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Alternative methods for delineating seed transfer zones: comparisons of genetic and common garden data

Principal Investigators (PIs):
Taylor Crow, PhD Candidate, University of Wyoming, tcrow3@uwyo.edu
Kristina Hufford, PhD, University of Wyoming, khufford@uwyo.edu

Introduction and Objectives

The distributions of many plant species span a large geographic range and populations within that range are often adapted to local soils and climate conditions. Plants from southern latitudes, for example, may be better adapted to dry conditions than plants of the same species from northern latitudes. Local adaptation represents a challenge in ecological restoration because little is known about the distance that native plant seeds can be transferred with reasonable assurance of planting success. Seed transfer zones represent one tool to determine regions within which seed sources may be transferred with no negative effects on restoration outcomes. However, few seed transfer zones have been developed for target species because of the costly, long-term field monitoring required. We are testing seed transfer zones for true mountain mahogany (*Cercocarpus montanus* Raf.: Rosaceae), an important reclamation species in the Rocky Mountain region. This widespread shrub grows in rocky shallow soils, hosts a nitrogen fixing actinobacteria, and is a key winter browse species for elk and mule deer. Our aim is to conduct traditional common garden studies to develop seed transfer zones and compare those results to molecular marker analyses, which are less time-consuming and costly.

Methods

Study Species & Common Garden

We sampled seeds and leaf tissue of mountain mahogany from Wyoming through New Mexico to represent a “latitudinal cline,” and installed four common gardens to test for local adaptation among populations (Fig. 1). Common gardens enable researchers to measure heritable trait variation among seed sources by growing plants in a common

environment. If seed sources are locally adapted to their environment of origin, we anticipate that local populations will have higher rates of survival over time relative to populations derived from ecologically or geographically distant sites.

Mountain Mahogany seedlings will undergo a two stage planting on Boulder County Open Space property. The first planting occurred in October 2015, and included 250 seedlings (Photo 1). The second batch of 500 seedlings will be planted in the spring of 2016. The two-stage approach will allow us to buffer plant mortality and also determine the best planting time for reestablishment of mountain mahogany. The second planting will increase the number of living plants and improve the common garden study.

