

Ecotypes of Native Species: How Local is Local in Restoration Plantings?

Epic E. Knapp and Kevin J. Rice

Department of Agronomy and Range Science and Center for Population Biology
University of CA, Davis, CA 95616-8515

The growing knowledge of the threats posed by exotic species to native biodiversity has generated a great deal of interest in planting native species and restoring native plant habitat. In addition, planting native species is often an integral part of exotic plant eradication efforts. By planting native species the hope is that these plants will take over the space formerly occupied by the exotics and thereby help reduce the chances of unwanted recolonization by the latter.

However, planting native species can bring a new set of "exotic vs. native" issues. If a seed source is used that evolved in a different region, in association with different climate and soil types, it can be argued that such a planted population is in reality no more "native" to that site than an exotic species would be. A basis for this argument is that extensive genetic differences are found among populations of most native species. These genetic differences can be caused by natural selection acting across a diverse array of environments, or by genetic drift, due to small population size and lack of genetic mixing (gene flow) among populations, or a combination of both.

Populations that differ as a result of natural selection are known as "ecotypes" and are often best adapted to the local environmental conditions. Thus, if non-local seed sources are used for a project, the plants may not be well adapted. This can not only lead to failure of the plants to persist (see discussion in Handel et al. 1994), but can also result in genetic contamination of existing local populations of the species (Millar and Libby 1989, Libby and Rodrigues 1992, Knapp and Rice 1994, Knapp and Dyer 1997).

Only recently, the importance of genetic variation within the species has been widely appreciated by restorationists (Padgett and Crow 1994). An example of this shift in thinking is that agencies such as the U.S. Forest Service (USFS) and the Bureau of Land Management (BLM) now require seed of native plants for projects on Federal lands to originate from "genetically local sources." However, the lack of information on patterns of natural genetic variation in many native species makes defining "local" difficult.

The USFS uses seed zones based on geographic and genetic information to dictate regions of transfer for commercially harvested conifer species (Buck et al. 1970). Rudimentary rules have been attempted for other species for which only limited, if any, genetic information is available. For example, Linhart (1995) suggested that seed of herbaceous species be collected not more than 100 meters away, and seed of woody species not more than 1 km away. However, it is not yet known for most native plant species over what geographic ranges seed can be successfully transferred and grown over the long term, or to what extent local populations are genetically superior to non-local populations.

In addition, experiments have not yet been carried out to determine what happens to seed from non-local plants. Do these plants eventually die? Can they spread like weedy species, and can these non-local genotypes contaminate local populations of the same native species? These are some of the questions we sought to answer when we initiated our research on patterns of genetic variation in native grass species. Our objective in this paper is to briefly summarize some of our findings to date and to explain the relevance of these findings to native plant restoration.

Materials and Methods

Patterns of genetic variation in *Nassella pulchra*

Seed was collected from populations of the commonly planted native perennial bunchgrass *Nassella pulchra* throughout its diverse environmental range in California. These included coastal sites from Marin County to

Southern California and interior sites in the Sacramento Valley and surrounding Sierra foothills. We evaluated up to 60 plants in each population for enzyme variation. These genetic markers, known as isozymes, were visualized using standard starch gel electrophoresis techniques and by staining for eleven different enzyme systems. Seven of the stains detected polymorphic banding patterns in the populations evaluated.

Seed from eight of these same populations was planted in a common garden at Davis, CA and plants scored over the course of two years for eleven different morphological and phenological (quantitative) traits, including plant height, plant shape, date of flowering, leaf length and width, average number of seeds per culm, seed weight, and awn weight. Cluster analysis was conducted for both isozymes and quantitative traits to determine the degree of genetic similarity among populations.

Patterns of variation for both quantitative traits and isozymes were compared to each other and to climatic differences and geographic distances between sites. Strength of relationships between different types of data were explored using a regression test incorporated within a matrix randomization computer program written by Bryan Manly (Manly 1994). A more thorough description of the methods used for both the isozyme analysis and the quantitative trait analysis can be found in Knapp and Rice (in press).

Reciprocal transplant experiments

We also sought to determine whether local populations are indeed better adapted, and to investigate the fate of plant populations translocated into new environments through another series of studies. Seed of *N. pulchra* and a second perennial grass *Elymus glaucus* (blue wildrye) were collected from native populations growing in the Sierra foothills east of Marysville, and at the coast near the town of Bodega Bay.

Plants of both populations and both species were planted in a reciprocal transplant experiment. Plants were established from seed and from three-month old greenhouse-grown seedlings at both the Sierra site and the Bodega Bay site. Location of the planted seeds was marked with colored toothpicks, and the three month old transplants were marked with a plastic pot label. After one growing season, survival and other measures of plant success, such as size and reproductive output were noted. Significance of results was determined using standard analysis of variance procedures.

