

THESIS

DISENTANGLING DRIVERS OF COLONIZATION SUCCESS IN LABORATORY AND
NATURAL SYSTEMS

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ABSTRACT

DISENTANGLING DRIVERS OF COLONIZATION SUCCESS IN LABORATORY AND NATURAL SYSTEMS

Understanding why colonizing populations successfully establish is important for predicting dynamics of invasive species. Propagule pressure, or the number of individuals in a founding group, is considered the most consistent predictor of establishment success, however, there remains considerable variance around predictions that demography alone cannot explain. The identity of individuals within a founding group (e.g. level of pre-adaptation to the recipient environment, diversity) as well as how individuals are introduced (e.g. frequency and timing of discrete introduction events) can influence establishment. The relative importance of these factors is unclear, and could vary across species and environmental contexts. To address these inconsistencies, we conducted two experiments: one with *Tribolium castaneum* (red flour beetle) populations maintained in controlled laboratory conditions, and one with *Bromus tectorum* (cheatgrass) founding populations introduced to a natural environment. For the *Tribolium* experiment, we varied the level of prior adaptation, diversity, and introduction frequency and timing for groups of eggs colonizing in a novel environment across three levels of propagule pressure ($n = 15, 30, 60$). Founding groups that were larger and more adapted to the novel environment survived the founding event better than smaller and less adapted groups. Further, we found that a high frequency of smaller introductions reduced initial survival. After a generation of mating, establishment success was driven predominantly by adaptation to the novel environment and diversity of founders. In the second experiment, we introduced groups of *B. tectorum* seeds at a constant propagule pressure ($n = 32$) to a common garden in Colorado, varying in source diversity (1, 2, 4, 8, or 16 source populations) and source region (Colorado = pre-adapted or Nevada = unadapted). We

evaluated establishment success by deriving the number of seeds produced by each founding group after one generation of growth and reproduction using a hierarchical Bayesian model. We found that increasing source diversity increased the number of seeds produced per founding group, but source region did not influence establishment success. Results from these experiments particularly speak to the context-dependency of the importance of pre-adaptation and diversity in predicting establishment success. This suggests that propagule pressure alone is not enough to explain why founding populations establish.

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PRIOR ADAPTATION, DIVERSITY, AND INTRODUCTION FREQUENCY MEDIATE THE RELATIONSHIP BETWEEN PROPAGULE PRESSURE AND ESTABLISHMENT SUCCESS¹

Introduction

Understanding why founding populations establish or fail to establish is of fundamental importance in explaining the abundance and distribution of organisms across landscapes (MacArthur & Wilson 1967). Within the field of invasion ecology, the total number of individuals introduced in a founding group, or “propagule pressure”, is the most consistent predictor of establishment success (Williamson 1996; Lockwood et al. 2005; Colautti et al. 2006; Simberloff 2009). However, groups founded with the same number of individuals can still vary widely in whether or not they successfully establish. It is critically important to better understand what drives differences in outcomes that numbers of individuals alone cannot predict, in order to guide effective management of invasive species (Gurevitch et al. 2011; Hufbauer et al. 2013). Crucial aspects influencing the positive effect of propagule pressure on establishment, as well as the variation around those predictions, include: if individuals within a founding group are adapted to the environment to which they are introduced (Hufbauer et al. 2011); how diverse founding groups are (Szűcs et al. 2014); and the timing, and frequency of discrete introduction events (Grevstad 1999).

Founding groups vary in the composition of their individuals. Principally, populations will fail to establish if none of the individuals within the founding group can survive in the recipient habitat (D’Antonio et al. 2001). If at least some individuals in a founding group can survive, the number of individuals in the founding group can shape establishment success (Lockwood et al. 2005). Prior adaptation to the recipient environment promotes higher fitness, as traits that were advantageous in the native environment are similarly advantageous

¹This chapter was submitted as a manuscript to *Biological Invasions*. Co-authors on the manuscript are Katriona Shea, Ciara Hovis, Brittany Teller, and Ruth Hufbauer.

in the recipient environment (Hufbauer et al. 2012). The fitness of individuals in the recipient habitat can mediate the magnitude and shape of the positive relationship between propagule pressure and success (D'Antonio 2001; Rouget & Richardson 2003; Thomsen et al. 2006; Duncan 2016), with fewer individuals needed to successfully establish a population if fitness is high (Crawley 1986; D'Antonio 2001; Szűcs et al. 2014). Thus, it seems advantageous for all individuals in a founding group to exhibit prior adaptation to the recipient environment to optimize establishment success.

In reality, founding groups often consist of a mix of adapted and unadapted individuals. As diversity increases from monocultures to polycultures, average establishment success can increase, as groups have an increased probability of containing an individual that exhibits prior adaptation to the recipient environment (i.e. sampling effect). This is considered an additive diversity effect because individuals in mixed groups perform the same as they would in monoculture. A diverse founding group acting through additive effects alone would not outperform a founding group of individuals that all exhibit prior adaptation to the recipient environment. However, if individuals perform better in mixture than they would in monoculture (non-additive diversity effect), more diverse founding groups could outperform a group that is adapted, but lacking diversity, if the non-additive effects outweigh the effect of pre-adaptation. Examples of positive non-additive effects of diversity include niche complementarity, admixture, and facilitation (Hughes et al. 2008).

Genetic diversity generally increases as propagule pressure increases, from a purely probabilistic standpoint (Ahlroth et al. 2003; Lockwood et al. 2005; Elam et al. 2007). Thus, to assess the impact of genetic diversity on the positive relationship between propagule pressure and establishment success, it is necessary to experimentally separate demography and genetics. A growing body of experimental work suggests that increased genetic diversity can match or even outweigh the effect of an increased number of individuals (Ahlroth et al. 2003; Elam et al. 2007; Hufbauer et al. 2013; Hedge et al. 2014; Szűcs et al. 2014; Szűcs et

al. 2017), but that sometimes larger founding groups, regardless of their diversity, are more likely to successfully establish than smaller groups (Wootton & Pfister 2013).

Beyond the composition of founding groups, how individuals are introduced can also influence establishment success. Propagule pressure is intrinsically the product of the number of individuals introduced in a single introduction event (hereafter: cohort) and the frequency of those events (Lockwood et al. 2005). Recent studies have addressed whether fewer introductions of larger cohorts are more successful in establishing as compared to more frequent introductions of smaller cohorts (Drake et al. 2005; Hedge et al. 2012; Britton & Gozlan 2013; Drolet & Locke 2016; Sinclair & Arnott 2016; Koontz et al. 2017). The optimal cohort size and frequency of introduction events is not consistent across studies as it can vary across species, environmental contexts, and whether introduction events are separated by space or time (e.g. Grevstad 1999). Further, the timing of introduction events can affect establishment success. For example, on a large temporal scale, annual fluctuations in temperature or precipitation moderate bioclimatic niches for invasive species (e.g. opening safe-sites) (Kowarik 1995). Similar patterns likely also exist on a smaller temporal scale; for example, introducing individuals of different ages at different times during the introduction process can affect intra-founder interactions, influencing establishment success (Jaremo & Bengtsson 2011).

To more fully understand the variation in establishment outcomes, it is necessary to jointly address the importance of adaptation to the recipient environment, diversity of the founding group, and introduction scenario (i.e. frequency and timing of introduction events), as these components could also interact. For example, less diverse populations could benefit from more frequent introduction events by mediating the reduction in diversity linked to bottlenecks in population size that many founding populations experience (Dlugosch & Parker 2008; Koontz et al. 2017). These potential interactions could explain the divergent conclusions regarding the importance of prior adaptation of individuals and diversity within founding groups and the importance of size and frequency of introduction events.

Here, we sought to better understand how the level of adaptation and diversity of founding groups, together with the timing, and frequency of introduction events, influence the relationship between propagule pressure and establishment success, using *Tribolium castaneum* (red flour beetle) as a model system. We assessed the influence of these factors on (1) initial founding success (survival after founding) and (2) establishment success (population size, growth rate, and avoidance of extinction after a generation of mating).

Materials and methods

Source Lineages

Large (500 to >2000 individuals) panmictic populations of six unique lineages of *T. castaneum* were reared in standard densities in incubators at 31°C for a minimum of 10 discrete generations prior to the start of the experiment. Lineages were maintained in replicate 4 x 4 x 6 cm plastic boxes, hereafter patches, containing 30 mL of medium (described below). Adults at standard densities were allowed 24 hours to mate and oviposit, and then were removed and discarded. Offspring were allowed 5 weeks to develop into adults, and then given fresh media to re-initiate the cycle. Three of these lineages were maintained on a nutritionally rich media (95% wheat flour, 5% brewers yeast) and three were maintained on a challenging media (98.2% corn flour, 1.71% wheat flour, 0.09% brewers yeast) that used a novel source of carbohydrate (corn flour) and had a reduced amount of nutritional yeast. Previous experiments using similarly nutritionally challenging corn media show that adaptation to a challenging corn environment can occur, and includes the evolution of increased cannibalism, faster development, and smaller body size (Agashe et al. 2011; Szűcs et al. 2014; Szűcs et al. 2017). Lineages that had been maintained on corn flour media will be hereafter referred to as “adapted” because the recipient environment in this experiment was mostly corn, and beetles that were maintained on the natal, wheat environment will be referred to as “unadapted”.

One generation before the experiment began, individuals within source lineages were introduced to a novel environment (99.0% corn flour, 0.95% wheat flour, 0.05% brewers yeast) at a constant density to standardize maternal environment carryover effects, which can be strong in *T. castaneum* populations (Van Allen & Rudolf 2013; Hufbauer et al. 2015; Van Allen & Rudolf 2016). The next generation of adults was then allowed to mate on fresh media for 24 hours and their eggs were sifted out of the media. These eggs were used to create experimental founding groups.

Experimental Design

We founded experimental groups of *T. castaneum* eggs on nutritionally challenging media (99.9% corn flour, 0.099% wheat flour, 0.01% brewers yeast), to simulate an introduction where successful establishment was difficult, potentially even for adapted lineages. Founding groups were composed either of eggs from a single, adapted lineage (one of 3 adapted source lineages), a single, unadapted lineage (one of 3 unadapted source lineages), or of eggs randomly drawn from a pool of all 6 lineages, with each lineage providing the same number of eggs to the pool. This allowed us to test the importance of adaptation on establishment success explicitly. We consider single and mixed treatments to be biologically representative of lower and higher genetic diversity treatments, respectively (as supported by data on microsatellite loci in Szűcs et al. 2014, 2017).

Groups were founded at three levels of propagule pressure: 15, 30, or 60 eggs, and were introduced to the patches in one of 6, week-long introduction scenarios, which varied in timing (which day introductions occurred), frequency (how many introduction events occurred), and cohort size (number of individuals at each introduction event) (Figure 1). For example, for introduction scenario 1 (Figure 1), for a propagule pressure of 15 eggs, five eggs were introduced each on Monday, Tuesday, and Wednesday. We thus could test the effect of varying cohort size and the number of introduction events jointly, as well as the timing of the introduction events. We implemented a full-factorial design (3 composition treatments

× 3 levels of propagule pressure × 6 introduction scenarios = 54 treatment combinations). As single line treatments were divided into single:adapted and single:unadapted post-hoc, we had 9 replicates of each treatment combination for single line treatments and 18 replicates of each treatment combination for mixed treatments.
























Introduction Scenario	Day of week					
I1	M 	T 	W 	R	F 	⇒ 
I2	M	T	W 	R 	F 	⇒ 
I3	M 	T 	W 	R 	F 	⇒ 
I4	M 	T	W	R	F 	⇒ 
I5	M	T	W 	R	F 	⇒ 
I6	M	T	W	R	F 	⇒ 

Figure 1: Experimental introduction scenarios each varying in frequency (number of discrete introduction events), size (proportion of individuals in each introduction event), and timing (what day(s) beetles are introduced). Note: figure depicts propagule pressure $n=15$, but in the experiment, 3 levels of propagule pressure were used ($n = 15, 30, 60$) using the same proportion of eggs for each introduction event.

Beetles that reached adulthood in each founding group were censused after a 5-week development period, placed on fresh media, allowed to mate for 24 hours, and were then removed. Offspring were censused an additional 5 weeks later. As there were few population extinctions across our treatments in the first generation, we define initial *founding success* as the proportion and number of initial individuals that survived to adulthood. We evaluate three aspects of *establishment success*: population size, growth rate of extant populations

after a generation of mating in the recipient environment, and the likelihood of population extinction by the end of the second generation.