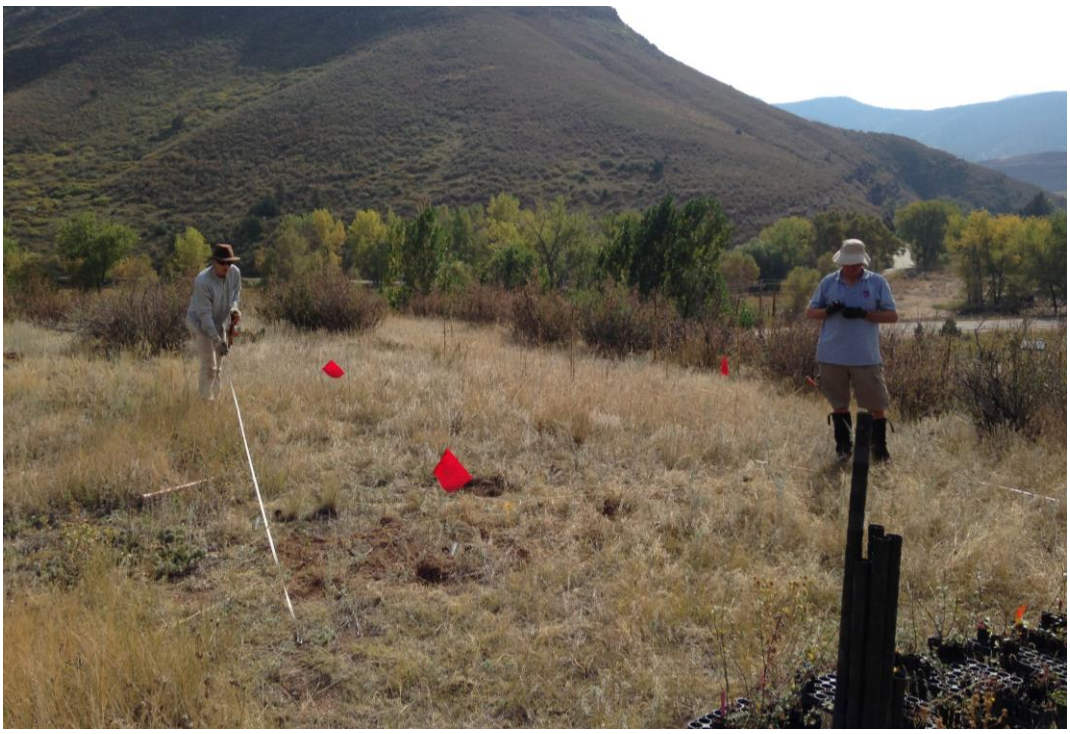

Photo 1: First planting at Hall Ranch, October 2015

Molecular markers

We sampled populations across the range of mountain mahogany to capture the geographic and environmental structure underlying DNA sequence variation (Fig. 1). Collections included leaf tissue from 30 individual plants in 50 populations of mountain mahogany for genetic analyses. Molecular markers were sequenced from the 25 populations planted in the common gardens, as well as 25 additional populations sampled across a north-south transect of mountain mahogany's range. We used next generation sequencing to identify over 6,000 single nucleotide polymorphisms (SNPs), and we plan to test whether allele frequencies are correlated with seedling growth and survival in the common gardens. Further data collection and sequencing is underway and will continue through 2016.

Results

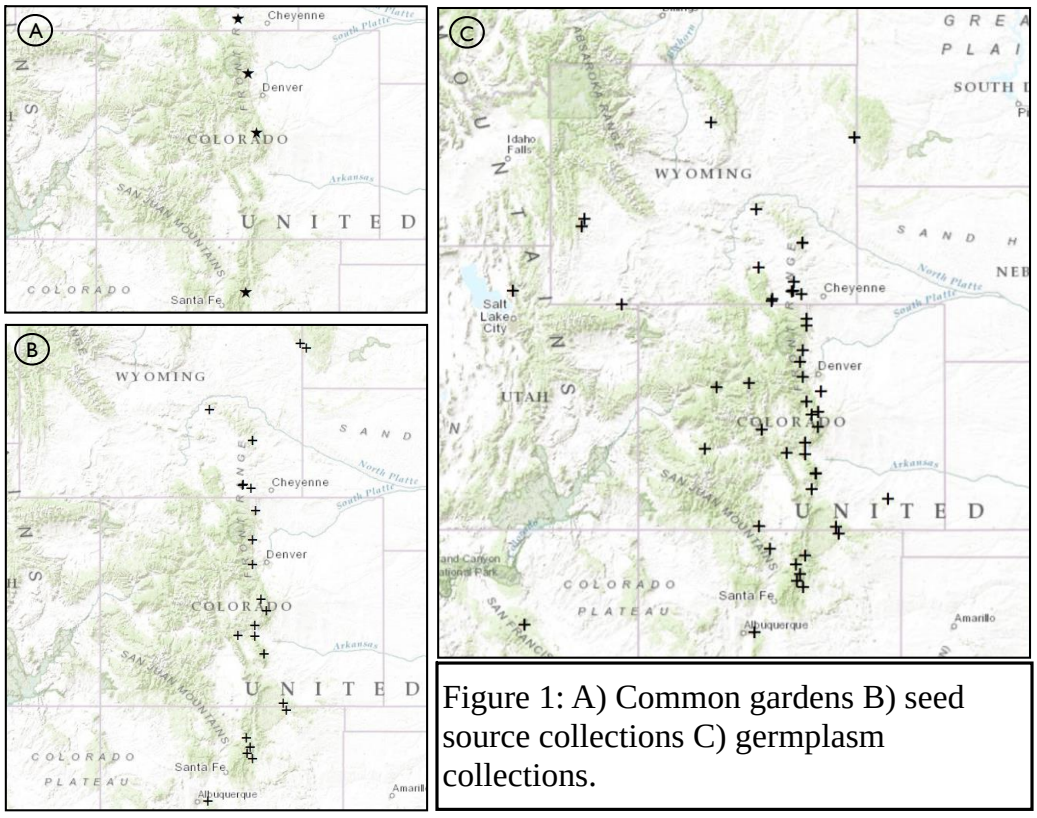

Figure 1: A) Common gardens B) seed source collections C) germplasm collections.

To date, we have discovered a strong correlation between pairwise genetic distance, and a combination of geographic and environmental distance measures. We used pairwise Nei's genetic distance as a measure of DNA sequence differentiation, geographic distance in kilometers, and created an environmental distance using degree days < 0°C and frost-free period climate data. We modeled genetic distance as a function of geographic and environmental distances to measure the amount of variation explained by each metric separately and together (Fig 2). Geographic distance explained 35% of the genetic variation, while geographic and environmental distance accounted for 38%.

