Assessing Potential Seed Transfer Zones for Five Forb Species from the Great Basin Floristic Region, USA

Author(s): Andrea T. Kramer , Daniel J. Larkin and Jeremie B. Fant
Source: Natural Areas Journal, 35(1):174-188.
Published By: Natural Areas Association
DOI: http://dx.doi.org/10.3375/043.035.0119
URL: http://www.bioone.org/doi/full/10.3375/043.035.0119

RESEARCH ARTICLE

Assessing Potential Seed Transfer Zones for Five Forb Species from the Great Basin Floristic Region, USA

Andrea T. Kramer[1,2]

[1]Chicago Botanic Garden
Department of Plant Science
and Conservation
1000 Lake Cook Road
Glencoe, IL 60022

Daniel J. Larkin[1]
Jeremie B. Fant[1]

[2] Corresponding author:
akramer@chicagobotanic.org

Natural Areas Journal 35:174–188

ABSTRACT: For plant species important in ecological restoration, seed transfer zones have been developed to maximize the probability that sown seed will germinate, establish, persist, and reproduce without negatively impacting the genetic composition of remnant plant populations. However, empirically based seed transfer zones have not been developed for most species. In their absence, maps based on ecological or climatic variables have been suggested as proxies. In the United States, these maps typically include the Environmental Protection Agency's Levels III and IV Ecoregion maps and the US Forest Service's Provisional Seed Zones. Maps of different spatial scales represent a compromise between economic and ecological considerations; those that delineate larger seed transfer zones are less costly to implement but impose more risk of poor adaptation to local conditions. To test the relative suitability of each map in delineating seed transfer zones, we conducted common garden experiments using five forb species found throughout the Great Basin and measured variation in traits thought to influence plant performance. We distinguished between environmentally and genetically controlled variation in measured traits and assessed how well this variation was explained by different candidate seed transfer zones. We found significant, population-level variation in all species for most measured traits. All tested seed transfer zones significantly explained some of this variation, but the proportion explained generally decreased with increasing zone size. Results suggest the intersection of Provisional Seed Zones and Level III Ecoregions was the best proxy for formal seed transfer zones developed based on common garden studies. This spatial scale captured 80% of the variation among source populations on average, and represents a viable compromise between ecological and economic considerations.

Index terms: common garden, *Eriogonum*, Great Basin, *Penstemon*, seed transfer zone

INTRODUCTION

Plant populations are often adapted to local environmental conditions (Hufford et al. 2008; Kronholm et al. 2012), which has important implications for sourcing native plant materials for restoration (McKay et al. 2005). Recent meta-analyses have shown that local plants outperform nonlocal plants at their home site in a majority (71%) of studies (Leimu and Fischer 2008; Hereford 2009), with the fitness of locally sourced plants 45% greater, on average, than nonlocally sourced plants (Hereford 2009). Restoration efforts that source plant material from environments as similar to the restoration site as possible are thus likely to have better outcomes (Joshi et al. 2001; Montalvo and Ellstrand 2001; Bennington et al. 2012). However, this can be complicated for species found in habitats undergoing rapid environmental change or in small populations (e.g., fewer than 1000 individuals), as they are less likely to exhibit local adaptation (McKay et al. 2005; Leimu and Fischer 2008; Shaw and Etterson 2012).

Common garden studies are used to identify variation between populations for potentially adaptive traits. Using differences identified within these studies, it is possible to delineate seed transfer zones, which are geographic regions where seed can be moved with minimal risk of being poorly adapted (Johnson et al. 2004). The primary goal of seed transfer zones is to help land managers maximize the potential that sown seed will germinate, establish, persist, and reproduce at a restoration site without negatively impacting the genetic composition of remnant native plant populations (e.g., outbreeding depression) (McKay et al. 2005). Seed transfer zones were first developed in the 1960s for timber species after unexpected losses and declines in productivity associated with the use of nonlocal material were seen in reforestation efforts years or sometimes even decades after establishment (Millar and Libby 1989; Johnson et al. 2004).

The extent and scale of local adaptation varies greatly among species, as it is determined by the strength and direction of natural selection and gene flow (Endler 1973). This means that seed transfer zones are not universal but must be developed on a species-by-species basis (Hufford and Mazer 2003; Johnson et al. 2004). For example, common garden studies carried out by the US Forest Service led to the delineation of very different seed transfer zones for two wind-pollinated timber species with overlapping distributions: western redcedar (*Thuja plicata* Donn ex D. Don) has 4 large seed transfer zones in Oregon, while the more narrowly adapted

Douglas-fir (*Pseudotsuga menziesii* (Mirb.) Franco) has 16 much smaller seed transfer zones covering the same area (Johnson et al. 2004). This difference in seed transfer zone size has implications for the costs and challenges associated with developing, producing, and using plant material for restoration (Ying and Yanchuk 2006). For example, sourcing species like Douglas-fir is more complicated and expensive than species like western redcedar (Johnson et al. 2004; G. R. Johnson et al. 2010).

Seed transfer zone development is time-consuming and expensive, thus efforts to date have focused on species with high economic and/or restoration value. This includes timber species and a growing number of common grasses and forbs (Erickson et al. 2004; St.Clair et al. 2005; R.C. Johnson et al. 2010; Johnson et al. 2012; Johnson et al. 2013; St. Clair et al. 2013). Still, seed transfer zones have not been developed for most species used in restoration. In their absence, land managers often conservatively source seed from the nearest source available (Saari and Glisson 2012). However, economic constraints make the use of very local seed sources for restoration challenging, particularly for regions where restoration is often needed over a large geographic scale, as in western North America (BLM 2000). For species without seed transfer zones, and in regions where locally sourced seed may be unavailable or prohibitively expensive, use of ecological boundaries as a proxy for seed transfer zones has been advocated (MacKay 1993; Johnson et al. 2004; G.R. Johnson et al. 2010).

In the United States, several existing ecological mapping systems could potentially serve as proxies for formal (empirically evaluated) seed transfer zones. One of the most extensive systems comes from the US Environmental Protection Agency (EPA), which has developed maps dividing the country into "ecoregions," or geographic areas of similar geology, soils, climate, hydrology, vegetation, and human land-use types (Omernik 1987). Four Ecoregional scales (Levels I through IV) represent increasingly fine-grained resolution and decreasing spatial extent. Level III and IV Ecoregions in particular have been suggested as potential proxies for seed transfer zones. Level III Ecoregions, large continuous zones that capture coarse-scale environmental variation, are often considered as minimum potential seed transfer zones. Level IV Ecoregions are much smaller, noncontinuous zones subsumed within each Level III Ecoregion.

As an alternative to the EPA's Ecoregions, the US Forest Service has recently developed Provisional Seed Zones (hereafter PSZ I), which incorporate high-resolution climate data on mean monthly minimum winter temperature, and aridity to create putative seed transfer zones for unstudied species (Bower et al. 2014). Bower et al. (2014) also recommend that Level III Ecoregions be used with PSZ I to further differentiate climatically similar regions existing within ecologically distinct areas (hereafter PSZ II). The PSZ II framework, thus, leads to zones of greater resolution and smaller size compared with PSZ I. Both PSZ I and PSZ II are of intermediate size between Level III and Level IV Ecoregions, and, thus, of intermediate cost and complexity to implement for restoration. For example, Nevada has 5 Level III Ecoregions containing 8 PSZ I, 16 PSZ II, and 43 Level IV Ecoregions.

To date, few studies have tested the extent to which these different proxies for seed transfer zones capture genetic variation that is potentially important in terms of restoration outcomes (but see Erickson et al. 2004; Miller et al. 2011; Bower et al. 2014). To address this, we established common garden experiments for five common forb species collected from multiple source populations across the Great Basin floristic region. Within common gardens, we then measured variation in traits expected to influence plants' establishment, survival, and reproduction, including seed germination, plant morphology, production, and phenology. We used two common garden sites to distinguish when variation in measured traits was environmentally controlled (phenotypic plasticity), genetically controlled (potentially adaptive variation), or due to interactions between these two factors. We then asked how well measured variation was explained under different candidate seed transfer zones, assessing the ability of increasingly coarse zones to adequately capture variation in source populations.