Statistical Analyses

All statistical models were fitted in the package *lme4* (version 1.1.12) in R version 3.2.3 (R core team). We used parametric bootstrap methods (with 10,000 iterations) to evaluate the significance of interactions and main effects in each model using the package *pbrkrtest* (version 0.4.6). We estimated p-values of each interaction by comparing the deviances of a full model (i.e. with interactions of the same order and below) to a model without the interaction of interest. To estimate p-values of main effects, we compared the deviances of a model with all main effects but no interactions, as compared to a model without the effect of interest (Halekoh & Højsgaard 2014). Confidence intervals (CI) were estimated using the adjusted bootstrap percentile method (with 10,000 iterations) using packages *pbrkrtest* (version 0.4.6) and *boot* (version 1.3.18) for all models except the extinction model (see below).

We used a generalized linear mixed model with a binomial error distribution (logit link) to assess how propagule pressure, composition, and introduction scenario influenced the proportion of individuals that survived founding. Fixed effects in the model were categorical factors of propagule pressure (15, 30, or 60), composition (single:adapted, single:unadapted, or mixed), introduction scenario (see Figure 1), and their interactions. Block was included as a fixed effect, as we have only 3 blocks, which is too few to estimate an appropriate variance of all possible blocks (Crawley 2002). To account for overdispersion in the model, we included an observation-level random effect. We multiplied predicted proportional survival by appropriate propagule pressure levels to estimate the number of beetles that survived for each treatment combination.

To assess total establishment success, we evaluated treatment differences for the final size of extant populations, growth rates of extant populations between the first and second

generations, and the likelihood of population extinction by the second census. We modeled the size of extant populations with a standard linear model with a Gaussian error distribution using propagule pressure, composition, introduction scenario, their interactions, and block as fixed effects. Population sizes were log-transformed to meet the assumption of homogeneity of variance.

We modeled the growth rates of extant populations using a standard linear model with a Gaussian error distribution. Growth rates were log-transformed to meet the assumption of homogeneity of variance. As the propagule pressure determined the possible number of adults at the first census and *T. castaneum* growth rates are strongly density-dependent (Birch et al. 1951; Halliday et al. 2015), we modeled growth rates of extant populations using composition, introduction scenario, number of adults at first census (density), their interactions, and block as fixed effects. A significant effect of treatment (composition or introduction scenario) or an interaction between these treatments and density would indicate that there is a mechanism moderating population growth beyond what is explained by the number of adults at the first census.

We assessed the probability of population extinction using a logistic regression (logit link) with a binary response of extinct or extant after two generations. Fixed effects were the same for the population size analysis. For some treatment combinations, no populations went extinct. Since we do not expect the probability of extinction to be truly zero for these treatment combinations, we implemented a penalized likelihood method in the model (in R, `method = "brglm.fit"`; i.e. the separation problem, Albert et al. 1984). We present Wald confidence intervals for factor means obtained using the *lsmeans* package in R.

Results

We evaluated initial founding success using the number and proportion of individuals that survived the founding event. The number of individuals that survived increased as propagule pressure increased (Figure 2b). However, at higher levels of propagule pressure, a

lower proportion of individuals survived founding ($p = 0.001$), suggesting negative density dependence (Figure 2a).

The introduction scenario, specifically differences in the frequency of founding, also influenced initial founding success. Fewer individuals survived in founding groups that were introduced most frequently and subsequently, in the smallest cohorts ($p = 0.001$; Figure 2c 2d), than in founding groups introduced less frequently and thus in larger cohorts.

A higher proportion of individuals survived when founding groups were composed of adapted individuals from a single lineage as compared to mixed founding groups. Unadapted founding groups had a significantly smaller proportion of individuals survive than both groups of mixed and adapted lineages, and mixed groups had a significantly smaller proportion of individuals survive than adapted lineages ($p = 0.001$; Figure 2a). Founding groups of unadapted individuals were less sensitive to negative density dependence, such that founding groups of sizes 15 and 30 did not have significantly different proportions of individuals survive (composition by propagule pressure interaction, $p = 0.004$; Figure 2a).

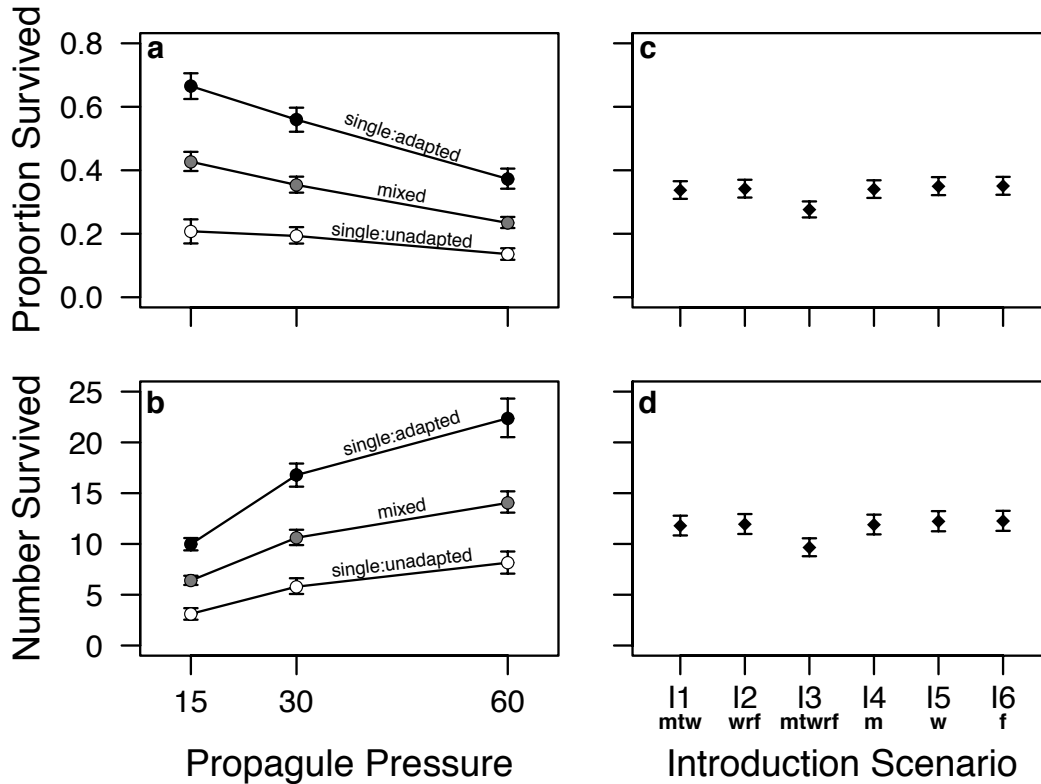


Figure 2: (a) Proportion and (b) number of adults that survived the initial introduction for each level of propagule pressure and composition combination averaged across all introduction scenarios. (c) Proportion and (d) number of individuals that survived each introduction scenario averaged across all levels of propagule pressure and composition. Back-transformed model means and 95% confidence intervals around model means are reported for ease of interpretation.

Final population sizes of mixed and adapted founding groups were larger on average than unadapted founding groups ($p = 0.0001$) (Figure 3). Interestingly, population sizes for mixed founding groups were 38.0% (CI 23.6-49.4%) smaller when initially founded with fewest number of individuals as compared to the largest number of individuals, however populations composed of a single line were relatively the same size regardless of initial propagule pressure (composition by propagule pressure interaction, $p=0.049$). The effect of introduction scenario on population size was lost by the second generation ($p=0.804$).

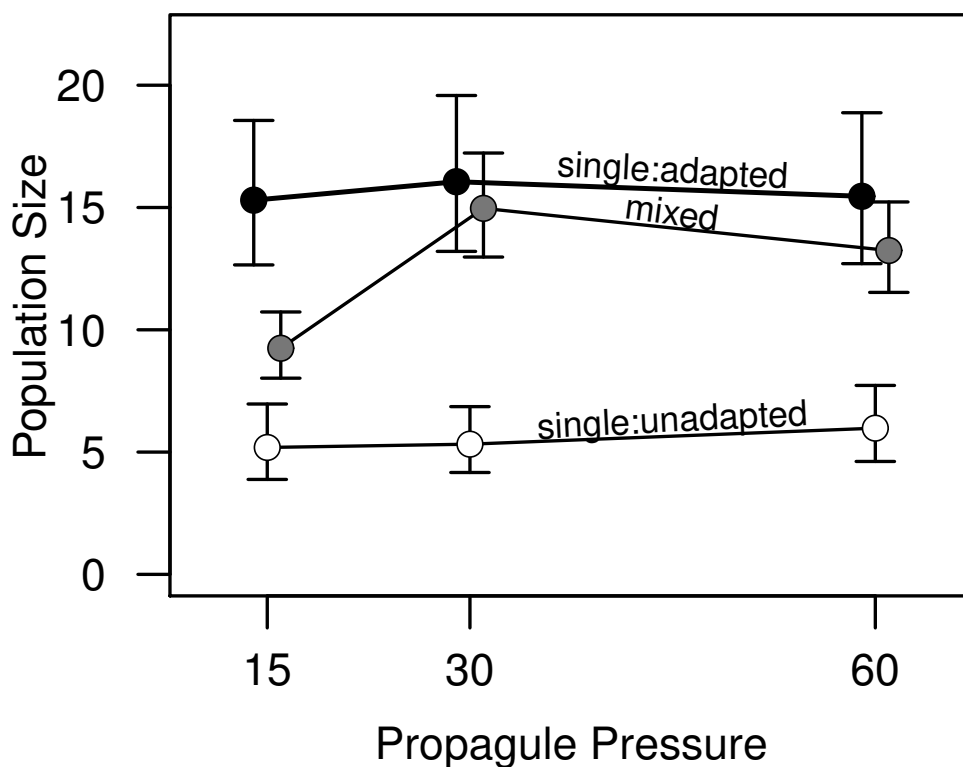


Figure 3: Final population size after introduction and one generation of mating for adapted single lineage, unadapted single lineage, and mixed founding groups at three levels of propagule pressure (15, 30, 60 eggs). Back-transformed model means and 95% confidence intervals are reported for ease of interpretation. Points for each composition group are jittered for ease of interpretation

Differences in population size were partially driven by negative density-dependent growth. As the number of individuals that survived the founding event increased, population growth rates decreased ($p = 0.0001$). Averaged across densities, mixed and adapted founding groups still exhibited 110.5% (CI 73.2-156.1%) and 128% (CI 82.3-181.3%), higher population growth rates respectively, as compared to unadapted founding groups ($p = 0.0001$), however there were no significant differences in growth rate between mixed and adapted founding groups.

Mixed founding groups and adapted founding groups were less likely than unadapted founding groups to go extinct by the second generation ($p = 0.0001$; Figure 4) and smaller founding groups were more likely to go extinct than larger founding groups ($p=0.0033$; Figure

4). There were no differences in extinction or growth rate between different introduction scenarios ($p = 0.469$, Figure 4; $p = 0.411$).

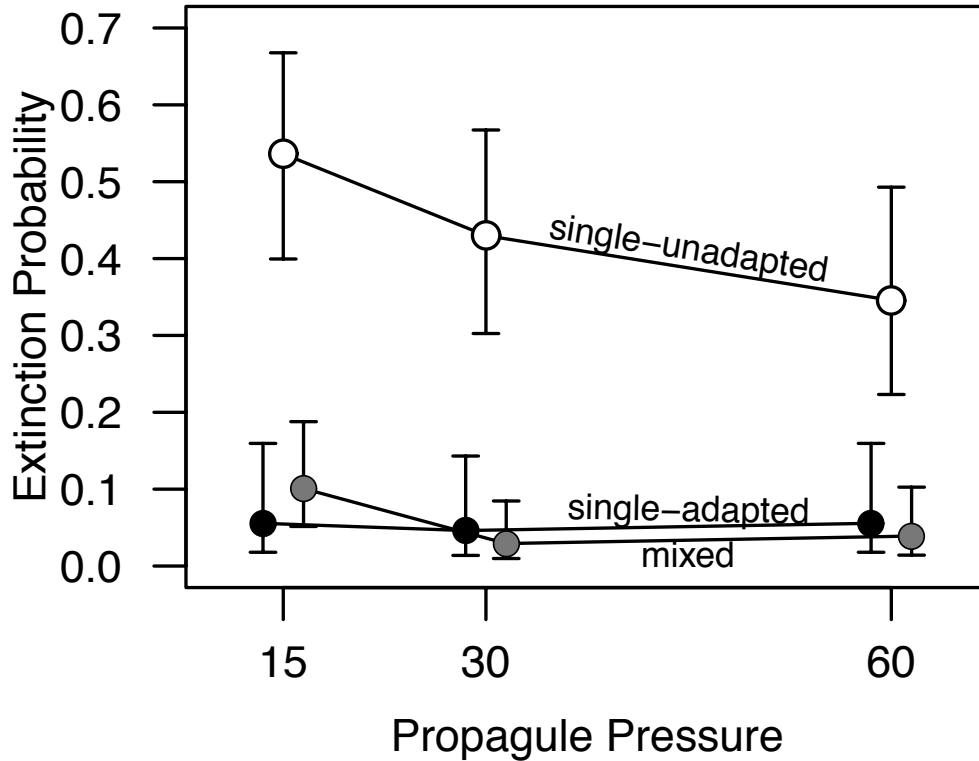


Figure 4: Probability of population extinction after introduction and one generation of mating for adapted single lineage, unadapted single lineage, and mixed founding groups at three levels of propagule pressure (15, 30, 60 eggs). Back-transformed model means and 95% confidence intervals are reported for ease of interpretation. Points for each composition group are jittered for ease of interpretation.

