transformed average ecotype biomass in a plot as a response variable, ecotypic richness as a covariate, fungicide treatment as a fixed factor, and ecotype identity as a random factor (Table S1). All two-way interaction terms were also included. We also used general linear models to estimate the effects of ecotypic richness and treatment on total overyielding, complementarity effect, and selection effect. Ecotypic richness was used as a covariate, and fungicide treatment was used as a fixed factor. An interaction term between richness and treatment was also included. We ran models both using estimates based on projected monoculture yields for 11 ecotypes (Table 1) and on actual monoculture yields for 6

ecotypes (Table S4). To determine how richness, climate, and monoculture yield affected overyielding, we used a general linear model with relative overyielding ( $\Delta RY$ ) as a response variable, and richness, CPCA1, and estimated monoculture yield as covariates. For replicates we used mean ecotype yield in each plot ( $n = 254$ ). To evaluate how soil nitrogen correlated with overyielding, we used Pearson correlations. Select correlations between complementarity effect, ecotypic richness, and N-normalized plant yield were evaluated (Fig. 3). We define N-normalized plant yield as total plot yield divided by soil available nitrogen.

SUPPLEMENTARY FIGURES

FIG. S1



Map of collection locations for the ecotypes used in this study.

SUPPLEMENTARY TABLES

TABLE S1

	df	MS	F	p
Intercept	1	47.248	21.201	0.001
Richness	1	8.282	13.261	< 0.001
Fungicide	1	0.157	0.254	0.615
Ecotype	11	2.501	4.053	< 0.001
Richness × Fungicide	1	0.343	0.549	0.459
Richness × Ecotype	11	1.376	2.203	0.015
Fungicide × Ecotype	11	0.547	0.876	0.565

Results of a linear model of natural log-transformed per-plant yield against ecotypic richness, fungicide treatment, and ecotypic identity.

TABLE S2

Ecotype	Lat	Long	Weather Station	July Max. (°C)	Days over 32.2 °C	Spring Precip	Summer Precip	Total Precip	HDD	CDD	CPCA 1	CPCA 2
1	39.05	-108.70	Colorado NM 051772	33.4	51.2	74	71	282	5671	2516	-1.227	0.536
2	46.51	-114.08	Stephensville 247894	29.6	20.7	79	86	316	7508	975	0.329	-1.016
3	43.67	-118.98	Burns 351176	29.6	21.0	66	41	294	7140	1200	-0.111	-1.138
4			Anatone 2S 450184	26.9	9.2	148	92	547	7506	870	1.535	0.878
5	44.35	-117.27	Huntington 354098	34.5	62.7	76	40	311	5631	2514	-1.553	0.707
6	46.60	-117.17	Pullman Exp Stn 456784	27.9	13.4	129	60	505	6614	1263	0.829	0.953
7	46.87	-113.95	Missoula 2 NE 245735	28.6	19.5	126	120	433	7443	1128	0.999	0.367
8	46.63	-118.20	Little Goose Dam 454702	32.6	46.5	65	40	289	4825	2309	-1.340	0.585
9	44.40	-118.95	Canyon City 351352	30.9	34.5	125	54	394	5810	1472	-0.096	0.959
10	47.48	-111.53	Great Falls Airpo 243751	28.6	18.7	124	136	379	7685	1277	0.973	0.089
11	43.64	-113.40	Arco 3 SW 100375 Pine Valley Baily Ranch	30.2	19.1	66	58	241	8426	1082	0.066	-1.979
12	40.00	-116.12	266242	32.6	48.0	98	43	272	7564	944	-0.403	-0.940
Garden 2010			Missoula Airport	84	14	84	95	363	7565	1108	0.998	-0.007
Garden 2011			Missoula Airport	85	26	98	150	418	7342	1084	0.403	-0.614

Ecotype locality data, including latitude and longitude, location of nearest COOP station, average July maximum temperature, number of days per year over 32.2 °C, spring precipitation, summer precipitation, yearly total precipitation, heating degree days below 65 °F (HDD), cooling degree days above 55 °F (CDD), and climate factor scores (CPCA1 and CPCA2). Precise coordinates of the locality for Ecotype 4 (Goldar) are unknown, but the collection is known to be from Asotic County, WA, USA. Common garden conditions are also shown for both years of the study (Garden 2010 and Garden 2011).

TABLE S3

	CPCA1	CPCA2
Average July Max. Temp.	-0.961	-0.003
Yearly Days Over 32.2 °C	-0.918	0.176
Spring Precipitation	0.764	0.549
Summer Precipitation	0.694	0.131
Yearly Precipitation	0.731	0.639
Heating Degree Days (65 °F)	0.698	-0.652
Cooling Degree Days (55 °F)	-0.825	0.441
Eigenvalue / Final Loading	4.531	1.377
% Variance Explained	65%	20%

Results of principal components analysis for climate variables.

TABLE S4

Term	df	Total Overyielding			Complementarity Effect			Selection Effect		
		MS	F	p	MS	F	p	MS	F	p
Intercept	1	88207	2.362	0.135	153960	2.887	0.100	9096	0.697	0.410
Richness	1	174949	4.684	0.039	263161	4.934	0.034	8972	0.688	0.413
Fungicide	1	13382	0.358	0.554	13813	0.259	0.615	3	0.000	0.987
Rich. × Fung.	1	8079	0.216	0.645	22231	0.417	0.523	3507	0.269	0.608
Error	32	37349			53332			13044		
Total	36									

Results of linear models of total overyielding, complementarity, and selection effect ( $\text{g/m}^2$ ) against main effects and interaction effects of ecotypic richness and fungicide treatment, using data only for the six ecotypes with known monoculture yield (compare to Table 1).