METHODS

Study Species and Populations

Eriogonum and *Penstemon* are two of North America's largest endemic genera. Many of the more common species in both genera are increasingly included in restoration seed mixes, but empirically based guidance on seed transfer zones is absent. Our five study species included *E. ovalifolium* Nutt. var. *purpureum* (Nutt.) Durand, *E. microthecum* Nutt. var. *laxiflorum* Hook, *P. deustus* Douglas ex Lindl. var. *pedicellatus* M.E. Jones, *P. pachyphyllus* A. Gray ex Rydb. var. *congestus* (M.E. Jones) N.H. Holmgren, and *P. rostriflorus* (Kellogg). All study species are animal-pollinated perennial forbs with gravity-dispersed seeds. They are relatively common and widely distributed throughout the Great Basin and elsewhere in the western United States (Kartesz 1999), occurring almost exclusively in sagebrush-steppe habitat at a range of mid- to high-elevations on mountain ranges. The extent of gene flow among populations of our *Eriogonum* study species is unknown. In the three *Penstemon* species, the extent of gene flow differs markedly, which is likely due to different primary pollinators: *P. rostriflorus* is hummingbird-pollinated and has extensive gene flow across the Great Basin, while *P. pachyphyllus* and *P. deustus* are both bee-pollinated and have much more restricted gene flow (Kramer et al. 2011).

Seed was collected in 2003 (three *Penstemon* species) and 2004 (two *Eriogonum* species). Six to eight study populations, each with at least 150 individuals, were identified for each species, representing the geographic and climatic ranges of their distribution in the Great Basin (Table 1, Figure 1, and Appendix 1). In general, we followed Seeds of Success protocols (BLM 2012), with the additional stipulation that we kept seed separated by maternal line. For each of 33 populations, we collected seed from 50 maternal plants, ensuring that sampled plants were at least 5 m apart to

Table 1. Number of source populations, potential seed transfer zones, average plants measured per population for each common garden between 2004 and 2008, and total plants measured for all five Great Basin study species. In general, source populations are grouped into fewer zones when moving from Level IV Ecoregions to Level III Ecoregions. However, this was not always the case, with some potential zones grouping source populations in exactly the same way (indicated with *). The same populations were used in the common garden and seed germination study for all species except *E. ovalifolium*, where one population was excluded from the seed germination study due to lack of available seeds (numbers shown in parenthesis).

Species Name	Source Populations	Level IV Ecoregions	PSZ II	PSZ I	Level III Ecoregions	Avg. Plants /Pop/CG	Total Plants Measured
Eriogonum microthecum	6	6*	5	5*	2	14	169
Eriogonum ovalifolium	6 (5)	5 (4)	4*	4*	3	17	206
Penstemon deustus	6	4	6*	4	2	17	204
Penstemon pachyphyllus	7	4	5	4**	2	12	171
Penstemon rostriflorus	8	6	5	3	2	16	249

* Potential seed transfer zone did not provide fewer groups than source population (*E. microthecum* : Level IV Ecoregions, *P. deustus* : PSZ II) or did not group source populations into fewer zones as expected, so excluded from results presented in Figures 3 and 4 (*E. microthecum* : PSZ I, *E . ovalifolium* : PSZ I [Figures 3 and 4] and PSZ II [Figure 3 only]).

** Number of zones in PSZ I = Level IV Ecoregions, but different source populations are assigned to each, so not excluded from results.

avoid likely siblings. Collected seed was cleaned and stored at room temperature and 20% relative humidity in laboratory facilities at Chicago Botanic Garden (CBG) prior to use.

Seed Germination

Seed germination studies were conducted at CBG in 2004 (three *Penstemon* spp.) and 2005 (two *Eriogonum* spp.) to determine population-level variation in germination under different growing conditions. For each population, a bulk collection was created with an equal number of seeds from each of 25 maternal lines. Bulked seed was divided into 12 replicates of 50 seeds each, for a total of 600 seeds per study population per species. All seeds were washed in 0.25% sodium hypochlorite (bleach) solution for one minute and rinsed twice in deionized water before being placed into 5.5-cm petri dishes with two layers of No. 5 Whatmann filter paper and dampened with deionized water. Petri dishes were placed in Percival incubators (Model I-36VL, Perry IA) that simulated winter (8 h at 10 ºC with light, 16 h at 4 ºC dark) and spring (12 h at 10 ºC dark, 12 h at 20 ºC with light) germination conditions. Petri dishes were randomly assigned to one of six germination treatments simulating different winter lengths (no winter, and 8, 10, 12, 14, and 16 weeks of winter). For the no winter treatment, two dishes were placed directly in spring germination conditions. The remaining 10 dishes were placed in winter germination conditions for eight weeks, at which point two petri dishes were moved every two weeks from winter to spring conditions to create each winter length treatment. All replicates were monitored three times per week, with any germinants (radicle emergence >1 mm) recorded and discarded following Meyer et al. (1995). Filter paper was kept moist throughout the study with regular application of deionized water. At 20 weeks, remaining seed was counted and recorded as either nonviable (empty or mushy) or viable but dormant (solid and indistinguishable from untreated seed).

Common Garden

Seeds were germinated at CBG under greenhouse conditions in the winters of 2004 (*Penstemon*) and 2005 (*Eriogonum*). The following springs, seedlings were transported to two common garden sites within the Great Basin floristic region: Utah Botanical Center near Salt Lake City, Utah (hereafter SLC), and Boise State University in Boise, Idaho (hereafter Boise). For each species, at least one seedling from each of 20 maternal lines per population (when available; see Table 1) was planted in randomized plots at both common garden sites. The common garden sites varied considerably in climatic characteristics and soil composition, with more natural rainfall and clay alluvial soil at SLC and drier conditions and fine sandy soil at Boise. Both sites were tilled prior to planting. Supplemental irrigation was added as needed, predominantly during establishment, and weeds were controlled by hand. From spring of the first year to fall of the third year, we measured quantitative traits that were highly heritable in related species (Mitchell and Shaw 1993), were thought to be associated with reproductive success or have adaptive significance, or which were highly variable between populations. Our measurements are grouped into three categories:

1. Morphology: Morphological traits measured for *Penstemon* spp. included leaf area, stem diameter, and stem architecture (the portion of flowering stems dedicated to floral display). For *Eriogonum* spp., leaf area, stem architecture, and inflorescence height were recorded.

2. Production and phenology: Six traits were measured across all *Eriogonum* and *Penstemon* species: number of flowering stems, number of flowers per stem, flowering phenology, maximum size (cubic centimeters; a volume measure), summer growth (changes in plant volume from spring to fall), and winter growth (changes

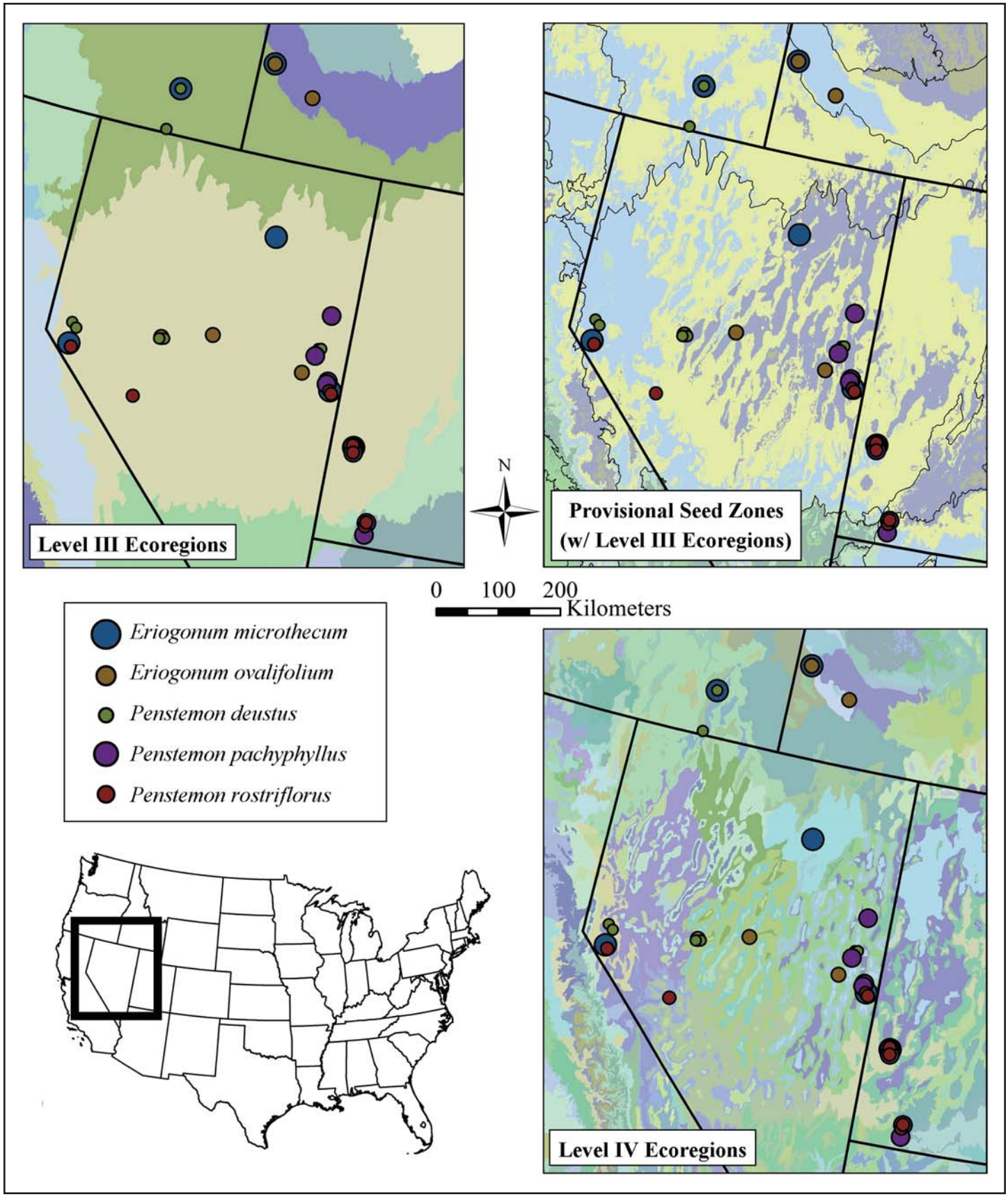

Figure 1. In 2003 and 2004, seeds from 33 source populations were collected from the Great Basin floristic region for common garden studies. Source populations are shown by species for all candidate seed transfer zones tested (Level III Ecoregion, Provisional Seed Zone, and Level IV Ecoregion).