Discussion

Propagule pressure consistently explains much of the variation in the establishment outcomes of founding populations (Lockwood et al. 2005; Simberloff 2009). However, not all founding groups of the same size have equal establishment success. These differences could be explained by other characteristics of the introduction, including the level of prior adaptation and diversity of the founding group, and how frequently individuals are introduced to the recipient environment. We confirmed that propagule pressure can be important in predicting

founding and establishment success, but at least in the proximate generations after founding, can be surpassed by the composition of the founding group (e.g. level of prior adaptation to the novel environment and diversity).

Propagule Pressure

Our results validate the theoretically and empirically supported positive relationship between propagule pressure and founding success: as the size of founding groups increased, initial founder success increased (Figure 2b). However, we found that the proportion of individuals that survived the founding event decreased as propagule pressure increased and that negative density dependence was strongest for adapted and mixed groups (Figure 2a). Negative density dependence is strong in our experimental system (Birch et al. 1951; Halliday et al. 2015); increases in density can reduce survival from egg to adult because there is proportionally lower nutrient availability during development (Wong et al. 2011). This effect can also be exacerbated by an increased likelihood of cannibalism to mitigate the nutritional deficit (Mertz & Cawthorn 1973). Although negative density dependence strongly influenced establishment outcomes in our experiment, this is a biologically reasonable scenario for some invasive species: when colonizing in a novel environment, competition for limited resources is common (Siepielski & McPeck 2010). By the end of the second generation, propagule pressure only weakly influenced the establishment success of founding groups. Only the smallest founding groups ($n = 15$) had a significantly lower likelihood of extinction (Figure 4) and only for diverse founding groups did smaller groups maintain smaller population sizes as compared to larger groups.

Prior Adaptation and Diversity

Adapted founding groups had more individuals survive founding as compared to mixed groups, which in turn had more individuals survive than unadapted groups. This suggests that increased survival in the mixed founder treatment as compared to the unadapted groups

can be attributed to the presence of one or more adapted founders drawn from the pool of mixed lines; likely an additive effect of diversity.

Diversity effects continued to be influential in predicting establishment success. Both adapted and mixed founding groups were less likely to go extinct as compared to unadapted groups and had a similarly low likelihood of extinction (on average $<5\%$), indicating that only a proportion of individuals in the founding group need be adapted to avoid extinction. Final population sizes were contingent on propagule pressure, the composition of founding groups, and the number of individuals that survived the initial founding event (density) as evidenced by the growth rate analysis. At the lowest level of propagule pressure, final population size reflected the patterns of founding group survival: adapted founding groups were larger than mixed groups, and mixed groups were larger than unadapted groups. However, for larger founding groups ($n=30, 60$), adapted and mixed groups performed similarly better than unadapted groups (Figure 3). Similarities in population sizes between mixed and adapted populations for larger founding groups can be explained by two possible, non-mutually exclusive, mechanisms in the context of our experiment.

First, a positive non-additive diversity effect from mixing unadapted and adapted individuals in this experiment could be more beneficial to founding groups of larger sizes. Adapted individuals could be more productive in a mixed group than in a monoculture via reduced competition, complementarity between lineages, or admixture. However, this more positive effect of genetic diversity on larger rather than smaller populations is contrary to theory and empirical evidence suggesting that increased diversity should be most advantageous to smaller populations that are prone to inbreeding (Lande 1988; Szűcs et al. 2014). Our experimental design restricts our ability to partition additive and non-additive diversity effects on founding and establishment success, as we randomly drew individuals from adapted and unadapted lines to compose mixed group replicates.

Second, the strong effect of negative density-dependent growth could explain why large, mixed founding groups were of similar final population size as large, adapted groups. As

mixed groups had significantly fewer individuals survive initially as compared to adapted groups, they would have higher growth rates, increasing their size relative to adapted populations. Further, theory suggests that increased diversity can decrease population sensitivity to density-dependent regulation (Bjørnstad & Hansen 1994). This has also been seen empirically with marine fish (Johnson et al. 2016). We see this pattern here, where within both adapted and unadapted founding groups, population sizes were similar across all levels of propagule pressure, but mixed groups exhibited final population sizes that increased with increasing propagule pressure, exhibiting similar patterns as the initial number of survivors. However, these hypotheses related to negative density-dependent growth are not supported by our statistical analyses; growth rates of adapted and diverse founding groups, controlling for density, were not significantly different from each other.

Regardless of the mechanism, we provide evidence that increasing the number of adapted individuals in a founding group increases founding and establishment success. Mixed groups avoid extinction to the same extent as adapted groups, and reach larger population sizes across all levels of propagule pressure as compared to unadapted groups, even reaching the sizes of adapted groups for larger founding group sizes.

Interestingly, we found that adaptation to the novel environment seemed to outweigh the overall effect of propagule pressure, particularly in the later stages of establishment. This result contributes to the growing body of literature assessing the relative importance of habitat suitability and propagule pressure in predicting establishment success (Maron 2006; Nunez et al. 2011; Erfmeier et al. 2013; Hufbauer et al. 2013; Szűcs et al. 2014; Duncan 2016). For example, Hufbauer et al. (2013) conducted a factorial experiment crossing host suitability and propagule size (the number of individuals in one introduction event) in *Bemisia* whitefly introductions, and found that host suitability alone explained probability of establishment and population growth rate. However, final population size was influenced by an interaction between propagule size and host suitability. Alternatively, propagule size was found to more important than habitat suitability, in the form of latitude matching,

for *Hypericum perforatum* colonization success (Maron 2006). Recently, models by Duncan (2016) verified the context-dependency of the effect of propagule pressure on establishment success for dung beetles. For one beetle species studied, there were drastic differences in the suitability of recipient environments, resulting in propagule pressure failing to predict establishment success; however, for a species for which there was less variation in habitat quality, propagule pressure was an important predictor of establishment success (Duncan 2016). Thus, the relative importance of prior adaptation and propagule pressure in our experiment is likely driven by the fact that our recipient environment was consistently very harsh and selective.

Introduction Scenario

Founding success was further influenced by introduction scenario: introducing many, small cohorts resulted in the fewest individuals surviving the founding event regardless of the total number of individuals introduced (Figure 2). This result contrasts classic work by Grevstad (1999), but corroborates some recent simulation studies (Wittmann et al. 2014; Drolet & Locke 2016). Grevstad (1999) suggested that when cohorts are released under environmentally variable conditions, the positive effect of an increased number of releases would be particularly strong. As the environment was stable in our experiment, we may not expect to see that an increased number of releases to be advantageous. Recently, Wittman et al. (2014) found that in difficult but homogenous introduction environments (where the average per capita growth rate is negative), having fewer but larger introduction events best promotes establishment success. Drolet & Locke (2016) also found that in the models including both demographic and environmental stochasticity (assuming Allee effects are present), increasing the number of introduction events decreases establishment success. However, when Allee effects were absent, they found the opposite result: increasing the number of introductions actually increased establishment (Drolet & Locke 2016).

Whether introducing many, small cohorts or few, large cohorts is optimal for colonization success can be context-dependent, varying by organism, environment, or the amount of time between introduction events. For example, a recent study by Koontz et al. (2017) using the same *Tribolium* system with a very similar recipient environment to this study, found that introducing many, small cohorts increased establishment success as compared to introducing fewer, larger cohorts. The important distinction between our studies is that we manipulated introduction scenarios using eggs within a generation while Koontz et al. (2017) introduced cohorts of adults across generations. Thus, the effect of increasing the number of introductions in their experiment was largely genetic while ours was likely not.

Interestingly, a parallel experiment with a deliberately similar design to our experiment, which introduced *Carduus nutans* (musk thistle) seeds to plots in a natural setting also found that plots with more frequent introductions (5 introduction events) had lower founding success than those with moderately frequent introductions (3 introduction events) (Hovis et al., *in prep*). Increased introduction frequency decreased founding success consistently across levels of propagule pressure and maternal diversity. The similarity in findings between our experiments is particularly striking given that *C. nutans* colonization was not regulated by density dependence and that seeds were introduced across weeks under natural environmental conditions that were temporally heterogenous, albeit relatively benign. These similarities between our studies, which used vastly different organisms and recipient habitats, provide promise for identifying emergent trends in how introduction frequency influences founding success when propagules are introduced in a single generation. Our study provides empirical evidence that having many frequent, but small introductions in a homogenous environment is consistently disadvantageous to founding success across multiple levels of propagule pressure, genetic diversity, and prior adaptation to the recipient environment, which corroborates the recent findings of Hovis et al. (*in prep*). The effect of introduction frequency found in our experiment was lost by the second generation as population composition and negative

density-dependent growth overwhelmed the differences in likelihood of extinction and final population size between groups.

Conclusion

Our results show that multiple factors contribute to the fate of founding populations above and beyond the positive effect of propagule pressure. Propagule adaptation to the recipient environment additionally increases the likelihood of establishment success. Increasing the number of individuals exhibiting prior adaptation to the recipient environment decreases the likelihood of extinction. Increasing the number of introduction events can alter interactions among founders, mediating establishment success, however, this effect is minor compared to those of population composition and propagule pressure, at least in the harsh environments in the present study. Given that many factors can influence establishment success, predictive models for invasive species colonization cannot rely on propagule pressure alone to predict if founding populations will successfully establish. Further, understanding the population dynamics of a particular invasive species (e.g. strength of density-dependent growth) may be critical in determining how important prior adaptation, diversity, and introduction scenario are in determining population size and likelihood of extinction in the generations after founding.

REFERENCES

- Agashe D, Falk JJ, Bolnick DI (2011) Effects of founding genetic variation on adaptation to a novel resource. *Evolution* 65:2481-2491.
- Ahroth P, Alatalo RV, Holopainen A, Kumpulainen T, Suhonen J (2003) Founder population size and number of source populations enhance colonization success in waterstriders. *Oecologia* 137:617-620.
- Albert A, Anderson JA (1984) On the existence of maximum-likelihood estimates in logistic-regression models. *Biometrika* 71:1-10.
- Birch LC, Park T, Frank MB (1951) The effect of intraspecies and interspecies competition on the fecundity of two species of flour beetles. *Evolution* 5:116-132.
- Bjørnstad ON, Hansen TF (1994). Individual variation and population dynamics. *Oikos*, 69, 167-171.
- Bossdorf O, Auge H, Lafuma L, Rogers WE, Siemann E, Prati D (2005) Phenotypic and genetic differentiation between native and introduced plant populations. *Oecologia* 144:1-11.
- Britton JR, Gozlan RE (2013) How many founders for a biological invasion? Predicting introduction outcomes from propagule pressure. *Ecology* 94:2558-2566.
- Colautti RI, Grigorovich IA, MacIsaac HJ (2006) Propagule pressure: A null model for biological invasions. *Biol Invasions* 8:1023-1037.
- Crawford KM, Rudgers JA (2012) Plant species diversity and genetic diversity within a dominant species interactively affect plant community biomass. *J Ecol* 100:1512-1521.