## CHAPTER 4

### EXTENDED ECOLOGICAL CONSEQUENCES OF INVADER-DRIVEN SELECTION

*Abstract:* Invasive plant species can drastically decrease the abundances of native plant species and in the process act as an ecological filter that selects for the survival of individuals that compete better with the invader. However, increased competitive ability in general is thought to incur costs or tradeoffs in performance in other situations, such as the ability to tolerate stress or defend against herbivores. Thus selection for the ability to compete with an invader may have “extended consequences” for other ecological traits. However, to my knowledge no studies have explored the extended consequences of invader-driven selection. I compared the performance of offspring of two native plant species and one exotic species collected from outside patches of the invasive forb *Euphorbia esula* to that of offspring from individuals collected inside *Euphorbia* patches. Individuals of the exotic annual *Bromus japonicus* inside *Euphorbia* patches allocated more resources to roots compared to individuals outside *Euphorbia* patches over a wide range of treatments. For the native perennial bunchgrass *Poa secunda*, the offspring of individuals growing inside *Euphorbia* patches showed lower plasticity in root allocation than those collected from outside *Euphorbia* patches. For the native perennial bunchgrass *Pseudoroegneria spicata*, we found no evidence that survivors of *Euphorbia* invasion differed in a heritable manner from populations that had not experienced invasion. Provided that *Euphorbia* was the agent of selection that produced these differences, and that they are not due to some other process such as selection by another agent or maternal effects, these results suggest that selection caused by an

invader may have consequences for how native populations decimated by invasions respond in specific ways to the invader, but also to other aspects of their biotic and abiotic environment.

*Key Words:* competition, drought, ecological-evolutionary feedback, geographic mosaic, herbivory, invasion, leafy spurge, natural selection, resources, traits

## INTRODUCTION

Exotic plant species often cause severe local losses in native species abundance and diversity (Ridenour and Callaway 2001; Braithwaite et al. 1989; Memmott et al. 2000; Grigulis et al. 2001; Maron and Marler 2008a; Hejda et al. 2009; Vilá et al. 2011) and reduced growth and reproduction of native survivors (Gordon 1998; Maron and Marler 2008b, Atwater et al. 2011). Correspondingly, there is broad evidence that mortality caused by invasive plants acts as a filter to select for increased competitive ability in the surviving individuals in native plant populations (Callaway et al. 2005, Lau 2006, Meador and Hild 2007, Cipollini and Hurley 2008, Leger 2008, Ferrero-Serrano et al. 2010, Rowe and Leger 2011, Goergen et al. 2011). However, only two studies have also investigated whether such disproportionate survival affects native plants in ways other than how they compete with the invader. Lau (2006) found that *Medicago sativa* selected for individuals of the native *Lotus wrangelianus* that were more susceptible to a non-native herbivore. Ferrero-Serrano et al. (2010) found that *Hesperostipa comata* and *Stipa airoides* individuals that survived *Acroptilon repens* invasion were better competitors against a different invasive species than those that did not experience invasion, but they did not measure whether these plants were also better competitors against *Acroptilon*. These studies raise the



intriguing possibility that invader-imposed selection may affect aspects of the ecology of natives other than the ability to compete with the invader.

Ecological theory also provides a strong context for why selection for increased competitive ability might have extended consequences for the native genotypes that survive invasion. The ability of plants to compete with their neighbors is thought to be constrained by fundamental “compromises between the conflicting selection pressures resulting from particular combinations of competition, stress, and disturbance” (Grime 1977). In other words, being a strong competitor may reduce a species’ ability to colonize new ground, tolerate abiotic stress (Grime 1977), or to defend itself against herbivores (Herms and Mattson 1992). Furthermore, for reasons that are poorly understood, in some cases species appear to specialize in competition against certain species or genotypes at the expense of their ability to compete against other species or genotypes (Taylor and Aarssen 1990, Fridley et al. 2007). For these reasons, selection for greater competitive ability in a native species might affect how that species interacts with other native or invasive competitors, responds to herbivory, and responds to variation in its abiotic environment. I refer to these ramifications of invader-imposed selection on natives as “extended consequences” of selection by invaders.

Here I compare the performance between offspring of grasses growing inside and outside of patches of the high-impact invader, *Euphorbia esula* L. (leafy spurge) and investigate whether offspring of adults that have survived *Euphorbia* invasion differ in their ability to 1) compete with *Euphorbia*; 2) compete with a native grass, *Festuca idahoensis*; 3) tolerate drought; 4) respond to fertilization; and 5) tolerate clipping. I chose these treatments because they represent a variety of conditions theorized to involve tradeoffs with competitive ability. I used two native grasses, *Pseudoroegneria spicata* (Pursh) A. Löve (bluebunch wheatgrass) and *Poa secunda* J.

Presl. (Sandberg's bluegrass), and one exotic grass, *Bromus japonicus* Thunb. (Japanese brome), as focal species. If the offspring of individuals that survive invasion differ in traits other than the ability to compete with *Euphorbia*, and in the context of alternative interpretations discussed below, then selection by *Euphorbia* may have broad consequences for the ecology and evolution of other species.

## METHODS

Study Species: *Euphorbia esula* is a clonal forb native to Eurasia, but highly invasive in the Northern Great Plains and Intermountain West. Through shading, root competition, nutrient sequestration, allelopathy, toxicity to native grazers, and a vigorous vegetative reproductive habit, *Euphorbia* is a strong invader *sensu* Ortega and Pearson (2005) and an exceptionally aggressive competitor (Dittberner *et al.* 1983; Cyr and Bewley 1989; Trammell and Butler 1995) that reduces native diversity in many different community types (Belcher and Wilson 1989). *Euphorbia* forms clonal patches with discrete boundaries, owing to a primarily vegetative reproductive habit (Best *et al.* 1980).