in plant volume from fall to spring).

3. Floral morphology: For *Penstemon* species only, three measures of floral morphology were calculated from the average of three flowers per plant (mouth diameter, anther exsertion, and corolla length). Flowers were collected the day after opening and preserved in ethyl alcohol until measurements could be performed at CBG.

Data Analysis

Seed collection sites for each species were assigned to their Ecoregion (Level III and Level IV), and Provisional Seed Zones with and without recommended Level III Ecoregion divisions (PSZ II and PSZ I, respectively) using the sp and rgdal packages in R 2.15.3 (Bivand, Keitt, and Rowlingson 2013; Bivand, Pebesma, and Gomez-Rubio 2013; R Development Core Team 2013).

Seed germination data were analyzed with survival analysis, a temporally explicit approach for modeling whether or not some event occurs (in this case, germination) and the time it takes to reach that event (Miller 1997). Survival models were developed separately for each species, with all treatments combined, and days to germination (or failure to germinate) as the dependent variable. Regression of survival models was then used to test for significance of and variance explained by effects of source population (or different potential seed transfer zones) and germination treatment. These analyses were performed using the survival and rms packages in R 2.15.3 (Therneau 2012; R Development Core Team 2013; Harrell Jr. 2014).

Common garden data were first analyzed with two-way ANOVA to test for effects of source population, common garden, and their interactions for each measured trait for each species. We also used multivariate methods to characterize overall responses of plants across multiple traits. First, the position of individual plants in multidimensional trait space was quantified for each species using Gower dissimilarity (Gower 1971). Gower distance is suitable for datasets that comprise a mix of quantitative, semiquantitative, and categorical data, and can be used to handle cases where some data points are missing (Laliberté and Legendre 2010). Using Gower dissimilarity as the dependent variable, we then partitioned the variance explained by different predictors (source population or different potential seed transfer zones). The joint effects of source population and common garden were partitioned using adjusted R^2 values derived from redundancy analysis (RDA) (Legendre and Legendre 2012). For different single levels of geographic resolution (source population vs. Level IV Ecoregion, PSZ II, PSZ I, or Level III Ecoregion), adjusted R^2 was calculated using distance-based RDA (dbRDA) (Legendre et al. 2011). These analyses were performed using the FD and vegan packages in R 2.15.3 (Laliberté and Shipley 2011; Oksanen et al. 2013; R Development Core Team 2013).

RESULTS

Site Characterization

Our 33 study populations were located in 4 Level III Ecoregions, 8 Provisional Seed Zones (PSZ I), 12 Provisional Seed Zones (including Level III Ecoregions; PSZ II) and 15 Level IV Ecoregions (Table 1). In general, the number of potential seed transfer zones for each species decreased in the following order: Level IV Ecoregions, PSZ II, PSZ I, and Level III Ecoregions.

Seed Germination

Survival analysis showed highly significant differences associated with source population, germination treatment, and their interactions ($P << 0.0001$ for all species). There was large variation among populations in total germination percentage and rate for all species (Figure 2). Germination response ranged from 0% to 100%, depending on simulated winter length and source population; a few populations reached nearly 100% germination across all winter lengths (including almost all *E. ovalifolium* populations, and *P. pachyphyllus* populations from Ecoregion 20), but most attained maximum germination only under specific conditions (Appendix 2). Source populations in the two *Eriogonum* spp. varied mostly in *rate* of germination rather than total germination. *Eriogonum ovalifolium* germinated more consistently regardless of source population or germination treatment, yet even this species showed highly significant variation in response to different winter lengths. For *E. microthecum*, a majority of seeds remained dormant at the end of our study regardless of winter treatment for four of the six study populations (Figure 2). In one population of *E. microthecum*, winter conditions induced dormancy in seeds, while seeds from another population did not germinate until they were exposed to at least 14 weeks of winter conditions (Appendix 2). In all three *Penstemon* spp., some populations germinated without any winter conditions, but the majority required at least 8 weeks of winter conditions. In most populations, half or more of all viable seeds were dormant at the end of our study regardless of winter treatment (Figure 2, Appendix 2).

The variability in seed germination responses was significantly explained by all potential seed transfer zones in every study species (combined across germination treatments; Figure 3). Source population best explained variation, and analyses using increasingly large potential seed transfer zones (Level IV Ecoregion < PSZ II < PSZ I < Level III Ecoregion) generally explained less (but still significant) variation. This was particularly true in *E. microthecum* and *P. rostriflorus*, where Level III Ecoregions explained around 33% less variation than source population for each species. The primary exception was *E. ovalifolium*, where all potential seed transfer zones were equally effective at capturing this variation, although this is likely due to there being little population-level variability in germination (Figure 2). In *P. deustus*, Level IV Ecoregions surprisingly captured less than half of the variation in source populations (Figure 3), while PSZ I was much more effective, capturing 77% of the variation in source populations (PSZ II was equivalent to source population, so was excluded). In *P. pachyphyllus*, all seed transfer zones tested captured nearly all of the variation in source population except PSZ I, which captured only 63% of source population variation.