Principal component analysis (PCA) used was used to visualize the structure of the genetic data and explore patterns of population allele frequencies. We found that the first PCA axis explained ~75% of the variation within our samples, while axis 2 explained ~16% (Fig. 3). The top left panel show two distinct clusters, the upper cluster are samples of *Cercocarpus ledifolius* collected near Ogden, Utah while the lower points are samples of *Cercocarpus montanus*. The lower panels of Figure 3 are the PC scores plotted over latitude to determine how the geographic location is correlated with genetic structure. The bottom left panel is evidence that there is more genetic variation in northern populations, suggesting the species' distribution in northern Colorado and Wyoming may represent unique resources for reclamation and restoration (Fig. 3). Our goal for this next year is to explore these data in conjunction with results from common gardens to allow comparisons of plant fitness with molecular data.

Discussion

We have preliminary evidence to indicate populations of mountain mahogany are differentiated along a latitudinal gradient. The geographic structure underlying genetic variation within *Cercocarpus montanus* warrants the use of seed transfer zones. Our data also indicate that incorporating spatial distance along with environmental characteristics will be important for guiding seed transfer of mountain mahogany.

Initial analysis of data from common gardens indicates that seeds are more likely to germinate and survive in local environmental conditions (data analyses underway), and genetic distances reflect common garden results. Our next step is to generate seed transfer zones using molecular marker data and compare results to empirical seed zones generated from the common gardens. Our aim is to determine whether molecular methods for delineating seed transfer zones reflect results from common gardens. Alternative methods for delineating seed zones can improve our understanding of the ecology of understudied native plant species used in restoration.