I tested the differences between grasses that survived *Euphorbia* invasion and grasses from uninvaded communities for three species: two native grasses (*Pseudoroegneria spicata* and *Poa secunda*) and one exotic grass (*Bromus japonicus*). *Pseudoroegneria* is a large, long-lived, dominant native perennial bunchgrass that reproduces sexually and is common throughout the semi-arid grasslands of the Western United States where it is a valuable rangeland species. It persists at low abundance in mono-dominant *Euphorbia* stands (Atwater *et al.* 2011). *Poa secunda* is a small native bunchgrass common in the prairies of western Montana that can reproduce sexually but that is facultatively apomictic (Kellogg 1987). It persists within

*Euphorbia* patches and can flower and set seed even in dense patches (D.Z. Atwater, *pers. obs.*). *Bromus japonicus* is a small exotic annual grass that grows in interstitial spaces in bunchgrass communities and also grows relatively well within *Euphorbia* stands (D. Z. Atwater, *pers. obs.*). *Bromus* is almost exclusively self-fertile with very high homozygosity (Oja and Jaaska 1998). One experimental treatment was competition with *Festuca idahoensis*. *Festuca* is a small but very common and widespread native cool-season bunchgrass that co-occurs with *Pseudoroegneria* at my study sites and throughout the intermountain grasslands of Montana (Atwater et al. 2011).

Experimental Design: I collected seeds of the three target species from inside and outside of *Euphorbia* patches at 7 sites in the Missoula Valley in Western Montana (Table 1). No site had a recent history of herbicide use or grazing (M. Valliant, B. Lindler, G. Thelen, *pers. comm.*). Although other invasive exotic plants were present at all of the study sites, we selected patches with minimal colonization of exotics other than *Euphorbia esula* and *Bromus japonicus*. Seeds were collected in 2008 and 2010 and were used in experiments designed to compare the abilities of progeny to compete with *Euphorbia* and respond to other experimental conditions.

In 2008 I collected seeds of *Pseudoroegneria*, *Poa*, and *Bromus* from five sites (Table 1). Each site was 1-5 km from the nearest other site. At each site I identified a large (approximately 0.25 ha) patch of *Euphorbia* and haphazardly collected seeds from 25 individuals of each species inside and > 25 plants of each species immediately outside of each *Euphorbia* patch. Seeds of each species from each environment were pooled separately for each site. I applied five treatments to seedlings of these species, but each species did not receive all treatments (Table 2). These treatments were the control, competition with *Euphorbia*, competition with *Festuca*,

drought, and clipping. Seeds were planted in 550 mL pots containing a 50/50 mix of sand and locally collected native soil and thinned to one seedling per plot three weeks after planting. In the control treatment, individuals of each species were grown alone ( $n = 7-11$  for each habitat-treatment-species combination). I administered the drought treatment by watering once per week, instead of three times per week as in the other treatments. This allowed the upper portion of the soil to dry to the touch between watering events. For the *Festuca* competition treatment, I planted *Festuca* seeds in each pot. Three weeks later, I thinned them to one seedling and then added seeds of the target plants. For the *Euphorbia* competition experiment, rhizomes from a *Euphorbia* stand, different from the stands from which seeds were collected, were collected from a site several kilometers from any sites where seed was collected. These rhizomes were cut into 3-4 cm pieces and transplanted these into pots where they vegetatively produced shoots. Four weeks after planting rhizomes I added seeds of *Pseudoroegneria*, *Poa*, and *Bromus*. In the treatment designed to mimic tolerance to herbivory (Newingham et al. 2005), after thinning to one individual per plot, all leaf tissue above 1 cm was clipped with scissors immediately after thinning, and one month later leaf tissue was removed again in the same manner. After four months I harvested all plants, dried roots and shoots separately at 60°C, and weighed them. In 2010, I used all of the 2008 sites and also sampled at two new sites (Table 1). I collected seeds for *Pseudoroegneria* as in 2008, but I did not collect seeds of *Poa* in 2010. For *Bromus* I collected entire adult plants at the same time I collected their seeds, and weighed shoot and seed mass. I controlled for genotype by assigning full siblings to each treatment. Because *Bromus* is almost entirely self-pollinated and has high homozygosity (Oja and Jaaska 1998), full siblings of *Bromus* are genetically nearly identical to their parents. Seedlings grown from this field-collected seed of both *Bromus* and *Pseudoroegneria* collected in 2010 were subjected to five

treatments (each treatment,  $n = 10$ ): control, competition with *Euphorbia*, fertilization, competition with *Festuca*, and clipping. In this experiment *Festuca* seeds were planted at the same time as *Pseudoroegneria* and *Bromus*. For the fertilizer treatment, each pot received 0.136 g of MiracleGro in a 100 mL solution immediately after thinning.