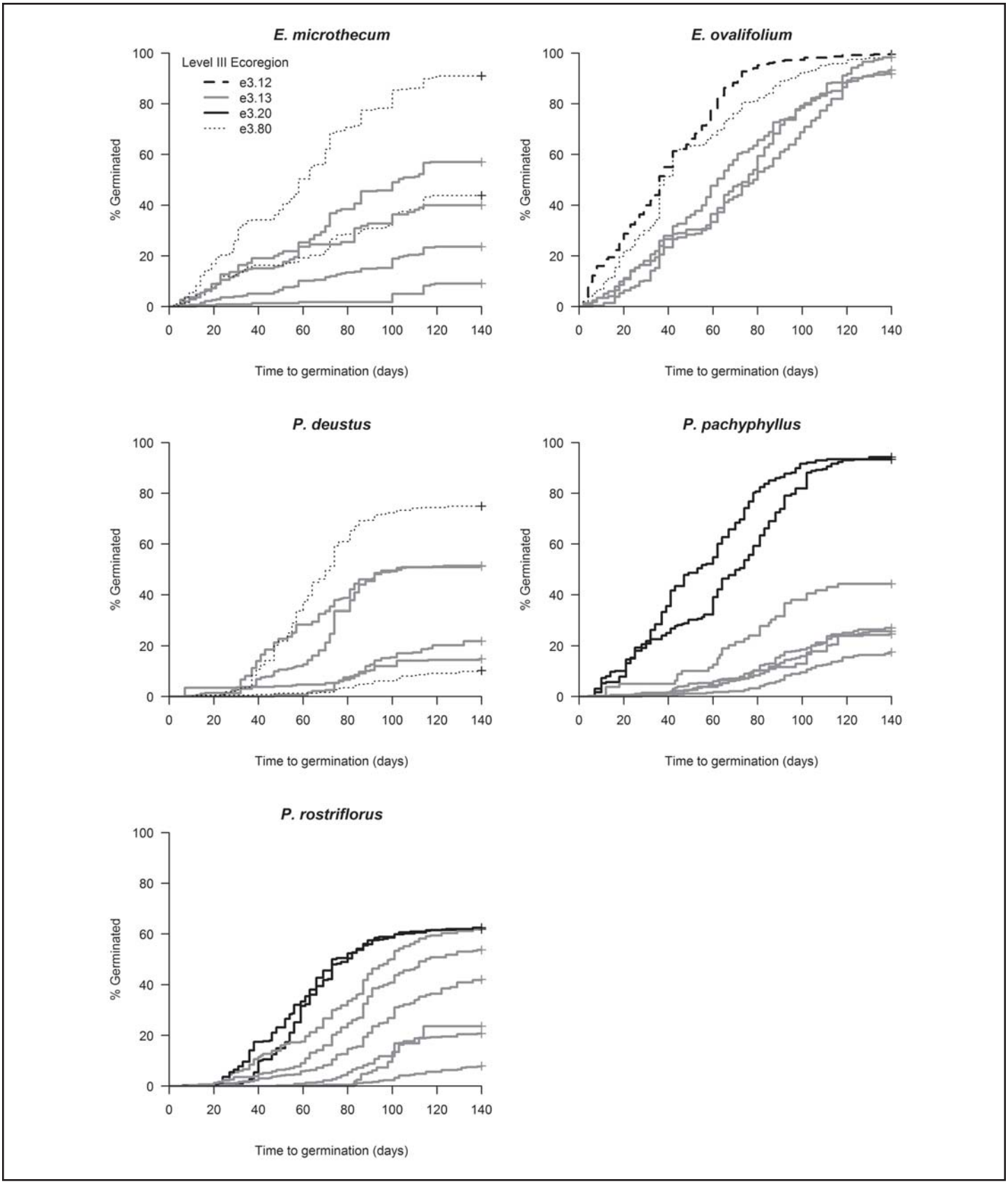

Figure 2. Germination curves showing total germination by source population for each species (data combined across germination treatments). Line styles indicate the Level III Ecoregion where each source population is located. Codes correspond to Level III Ecoregion colors on Figure 1 as follows: e3.12 = purple; e3.13 = tan; e3.20 = gray; e3.80 = olive.

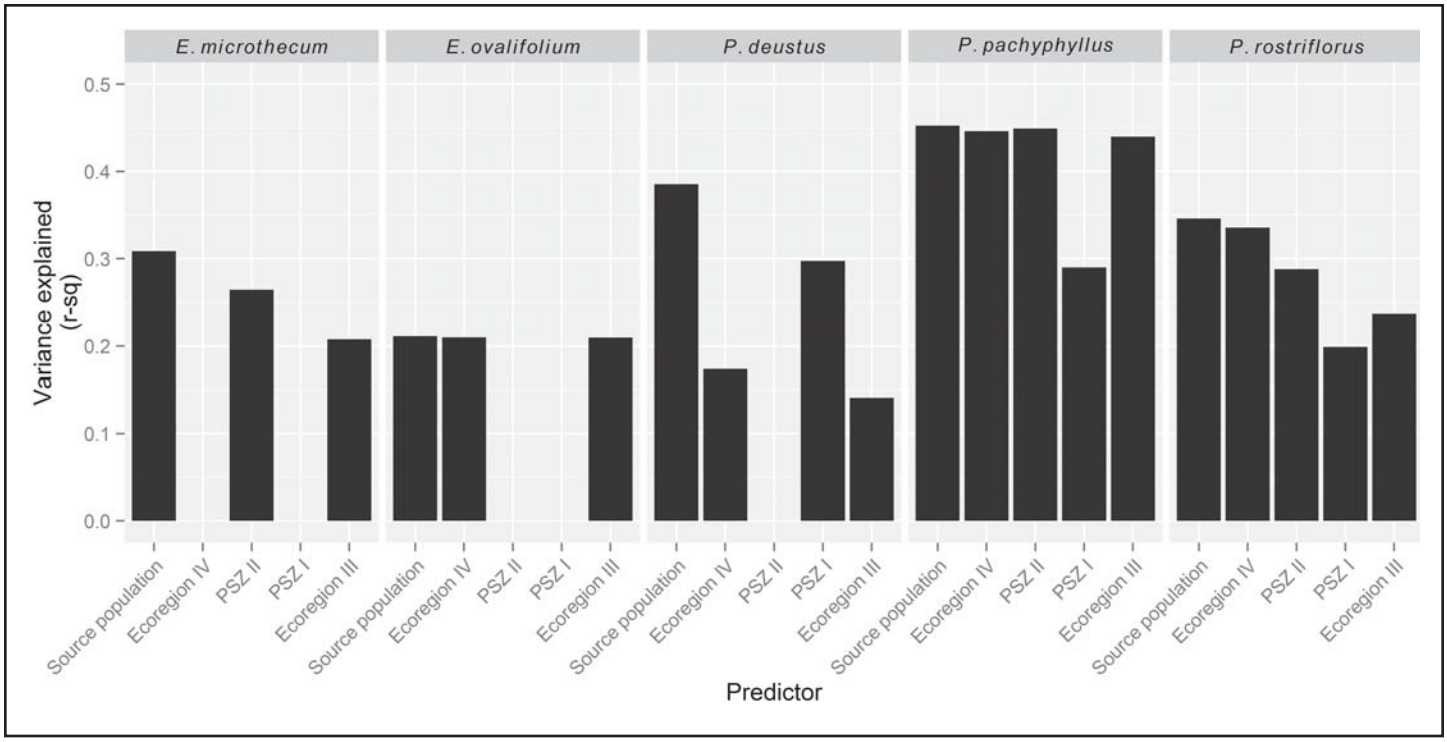

Figure 3. Variance in seed germination for each species (all germination treatments combined) explained by source population and three potential seed transfer zones of different geographic resolution in the Great Basin. Variance explained for all species, sites, and potential seed transfer zones was highly significant ($P < 0.001$). In general, the number of potential seed transfer zones decreases from left to right for each species (e.g., there are more source populations than Level IV Ecoregions; Table 1). In *E. microthecum, E. ovalifolium*, and *P. deustus* some potential seed transfer zones grouped source populations in exactly the same way (e.g., all six *E. microthecum* source populations were grouped into six Level IV Ecoregions and the same five zones for PSZ II and PSZ I; Table 1). In these cases, results from the larger potential seed transfer zone (e.g., PSZ I in the case of *E. microthecum*) are not shown in the figure.

Common Garden

Each quantitative trait measured showed a significant response to either common garden, source population, or their interaction in at least two species (Table 2). Variation in morphological traits was significantly explained by source population for each species, except in the case of stem architecture in *E. microthecum*. For growth and phenology traits, source population significantly explained variation in at least half of all measured traits for each species. Finally, all floral morphology traits measured in *Penstemon* spp. varied significantly by source population. In *P. deustus,* anther exsertion also had a significant common garden effect, and in *P. rostriflorus,* flower opening and corolla length had significant interaction terms. Significant variation by common garden was found in at least half of all traits for each species, and a number of significant interactions between source population and common garden were found for each species except *E. microthecum*.

Multivariate analyses combining all traits showed that, for all species and common gardens, source population best explained variation (ranging from 10% in *E. microthecum* to 22% in *P. pachyphyllus*, both at Boise; Figure 4), although there was also substantial variation within each population. Analyses using increasingly large candidate seed transfer zones (Level IV Ecoregion < PSZ II < PSZ I < Level III Ecoregion) generally explained less (but still significant) variation. Level IV Ecoregions and PSZ II, the two smallest zones, provided the best fit among tested seed transfer zones, capturing an average of 80% of the variation explained by source populations. Larger seed transfer zones such as PSZ I captured a significant amount of measured genetic variation for all species and common gardens (except PSZ I for *P. rostriflorus* at Boise; Figure 4), but only explained an average of 59% of the variation explained by source populations. Finally, the largest seed transfer zones (Level III Ecoregions) explained the lowest percentage of source population variation across all species (only 39% of variation explained by source population). However, results were not consistent across all species, potential seed transfer zones, or common gardens for any species. For example, Level IV Ecoregion captured 94% of the variation explained by source populations for *E. ovalifolium* at Boise, but only 53% in *P. deustus* at SLC. In *P. pachyphyllus*, PSZ I and Level III Ecoregion both explained similar and significant variation, but this accounted for only about 55% of the variation explained by source population. In *P. rostriflorus*, PSZ I actually explained less of the variation explained by source population than Level III Ecoregions (16% vs. 49%, respectively) even though PSZ I had three potential seed transfer zones and Level III Ecoregions had only two.

DISCUSSION

We found significant genetic differences between source populations for many germination, morphological, phenologi-

cal, and production-related traits in all five study species, suggesting that, when feasible, a local sourcing approach may be the best option for restoration efforts (Hufford and Mazer 2003). However, local sourcing will often be cost- and time-prohibitive for large-scale restoration projects, especially those with short time frames such as post-disturbance restorations (e.g., revegetation following fire). Our results indicate that tested seed transfer zones can be useful guides for moving seed when local sourcing is not a viable option, but that the effectiveness of the seed transfer zones decreases as zone size increases (Figures 3, 4).