- Crawford KM, Whitney KD (2010) Population genetic diversity influences colonization success. *Mol Ecol* 19:1253-1263.
- Crawley MJ (1986) The population biology of invaders. *Philos T Roy Soc B* 314: 711-729.
- Crawley MJ (2002) Statistical computing: An introduction to data analysis using S-Plus. John Wiley & Sons, West Sussex, England.
- Crutsinger GM, Collins MD, Fordyce JA, Gompert Z, Nice CC, Sanders NJ (2006) Plant genotypic diversity predicts community structure and governs an ecosystem process. *Science* 313:966-968.
- D'Antonio CM, Levine J, Thomsen M (2001) Ecosystem resistance and the role of propagule supply: a California perspective. *Journal of Mediterranean Ecology* 2:233-245.
- Dlugosch KM, Parker IM (2008) Founding events in species invasions: genetic variation, adaptive evolution, and the role of multiple introductions. *Mol Ecol* 17:431-449.
- Drake JM, Baggenstos P, Lodge DM (2005) Propagule pressure and persistence in experimental populations. *Biol Letters* 1:480-483.
- Drolet D, Locke A (2016) Relative importance of propagule size and propagule number for establishment of non-indigenous species: a stochastic simulation study. *Aquatic Invasions* 11:101-110.
- Duncan RP (2016) How propagule size and environmental suitability jointly determine establishment success: a test using dung beetle introductions. *Biol Invasions* 18:985-996.
- Elam DR, Ridley CE, Goodell K, Ellstrandt NC (2007) Population size and relatedness affect fitness of a self-incompatible invasive plant. *P Natl Acad Sci USA* 104:549-552.
- Erfmeier A, Hantsch L, Bruelheide H (2013) The role of propagule pressure, genetic diversity and microsite availability for *Senecio vernalis* invasion. *PLOS ONE* 8: e57029

- Estoup A, Ravigne V, Hufbauer R, Vitalis R, Gautier M, Facon B (2016) Is there a genetic paradox of biological invasion? *Annu Rev Ecol Evol S* 47:51-72.
- Facon B, Jarne P, Pointer JP, David P (2005) Hybridization and invasiveness in the freshwater snail *Melanoides tuberculata*: hybrid vigour is more important than increase in genetic variance. *J Evol Biol* 18:524-535.
- Forsman A, Wennersten L (2016) Inter-individual variation promotes ecological success of populations and species: evidence from experimental and comparative studies. *Ecography* 39:630-648.
- Grevstad FS (1996) Establishment of weed control agents under the influences of demographic stochasticity, environmental variability and Allee effects. *Proceedings of the IX International Symposium on Biological Control of Weeds* 261-267.
- Grevstad FS (1999) Factors influencing the chance of population establishment: implications for release strategies in biological control. *Ecol Appl* 9:1439-1447.
- Gurevitch J, Fox GA, Wardle GM, Inderjit, Taub D (2011) Emergent insights from the synthesis of conceptual frameworks for biological invasions. *Ecol Letters* 14:407-418.
- Halekoh U, Højsgaard S (2014) Kenward-Roger approximation and parametric bootstrap methods for tests in linear mixed models - the R package pbkrtest. *J Stat Softw* 59: 1-32.
- Halliday WD, Thomas AS, Blouin-Demers G (2015) High temperature intensifies negative density dependence of fitness in red flour beetles. *Ecol Evol* 5:1061-1067.
- Hedge LH, OConnor WA, Johnston EL (2012) Manipulating the intrinsic parameters of propagule pressure: implications for bio-invasion. *Ecosphere* 3:1-13.

- Hedge LH, Leung B, O'Connor WA, Johnston EL (2014) The interacting effects of diversity and propagule pressure on early colonization and population size. *J Anim Ecol* 83:168-175.
- Hovis CL, Keller JA, Teller BJ, Hufbauer RA, Shea K. Parsing propagule pressure: The effects of multiple aspects of propagule pressure on the success of an invasive species. Manuscript in preparation.
- Hufbauer RA, Facon B, Ravigné V, Turgeon J, Foucaud J, Lee CE, Estoup A (2011) Anthropogenically induced adaptation to invade (AIAI): contemporary adaptation to human-altered habitats within the native range can promote invasions. *Evol Appl* 5:89-101.
- Hufbauer RA, Rutschmann A, Serrate B, de Conchard HV, Facon B (2013) Role of propagule pressure in colonization success: disentangling the relative importance of demographic, genetic and habitat effects. *J Evol Biol* 26:1691-1699.
- Hufbauer RA, Szűcs M, Kasyon E, Youngberg C, Koontz MJ, Richards C, Tuff T, Melbourne BA (2015) Three types of rescue can avert extinction in a changing environment. *P Natl Acad Sci USA* 112:10557-10562.
- Jaremo J, Bengtsson G (2011) On the importance of life history and age structure in biological invasions. *Ecol Model* 222:285-492.
- Johnson DW, Freiwald J, Bernardi G (2016) Genetic diversity affects the strength of population regulation in a marine fish. *Ecology* 97:627-639.
- Kolbe JJ, Glor RE, Schettino LR, Lara AC, Larson A, Losos JB (2004) Genetic variation increases during biological invasion by a Cuban lizard. *Nature* 431:177-181.
- Kolbe JJ, Glor RE, Schettino LR, Lara AC, Larson A, Losos JB (2007) Multiple sources, admixture, and genetic variation in introduced *Anolis* lizard populations. *Conserv Biol* 21:1612-1625.

- Koontz MJ, Oldfather MF, Melbourne BA, Hufbauer RA (2017) Parsing propagule pressure: Number, not size, of introductions drives colonization success in a novel environment. *bioRxiv* 108324; doi: <http://doi.org/10.1101/108324>.
- Kowarik I (1995) Time lags in biological invasions with regard to the success and failure of alien species. In: Pysek P, Prach, K, Rejmanek, M, Wade, M (eds) Plant invasions, general aspects and special problems. SPB Academic Publishers, Amsterdam, pp 15-38.
- Lande R (1988) Genetics and demography in biological conservation. *Science* 241:1455-1460.
- Lockwood JL, Cassey P, Blackburn T (2005) The role of propagule pressure in explaining species invasions. *Trends Ecol Evol* 20:223-228.
- Loreau M, Hector A (2001) Partitioning selection and complementarity in biodiversity experiments. *Nature* 412:72-76.
- MacArthur RH, Wilson EO (1967) The theory of island biogeography. Princeton University Press, Princeton, NJ.
- Maron JL (2006) The relative importance of latitude matching and propagule pressure in the colonization success of an invasive forb. *Ecography* 29:819-826.
- Mertz DB, Cawthon DA (1973) Sex differences in the cannibalistic roles of adult flour beetles. *Ecology* 54:1400-1402.
- Newman D, Pilson D (1997) Increased probability of extinction due to decreased genetic effective population size: Experimental populations of *Clarkia pulchella*. *Evolution* 51:354-362.
- R Core Team (2016) R: A language and environment for statistical computing. R Foundation for Statistical Computing, Vienna, Austria. <https://www.R-project.org/>.
- Roman J, Darling JA (2007) Paradox lost: Genetic diversity and the success of aquatic invasions. *Trends in Ecology and Evolution* 22:454-464.

- Rouget M, Richardson DM (2003) Inferring process from pattern in plant invasions: a semimechanistic model incorporating propagule pressure and environmental factors. *Am Nat* 162:713-742.
- Siepielski AM, McPeck MA (2010) On the evidence for species coexistence: a critique of the coexistence program. *Ecology* 91:3153-3164.
- Simberloff D (2009) The Role of Propagule Pressure in Biological Invasions. *Annu Rev Ecol Evol S* 40:81-102.
- Sinclair JS, Arnott SE (2016) Strength in size not numbers: propagule size more important than number in sexually reproducing populations. *Biol Invasions* 18:497-505.
- Szűcs M, Melbourne BA, Tuff T, Hufbauer RA (2014) The roles of demography and genetics in the early stages of colonization. *P Roy Soc B-Biol Sci* 281:1-8.
- Szűcs M, Melbourne BA, Tuff T, Weiss-Lehman C, Hufbauer RA (2017) Genetic and demographic founder effects have long-term fitness consequences for colonising populations. *Ecology Letters* doi:10.1111/ele.12743.
- Thomsen MA, Corbin JD, D'Antonio CM (2006) The effect of soil nitrogen on competition between native and exotic perennial grasses from northern coastal California. *Plant Ecol* 186:23-35.
- Tilman D, Wedin D, Knops J (1996) Productivity and sustainability influenced by biodiversity in grassland ecosystems. *Nature* 379:718-720.
- Van Allen BG, Rudolf VHW (2013) Ghosts of habitats past environmental carry-over effects drive population dynamics in novel habitat. *Am Nat* 181:596-608.
- Van Allen BG, Rudolf VHW (2016) Carryover effects drive competitive dominance in spatially structure environments. *P Natl Acad Sci USA* 113:6939-6944.

Williamson M (1996) *Biological Invasions*. Chapman Hall, London.

Wittman MJ, Metzler D, Gabriel W, Jeschke JM (2014) Decomposing propagule pressure: the effects of propagule size and propagule frequency on invasion success. *Oikos* 123:441-450.

Wong N, Lee C (2011) Relationship between population growth of the red flour beetle *Tribolium castaneum* and protein and carbohydrate content in flour and starch. *J Econ Entomol* 104:2087-2094.

Wootton JT, Pfister CA (2013) Experimental separation of genetic and demographic factors on extinction risk in wild populations. *Ecology* 94:2117-2123.

SOURCE DIVERSITY PROMOTES ESTABLISHMENT SUCCESS OF *B. TECTORUM* FOUNDING POPULATIONS²

Introduction

A central goal in ecology is to understand the forces that promote successful establishment of founding populations (Mayr 1965; MacArthur & Wilson 1967). Propagule pressure, or the number of individuals in a founding group, has been considered a strong predictor of establishment success (Lockwood et al. 2005; Simberloff 2009). However, propagule pressure is often confounded with intraspecific diversity because diversity increases with increasing propagule pressure (Ahlroth et al. 2003; Lockwood et al. 2005; Elam et al. 2007). The importance of intraspecific diversity within founding populations has traditionally focused on the later stages of colonization when diversity is necessary for local adaptation and rapid evolution. Models by Lande (1988) suggested that demography should mediate initial establishment success, while diversity should become more important in moderating population growth and persistence once established. However, growing evidence suggests that increased intraspecific diversity drives the positive relationship observed between propagule pressure and initial establishment success (reviewed in Forsman 2014; Forsman & Wennersten 2016).

Intraspecific diversity can enhance the chance of surviving the founding event (Ahlroth et al. 2003; Szűcs et al. 2017), the size of the population after founding (Gamfeldt et al. 2005; Forsman et al. 2012; Smee et al. 2013; Hanley et al. 2016), its biomass (Hughes & Stachowicz 2004; Crawford & Whitney 2010; Drummond & Vellend 2012; Erfmeier et al. 2013; Tomimatsu et al. 2014), and growth rate (Hufbauer et al. 2013; Szűcs et al. 2014, 2017). The immediate effects of founding group diversity can have cascading effects, because they mediate longer term evolutionary responses after populations become established in the introduced range (Parker et al. 2003; Hughes et al. 2008).

²This is the working title for the manuscript that will be submitted based off of the work in this chapter. N. Thompson Hobbs will be a co-author on the manuscript.

Although increasing intraspecific diversity generally increases establishment success (Forsman 2014; Forsman & Wennersten 2016), the benefits of increasing diversity appear context-dependent. Theory predicts that diversity effects might be magnified in favorable or unfavorable environments (Charmantier & Grant 2005; Drummond & Vellend 2012; Hedge et al. 2013), and empirical support remains inconclusive. Hanley et al. (2016) manipulated the diversity of founding populations of *Crassostrea* oysters and introduced them to habitats either with or without predators, and only found positive effects of diversity in the absence of predators. Hughes and Stachowicz (2004) found in an experiment manipulating intraspecific diversity of seagrass, that the effect of diversity on colonization was only realized after a grazing disturbance (Hughes & Stachowicz 2004). Other studies have alternatively found no interactions between environmental context and diversity treatment (Gamfeldt et al. 2005, Agashe 2009).

Understanding the role of increased intraspecific diversity is also challenging as increased establishment success may result from two different mechanisms. More diverse founding groups have a higher likelihood of containing an individual or individuals that are pre-adapted to the recipient environment (Loreau & Hector 2001) (i.e. sampling effects). We use the term pre-adapted to signify the process of prior adaptation alone (*sensu* Hufbauer et al. 2012), rather than encompassing prior adaptation and exaptation (co-opting traits that were advantageous in the native environment for a different purpose than in the introduced environment). Increasing diversity can also promote niche partitioning, reduce competition, or increase facilitation via non-additive diversity effects, where individuals perform differently in diverse founding groups than they would in monoculture (Hughes et al. 2008). These two mechanisms are not mutually exclusive (2008).