Data Analysis: I used a variance partitioning approach to investigate whether invasion status (i.e. seeds collected inside or outside of *Euphorbia* patches) had significant effects on performance, and to identify whether the extended consequences of selection by *Euphorbia* were consistent across treatments. A significant main effect of invasion status was interpreted as evidence for selection by *Euphorbia*. A significant invasion status  $\times$  treatment interaction indicated that selection by *Euphorbia* affected plants differently in different treatments. For all data, I used ANOVA with site as a random factor, and invasion status (inside or outside of *Euphorbia* patch), and treatment as fixed factors. I modeled the main effects for each factor, plus all second order interactions. Because the full factorial models contained a large number of interaction terms, which can cause problems of over-fitting, I decided *a priori* to use a stepwise removal approach to arrive at each final, reported model. Briefly, I began with a full factorial model containing all main effects and possible second order terms. Next, the second-order factor with the lowest  $P$ -value was removed. When all second order factors were  $P \leq 0.100$ , no more factors were removed. Main effects were retained even if they were not significant. Such an approach does not always arrive at the best-fitting model, but it is an efficient way to pare poorly fitting terms (Kadane & Lazar 2004, Whittingham *et al.* 2006). Because I controlled for *Bromus* genotype in 2010, the 2010 data for *Bromus* could have been analyzed using treatment as a within-subject factor. However, due to low seed availability, only about half of the families produced

successfully germinated *Bromus* seedlings in all treatments. Because this halved my sample size when treatment was modeled as a within-subject factor, I chose not to model treatment as a within-subject factor, instead treating it as a typical fixed factor. This enabled me to use all of my data despite missing variables.

To determine whether genetic constraints influenced how individual *Bromus* genotypes responded to different treatments, I used a principal components analysis of performance traits for *Bromus* siblings growing in each treatment. I used a “varimax” rotation to identify orthogonal factors and performed the analysis on correlation matrices. The Kaiser criterion (retaining factors with initial Eigenvalues > 1) was used for factor selection. Missing values were filled with within-treatment means. If responses to different treatments cluster into the same component factors, it suggests the presence of underlying genetic correlations that simultaneously influence the performance of *Bromus* in multiple treatments. Thus this analysis allowed investigation of correlations in performance among treatments and whether these correlations had a genetic basis. This analysis was available only for *Bromus* because *Bromus* was the only species for which I subjected siblings to each treatment. Because *Bromus* did not differ in performance with respect to invasion status in 2010, I decided *post hoc* to include all *Bromus* irrespective of invasion status in the PCA model. I performed this and all other analyses using the PASW Statistics version 18.0.0 GradPack (SPSS, IBM Software, July 30, 2009).

## RESULTS

In 2008, two species (*Poa secunda* and *Bromus japonicus*) performed differently depending upon whether they were collected inside or outside of patches of *Euphorbia esula*. More specifically, in 2008, *Poa* plants produced from seed collected from *Euphorbia* patches

showed treatment-specific changes in root mass in comparison to conspecifics collected outside *Euphorbia* patches. In the control and drought treatments, *Poa* from inside *Euphorbia* patches had a greater root mass ratio (RMR), but in competition with *Euphorbia* they had a lower RMR. Thus there was no main effect of *Euphorbia* invasion status on *Poa* RMR ( $p = 0.353$ ), but there was a significant treatment  $\times$  invasion status interaction ( $p = 0.027$ , Table 3). Conversely, for *Bromus* in 2008, there was a significant main effect of invasion status on RMR ( $p = 0.042$ ) and a strong trend for root mass ( $p = 0.062$ ), but no significant interaction between treatment and invasion status (Table 3). In other words, *Bromus* from inside of *Euphorbia* patches demonstrated increased root investment in the control, drought, *Euphorbia* competition, *Festuca* competition, and artificial herbivory treatments, compared to *Bromus* produced from uninvaded communities. For both *Bromus* and *Poa* in 2008, the performance of greenhouse-reared seedlings also differed depending upon which site seeds were collected from (significant site or site  $\times$  treatment interaction, Table 3). I was unable to analyze *Pseudoroegneria* in 2008 due to low sample sizes caused by poor germination, and in 2010, I found no differences in the performance of either *Pseudoroegneria* or *Bromus* seedlings depending upon whether they were collected inside or outside of *Euphorbia* patches despite adequate germination rates for both species (Table 4), although performance again varied significantly among sites ( $p < 0.050$ ).

Principal Components Analysis (PCA) on *Bromus* performance data collected in 2010 produced five factors that met my retention criterion (Eigenvalues  $> 1$ ), an outcome supported by a scree plot of the Eigenvalues. Once rotated, all factors contributed between 17.3% and 19.4% to the total variance explained by the analysis, and together they explained 92.6% of the variance present in the initial ten variables. In the rotated factor solution, root and shoot masses of *Bromus* in each treatment loaded strongly into a single factor score representing primarily that

treatment and with virtually no loading into other scores (Table 5). Thus, performance in any given treatment was almost completely independent of performance in other treatments. In other words, genotypes with large roots in a given treatment also produced large shoots in that treatment, but performance in that treatment did not correlate with performance in any other treatment.

## DISCUSSION

My results indicated that heritable changes in the performance of the offspring of a native species and an exotic invasive species have occurred during *Euphorbia* invasion. Such changes have been typically interpreted as evidence of selection caused by invasive plants (see Callaway et al. 2005, Lau 2006, Meador and Hild 2007, Cipollini and Hurley 2008, Leger 2008, Ferrero-Serrano et al. 2010, Rowe and Leger 2011, Goergen et al. 2011), particularly when the offspring of plants in invaded communities compete better with the invader than those from uninvaded communities. However, selection by invasive plants is not the only process that could result in these differences in performance. For example, pre-existing micro-site conditions may have constrained the distribution of the invader and simultaneously caused selection in *Bromus* and *Poa*. This concern is common to most, if not all, studies that have investigated selection caused by invasive plants (Callaway et al. 2005, Lau 2006, Meador and Hild 2006, Meador and Hild 2007, Cipollini and Hurley 2008, Leger 2008, Ferrero-Serrano et al. 2010, Rowe and Leger 2011, Goergen et al. 2011), because all of these studies have used naturally, rather than experimentally, invaded communities. Importantly, *Euphorbia* reproduces vegetatively creating very distinct patch boundaries and all *Euphorbia* patches in the study area have been present for less than 20 years. Furthermore, all un-invaded native communities sampled in 2008 were



colonized by *Euphorbia* by 2010, confirming that *Euphorbia* was not limited by micro-site conditions at these study sites. This makes it less likely that micro-site variation confounded the selective effects of *Euphorbia*.