Results and Discussion

Patterns of genetic variation in *Nassella pulchra*

Genetically based differences were found among populations regardless of the trait evaluated (Knapp and Rice, in press). A cluster analysis of the isozyme data demonstrated that the strongest genetic differences for these markers were between populations growing in southern California and populations growing in northern and central California. Data for whole plant traits relating to morphology and phenology detected different clustering of populations. For these quantitative traits, the strongest difference was between three populations collected from interior sites in or adjacent to the Sacramento valley, and all of the remaining populations from regions closer to the Pacific coast. In fact, the regression of isozyme differences between populations and quantitative trait value differences between populations was not statistically significant ($p = 0.211$), showing that these two types of traits are detecting different patterns of genetic variation. This suggests that it is probably not a good idea to base guidelines for seed transfer on data for one type of trait.

Development of molecular marker techniques has generated interest in using these markers as surrogates for whole plant traits, since genetic marker data are generally more readily and rapidly obtainable than whole plant traits. Our results showed that patterns of isozyme variation are poor predictors of variation for quantitative traits in *N. pulchra*. Both isozyme and quantitative trait data are interesting for visualizing patterns of genetic variation, but the quantitative traits are likely more closely allied with adaptation and are thus probably better predictors of how well a population will do in its new location.

Relative degree of genetic difference for the morphological and phenological traits was strongly correlated to climatic difference between the sites from which the populations were collected ($p = 0.004$) (Knapp and Rice, in press). This illustrates the role of natural selection in shaping population differences for these quantitative traits, and suggests that choosing a well adapted seed might be accomplished by matching the environment of the

planting site with the environment at the site where the seed was obtained. The advantage of climatic data is that this information is already available and in many cases has been condensed into easily interpretable information, such as horticultural climate zones (for example, see Sunset Western Garden Book 1995). Obtaining seed from the same or similar climate zones would help ensure that seeds planted would likely be at least roughly adapted to local conditions at the planting site. Another factor that would be useful to consider when matching the seed source to local environmental conditions is the soil type, because edaphic conditions also often produce strong selection pressures on plant populations (Linhart and Grant 1996). For example, if the site to be planted is on serpentine soils, it would be beneficial to obtain seed from a source population also growing on serpentine soils within a similar climate zone.

Reciprocal transplant experiments

What we have discussed so far are genetic differences between populations. But what do these genetic differences mean? The association of quantitative traits with climatic variation suggests that adaptation to local climatic conditions has occurred, but to what extent are populations of native species really locally adapted? And what happens when plants are moved into a new environment?

When seed from a coastal and a Sierra foothill (interior) population of the commonly planted grass, *Elymus glaucus*, were reciprocally transplanted, we found that after 6 months, percent survival was greatest for the Bodega population at the Bodega site, and greatest for the Sierra population at the Sierra foothill site (Fig. 1). These higher rates of survival of the local population at both sites provide evidence for local adaptation. A similar trend was evident for *N. pulchra* populations at the same two sites, and the trend approached statistical significance ($p = 0.075$, data not shown).

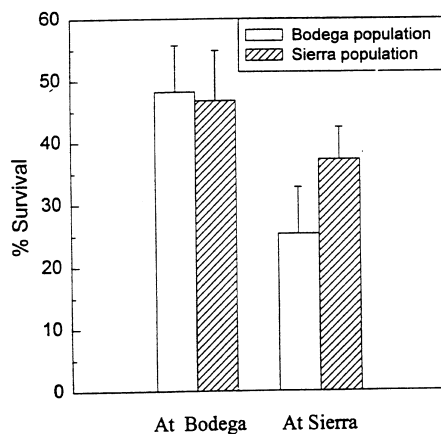


Fig. 1. Percent survival after six months of seedlings from Bodega and Sierra *Elymus glaucus* sites. The location \times population interaction was statistically significant ($P = 0.045$).

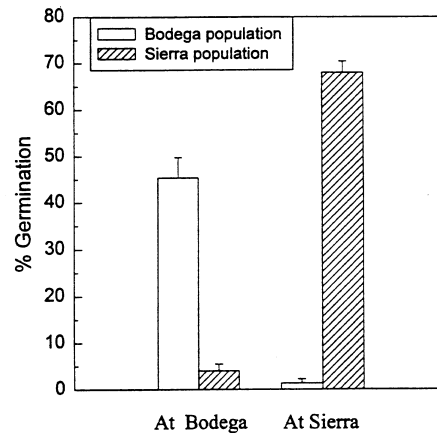


Fig. 2. Percent germination of seed grown at Bodega and Sierra on plants from both the Bodega and Sierra *Elymus glaucus* populations. The location \times population interaction was statistically significant ($P = <0.0001$).

Adaptation can occur at various stages of the plant life cycle. The results described above demonstrate adaptation for survival from the seed to seedling stage. Other stages of the plant life cycle may be more or less susceptible to local selection. For example, we found evidence for very strong selection against the non-local source at the seed fill stage. Nearly all plants from both the Sierra and the Bodega populations produced abundant seed when grown to maturity at the Bodega site, yet much of the seed from the Sierra population was not filled and did not germinate in germination tests (Fig. 2). Similarly, when plants from both populations were grown at the Sierra site, only a small proportion of the seed produced by plants from the non-local Bodega population was viable (Fig. 2).