Level IV Ecoregions were almost always the most effective seed transfer zone at capturing genetic variation in our source populations (with the exception of seed germination in *P. deustus*; Figure 3). However, Level IV Ecoregions were also the smallest zones tested, making them the least economical sourcing option. Provisional Seed Zones were nearly as effective as Level IV Ecoregions at capturing variation when Level III Ecoregions were also incorporated (PSZ II). Because PSZ II zones are fewer and larger than Level IV Ecoregions, their use may be an appropriate compromise when developing native plant materials for unstudied forbs. Level III Ecoregions represent the largest and therefore the most economically attractive seed transfer zones tested in this study. Our results suggest they may be effective *minimum* seed transfer zones, as they captured a significant but relatively small amount (39% on average) of genetic variation measured in our source populations. However, using Level III Ecoregions may increase the risk of a mismatch between seed source and restoration site (Bennington et al. 2012). This may also increase the risk of outbreeding depression when restorations are located near native stands of the same species (Frankham et al. 2011).

The degree of population divergence in any species is determined by a combination of adaptation, genetic drift and gene flow (Hufford and Mazer 2003). It is generally expected that species with extensive gene flow will have few genetic differences between populations, and, therefore, fewer,

Table 2. Two-way ANOVA results of source population, common garden, and interaction effects shown by species and trait for five selected Great Basin forb species. Significant effects are indicated by asterisks **** $P < 0.0001$; *** $P < 0.001$; ** $P < 0.01$; * $P < 0.05$, while ns means nonsignificant and "–" means the measure was not taken for that species/trait. EM = *Eriogonum microthecum*, EO = *E. ovalifolium*, PD = *Penstemon deustus*, PP = *P. pachyphyllus*, and PR = *P. rostriflorus*.

	Source Population					Common Garden					Source Population × Common Garden				
Trait	EM	EO	PD	PP	PR	EM	EO	PD	PP	PR	EM	EO	PD	PP	PR
Morphology															
Leaf area	***	****	**	**	****	****	***	***	****	****	ns	ns	ns	ns	ns
Stem diameter	–	–	**	*	****	–	–	****	ns	**	–	–	ns	ns	ns
Stem architecture	ns	****	*	*	****	****	ns	ns	ns	ns	*	ns	**	ns	ns
Flower stalk height	**	****	–	–	–	****	****	–	–	–	ns	ns	–	–	–
Production and Phenology															
Flowers per stem	****	*	ns	ns	**	***	****	****	****	****	ns	*	ns	*	ns
Number flowering stems	**	ns	*	*	**	***	***	***	***	***	ns	ns	ns	*	ns
Flowering phenology	****	****	**	****	****	ns	****	****	****	ns	ns	ns	**	ns	ns
Maximum size	ns	****	****	****	****	ns	ns	****	ns	****	ns	ns	ns	*	ns
Summer growth	**	ns	*	ns	***	ns	ns	**	ns	ns	ns	ns	ns	ns	ns
Winter growth	****	ns	ns	****	ns	**	ns	*	****	****	ns	ns	ns	****	***
Floral Morphology															
Flower opening	–	–	**	****	**	–	–	ns	ns	ns	–	–	ns	ns	****
Flower anther exsertion	–	–	****	**	****	–	–	***	ns	ns	–	–	ns	ns	ns
Flower corolla length	–	–	****	****	****	–	–	ns	ns	ns	–	–	ns	ns	***

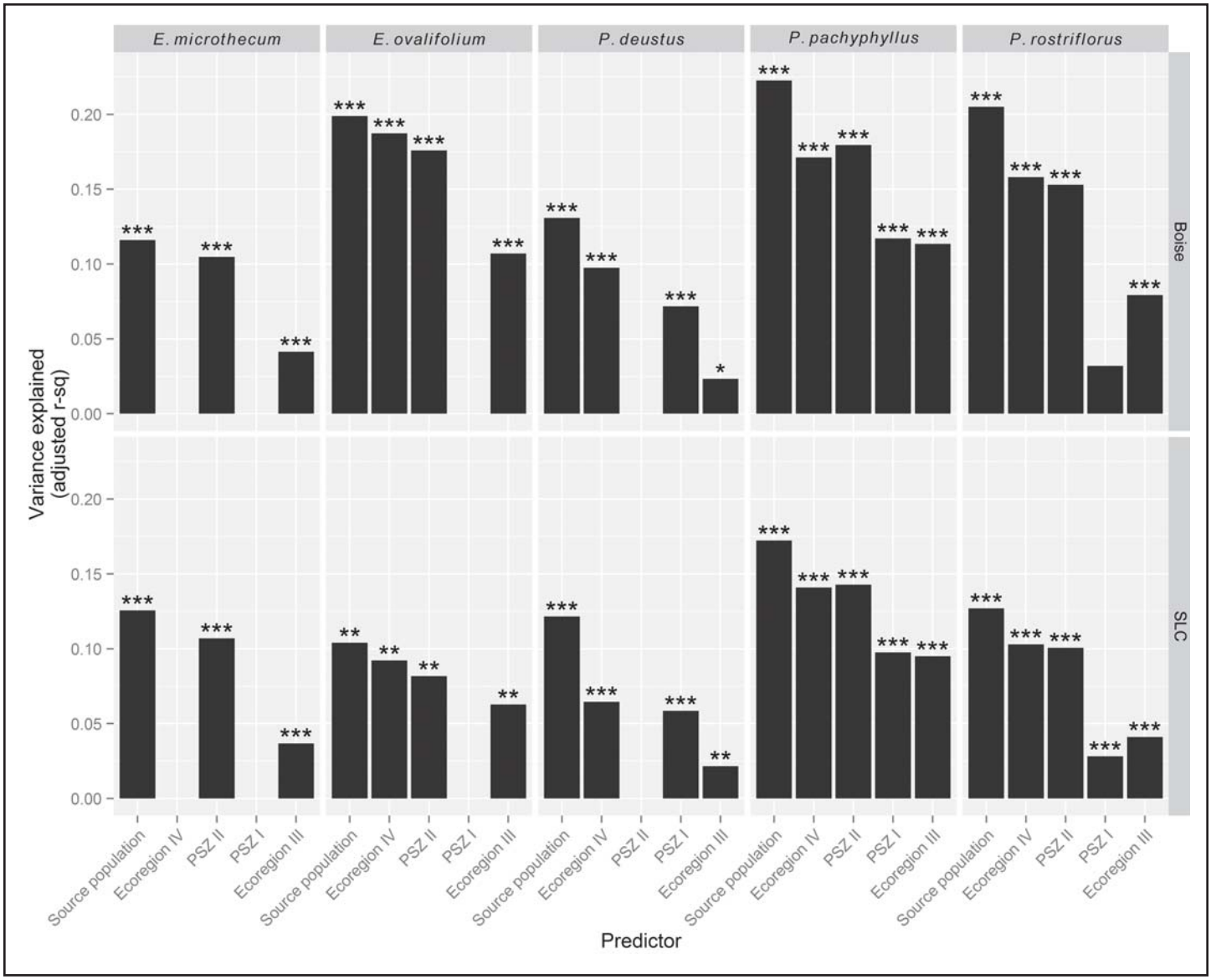

Figure 4. Variance in multivariate trait space (for each species × common garden combination) explained by source population and potential seed transfer zones of different geographic resolution in the Great Basin. In general, the number of potential seed transfer zones decreases from left to right for each species (e.g., there are more source populations than Level IV Ecoregions; Table 1). Results are shown for the Boise common garden in the top portion of the figure, and for the Salt Lake City (SLC) common garden in the lower portion of the figure. As in Figure 3, results are excluded if the smaller seed transfer zones do not show different groupings than the larger seed transfer zones. Significance levels for each model: *** $P < 0.001$; ** $P < 0.01$; * $P < 0.05$.

larger seed transfer zones can be used for these species than for species with limited gene flow (Hufford and Mazer 2003). Previous research has shown that *P. rostriflorus* has much higher among-population gene flow than *P. deustus* and *P. pachyphyllus* (Kramer et al. 2011), likely because the red flowers of *P. rostriflorus* attract hummingbirds that are effective at moving pollen long distances, while the white and purple flowers of *P. deustus* and *P. pachyphyllus* attract bees that generally move pollen over much shorter distances. We would, therefore, expect *P. rostriflorus* to have fewer genetic differences among populations, and these differences would be effectively captured by large seed transfer zones like Level III Ecoregions. Our results did not meet these expectations. While results varied slightly by species, the genetic differences we identified among populations (Table 2) and the relative variation explained by different candidate seed transfer zones (Figures 3, 4) were surprisingly similar among all study species.