If the differences between conspecific individuals reported here are caused by *Euphorbia* and not microsite conditions, these differences may still not be due to selective mortality on certain genotypes, but instead to maternal effects. For example, poor conditions in dense *Euphorbia* patches may result in reduced maternal investment in seed production, resulting in smaller seeds being produced by plants in *Euphorbia* patches. Although we did not observe differences in seed mass for the progeny of *Poa* and *Bromus* collected inside and outside *Euphorbia* patches in 2008 (T-test;  $t_4 = 0.457$ ,  $P = 0.672$ ;  $t_4 = 0.631$ ,  $P = 0.563$ ), when selection was observed, epigenetic and transgeneration inheritance can occur in ways that are not reflected in seed mass and that we could not measure in this study. Such effects can involve transmission of cytoplasmic genetic information, differential partitioning of endosperm resources, and environmental effects that alter seed morphology and energy storage, and even DNA and chromosome modification by either parent plant (Roach and Wulff 1987, Rapp and Wendel 2005).

I hypothesized that *Euphorbia* would select for increased competitive ability in the offspring of individual native and exotic grasses that survived invasion, and that extended consequences of selection by *Euphorbia* would reflect tradeoffs between competitive ability, response to abiotic stress, and tolerance to herbivores (see Grime 1977, Herms and Mattson 1992), indicated by a significant interaction between treatment and invasion status (i.e. whether the seedling came from a *Euphorbia*-invaded community or not). For example, selection might favor competitive performance against *Euphorbia* but also lead to a decline in tolerance to

drought or herbivory. For *Poa*, there was a significant treatment  $\times$  invasion status interaction for root mass ratio, although not one that was consistent with the hypothesized predictions.

Specifically, in the competition and drought treatments, seedlings from *Euphorbia*-invaded patches had lower root mass ratio than those from un-invaded communities, but root allocation did not vary in the *Euphorbia* competition treatments (Fig. 1B). As a result, root investment for *Poa* that survived invasion was much more uniform across treatments than root investment for *Poa* from uninvaded communities. One possible explanation for this pattern is that *Euphorbia* selected for variants of *Poa* with lower plastic responses to drought and competition.

For *Bromus*, on the other hand, invasion status was only important as a main effect, meaning that selection by *Euphorbia* had similar effects across all of the different treatments I applied. Thus, my results suggest the possibility that selection by *Euphorbia* has extended consequences, in the sense that it affects how plants respond to different experimental treatments (Fig. 1D). However, the performance of *Poa* and *Bromus* in these experiments did not clearly indicate that the hypothesized ecological or physiological tradeoffs mediate these responses.

Tradeoff theory concerning competitive ability is based on the notion that genetic and physiological constraints prevent a single species or genotype from outperforming all other species in a community under all conditions. In addition to influencing mean performance in different conditions, such constraints should also result in covariance in the performance of species or genotypes grown under different conditions. For example, if underlying genetic or physiological constraints prevent plants from competing well with *Euphorbia* and also responding well to clipping, this would cause negative covariance in performance in the clipping and *Euphorbia* competition treatments. Selection caused by a competitor would therefore be expected to increase competitive ability but a concomitant decrease in tolerance of herbivory. In

this context the PCA analysis I conducted on the performance of *Bromus* siblings collected in 2010 indicated almost total independence in the expression of traits among treatments. In other words, families that performed well in a given treatment were no more or no less likely to perform well in any other treatment. Thus in the common greenhouse conditions in this study, the performance of families across the treatments used in this study was not inherently constrained by performance tradeoffs among the various environments that were tested. Also, the PCA results suggest that selection for increased performance in the *Euphorbia* competition treatment should not affect performance in the other treatments. However, in 2008 *Bromus* that survived *Euphorbia* invasion showed increased root growth in all treatments. If the genetic structure of the *Bromus* population was similar in 2008 and 2010, I would have expected to see changes in root allocation only in the *Euphorbia* competition treatment. This result suggests either 1) that greenhouse did not effectively mimic natural conditions, or 2) that in nature *Euphorbia* causes selection on multiple independent traits that not directly related to competitive ability. This could occur, for example, if *Euphorbia* simultaneously competes with associated plants, influences herbivore attacks (Atwater *et al.* 2011), and changes their edaphic environment. Thus, “extended” evolutionary consequences of invasion might not be influenced by tradeoffs at all, but instead respond to a complex suite of environmental changes created by an invader that can affect multiple independent traits. It is important to note that this interpretation is based on a PCA analysis performed only on *Bromus* plants collected in 2010 when I did not detect evidence of selection caused by *Euphorbia*, and thus should be considered with caution.