Environmental context can mediate the magnitude of the effect of increased diversity on establishment success. For example, increasing the chance of containing a pre-adapted individual through additive sampling effects may be of little importance if most individuals from source populations are pre-adapted to the recipient environment. Sampling effects could

be crucial in obtaining a rare individual that is more pre-adapted to the recipient environment for source populations that are, on average, less pre-adapted to the recipient environment (Figure 5). Non-additive effects can also be amplified when individuals in a founding group are not pre-adapted to the recipient environment: the variability in response to the recipient environment decreases as different genotypes or phenotypes respond differently to a new environmental context (i.e. insurance hypothesis *sensu* Yachi & Loreau 1999)

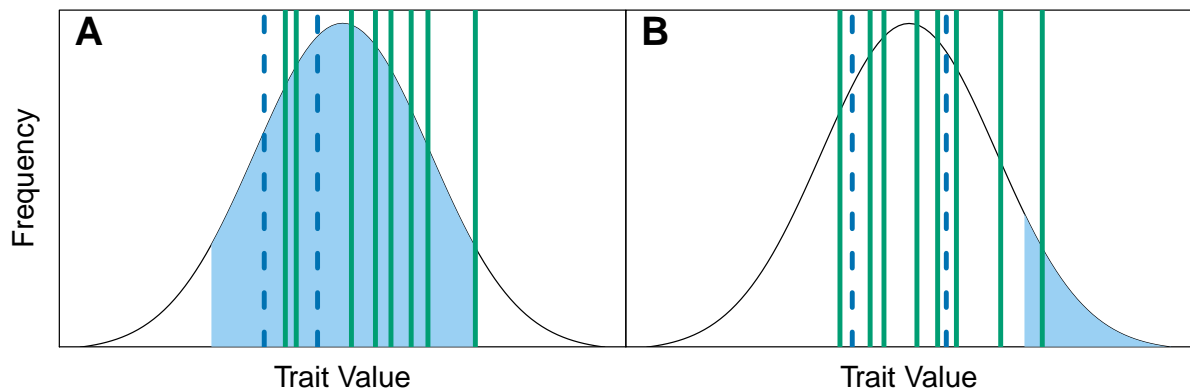


Figure 5: Conceptual model of the how additive sampling effects may vary in importance depending on the average level of pre-adaptation of source populations. Shaded regions represent pre-adapted trait values that are advantageous in the introduced environment. Green lines represent 8 random draws from the distribution and dark blue lines represent 2 random draws from the distribution. (A) There is a high likelihood of sampling an individual that is pre-adapted to the recipient environment so the level of pre-adaptation across diversity treatments is similar. (B) There is a low likelihood of sampling an individual that is pre-adapted to the recipient environment so increasing diversity would increase the probability of including an individual that is pre-adapted in the founding group.

Determining the importance of intraspecific diversity in establishment is particularly important in a management context when individuals from source populations colonize in a natural setting, where environmental context is often highly variable. Only six of fourteen studies manipulating intraspecific diversity of colonizing populations reviewed in Forsman & Wennersten 2016 were conducted in a natural setting. Only one of those studies, Drummond and Vellend (2012), assessed the relative importance of intraspecific diversity across habitats that vary in their conditions or context. Further, the climate-matching or environment-matching paradigm is pervasive throughout invasion ecology, yet does not consistently ex-

plain patterns of invasion (Parker et al. 2003; Sol 2007; Nuñez & Medley 2011). Using manipulative experiments to distinguish between pre-adaptation and phenotypic plasticity could bring greater clarity to how environment-matching influences establishment success (Bock et al. 2015).

Here, we conducted a common garden experiment in Northern Colorado to investigate how environmental context (via level of pre-adaptation) mediates the establishment success of a notorious invasive grass, *Bromus tectorum* L., across multiple levels of intraspecific diversity. We used reproductive effort (*sensu* Thompson & Stewart 1981) as a metric for establishment success. The number of source populations represented in a founding population (hereafter: source diversity) was used as a proxy for intraspecific diversity (from least to most diverse: 1, 2, 4, 8, or 16 source populations). We founded two sets of populations: those comprised of seed collected from Colorado or from Nevada. Thus, our two sets of populations represent a pre-adapted and an unadapted region with respect to the recipient common garden in Colorado. There is substantial ecologically-relevant genetic variation between and within natural populations of *B. tectorum* in the Intermountain West and Great Basin (Novak & Mack 2016, see Methods). Also, source populations within regions were within distances in which they could feasibly interact (see Methods), thus we assess the consequence of realistic population mixing via animal or human-mediated transport.

We predicted that founding populations composed of seeds from Colorado would have greater establishment success than populations composed of seeds from Nevada, as seeds from Colorado are more likely to be pre-adapted to the common garden environment. We also predicted that increased source diversity would result in a linear increase in reproductive effort, following the pattern shown by most studies reviewed in Forsman and Wennersten (2016). We predicted that the effect of source diversity would be context-dependent, such that the effect of diversity would be stronger for experimental plots composed of Nevada seeds, either by increased importance of sampling effects as discussed above (Figure 5), or because non-additive diversity effects can be magnified in less suitable environmental

conditions as genotypes and phenotypes are pushed to their limits in a novel environmental context (Yachi & Loreau 1999; Drummond & Vellend 2012).

Materials and methods

Study System

Bromus tectorum L. (cheatgrass) is a highly autogamous, winter annual grass native to Eurasia. Cheatgrass rapidly established populations across the contiguous United States following its introduction in the mid 19th century (Mack 1981), achieving dominance by outcompeting native vegetation (Stewart & Hull 1949) and increasing the frequency of fires in its introduced range (Knapp 1996). Cheatgrass is currently a pervasive threat to natural shrub-steppe and mountain ecosystems, covering over 100 million hectares of land in the Western United States (Pellant 1996).

Mechanisms controlling colonization and spread of cheatgrass to new habitats remain poorly understood (Meyer & Leger 2010). Successful establishment and persistence may be due to phenotypic plasticity; cheatgrass exhibits multiple “general-purpose-genotypes” that can thrive across a wide range of environmental conditions (Ramakrishnan et al. 2006). However, local adaptation drives considerable variation among North American populations (Ramakrishnan et al. 2006; Kao et al. 2008; Leger et al. 2009; Merrill et al. 2012). Introduced populations of cheatgrass also exhibit high within-population diversity, likely due to multiple introduction events involving different native genotypes (Kao et al. 2008; Ashley & Longland 2009; Pawlak et al. 2015; Novak & Mack 2016). Introduced cheatgrass populations often do not experience reductions in diversity as compared to native populations, unlike many invasive species (Dlugosh & Parker 2008). Heritable phenotypic variation within and between populations has been shown for ecologically important traits including aboveground biomass (Rice et al. 1992; Scott et al. 2010) and resistance to the fungal pathogen, *Ustilago bullata* (Meyer et al. 2010). Cheatgrass thus serves as a practical and ecologically relevant

system for understanding the role of intraspecific diversity in driving the establishment of colonizing populations.

Sampling

We collected cheatgrass seeds from 32 populations in Colorado and 32 populations in Nevada in June 2015. Cheatgrass seeds collected were technically seed structures that contain one filled caryopsis, potentially one or multiple sterile caryopses, and one or multiple awns (Monty et al. 2013), but for simplicity, these seed structures will be referred to as seeds hereafter. Seeds were collected from 15-20 individuals in each sample population and were stored at room temperature before planting. Within each region (CO or NV), we sampled from source populations haphazardly, but ensured that populations were at least 0.5 km apart (Table 1; mean and [range] of distances for CO: 68.71 km [0.67 - 207.10 km]; NV: 88.80 km [0.84 - 321.25 km]). All populations within each region could reasonably interact (i.e. seeds could be transported between populations via “hitchhiking” on tires of automobiles or on human clothing [Banks & Baker 2011]), allowing us to test if source diversity at a regional scale influenced establishment success.

Common Garden Experiment

We established experimental plots in a common garden in Fort Collins, CO (40.65284°N, 104.99756°W) at Colorado State University’s Agricultural Research, Development and Education Center in August 2015. We used a 2 x 5 factorial design (2 source regions x 5 diversity treatments). Each plot contained 32 seeds from Colorado or Nevada, and represented 1, 2, 4, 8, or 16 source populations. For example, a plot representing 4 source populations from Nevada would contain 8 seeds from each of 4 individuals, each individual originating from a different source population in Nevada. Source populations were randomly drawn from the pool of 32 total populations, without replacement, for each experimental plot. We removed replicates post-hoc to alleviate variance-reduction effects with increasing diversity (Huston

Table 1: Source population GPS locations in Colorado and Nevada.

Region	Location	N	W	Region	Location	N	W
CO	Onaledge	38.8591	104.9019	NV	San Rafael Ranch	39.5461	119.8265
CO	Air Force	38.9983	104.8617	NV	Panther Valley	39.5909	119.8264
CO	IKEA Blvd	40.6076	104.8750	NV	Hoge Road	39.5741	119.8547
CO	Poudre Trail	39.5719	105.1141	NV	Evan’s Creek	39.5510	119.8339
CO	Horsetooth	40.5562	105.1480	NV	Pembroke Dr	39.4980	119.7314
CO	Carter Lake 1	40.3500	105.2090	NV	Hidden Valley	39.4980	119.7090
CO	Carter Lake 2	40.3408	105.2052	NV	Springs Lutheran	39.6205	119.7258
CO	Larimer Co Office	40.3750	105.2277	NV	Pebble Creek	39.6927	119.6733
CO	S County Rd 31	40.3606	105.2679	NV	Pyramid Lake	39.9022	119.5658
CO	CSU Shadehouse	40.5710	105.0806	NV	Coyote Spring	39.7838	119.6829
CO	Remington St	40.5760	105.0760	NV	Lockwood	39.5103	119.6527
CO	Lower Poudre	40.6653	105.2001	NV	Lovelock	40.1891	118.4616
CO	Greyrock	40.6955	105.2859	NV	Oreana	40.3338	118.3182
CO	Hewlett’s Gulch	40.6893	105.3103	NV	Dun Glen	40.6976	118.0444
CO	N County Rd 25G	40.6092	105.1783	NV	Paradise Dunes	41.0909	117.6971
CO	Cleveland Ave	40.3891	105.0743	NV	Paradise Ranchos	41.2512	117.6864
CO	Mountain River Rd	40.2698	105.1173	NV	Rye Patch	40.4673	118.2874
CO	Fairgrounds Lake	40.1604	105.1290	NV	Galen County Est	39.3916	119.7792
CO	Boulder Reservoir	40.0782	105.2381	NV	Galena Creek	39.3405	119.8663
CO	Flatirons	39.9977	105.2847	NV	Bellevue	39.2381	119.8121
CO	Sawhill Ponds	40.0499	105.1829	NV	Genoa	39.0018	119.8365
CO	Northglenn Reservoir	39.8975	105.0116	NV	Fairfield	38.9386	119.7284
CO	Mead	40.2363	104.9990	NV	Davis Creek Park	39.3048	119.8315
CO	Windsor Ditch	40.7116	104.9716	NV	Six Mile Canyon	39.3086	119.6209
CO	Rocky Arsenal	39.8078	104.8956	NV	Silver City	39.2746	119.6519
CO	Watkins	39.7413	104.6606	NV	Curry Street	39.1338	119.7724
CO	Front Range Airport	39.7467	104.5866	NV	Windmill Rd	38.9555	119.6303
CO	Bennett	39.7573	104.4280	NV	CA Border	38.8631	119.7789
CO	Upper Flatirons	39.9866	105.2938	NV	Centennial Park	39.1892	119.7073
CO	ARDEC	40.6529	104.9997	NV	Robb Dr	39.5175	119.8830
CO	Bacon Elementary	40.5036	105.0375	NV	Sommerset	39.5329	119.9151
CO	Douglas Reservoir	40.7014	105.0744	NV	Verdi	39.5159	119.9906

1997) using the following criteria (following Crawford & Whitney 2010): 16 source treatment replicates were allowed no more than 8 sources in common, 8 source treatment replicates were allowed no more than 4 sources in common, 4 source treatment replicates were allowed no more than 2 sources in common, and 2 source treatment replicates were allowed no more than 1 source in common. Each diversity treatment originally had 20 replicates, except for the 1-source plots (monocultures), which had 32 replicates. One of the replicate pairs was randomly chosen to be removed for pairs of replicates that shared more sources than the above criteria. There were 16-32 replicates for each of the region by source treatment combinations, for a total of 204 experimental plots.

A week before introducing cheatgrass seed, the garden was treated with 2,4-D and Roundup herbicide to remove any existing vegetation. Plots were delineated by cut sections of 4-inch diameter drain pipe, spaced 0.75 meters apart, that were partially inserted into the ground with a \sim 1 inch edge of pipe remaining aboveground (following Scott et al. 2010). Experimental plots were randomly assigned to locations in a grid. Seeds for each experimental plot were placed within the drain pipe and then the top of the pipe was covered with a piece of thin mesh to ensure that seeds did not escape. We estimated background cheatgrass contamination by establishing 17 additional control plots for which drain pipe was inserted into the ground and covered, but no cheatgrass seeds were added. The thin mesh was removed after seeds in most of the plots had germinated. Plots and space between plots were weeded biweekly to remove any non-experimental plants until early spring 2016, and were then weeded weekly for the remainder of the experiment.