These results suggest that when seed of this species is translocated into very different environments, strong selection occurs against the non-local populations at various times throughout the life cycle of the plants. As a

consequence, one would expect that non-local populations are rapidly eliminated by selection. It remains to be seen whether similar results will be found over different spatial scales and for populations growing in environments that do not differ as greatly as those in this study.

The occurrence of local adaptation and selection against non-local populations is not surprising, given the results from many prior studies done on plant adaptation (Linhart and Grant 1996). The scientific literature suggests that local adaptation is the norm, and that in most cases, genes from poorly adapted seed sources are expected to be eliminated by natural selection over time. Existing data also leads us to predict that it is unlikely that non-local populations of native species can become invasive in the way often seen with introduced non-native weedy species. This does not mean we should not be cautious when planting native species.

Little is known about what would actually happen, because so few restoration plantings using non-local genetic material have actually been monitored over long periods of time. However, if non-local populations are used, there is obviously a danger that the planted populations will not survive or grow well, thereby not performing the desired ecosystem function (e.g. erosion control). In addition, it is possible that non-local sources could genetically contaminate existing native populations, leading to potentially significant negative impacts on the existing local populations (Millar and Libby 1989, Knapp and Rice 1994, Knapp and Dyer 1997).

The potential for negative impacts resulting from genetic contamination may depend on the number of non-local plants planted and the size of the existing local native population. If a few non-native seeds are planted into a much larger native population, the impact may not be great in the long term, especially if seed from this non-local source is rapidly eliminated by selection. However, large volumes of seed can now be purchased for several native plant species, due to the relatively recent development of methods to grow native plants on a large scale in agricultural settings. If large numbers of non-local seed are sown, swamping or overwhelming local populations of the same species growing naturally at the planting site is possible. The impact of this genetic contamination is especially of concern for plant species with a natural mating system of outcrossing, because hybridization of local plants with non-local pollen can result in progeny that are poorly adapted to local conditions. However, the potential for genetic contamination remains poorly understood, because very little data has been gathered on this topic. We are in the process of setting up several experiments that we hope will provide a better idea of the potential for genetic contamination. At this point we can only predict that genetic contamination will have greater potential for negative impacts in outcrossing species than in inbreeding species.

Conclusions

Populations of native species growing in different environments and separate regions are often genetically distinct. These genetic differences should be considered when source material of native species is obtained for restoration and revegetation. We investigated patterns of genetic variation for different types of traits among populations of a popularly planted native grass (*Nassella pulchra*), and determined the extent to which local populations of *Elymus glaucus*, and *N. pulchra* are better adapted than non local populations. We found strong genetic differences among populations of *N. pulchra* for both isozyme markers and quantitative traits, but the patterns of genetic differentiation visualized by the two methods were not similar. Thus, management recommendations and seed transfer guidelines should not be based on data for only one type of trait. In addition, patterns of quantitative trait variation in *N. pulchra* were strongly correlated with climatic variation among sites, suggesting that it may be possible to obtain a rough match between seed sources and planting sites by using readily available climate zone data.

Reciprocal transplant studies with *E. glaucus* populations from a coastal and an interior site demonstrated local adaptation. The strength of apparent selection against the non-local seed depended on the trait and the life cycle stage evaluated. Results suggested that for these two populations, genes from non-local sources will be rapidly and selectively eliminated.

When guidelines call for using "local" plant sources in restoration and revegetation efforts, the definition of the spatial scales that constitute a local population will vary depending on the situation and the species. For example, if the climatic and/ or edaphic environment is highly heterogeneous over small areas (e.g. among serpentine soil patches, or from coastal bluff habitat to adjacent sites just inland), a definition of "local" might be

more spatially restricted than in regions where the climate and/or soils vary little over larger geographic areas (e.g. the Central Valley of California). In addition, a definition of "local" may depend on the species. Plants that reproduce primarily through self-pollination often show greater genetic differentiation among populations than plants that cross-pollinate. This is due to the greater mixing of genes (gene flow) among populations and the resulting homogenization of genetic differences. Seed transfer across large areas is therefore more likely to result in poor adaptation and planting failure in self-pollinating species than in cross-pollinating species.

Although non-local genes may be at a selective disadvantage and might be rapidly eliminated in natural environments, this does not mean that caution should not be used when obtaining plant material for a project. If the planting fails due to lack of adaptation, the ecological objectives of the planting will not be met. In addition, introducing genes of non-local sources into areas where local native populations of the same species already exist risks genetic contamination of the local population. Such genetic contamination is more likely if the number of introduced plants is large in relation to the size of the pre-existing native population. Genetic contamination is also more likely to be a problem in species that have a mating system of cross-pollination.

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