In sum, found evidence for a response to selection by *Euphorbia esula* for two species. To my knowledge, this is the first evidence that an exotic invasive plant species has selected for

increased competitive ability in another invasive species. These results also demonstrate limited extended consequences, including how native and invasive species may respond to competition with other native species, to herbivory, and to altered resource availability. These results suggest that the evolutionary consequences of plant invasions could have subtle and complex effects on the ecology of native plants, beyond simply affecting their ability to compete with the invader causing selection. Moreover, these “extended” consequences of invasion may not necessarily follow classic tradeoffs such as the CSR tradeoffs (Grime 1977) or “grow or defend” tradeoffs (Herms and Mattson 1992). Instead, invader-driven selection appears to affect both native and invasive plants in complex and unintuitive ways. Understanding the extended consequences of selection may therefore be crucial for our understanding of the long term consequences of plant invasions, and of the evolutionary consequences of plant interactions in general

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#### LITERATURE CITED

- Atwater, D. Z., C. M. Bauer and R. M. Callaway. 2011. Indirect positive effects ameliorate strong negative effects of *Euphorbia esula* on a native plant. *Plant Ecology* 212:1655-1662.
- Belcher, J. W. and S. D. Wilson. 1989. Leafy spurge and the species composition of a mixed-grass prairie. *Journal of Range Management* 42:172-175.

- Best K.F., G. G. Bowes, A. G. Thomas and M. G. Maw. 1980. The biology of Canadian weeds  
.39. *Euphorbia esula* L. Canadian Journal of Plant Science 60:651-663.
- Braithwaite, R. W., W. M. Lonsdale and J. A. Estbergs. 1989. Alien vegetation and native biota  
in tropical Australia: The spread and impact of *Mimosa pigra*. Biological Conservation  
48:189-201.
- Callaway, R. M., W. M. Ridenour, T. Laboski, T. Weir and J. M. Vivanco JM. 2005. Natural  
selection for resistance to the allelopathic effects of invasive plants. Journal of Ecology  
93:576-583.
- Cipollini, D., and K. A. Hurley. 2008. Variation in resistance of experienced and naïve seedlings  
of jewelweed (*Impatiens capensis*) to invasive garlic mustard (*Alliaria petiolata*). Ohio  
Journal of Science 108:47-49.
- Cyr, D. R. and J. D. Bewley. 1989. Carbon and nitrogen reserves of leafy spurge (*Euphorbia  
esula*) roots as related to overwintering strategy. 77:67-72.
- Dittberner, P. L. and M. R. Olson MR. 1983. The plant information network (PIN) data base:  
Colorado, Montana, North Dakota, Utah, and Wyoming. FWS/OBS-83/86. Washington,  
DC: U.S. Department of the Interior, Fish and Wildlife Service. pp. 786.
- Ferrero-Serrano A, A. Hild and B. Meador. 2010. Can invasive species enhance competitive  
ability and restoration potential in native grass populations? Restoration Ecology 19:545-  
551.
- Fridley, J. D., J. P. Grime and M. Bilton M. 2007. Genetic identity of interspecific neighbours  
mediates plant responses to competition and environmental variation in a species-rich  
grassland. Journal of Ecology 95:908-915.

- Goergen, E. M., E. A. Leger and E. K. Espeland. 2011. Native perennial grasses show evolutionary response to *Bromus tectorum* (cheatgrass) invasion. PLoS ONE 6:e18145.
- Gordon, D. 1998. Effects of invasive, non-indigenous plant species on ecosystem processes: Lessons from Florida. *Ecological Applications* 8:975-989.
- Grigulis, K., A. W. Sheppard, J. E. Ash and R. H. Groves. 2001. The comparative demography of the pasture weed *Echium plantagineum* between its native and invaded ranges. *Journal of Applied Ecology* 38:281-290.
- Grime, J. P. 1977. Evidence for the existence of three primary strategies in plants and its relevance for ecological and evolutionary theory. *American Naturalist* 111:1169-1194.
- Heijda M., P. Pyšek and V. Jarošík. 2009. Impact of invasive plants on the species richness, diversity and composition of invaded communities. *Journal of Ecology* 97:393-403.
- Hermes, D. A. and W. J. Mattson. 1992. The dilemma of plants: to grow or defend. *The Quarterly Review of Biology* 67:283-335.
- Kadane J. B. and N. A. Lazar. 2004. Methods and criteria for model selection. *Journal of the American Statistical Association* 99:279-290.
- Kellogg E. A. 1987. Apomixis in the *Poa secunda* complex. *American Journal of Botany* 74:1431-1437.
- Lau, J.A. 2006. Evolutionary responses of native plants to novel community members. *Evolution* 60:56-63.
- Leger, E. A. 2008. The adaptive value of remnant native plants in invaded communities: an example from the great basin. *Ecological Applications* 18:1226-1235.
- Maron J. L. and M. Marler. 2008a. Effects of native species diversity and resource additions on invader impact. *American Naturalist* 172:S18-S33.

- Maron J. L. and M. Marler. 2008b. Field-based competitive impacts between invaders and natives at varying resource supply. *Journal of Ecology* 96:1187-1197.
- Mealor BA and Hild AL. 2006. Potential selection in native grass populations by exotic invasion. *Molecular Ecology* 15:2291-2300.
- Mealor, B. A. and A. L. Hild. 2007. Post-invasion evolution of native plant populations: a test of biological resilience. *Oikos* 116:1493-1500.
- Memmott, J., S. V. Simon, Q. Paynter, A. W. Sheppard and P Syrett. 2000. The invertebrate fauna on broom, *Cystus scoparius*, in two native and two exotic habitats. *Acta Oecologia* 21:213-222.
- Newingham, B. A., G. Boquien, P. Choler and R. M. Callaway. 2005. Effects of *Festuca paniculata* on the compensatory growth response of *Centaurea uniflora* in the French Alps. *Plant Ecology* 176:237-244.
- Oja T. and V. Jaaska. 1998. Allozyme diversity and phylogenetic relationships among diploid annual bromes (*Bromus*, Poaceae). *Annales Botanici Fennici* 35:123-130.
- Ortega, Y. K., and D. E. Pearson. 2005. Weak vs. strong invaders of natural plant communities: assessing invasibility and impact. *Ecological Applications* 15:651-661.
- Rapp R. A. and F. Wendel. 2005. Epigenetics and plant evolution. *New Phytologist* 168:81-91.
- Ridenour, W. M. and R. M. Callaway. 2001. The relative importance of allelopathy in interference: the effects of an invasive weed on a native bunchgrass. *Oecologia* 126:444-450.
- Roach D. A. and R. D. Wulff. 1987. Maternal effects in plants. *Annual Review of Ecology and Systematics* 18:209-235.