We were not directly able to test whether the genetic differences we identified are adaptive (driven by different selection pressures at different sites) and, therefore, important to restoration-sourcing decisions, or whether they are random (driven by genetic drift). However, because populations connected by gene flow are less likely to accumulate random differences from drift (Slatkin 1987), there is a high likelihood that the differences we measured in *P. rostriflorus* are adaptive and result

from strong natural selection that has overridden the homogenizing effects of gene flow (Endler 1973). In the other *Penstemon* species, and in both *Eriogonum* species, random genetic drift may be contributing to some of the differences we identified, as gene flow is likely less extensive than in *P. rostriflorus*. However, all of our populations had at least 200 reproductive individuals (in many cases more than 500), minimizing the chance that random genetic drift is the only force driving the differences we measured.

We found significant differences in germination by source population in response to different winter length treatments for all species (Figure 2). In *Penstemon* species, germination requirements have been shown to be heritable and adaptive, as they are directly related to winter conditions at the source site (Meyer 1992). For *E. ovalifolium* (a spring-flowering species), 100% germination was achieved for most source populations and treatments, while in *E. microthecum* (a summer-flowering species), 100% germination for some source populations was only achieved after a second round of winter conditions (data not shown), suggestive of a bet-hedging strategy (Evans et al. 2007). Seed germination is a critical step in restoration establishment (James et al. 2011; James et al. 2012), yet seed germination requirements are rarely explicitly incorporated in common garden studies geared toward delineating seed transfer zones (but see Erickson et al. 2004; Wilson et al. 2008; St. Clair et al. 2013). Numerous studies have shown that seed germination requirements are often adaptive and under genetic control (Li and Foley 1997; Foley and Fennimore 1998; Gu et al. 2004). Using seed transfer zones that best capture variation in seed germination requirements can, therefore, minimize the risk that seeds will be used at a restoration site where they germinate at the wrong time or are not able to germinate at all.

In our common garden study we measured considerable variation within source populations (60% to almost 90%, depending on species and trait; Figures 3, 4). This variation was not explained by differences between common gardens, and was likely due to genetic diversity within and between the maternal lines used for each study population. Genetic variation within populations is an important consideration in restoration-sourcing decisions, as it can improve establishment success under different conditions and make populations more resistant to pests, pathogens, disturbance, and climate extremes (Hughes and Stachowicz 2004; Reusch et al. 2005; Crawford and Whitney 2011; Tooker and Frank 2012). Populations with high genetic diversity are also more likely to be able to survive and adapt to changing conditions (Jump et al. 2009), are more productive, and support more diverse animal communities than low-diversity populations (Reynolds et al. 2012). Conversely, populations established with limited genetic diversity may have low fitness associated with inbreeding depression, including poor seed set and seedling vigor (Keller and Waller 2002; Vilas et al. 2006), which may increase the probability of extinction.

The large differences seen between common garden sites also reveal high levels of phenotypic plasticity in our study species for many traits (significant common garden and germination treatment effects; Table 2). Although phenotypic plasticity is considered predominantly an environmental response, it can also be genetically controlled and heritable (Bradshaw 2006; Nicotra et al. 2010). It is an important consideration for restoration-sourcing because it may allow a population to survive in a broader range of habitats and provides some flexibility in the face of changing climates (Nicotra et al. 2010). Our Boise common garden site had conditions that were generally more stressful and more typical of field conditions (sandier soils, lower annual precipitation, no irrigation) than our SLC site, and it was there that genetic differences among source populations were most pronounced (especially for *E. ovalifolium*, *P. pachyphyllus* and *P. rostriflorus*; Figure 4). This suggests that the genetic variation we measured may be most important under more stressful conditions (Hoffmann and Parsons 1991). The general lack of interaction between source population and common garden indicates that most individuals responded similarly to the very different growing conditions in each common garden.

While generalizations are not warranted based only on results from five species, our approach may be useful to employ with other common garden datasets to better understand how genetic variation is captured by candidate seed transfer zones. If carried out for enough species, this could ultimately support broader generalizations about the best seed transfer zones to use for unstudied species. Additionally, there is substantial information to be gained by monitoring how current sourcing decisions impact restoration outcomes. Restoration practitioners effectively test potential seed transfer zones every time seed sourcing decisions are made, presenting ongoing opportunities to capture useful data from restorations to inform future sourcing decisions. To take full advantage of these opportunities, researchers and practitioners should work together to incorporate an experimental element into future restoration efforts.

CONCLUSION

We have shown that all of our study populations harbor potentially important genetic variation that should be considered in seed-sourcing decisions for restoration. The three candidate seed transfer zones tested captured much of this variation, but their effectiveness varied by zone size. Provisional Seed Zones incorporating Level III Ecoregions were largely effective at capturing most of the variation in our source populations, and may be a useful guide for matching seed sources to restoration sites.

ACKNOWLEDGMENTS

We are grateful to Becky Tonietto for her diligence in collecting all floral trait measurements and to Shannon Still for spatial analysis assistance. We thank the Utah Botanical Center and Boise State University for hosting our common garden studies, as well as the agencies and foundations who have supported our work on seed transfer zones and research related to seed sourcing, including the Bureau of Land

Management Plant Conservation Program and US Environmental Protection Agency (STAR Fellowship to AK).

Andrea Kramer is a Conservation Scientist at the Chicago Botanic Garden and an adjunct assistant professor at Northwestern University, where she teaches and mentors students through the Graduate Program in Plant Biology and Conservation. Her research interests include ecological genetics, including applications to native plant materials development and ecological restoration on public lands in the western United States.

Dan Larkin is a Conservation Scientist and the David Byron Smith Family Curator of Native Habitats at the Chicago Botanic Garden and an adjunct assistant professor at Northwestern University, where he teaches and mentors students through the Graduate Program in Plant Biology and Conservation. He conducts research in restoration ecology, community ecology, and invasion biology, primarily in wetland and terrestrial habitats of the upper Midwest.

Jeremie Fant is a Conservation Scientist and Molecular Ecology Lab Manager at Chicago Botanic Garden and an adjunct assistant professor at Northwestern University, where he teaches and mentors students through the Graduate Program in Plant Biology and Conservation. His research interests include restoration and conservation genetics and understanding the role pollinators play in driving the genetic structure of plant populations.

LITERATURE CITED

Bennington, C.C., N. Fetcher, M.C. Vavrek, G.R. Shaver, K.J. Cummings, and J.B. McGraw. 2012. Home site advantage in two long-lived arctic plant species: results from two 30-year reciprocal transplant studies. Journal of Ecology 100:841-851.

Bivand, R., T. Keitt, and B. Rowlingson. 2013. rgdal: Bindings for the Geospatial Data Abstraction Library. R package version 0.8-14. <http://CRAN.R-project.org/package=rgdal>.

Bivand, R.S., E. Pebesma, and V. Gomez-Rubio. 2013. Applied spatial data analysis with R, 2nd ed. Springer, NY.

BLM. 2000. The Great Basin: Healing the Land. Bureau of Land Management, Boise, ID.

BLM. 2012. Technical Protocol for the Collection, Study, and Conservation of Seeds from Native Plant Species for Seeds of Success. Bureau of Land Management, Washington, DC.

Bower, A.D., J.B. St. Clair, and V. Erickson. 2014. Generalized provisional seed zones for native plants. Ecological Applications 24:913-919.

Bradshaw, A.D. 2006. Unravelling phenotypic plasticity – why should we bother? New Phytologist 170:644-648.

Crawford, K.M., and K.D. Whitney. 2011. Population genetic diversity influences colonization success. Molecular Ecology 19:1253-1263.

Endler, J.A. 1973. Gene flow and population differentiation. Science 179:243-250.

Erickson, V.J., N.L. Mandel, and F.C. Sorensen. 2004. Landscape patterns of phenotypic variation and population structuring in a selfing grass, *Elymus glaucus* (blue wildrye). Canadian Journal of Botany 82:1776-1790.

Evans, Margaret E.K., R. Ferrière, M.J. Kane, and D.L. Venable. 2007. Bet hedging via seed banking in desert evening primroses (*Oenothera*, Onagraceae): Demographic evidence from natural populations. The American Naturalist 169:184-194.

Foley, M.E., and S.A. Fennimore. 1998. Genetic basis for seed dormancy. Seed Science Research 8:173-182.

Frankham, R., J.D. Ballou, M.D.B. Eldridge, R.C. Lacy, K. Ralls, M.R. Dudash, and C.B. Fenster. 2011. Predicting the probability of outbreeding depression. Conservation Biology 25:465-475.

Gower, J.C. 1971. A general coefficient of similarity and some of its properties. Biometrics:857-871.

Gu, X.Y., S.F. Kianian, and M.E. Foley. 2004. Multiple loci and epistases control genetic variation for seed dormancy in weedy rice (*Oryza sativa*). Genetics 166:1503-1516.

Harrell, F.E. Jr. 2014. rms: Regression Modeling Strategies. R package version 4.1-1.