All aboveground biomass was removed for each plot in mid June 2016. Aboveground biomass samples were dried for at least 72 hours at 60°C. We considered reproductive effort to be the strongest metric of establishment success of founding groups. We thus separated cheatgrass seeds (i.e. seed structures as previously noted) from the vegetative biomass by hand to obtain a biomass that represented reproductive effort. 37% of our experimental plots were infected to some degree with *Ustilago bullata*, a fungal pathogen that infects cheatgrass

during the seedling stage (Fischer 1940), resulting in a proportion of inflorescences that do not produce viable seed. These easily identifiable inflorescences were not included in the seed biomass measurements. We considered this variation important in assessing the role of source diversity in an ecologically complex field environment as there is evidence for heritable phenotypic variation for resistance to *U. bullata* in cheatgrass populations (Meyer et al. 2010), and included infected plots in our analyses.

Statistical Analyses

We constructed a fully Bayesian hierarchical model to explain how source diversity and region influenced establishment success (i.e. number of seeds produced) in our experimental plots. The Bayesian approach was needed to account for uncertainty in observation of response variables and to model spatial structure in the data.

Model structure We used a process model to explain the ecological process of establishment as a function of designed covariates. We used a data model to explain the variance around the unobserved number of seeds produced per plot, which arose from observing seed biomass per plot rather than the response of interest. The process and data models can be represented as:

$$[\boldsymbol{\theta}_d, \boldsymbol{\theta}_p, \mathbf{z} | \mathbf{y}] \propto \underbrace{[\mathbf{y} | \boldsymbol{\theta}_d, \mathbf{z}]}_{\text{data}} \overbrace{[\mathbf{z} | \boldsymbol{\theta}_p]}^{\text{process}} \underbrace{[\boldsymbol{\theta}_d, \boldsymbol{\theta}_p]}_{\text{parameters}}.$$

A vector of observed seed biomass across plots is represented by \mathbf{y} , and \mathbf{z} is a vector of unobserved seed counts across plots. The vector $\boldsymbol{\theta}_d$ represents parameters in the data model and $\boldsymbol{\theta}_p$ is a vector of parameters in the process model. Brackets signify probability densities (all parameters and latent states are continuous) and vertical bars (|) within brackets denote conditionality, such that $[\mathbf{z} | \boldsymbol{\theta}_p]$ means the probability density of \mathbf{z} is conditional on parameters $\boldsymbol{\theta}_p$.

Seed weight and number calibration model We conducted a calibration experiment in which we counted and weighed samples of 100, 150, 250, 500, and 1000 seeds for a subsample of 20 experimental plots (two experimental plots for each region by diversity treatment combination), to inform parameters in the data model, $\boldsymbol{\theta}_d = \alpha_c, \sigma_c^2$. We modeled seed weight (w_o) as a function of seed count (in hundreds of seeds, c_o), for each observation (o), such that

$$\log(w_o) \sim \text{normal}(\log(\alpha_c c_o), \sigma_c^2)$$

where α_c describes the slope of the relationship between seed weight and count (in hundreds of seeds), and σ_c^2 is the calibration variance on the log-scale. We informed the calibration model with vague prior distributions of $\alpha_c \sim \text{normal}(0, 1000)$ and $\sigma_c^2 \sim \text{inverse gamma}(0.001, 0.001)$. The full posterior distribution is described in *APPENDIX A*.

Hierarchical regression model Estimates of the mean and variance from the marginal posterior distributions of $\boldsymbol{\theta}_d$ were used to inform parameters in the hierarchical model describing the relationship between observed seed biomass, y_{ijk} , and the latent variable, z_{ijk} , the true number of seeds produced per plot (in hundreds of seeds),

$$\log(y_{ijk}) \sim \text{normal}(\log(\alpha_c z_{ijk}), \sigma_c^2)$$

where i indexes source region (CO or NV), j indexes source diversity level (1, 2, 4, 8, or 16), and k indexes the number of replicates in each source region by diversity treatment combination, from 1 to n_{ij} . We used a regression process model to delineate the effects of region, diversity, and their interaction on the latent number of seeds produced in each plot, (z_{ijk}),

$$\log(z_{ijk}) \sim \text{multivariate normal}(\log(\mathbf{X}\boldsymbol{\beta}), \boldsymbol{\Sigma})$$

where \mathbf{X} is a design matrix including columns for an intercept and for experimental indicators of source region (0=CO, 1=NV), source diversity (1, 2, 4, 8, or 16), and an interaction

between region and diversity and $\boldsymbol{\beta}$ is a vector of regression coefficients ($\beta_0, \beta_{\text{region}}, \beta_{\text{diversity}}, \beta_{\text{region*diversity}}$) as a component of $\boldsymbol{\theta}_p$. Seed counts per plot were very high (often in the tens of thousands), and were described in hundreds of seeds, thus we allowed the distribution for the number of seeds to be continuous, rather than discrete.

We created a spatial covariance matrix, $\boldsymbol{\Sigma}$, to account for the spatial autocorrelation in the observed data (Figure 6; Mantel's test, $r^2 = 0.154$, $p < 0.001$). The covariance matrix accounted for structured variance (σ_η^2) and unstructured variance (σ_p^2) on the log-scale. We used an exponential covariance function, $R(\phi)$, to describe the spatial dependency,

$$\boldsymbol{\Sigma} = \sigma_p^2 \mathbf{I} + \sigma_\eta^2 R(\phi)$$

$$R(\phi) = \exp(-\mathbf{D}\phi)$$

where \mathbf{D} is a matrix of pairwise distances between experimental plots and ϕ is a scaling parameter. Thus, $\boldsymbol{\theta}_p$ represents $\boldsymbol{\beta}$, σ_η^2 , and σ_p^2 .

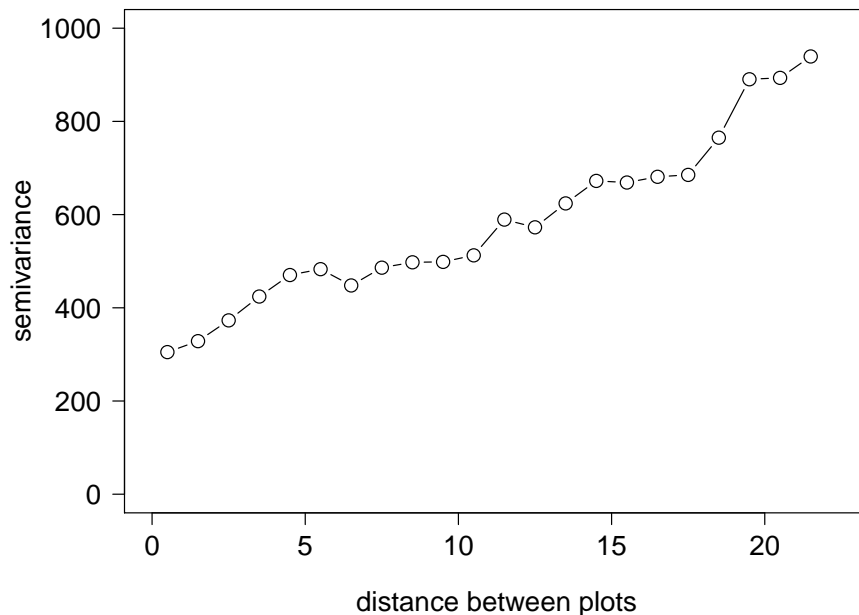


Figure 6: Semivariogram of observed biomass measurements from experimental plots. Distance between experimental plots is measured in meters. The positive slope between distance and semivariance indicates spatial autocorrelation in the data.

We used mean and variance estimates of parameters from the calibration model to inform α_c and σ_c^2 priors in the hierarchical model using moment matching:

$$\alpha_c \sim \text{gamma}(4405.47, 16244.21)$$

$$\sigma_c^2 \sim \text{inverse gamma}(49.25, 1.11)$$

We used vague priors for other variance parameters, regression coefficients, and the shape parameter for the exponential covariance function. The full posterior distribution is described in *APPENDIX A*.

$$\sigma_p^2 \sim \text{inverse gamma}(0.001, 0.001)$$

$$\sigma_\eta^2 \sim \text{inverse gamma}(0.001, 0.001)$$

$$\beta_l \sim \text{normal}(0, 1000), \text{ where } l = 0, \dots, 3$$

$$\phi \sim \text{uniform}(1 \times 10^{-10}, 5)$$

Model estimation and evaluation We approximated marginal posterior distributions for parameters using Markov chain Monte Carlo (MCMC) methods implemented in JAGS using the package ‘rjags’ in the R computing environment (R Core Team). We accumulated 35,000 iterations after 7,500 iterations of burn-in. Convergence was assessed visually from trace plots and by conducting Gelman and Rubin convergence diagnostics (Gelman & Rubin 1992). We conducted posterior predictive checks to evaluate model fit. At each MCMC iteration, we calculated the residual sums of squares for both the observed data and simulated data from the model. We then calculated a Bayesian p-value (P_B): the probability that the residual sums of squares of the observed data was greater than that of the simulated data across converged simulations. Bayesian p-values that are close to 0 or 1 indicate lack of model fit.

Results

Only three of 204 experimental plots (all Nevada, monoculture plots) failed to produce any seeds over the course of the experiment. We used only the extant populations in our statistical model. There was no cheatgrass biomass in any of the 17 control plots, indicating there was likely no background cheatgrass growth (e.g. seedbank) in the common garden.

Visual assessment of trace plots of marginal posterior distributions indicated model convergence for both calibration and hierarchical models. The upper confidence limits of all Gelman-Rubin diagnostics were <1.1 , further verifying convergence. Posterior predictive checks of residual sums of squares showed no lack of model fit (calibration model: $P_B=0.503$; hierarchical model: $P_B=0.496$, Figure 7).

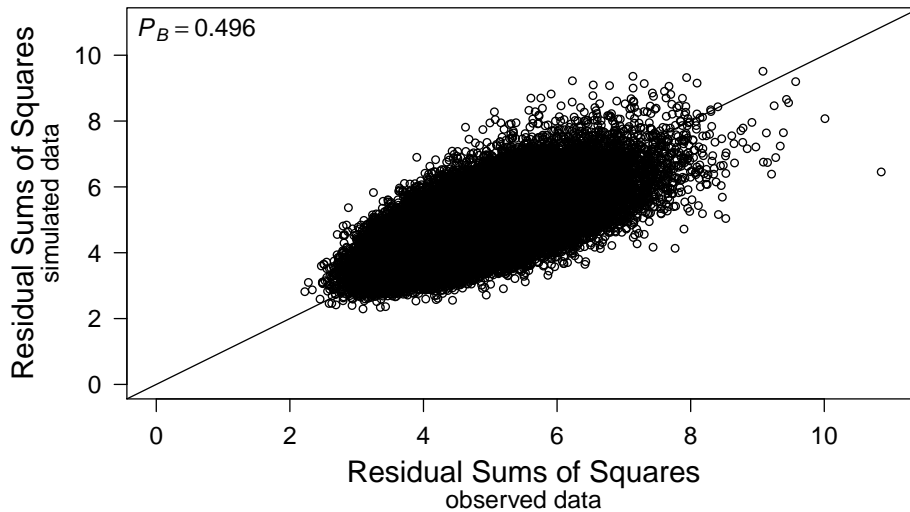


Figure 7: Residual sums of squares for hierarchical model simulated data showed no lack of fit ($P_B = 0.496$).

Source diversity enhanced establishment success of colonizing cheatgrass populations (Figure 8, $\beta_{\text{diversity}} = 3.09$, 95% Bayesian credible interval, BCI: 0.04 - 7.40). The positive effect of increased diversity was not influenced by whether seeds were from Colorado or Nevada ($\beta_{\text{region*diversity}} = 0.41$, 95% BCI: -4.34, 5.15). There was also no main effect of source region on reproductive effort ($\beta_{\text{region}} = -5.84$, 95% BCI: -35.14, 22.71).

Table 2: Summary statistics for parameters of hierarchical model calculated from marginal posterior distributions. σ_η^2 , σ_p^2 , and σ_c^2 summary statistics are reported on the log-scale.