- Rowe, C. L. J. and E. A. Leger. 2011. Competitive seedlings and inherited traits: a test of rapid evolution of *Elymus multisetus* (big squirreltail) in response to cheatgrass invasion. *Evolutionary Applications* 4:485-498.
- Taylor D. R. and Aarssen L. W. 1990. Complex competitive relationships among genotypes of three perennial grasses: implications for species coexistence. *American Naturalist* 136:305-327.
- Trammell, M.A. and J. L. Butler. 1995. Effects of exotic plants on native ungulate use of habitat. *Journal of Wildlife Management* 59:808-816.
- Vilá, M., J. L. Espinar, M. Hejda, P. E. Hulme, V. Jaršík, J. L. Maron, J. Pergl, U. Schaffner, Y. Sun and P. Pyšek. 2011. Ecological impacts of invasive alien plants: a meta-analysis of their effects on species, communities and ecosystems. *Ecology Letters* 14:702-708.
- Whittingham M. J., P. A. Stephens, R. B. Bradbury, R. P. Freckleton. 2006. Why do we still use stepwise modeling in ecology and behavior? *Journal of Animal Ecology* 75:1182-1189.



TABLE 1: Site data

Site	2008	2010		Latitude	Longitude	Elevation (m)	Slope	Aspect
		<i>P. spicata</i>	<i>B. japonicus</i>					
1	X	X	X	46°53'16.37"	113°59'44.83"	1029	24%	230°
2	X	X		46°53'34.32"	113°58'45.18"	1209	25%	70°
3	X	X	X	46°52'53.91"	113°59'10.08"	1083	37%	80°
4	X	X	X	46°51'59.40"	113°58'23.07"	1005	50%	200°
5	X	X		46°56'14.33"	114°00'58.38"	1157	40%	280°
6		X	X	46°53'36.25"	113°56'43.73"	1217	11%	290°
7		X	X	46°50'23.96"	113°58'19.61"	1284	21%	180°

An “X” indicates whether the specified site was used for that year of study.

TABLE 2: Experimental design

Species	Control	Competition:	Competition:	Clipping	Drought	Fertilizer	Sites	Control	Sampling Design
		<i>Euphorbia</i>	<i>Festuca</i>					for Genotype	
<i>B. japonicus</i>	X	X	X	X	X		5	no	broad
<i>P. spicata</i>	X	X			X		5	no	broad
<i>P. secunda</i>	X	X			X		5	no	broad
<i>B. japonicus</i>	X	X	X	X		X	5	yes	1 pair of 1 m <sup>2</sup> plots
<i>P. spicata</i>	X	X	X	X		X	7	no	broad

An “X” indicates whether the specified treatment was performed for that species.

TABLE 3: Results of ANOVA for 2008 experiment

		Shoot Mass			Root Mass			Total Mass			Root Mass Ratio		
		df	<i>F</i>	<i>P</i>	df	<i>F</i>	<i>P</i>	df	<i>F</i>	<i>P</i>	df	<i>F</i>	<i>P</i>
<i>P. secunda</i>	Invaded	1	1.316	0.253	1	0.035	0.851	1	0.500	0.514	1	1.068	0.353
	Treatment	2	5.694	0.004	2	1.775	0.173	2	2.523	0.084	2	3.692	0.027
	Site	4	1.924	0.110	4	5.161	0.001	4	1.257	0.415	4	6.305	0.051
	Invaded × Treat.		removed			removed		2	2.050	0.091	2	3.711	0.027
	Invaded × Site		removed			removed			removed		4	2.414	0.052
	Treatment × Site		removed			removed			removed			removed	
	Error	142			142			138			136		
<i>B. japonicus</i>	Invaded	1	0.002	0.966	1	3.501	0.062	1	1.240	0.266	1	4.151	0.042
	Treatment	4	63.657	<0.001	4	101.786	<0.001	4	80.221	<0.001	4	9.225	<0.001
	Site	4	4.797	0.037	4	2.111	0.079	4	4.576	0.011	4	2.862	0.023
	Invaded × Treat.		removed			removed			removed			removed	
	Invaded × Site	4	2.033	0.089		removed			removed			removed	
	Treatment × Site	16	1.882	0.021		removed			removed			removed	
	Error	367			387			367			367		

TABLE 4: Results of ANOVA for 2010 experiment

		df	Shoot Mass		Root Mass		Total Mass		RMR	
			<i>F</i>	<i>P</i>	<i>F</i>	<i>P</i>	<i>F</i>	<i>P</i>	<i>F</i>	<i>P</i>
<i>P. spicata</i>	Invaded	1	0.011	0.917	1.108	0.293	0.325	0.569	1.295	0.257
	Treatment	4	45.064	< 0.001	45.152	< 0.001	50.421	< 0.001	1.565	0.185
	Site	6	2.102	0.053	2.508	0.022	2.491	0.023	1.062	0.387
	Error	279								
<i>B. japonicus</i>	Invaded	1	0.003	0.954	0.821	0.366	0.289	0.591	2.208	0.138
	Treatment	4	236.789	< 0.001	259.44	< 0.001	279.614	< 0.001	23.373	< 0.001
	Site	4	0.402	0.807	1.105	0.354	0.677	0.608	2.487	0.044
	Error	319								