Hereford, J. 2009. A quantitative survey of local adaptation and fitness trade-offs. The American Naturalist 173:579-588.

Hoffmann, A.A., and P.A. Parsons. 1991. Evolutionary Genetics and Environmental Stress. Oxford University Press, NY.

Hufford, K., and S.J. Mazer. 2003. Plant ecotypes: Genetic differentiation in the age of ecological restoration. Trends in Ecology & Evolution 18:147-155.

Hufford, K., S.J. Mazer, and M.D. Camara. 2008. Local adaptation and effects of grazing among seedlings of two native California bunchgrass species: Implications for restoration. Restoration Ecology 16:59-69.

Hughes, A.R., and J.J. Stachowicz. 2004. Genetic diversity enhances the resistance of a seagrass ecosystem to disturbance. Proceedings of the National Academy of Sciences of the United States of America 101:8998-9002.

James, J.J., M.J. Rinella, and T. Svejcar. 2012. Grass seedling demography and sagebrush steppe restoration. Rangeland Ecology & Management 65:409-417.

James, J.J., T.J. Svejcar, and M.J. Rinella. 2011. Demographic processes limiting seedling recruitment in arid grassland restoration. Journal of Applied Ecology 48:961-969.

Johnson, G.R., F.C. Sorensen, J.B. St Clair, and R.C. Croon. 2004. Pacific Northwest forest tree seed zones: A template for native plants? Native Plants Journal 5:131-140.

Johnson, G.R., L. Stritch, P. Olwell, S. Lambert, M.E. Horning, and R. Cronn. 2010. What are the best seed sources for ecosystem restoration on BLM and USFS lands? Native Plants Journal 11:117-131.

Johnson, R.C., M.J. Cashman, and K. Vance-Borland. 2012. Genecology and seed zones for indian ricegrass collected in the southwestern United States. Rangeland Ecology & Management 65:523-532.

Johnson, R.C., V.J. Erickson, N.L. Mandel, J.B.S. Clair, and K.W. Vance-Borland. 2010. Mapping genetic variation and seed zones for *Bromus carinatus* in the Blue Mountains of eastern Oregon, USA. Botany 88:725-736.

Johnson, R.C., B.C. Hellier, and K.W. Vance-Borland. 2013. Genecology and seed zones for tapertip onion in the US Great Basin. Botany-Botanique 91:686-694.

Joshi, J., B. Schmid, M.C. Caldeira, P.G. Dimitrakopoulos, J. Good, R. Harris, A. Hector, K. Huss-Danell, A. Jumpponen, A. Minns, C.P.H. Mulder, J.S. Pereira, A. Prinz, M. Scherer-Lorenzen, A.-S.D. Siamantziouras, A.C. Terry, A.Y. Troumbis, and J.H. Lawton. 2001. Local adaptation enhances performance of common plant species. Ecology Letters 4:536-544.

Jump, A.S., R. Marchant, and J. Peñuelas. 2009. Environmental change and the option value of genetic diversity. Trends in Plant Science 14:51-58.

Kartesz, J.T. 1999. A synonymized checklist and atlas with biological attributes for the vascular flora of the United States, Canada, and Greenland. *In* J.T. Kartesz, and C.A. Meacham, eds., Synthesis of the North

American Flora. [CD ROM]. Version 1.0, North Carolina Botanical Garden, Chapel Hill.

Keller, L.F., and D.M. Waller. 2002. Inbreeding effects in wild populations. Trends in Ecology & Evolution 17:230-241.

Kramer, A.T., J.B. Fant, and M.V. Ashley. 2011. Influences of landscape and pollinators on population genetic structure: Examples from three *Penstemon* (Plantaginaceae) species in the Great Basin. American Journal of Botany 98:109-121.

Kronholm, I., F.X. Picó, C. Alonso-Blanco, J. Goudet, and J. de Meaux. 2012. Genetic basis of adaptation in *Arabidopsis thaliana*: Local adaptation at the seed dormancy QTL DOG1. Evolution 66:2287-2302.

Laliberté, E., and P. Legendre. 2010. A distance-based framework for measuring functional diversity from multiple traits. Ecology 91:299-305.

Laliberté, E., and B. Shipley. 2011. FD: Measuring functional diversity from multiple traits, and other tools for functional ecology. R package version 1.0-11.

Legendre, P., and L. Legendre. 2012. Numerical Ecology. Elsevier, Oxford, UK.

Legendre, P., J. Oksanen, and C.J.F. ter Braak. 2011. Testing the significance of canonical axes in redundancy analysis. Methods in Ecology and Evolution 2:269-277.

Leimu, R., and M. Fischer. 2008. A meta-analysis of local adaptation in plants. PLoS ONE 3:e4010.

Li, B., and M.E. Foley. 1997. Genetic and molecular control of seed dormancy. Trends in Plant Science 2:384-389.

MacKay, J. 1993. Seed source selection and genetic improvement of red oak (*Quercus rubra* L) in Québec. Annals of Forest Science 50:420s-424s.

McKay, J.K., C.E. Christian, S. Harrison, and K.J. Rice. 2005. "How local Is local?" A review of practical and conceptual issues in the genetics of restoration. Restoration Ecology 13:432-440.

Meyer, S.E. 1992. Habitat-correlated variation in firecracker penstemon (*Penstemon eatonii*: Scrophulariaceae) seed germination patterns. Bulletin of the Torrey Botanical Club 119:268-279.

Meyer, S.E., S.G. Kitchen, and S.L. Carlson. 1995. Seed germination timing patterns in intermountain *Penstemon* (Scrophulariaceae). American Journal of Botany 82:377-389.

Millar, C.I., and W.J. Libby. 1989. Disneyland or native ecosystem: Genetics and the restorationist. Restoration and Management Notes 7:18-24.

Miller, R.G. 1997. Survival Analysis. Wiley & Sons, NY.

Miller, S.A., A. Bartow, M. Gisler, K. Ward, A.S. Young, and T.N. Kaye. 2011. Can an ecoregion serve as a seed transfer zone? Evidence from a common garden study with five native species. Restoration Ecology 19:268-276.

Mitchell, R.J., and R.G. Shaw. 1993. Heritability of floral traits for the perennial wild flower *Penstemon centranthifolius* (Scrophulariaceae): Clones and crosses. Heredity 71:185-192.

Montalvo, A.M., and N.C. Ellstrand. 2001. Nonlocal transplantation and outbreeding depression in the subshrub *Lotus scoparius* (Fabaceae). American Journal of Botany 88:258-269.

Nicotra, A.B., O.K. Atkin, S.P. Bonser, A.M. Davidson, E.J. Finnegan, U. Mathesius, P. Poot, M.D. Purugganan, C.L. Richards, F. Valladares, and M. van Kleunen. 2010. Plant phenotypic plasticity in a changing climate. Trends in Plant Science 15:684-692.

Oksanen, J., F.G. Blanchet, R. Kindt, P. Legendre, P.R. Minchin, R.G. O'Hara, G.L. Simpson, P. Solymos, M.H.H. Stevens, and H. Wagner. 2013. vegan: Community Ecology Package. R package version 2.0-9.

Omernik, J.M. 1987. Ecoregions of the conterminous United States (map supplement). Annals of the Association of American Geographers 77:118-125.

R Development Core Team. 2013. R: A language and environment for statistical computing. R version 2.15.3. The R Foundation for Statistical Computing, Vienna, Austria.

Reusch, T.B.H., A. Ehlers, A. Hammerli, and B. Worm. 2005. Ecosystem recovery after climatic extremes enhanced by genotypic diversity. Proceedings of the National Academy of Sciences 102:2826-2831.

Reynolds, L.K., K.J. McGlathery, and M. Waycott. 2012. Genetic diversity enhances restoration success by augmenting ecosystem services. PLoS ONE 7:e38397.

Saari, C., and W. Glisson. 2012. Survey of Chicago region restoration seed source policies. Ecological Restoration 30:162-165.

Shaw, R.G., and J.R. Etterson. 2012. Rapid climate change and the rate of adaptation: Insight from experimental quantitative genetics. New Phytologist 195:752-765.

Slatkin, M. 1987. Gene flow and the geographic structure of natural populations. Science 236:787-792.

St. Clair, J.B., F.F. Kilkenny, R.C. Johnson, N.L. Shaw, and G. Weaver. 2013. Genetic variation in adaptive traits and seed transfer zones for *Pseudoroegneria spicata* (bluebunch wheatgrass) in the northwestern United States. Evolutionary Applications 6:933-948.

St.Clair, B.J., N.L. Mandel, and K.W. Vance-Borland. 2005. Genecology of Douglas Fir in Western Oregon and Washington. Annals of Botany 96:1199-1214.

Therneau, T. 2012. A Package for Survival Analysis in S. R package version 2.37-2.