Variable	Mean	Median	95% BCI
β_0	139.68	134.18	(86.08, 227.54)
β_{region}	-5.84	-5.72	(-35.14, 22.71)
$\beta_{\text{diversity}}$	3.09	2.89	(0.04, 7.40)
$\beta_{\text{region}*\text{diversity}}$	0.41	0.42	(-4.34, 5.15)
σ_η^2	0.31	0.20	(0.09, 1.21)
σ_p^2	0.16	0.17	(0.08, 0.23)
ϕ	0.28	0.23	(0.02, 0.84)
α_c^2	0.27	0.27	(0.26, 0.28)
σ_c^2	0.023	0.023	(0.017, 0.031)

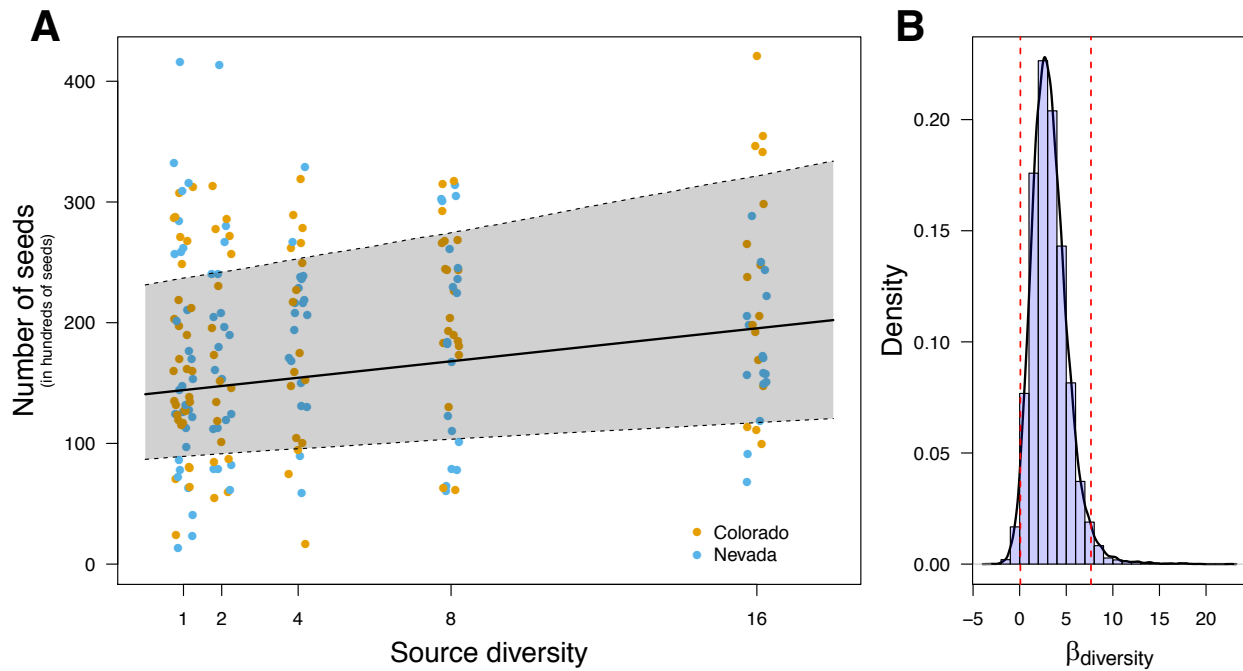


Figure 8: (A) Predicted number of seeds given number of sources. Bolded line represents median of posterior predictive distribution. Points represent individual experimental plots. Shaded area is bounded by 0.025 and 0.975 quantiles. (B) Marginal posterior distribution of $\beta_{\text{diversity}}$. Vertical red, dashed lines are 0.025 and 0.975 quantiles.

Discussion

Recent theoretical and empirical work suggests that a positive relationship between propagule pressure and establishment success is partly driven by increases in intraspecific diversity as propagule pressure increases (Forsman 2014; Forsman & Wennersten 2016). We further validate this pattern by showing that increasing source diversity in founding populations of *B. tectorum* increased reproductive effort at the plot-level. The most diverse founding groups (16 sources) produced 35% more seeds than monocultures. Source region did not influence seed production in founding groups: plots composed of seeds collected from source populations in Colorado exhibited similar reproductive effort across diversity treatments as compared to plots composed of seeds collected from source populations in Nevada. The strong spatial autocorrelation in our biomass measurements suggest that environmental context is very important to establishment success, but acts a microsite scale.

The magnitude of the effect of diversity on reproductive effort found in this experiment was comparable to other empirical studies (reviewed in Tomimatsu et al. 2014). For example, Crawford & Whitney (2010) found a $\sim 17\%$ increase in the number of fruits produced at the population-level when comparing monocultures and polycultures of 8 ecotypes of *Arabidopsis thaliana*. When comparing to similar diversity groups in our experiment (monocultures and founding populations with eight sources), we found a similar effect of diversity on reproductive effort (16% increase). This similarity is surprising given the considerable differences between our experiments. Crawford & Whitney (2010) conducted their experiment in a controlled greenhouse setting, which may not represent the effect of diversity in more complex ecological scenarios. The source genotypes used to construct experimental founding groups in that study were from a wide, global geographic range, which may magnify both additive and non-additive diversity effects (Crawford & Whitney 2010). We find the similar effect sizes in reproductive effort between these two experiments unexpected, particularly given the geographic proximity of source populations used for experimental plots in our experiment. The finding that diversity from source populations collected at a regional scale

can influence establishment success suggest there are consequences to mixing populations via human-mediated transport.

Diversity could have enhanced establishment through either, or both, additive and non-additive effects. Positive effects of diversity on population growth and stability were attributed solely to additive effects of diversity in some previous experiments (Hughes & Stachowicz 2004; Hughes 2014). Most studies, however, find that non-additive effects drive the relationship between increased diversity and establishment success or population persistence. These studies attributed non-additive effects to mechanisms such as complementarity between genotypes (Drummond & Vellend 2012; Smee et al. 2013), sexual recombination (Agashe 2009), niche or resource partitioning (Crawford & Whitney 2010; Wang et al. 2012), and selection effects (Drummond & Vellend 2012), where particular genotypes performed better in mixtures than in monoculture.

Reproductive effort strongly depended on where plots were located in the garden in our experiment, and we only had one monoculture replicate in the common garden for each source population. We could thus not use the standard method of comparing the expected additive yield of a diverse plot replicate based on the productivity of the monoculture plots to the observed reproductive effort of each diverse replicate (Loreau & Hector 2001; Crawford & Whitney 2010). As a result, we were unable to conclusively determine if the positive effect of diversity was additive (via sampling effects) or non-additive (via complementarity or facilitation). The strong spatial dependency in our biomass measurements likely means that non-additive effects such as complementarity played an important role in the increased success of more diverse populations. Sampling effects appear less likely to explain the diversity effect, as failure to produce any seeds for monoculture plots was relatively low (only three monoculture plots out of 64) and there is considerable evidence that cheatgrass populations maintain high within-population diversity after establishment (Kao et al. 2008; Ashley & Longland 2009; Pawlak et al. 2015; Novak & Mack 2016).

The only experimental plots that did not have any individuals survive founding were monocultures composed of seeds from Nevada, suggesting that source region interacted with diversity to influence the probability of successful founding. We predicted that seeds from Colorado would be more likely to be pre-adapted to the recipient environment in the common garden as compared to those from Nevada. However, we did not find that source region influenced establishment success (i.e. number of seeds produced) of successfully founded populations.

One major assumption underlying the prediction that source region would influence reproductive effort was that adaptation would be strong at the between-region scale. Although there are considerable differences in the regional climate between source populations sampled in Colorado and that of populations sampled in Nevada, it is possible that local adaptation was more important in determining establishment success at smaller geographic scale. Scott et al. (2010) found considerable local differentiation across cheatgrass populations that were separated by only tens of kilometers in Utah. Leger et al. (2009) also found evidence of local adaptation in populations that are were geographically close, but differed considerably ecologically and in elevation. Source populations sampled from Colorado in our experiment were, on average, higher in elevation than the those from Nevada, but there was some overlap in the range of elevations sampled between the two regions (mean \pm standard deviation of elevations for CO: 1675.05m \pm 169.42m; NV: 1466.88m \pm 180.11m). This means that within-region variation was possibly considerably greater than between-region variation.

Further, pre-adaptation to the recipient environment also encompasses adaptive phenotypic plasticity. If some individuals exhibited adaptive phenotypic plasticity, differences in establishment success between Colorado and Nevada plots in the common garden might not be realized. There is evidence that phenotypic plasticity can be adaptive in cheatgrass (Meyer et al. 2001; Meyer & Leger 2010), so there is a possibility that pre-adaptive plasticity in Nevada individuals could mask the lack of pre-adaptation to climatic conditions in the Colorado common garden.

We also predicted that intraspecific diversity would be more important in driving establishment success for plots composed of seeds from Nevada as compared to those from Colorado because increased diversity could increase the likelihood of containing a rare pre-adapted individual in a novel environment (additive sampling effect) or could include positive interactions between genotypes that are strengthened as differences between individuals from different sources increase in a stressful environment (non-additive complementarity effect; Drummond & Vellend 2012). The lack of interaction between source region and intraspecific diversity could be due to the unknown differentiation of the 32 source populations sampled in Colorado relative to the differentiation of the 32 populations sampled in Nevada. If source populations were more genetically distinct in one source region as compared to the other, differences in the strength of the diversity effect between source regions might not be realized. Both additive and non-additive diversity effects are expected to most strongly affect establishment success when differences between source populations are largest (Hughes et al. 2008). It is clear, however, that increasing diversity from monoculture to just two source populations resulted in increased survival (i.e. at least one individual in a group survives the founding event) for Nevada populations.

Our finding that the effect of intraspecific diversity on establishment success did not depend on the source region (a proxy for level of pre-adaptation), corroborates some other studies manipulating intraspecific diversity and some other environmental context. Gamfeldt et al. (2005) manipulated cohort diversity of settling oysters both with and without salinity stress, and for both salinity treatments, found a similar positive effect of diversity on establishment success. Experimental *Tribolium* populations exhibited similar increases in population size and stability in the first few generations of founding, regardless of whether they were introduced to a novel or natal media environment (Agashe 2009). Contrary to our expectations and the outcome of our experiment, Drummond & Vellend (2012) found that the positive effect of diversity on *Taraxacum officinale* establishment success was stronger when environmental conditions were favorable (i.e. without disturbance), rather than unfa-

vorable (i.e. with recurring disturbance).

The strong spatial autocorrelation found in our study suggests that spatial scale is critical to interpreting how environmental context modulates the effect of diversity on establishment success. In our experiment, the recipient habitat in the common garden was, on average, equally suitable for established plots composed of seeds from Colorado or to Nevada. However, within the common garden, spatially dependent environmental heterogeneity contributed greatly to observed differences in establishment success across plots. Future studies manipulating intraspecific diversity and other ecological factors important to colonization are needed to resolve in what ecological scenarios diversity will be critical in predicting establishment success within the first few generations of founding.

Conclusion and implications

Source diversity was more important than source region in predicting establishment success within our common garden. We corroborated previous findings that intraspecific diversity is a significant driver of establishment success, and show that increasing source diversity of *Bromus tectorum* founding groups increases reproductive effort in a natural setting. The magnitude of increased reproductive effort of diverse founding groups as compared to monoculture founding groups was larger than anticipated (35% increase between monoculture and most diverse plots) given that source populations used to construct diversity plots were not necessarily genetically distinct. The result that increased source diversity can drastically increase reproductive effort of colonizing cheatgrass populations implies that mixing of cheatgrass populations across the Intermountain West and Great Basin has ecological consequences. Cheatgrass disperses by attaching to animal fur and human clothing (Mack 1981), meaning there is a great potential for individuals to disperse between source populations at a geographic scale similar to the distribution of source populations sampled within regions in this experiment. As managers seek to eradicate, control, or mitigate the spread of cheatgrass populations, reduced mixing of source populations should be considered.

REFERENCES

- Agashe D (2009) The stabilizing effect of intraspecific genetic variation on population dynamics in novel and ancestral habitats. *Am Nat* 174:255-267.
- Ahlroth P, Alatalo RV, Holopainen A, Kumpulainen Y, Suhonen J (2003) Population size and number of source populations enhance establishment success in waterstriders. *Oecologia* 137:617-620.
- Ashley M, Longland W (2009) Assessing cheatgrass genetic diversity and population structure using RAPD and microsatellite molecular markers. *Western North American Naturalist* 69:63-74.
- Baker HG (1965) Characteristics and modes of origin of weeds. Pages 147-148 in H.G. Baker and 301 G.L. Stebbins. *The genetics of colonizing species*. Academic Press, New York.
- Banks ER, Baker WL (2011) Scale and pattern of cheatgrass (*Bromus tectorum*) invasion in Rocky Mountain National Park. *Natural Areas Journal* 31:377-390.
- Bock DG, Caseys C, Cousens RD, Hahn MA, Heredia SM, Hubner S, Turner KG, Whitney KD, Rieseberg LH (2015) What we still don't know about invasion genetics. *Mol Ecol* 24:2277-2297.
- Charmantier A, Grant D (2005) Environmental quality and evolutionary potential: lessons from wild populations. *Proc R Soc B* 272:1415-1425.
- Crawford KM, Whitney KD (2010) Population genetic diversity influences establishment success. *Mol Ecol* 19:1253-1263.
- Dlugosch KM, Parker IM (2008). Founding events in species invasions: genetic variation, adaptive evolution, and the role of multiple introductions. *Conserv Biol* 17:59-72.