TABLE 5: Results of PCA for performance of *Bromus japonicus* in different treatments in 2010

Treatment	Variable	Communality	Component				
			1	2	3	4	5
Control	Shoot Mass	0.939	-0.003	-0.055	<b>0.954</b>	0.093	0.129
	Root Mass	0.944	0.078	0.028	<b>0.968</b>	0.021	0.005
Fertilized	Shoot Mass	0.897	-0.007	-0.004	0.067	<b>0.944</b>	-0.021
	Root Mass	0.891	0.068	-0.001	0.041	<b>0.933</b>	-0.117
<i>Euphorbia</i> competition	Shoot Mass	0.960	0.112	<b>0.971</b>	0.025	0.001	0.058
	Root Mass	0.961	0.069	<b>0.977</b>	-0.049	-0.006	0.002
<i>Festuca</i> competition	Shoot Mass	0.956	<b>0.968</b>	0.087	0.010	0.055	0.098
	Root Mass	0.947	<b>0.943</b>	0.105	0.070	0.011	0.202
Clipping	Shoot Mass	0.892	0.033	0.074	0.093	-0.070	<b>0.934</b>
	Root Mass	0.872	0.298	-0.019	0.038	-0.076	<b>0.880</b>
Initial Eigenvalue			2.689	2.046	1.884	1.538	1.102
Rotated Contribution			19.4%	19.3%	18.7%	17.9%	17.3%
Total Contribution			19.4%	38.7%	57.4%	75.3%	92.6%

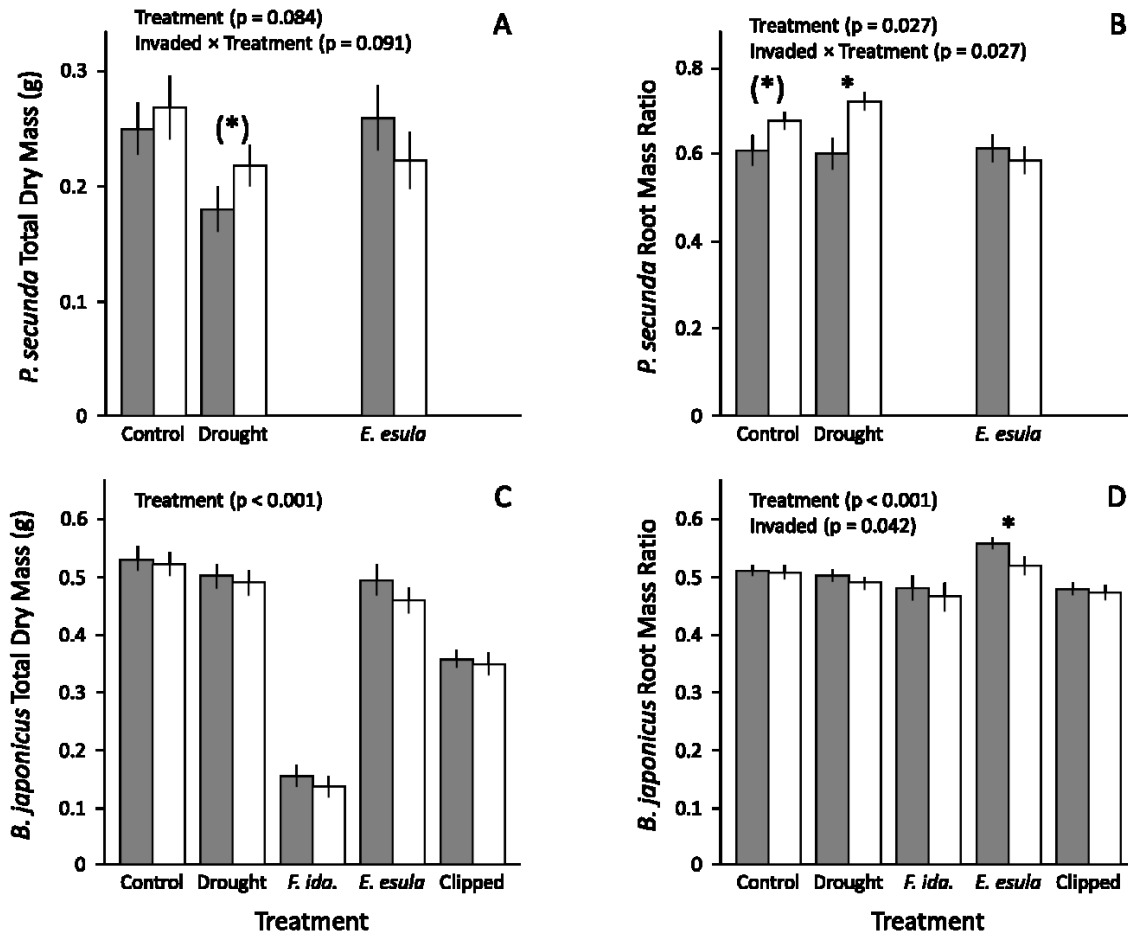


FIGURE 1. Effect of treatment and invasion status on total biomass and RMR of *P. secunda* (A,B), and *B. japonicus* (C,D) in 2008. Color indicates whether the parent plants were inside (grey) or outside (white) of *E. esula* patches. Error bars show standard error. Significant differences in mean performance between inside and outside plants in each treatment are indicated by \* ( $p < 0.050$ ) or (\*) ( $p < 0.100$ ). ANOVA terms with  $p < 0.100$  are displayed above each graph.