Tooker, J.F., and S.D. Frank. 2012. Genotypically diverse cultivar mixtures for insect pest management and increased crop yields. Journal of Applied Ecology 49:974-985.

Vilas, C., E. San Miguel, R. Amaro, and C. Garcia. 2006. Relative contribution of inbreeding depression and eroded adaptive diversify to extinction risk in small populations of shore Campion. Conservation Biology 20:229-238.

Wilson, B.L., D.C. Darris, R. Fiegener, R. Johnson, M.E. Horning, and K. Kuykendall. 2008. Seed transfer zones for a native grass (*Festuca roemeri*): Genecological evidence. Native Plants Journal 9:287-303.

Ying, C.C., and A.D. Yanchuk. 2006. The development of British Columbia's tree seed transfer guidelines: Purpose, concept, methodology, and implementation. Forest Ecology and Management 227:1-13.

Appendix 1. Population location descriptions in the Great Basin for all five study species, including Level III Ecoregion, Provisional Seed Zone, and Level IV Ecoregion details.

Species	Site Code	Mountain Range	Level III Ecoregion	Provisional Seed Zone (2013)	Level IV Ecoregion	Lat	Long	Elevation (m)
Eriogonum microthecum	EM-AR	Adobe Range	13	15 - 20 Deg. F. / 6 - 12	13m	40.94	-115.93	1924
	EM-OM	Owyhee Mountains	80	20 - 25 Deg. F. / 6 - 12	80f	43.25	-116.71	1262
	EM-PN	Pine Nut Mountains	13	15 - 20 Deg. F. / 2 - 3	13y	38.89	-119.49	2535
	EM-SM	Steens Mountains	80	20 - 25 Deg. F. / 3 - 6	80j	42.63	-118.53	2522
	EM-SR	Snake Range	13	15 - 20 Deg. F. / 3 - 6	13q	39.03	-114.26	2522
	EM-WWM	Wah Wah Mountains	13	15 - 20 Deg. F. / 3 - 6	13d	38.33	-113.59	1636
Eriogonum ovalifolium	EO-DM	Desatoya Mountains	13	15 - 20 Deg. F. / 6 - 12	13s	39.27	-117.74	1993
	EO-ER	Egan Range	13	10 - 15 Deg. F. / 6 - 12	13p	39.2	-114.87	2100
	EO-OM	Owyhee Mountains	80	20 - 25 Deg. F. / 6 - 12	80f	43.25	-116.71	1263
	EO-SCR	Schell Creek Range	13	10 - 15 Deg. F. / 6 - 12	13p	39.44	-114.69	2144
	EO-SRV	Snake River Valley	12	20 - 25 Deg. F. / 12 - 30	12j	42.9	-115.79	891
	EO-TR	Toquima Range	13	15 - 20 Deg. F. / 6 - 12	13r	39.45	-116.75	2018
Penstemon deustus	PD-DM1	Desatoya Mountains	13	15 - 20 Deg. F. / 6 - 12	13s	39.25	-117.68	2025
	PD-PNM1	Pine Nut Mountains	13	20 - 25 Deg. F. / 3 - 6	13x	39.18	-119.53	1861
	PD-PNM2	Pine Nut Mountains	13	20 - 25 Deg. F. / 6 - 12	13x	39.12	-119.42	1834
	PD-SCR1	Schell Creek Range	13	15 - 20 Deg. F. / 3 - 6	13e	39.56	-114.64	2649
	PD-SM1	Steens Mountains	80	20 - 25 Deg. F. / 3 - 6	80j	42.63	-118.53	1793
	PD-SM2	Steens Mountains	80	20 - 25 Deg. F. / 6 - 12	80j	42.05	-118.62	1368
Penstemon pachyphyllus	PP-AR1	Antelope Range	13	10 - 15 Deg. F. / 6 - 12	13q	40.04	-114.51	1995
	PP-MP1	Markagunt Plateau	20	15 - 20 Deg. F. / 3 - 6	20e	37.34	-113.08	2122
	PP-MP2	Markagunt Plateau	20	25 - 30 Deg. F. / 6 - 12	20h	37.17	-113.08	1119
	PP-SR1	Snake Range	13	15 - 20 Deg. F. / 3 - 6	13q	39.11	-114.35	2323
	PP-SR2	Snake Range	13	15 - 20 Deg. F. / 6 - 12	13q	39.15	-114.33	2227
	PP-WWM1	Wah Wah Mountains	13	15 - 20 Deg. F. / 3 - 6	13d	38.33	-113.59	2560
	PP-WWM3	Wah Wah Mountains	13	15 - 20 Deg. F. / 3 - 6	13d	38.25	-113.58	2216

Continued

Appendix 1. (*Continued*)

Species	Site Code	Mountain Range	Level III Ecoregion	Provisional Seed Zone (2013)	Level IV Ecoregion	Lat	Long	Elevation (m)
Penstemon rostriflorus	PR-MP1	Markagunt Plateau	20	15 - 20 Deg. F. / 3 - 6	20e	37.35	-113.08	2430
	PR-MP2	Markagunt Plateau	20	20 - 25 Deg. F. / 6 - 12	20e	37.29	-113.1	1632
	PR-PM1	Pilot Mountains	13	15 - 20 Deg. F. / 6 - 12	13v	38.39	-118.03	1919
	PR-PNM1	Pine Nut Mountains	13	20 - 25 Deg. F. / 6 - 12	13x	38.85	-119.44	1678
	PR-SR1	Snake Range	13	15 - 20 Deg. F. / 3 - 6	13e	39.02	-114.27	2768
	PR-SR2	Snake Range	13	15 - 20 Deg. F. / 6 - 12	13q	38.99	-114.22	2147
	PR-WWM1	Wah Wah Mountains	13	15 - 20 Deg. F. / 3 - 6	13d	38.35	-113.61	2510
	PR-WWM2	Wah Wah Mountains	13	15 - 20 Deg. F. / 3 - 6	13d	38.26	-113.58	2455

Appendix 2. Percent germination for each species and source population shown by winter length treatment, and the average germination for all treatments combined. The Level III Ecoregion where each population is located is also shown (as in Figure 2).

Population	Level III Ecoregion	No Winter	8 Week Winter	10 Week Winter	12 Week Winter	14 Week Winter	16 Week Winter	Average Germination
Eriogonum microthecum								
EM-OM	80	98%	80%	98%	100%	94%	74%	91%
EM-SM	80	86%	18%	42%	41%	60%	38%	47%
EM-AR	13	22%	25%	20%	22%	30%	23%	24%
EM-PN	13	67%	40%	24%	32%	56%	–	44%
EM-SR	13	0%	0%	0%	0%	22%	20%	7%
EM-WWM	13	83%	52%	53%	65%	47%	55%	59%
Eriogonum ovalifolium								
EO-OM	80	100%	100%	100%	98%	100%	92%	98%
EO-ER	13	71%	100%	98%	98%	91%	98%	93%
EO-SCR	13	74%	90%	100%	100%	86%	95%	91%
EO-TR	13	100%	100%	100%	98%	97%	96%	98%
EO-SRV	12	100%	100%	100%	100%	99%	97%	99%
Penstemon deustus								
PD-SM1	80	4%	3%	18%	5%	21%	18%	12%
PD-SM2	80	4%	70%	76%	95%	93%	98%	73%
PD-DM1	13	0%	3%	21%	51%	30%	35%	23%
PD-PNM1	13	13%	40%	63%	58%	78%	70%	54%
PD-PNM2	13	9%	36%	51%	54%	87%	71%	51%
PD-SCR1	13	19%	2%	15%	5%	23%	25%	15%
Penstemon pachyphyllus								
PP-MP1	20	97%	95%	82%	93%	92%	100%	93%
PP-MP2	20	100%	83%	96%	97%	100%	97%	96%
PP-AR1	13	14%	12%	19%	13%	17%	33%	18%
PP-SR1	13	12%	14%	16%	36%	18%	53%	25%
PP-SR2	13	33%	29%	24%	19%	25%	35%	27%
PP-WWM1	13	43%	64%	10%	50%	47%	50%	44%
PP-WWM3	13	30%	0%	11%	29%	37%	46%	25%
Penstemon rostriflorus								
PR-MP1	20	12%	62%	75%	79%	81%	90%	66%
PR-MP2	20	4%	58%	55%	77%	81%	91%	61%
PR-PM1	13	11%	40%	57%	62%	69%	86%	54%
PR-PNM1	13	38%	39%	59%	81%	79%	74%	62%
PR-SR1	13	1%	4%	6%	22%	11%	8%	9%
PR-SR2	13	2%	2%	2%	21%	37%	67%	22%
PR-WWM1	13	4%	6%	3%	25%	50%	39%	21%
PR-WWM2	13	10%	12%	49%	59%	54%	65%	42%