- Drummond EBM, Vellend M (2012) Genotypic diversity effects on the performance of *Taraxacum officinale* population increase with time and environmental favorability. *PLoS ONE* 7:e30314.
- Elam DR, Ridley CE, Goodell K, Ellstrandt NC (2007) Population size and relatedness affect fitness of a self-incompatible invasive plant. *P Natl Acad Sci USA* 104:549-552.
- Erfmeier A, Hantsch L, Bruelheide H (2013) The role of propagule pressure, genetic diversity and microsite availability for *Senecio vernalis* invasion. *PLoS ONE* 8:e57029.
- Fischer GW (1940). Host specialization in the head smut of grasses, *Ustilago bullata*. *Phytopathology* 30:991-1017.
- Forsman A, Wennersten L, Karlsson M, Caesar S (2012) Variation in founder groups promotes establishment success in the wild. *Proc R Soc B* 279:2800-2806.
- Forsman, A (2014) Effects of genotypic and phenotypic variation on establishment are important for conservation, invasion, and infection biology. *Proc Natl Acad Sci* 111:302-307.
- Forsman A, Wennersten L (2016) Inter-individual variation promotes ecological success of populations and species: evidence from experimental and comparative studies. *Ecography* 39:630-648.
- Gamfeldt L, Wallen J, Jonsson PR, Berntsson KM, Havenhand JN (2005) Increasing intraspecific diversity enhances settling success in a marine invertebrate. *Ecology* 86:3219-3224.
- Gelman A, Rubin DB. Inference from iterative simulation using multiple sequences. *Stat Sci* 7:457-511.
- Hanley TC, Hughes AR, Williams B, Garland H, Kimbro DL (2016) Effects of intraspecific diversity on survivorship, growth, and recruitment of the eastern oyster across sites. *Ecology* 97:1518-1529.

- Hedge LH, Leung B, OConnor WA, Johnston EL (2013) The interacting effects of diversity and propagule pressure on early colonization and population size. *J Anim Ecol* 83:168-175.
- Hufbauer RA, Facon B, Ravigne V, Turgeon J, Foucaud J, Lee CE, Rey O, Estoup A (2012) Anthropogenically induced adaptation to invade (AIAI): contemporary adaptation to human-altered habitats within the native range can promote invasions. *Evol Appl* 5:89-101.
- Hufbauer RA, Rutschmann A, Serrate B, Vermeil de Conchard H, Facon B (2013) Role of propagule pressure in establishment success: disentangling the relative importance of demographic, genetic and habitat effects. *J Evol Biol* 26:1691-1699.
- Hughes AR, Stachowicz J (2004) Genetic diversity enhances the resistance of a seagrass ecosystem to disturbance. *Proc Natl Acad Sci* 101:8998-9002.
- Hughes AR, Inoye BD, Johnson MTJ, Underwood N, Vellend M (2008) Ecological consequences of genetic diversity. *Ecol Lett* 11:609-623.
- Huston, MA (1997) Hidden treatments in ecological experiments: re-evaluating the ecosystem function of biodiversity. *Oecologia* 110:449-460.
- Kao RH, Brown CS, Hufbauer RA (2008) High phenotypic and molecular variation in downy brome (*Bromus tectorum*). *Invasive Plant Sci Manag* 1:216-225.
- Knapp PA. Cheatgrass (1996) (*Bromus tectorum* L.) Dominance in the Great Basin Desert: History, persistence, and influence to human activities. *J Range Manage* 25:194-201.
- Lande R (1988) Genetics and demography in biological conservation. *Science* 241:1455-1460.
- Leger EA, Espeland EK, Merrill KR, Meyer SE (2009) Genetic variation and local adaptation at a cheatgrass (*Bromus tectorum*) invasion edge in western Nevada. *Mol Ecol* 18:4366-4379.

- Lockwood JL, Cassey P, Blackburn T (2005) The role of propagule pressure in explaining species invasions. *Trends Ecol Evol* 20:223-228.
- Loreau M, Hector A (2001) Partitioning selection and complementarity in biodiversity experiments. *Nature* 412:72-76.
- MacArthur RH, Wilson EO (1967) The theory of island biogeography. Princeton, NJ: Princeton University Press.
- Mack RN (1981) Invasion of *Bromus tectorum* L into western North America: An ecological chronicle. *Agro-Ecosystems* 7:145-165.
- Mayr E (1965) The nature of colonization in birds. Pages 29-47 in HG Baker and GL Stebbins, editors. The genetics of colonizing species. Academic Press, New York.
- Merrill KR, Meyer SE, Coleman CE (2012) Population genetic analysis of *Bromus tectorum* indicates recent range expansion may be facilitated by specialist genotypes. *Am J Bot* 99:529-537.
- Meyer SE, Nelson DL, Clement S (2001) Evidence for resistance polymorphism in the *Bromus tectorum-Ustilago bullata* pathosystem: implications for biocontrol. *Canadian Journal of Plant Pathology* 23:19-27.
- Meyer SE, Nelson DL, Clement S, Ramakrishnan A (2010) Ecological genetics of the *Bromus tectorum* (Poaceae)-*Ustilago bullata* (Ustilaginaceae) pathosystem: A role for frequency-dependent selection? *Am Journal Bot* 97:1304-1312.
- Meyer SE, Leger EA (2010) Inbreeding, genetic variation, and invasiveness: The strange case of *Bromus tectorum*. *Rangelands* 32:6-11.
- Monty A, Brown CS, Johnston DB (2013) Fire promotes downy brome (*Bromus tectorum* L.) seed dispersal. *Biol Invasions* 15:1113-1123.

- Novak SJ, Mack RN (2001) Tracing plant introduction and spread: Genetic evidence from *Bromus tectorum* (cheatgrass). *BioScience* 51:114-122.
- Novak SJ, Mack RN (2016) Mating system, introduction and genetic diversity of *Bromus tectorum* in North America, the most notorious product of evolution within *Bromus* section *Genea*. In Exotic Brome grasses in arid and semiarid ecosystems of the Western US. pp. 193-221.
- Nuñez MA, Medley KA (2011) Pine invasions: climate predicts invasion success; something else predicts failure. *Divers Distrib* 17:703-713.
- Parker IM, Rodriguez J, Loik ME (2003) An evolutionary approach to understanding the biology of invasions: local adaptation and general-purpose genotypes in the weed *Verbascum thapsus*. *Conserv Biol* 17:59-72.
- Pawlak AR, Mack RN, Busch JW, Novak SJ (2015) Invasion of *Bromus tectorum* (L.) into California and the American Southwest: rapid, multi-directional and genetically diverse. *Biol Invasions* 17:287-306.
- Pellant M (1996) Cheatgrass: the invader that won the West. Unpublished Report. Interior Columbia Basin Ecosystem Management Project. Bureau of Land Management, Idaho State Office. 23 p.
- R Core Team *R: A Language and Environment for Statistical Computing* (R Foundation for Statistical Computing, 2015) <https://www.R-project.org/>.
- Ramakrishnan AP, Meyer SE, Fairbanks DJ, Coleman CE (2006) Ecological significance of microsatellite variation in western North American populations of *Bromus tectorum*. *Plant Species Biol* 21:61-73.

- Rice KJ, Black RA, Rademaker G, Evans RD (1992) Photosynthesis, growth, and biomass allocation in habitat ecotypes of cheatgrass (*Bromus tectorum*). *Functional Ecology* 6:32-40.
- Scott JW, Meyer SE, Merrill KR, Anderson VJ (2010) Local population differentiation in *Bromus tectorum* L. in relation to habitat-specific selection regimes. *Evol Ecol* 24:1061-1080.
- Simberloff D (2009) The role of propagule pressure in biological invasions. *Annu Rev Ecol Syst* 40:81-102.
- Smee SL, Overath D, Johnson KD, Sanchez JA (2013) Intraspecific variation influences natural settlement of eastern oysters. *Oecologia* 173:947-953.
- Sol D (2007) Do successful invaders exist? Pre-adaptations to novel environments in terrestrial invertebrates. In *Ecological Studies, 193: Biological Invasions*. (ed W Nentwig).
- Stewart G, Hull AC (1949) Cheatgrass (*Bromus tectorum* L.) — An ecologic intruder in Southern Idaho. *Ecology* 30:58-74.
- Szűcs M, Melbourne BA, Tuff T, Hufbauer RA (2014) The roles of demography and genetics in the early stages of colonization. *Proceedings of the Royal Society B* 281: 20141073.
- Thompson K, Stewart AJA (1981) The measurement and meaning of reproductive effort in plants. *Am Nat* 117:205-211.
- Tilman D, Wedin D, Knops J (1996) Productivity and sustainability influenced by biodiversity in grassland ecosystems. *Nature* 379:718-720.
- Tomimatsu H, Nakano K, Yamamoto N, Suyama Y (2014) Effects of genotypic diversity of *Phragmites australis* on primary productivity and water quality in an experimental wetland. *Oecologia* 175:163-172.

Yachi S, Loreau M (1999) Biodiversity and ecosystem productivity in a fluctuating environment: the insurance hypothesis. *Proc Natl Acad Sci USA*. 96:1463-1468.

APPENDIX A : POSTERIOR DISTRIBUTIONS

Notation

We use bracket notation when writing the derivations of posterior distributions. Anything enclosed in brackets ($[]$) is a probability (discrete) or a probability density (continuous). A bar ($|$) within brackets denotes conditionality, such that $[y|\theta]$ means y is conditional on the parameter θ . The posterior distribution is proportional to the likelihood multiplied by the prior distribution(s).

$$\overbrace{[\theta | y]}^{\text{posterior}} \propto \overbrace{[y | \theta]}^{\text{likelihood}} \overbrace{[\theta]}^{\text{prior}}$$

Calibration model

We expressed the full posterior distribution of the calibration model as

$$[\alpha_c, \sigma_c^2 | \mathbf{w}] \propto \prod_{o=1}^{120} [w_o | \alpha_c, \sigma_c^2] [\alpha_c] [\sigma_c^2]$$

where w_o represents total seed weight for samples indexed o (from 1 to 120), α_c describes the slope of the relationship between seed weight and seed count (in hundreds of seeds), and σ_c^2 is the calibration variance.

Hierarchical regression model

We expressed the full posterior distribution of the hierarchical regression model as

$$\begin{aligned}
 [\boldsymbol{\beta}, \phi, \sigma_p^2, \sigma_\eta^2, \mathbf{z}, \alpha_c, \sigma_c^2 \mid \mathbf{y}] &\propto \prod_{i=1}^2 \prod_{j=1}^5 \prod_{k=1}^{n_{ij}} \prod_{l=0}^3 [y_{ijk} \mid \alpha_c z_{ijk}, \sigma_c^2] [z_{ijk} \mid \beta_l, \sigma_p^2 \mathbf{I} + \sigma_\eta^2 \mathbf{R}(\phi)] \\
 &\times [\beta_l] [\phi] [\sigma_p^2] [\sigma_\eta^2] [\alpha_c] [\sigma_c^2]
 \end{aligned}$$

$$\mathbf{R}(\phi) = \exp(-\mathbf{D}\phi)$$

where y_{ijk} is the observed seed biomass per plot and z_{ijk} is the latent number of seeds (in hundreds of seeds) produced per plot, indexed by i (source region: Colorado or Nevada), j (source diversity: 1, 2, 4, 8, or 16 sources), and k (replicate from 1 to n_{ij}). The parameter α_c describes the relationship between seed weight and count (in hundreds of seeds) and σ_c^2 is the calibration variance. The regression parameters are represented as β_l , indexed from 0 to 3. There is an unstructured variance component, σ_p^2 , that explains variance that does not coincide with the distance between plots and a variance component, σ_η^2 , that explains variance that does coincide with the distance between plots. The covariance matrix, $\mathbf{R}(\phi)$, is described using a matrix of pairwise distances between plots, \mathbf{D} , and a scaling parameter, ϕ .