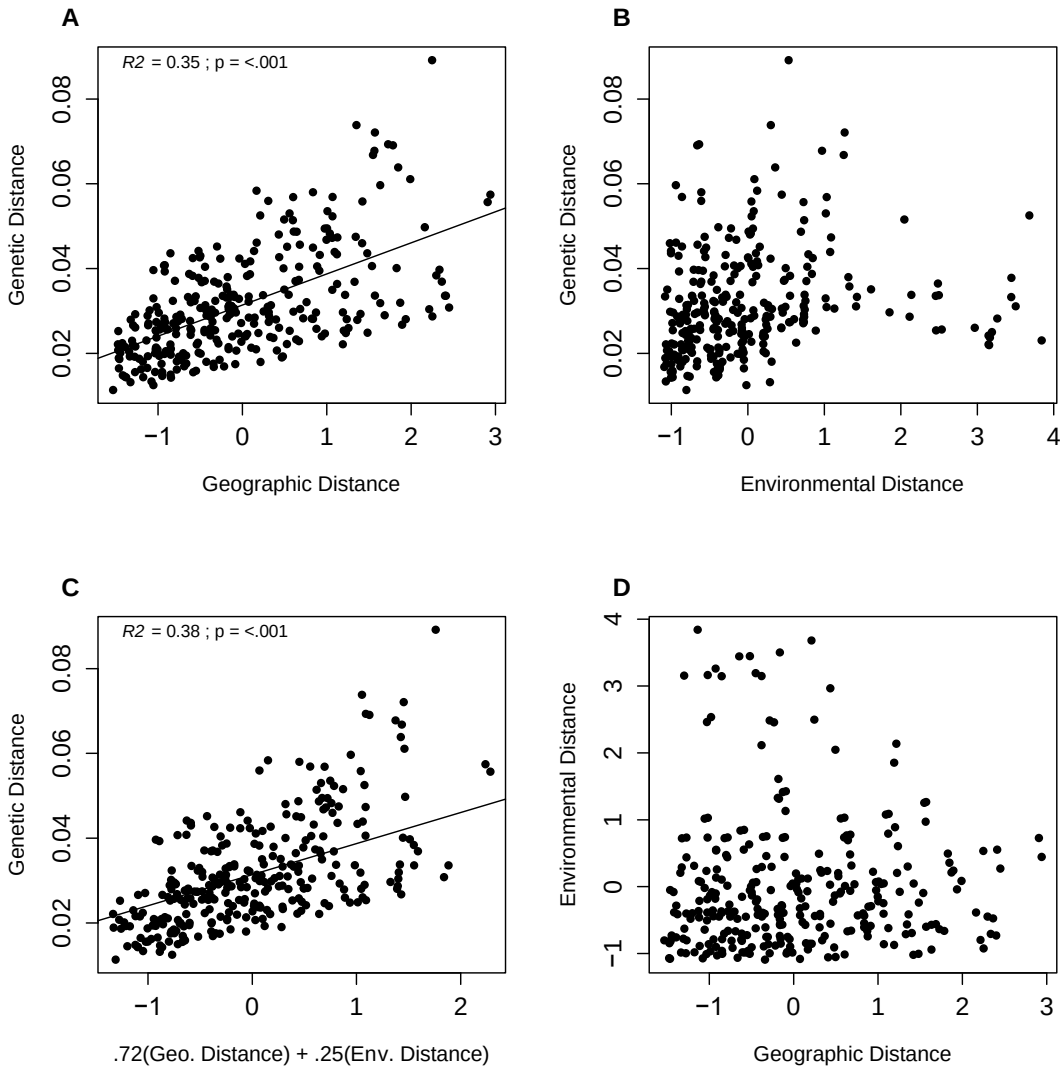

Figure 2: Relationship between geographic, environmental, and genetic distance metrics among populations of true mountain mahogany. Panel A and B represent genetic distance as a function of geographic and environmental distances respectively.

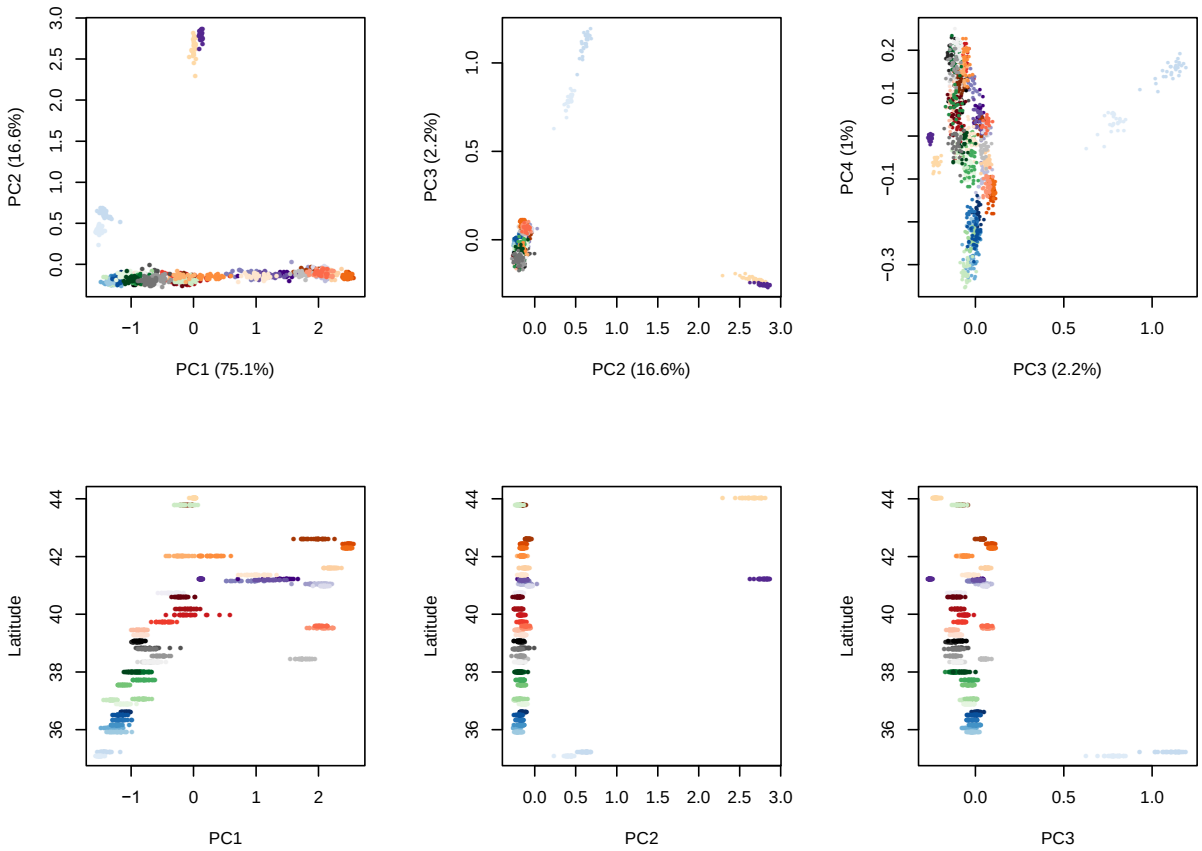

Figure 3: Panel figure of PCA for mountain mahogany. Different colors represent different populations and results highlight the latitudinal cline for genetic diversity in this species. Note that light tan and dark purple (at top of first graph and repeated in all graphs) represent *C. ledifolius* populations, a related species included in analyses for comparison.

Photo 2: *Cercocarpus montanus* in flower

Photo 3: Ongoing genetic research.