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RESEARCH ARTICLE

Latitude of seed source impacts flowering phenology and fitness in translocated plant populations

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Seed sourcing strategies have received considerable attention in the restoration literature and are a key component of effective management for restoration and conservation of natural areas. Research and discussion tend to focus on optimal distances and environmental similarities between seed sources and planting sites. However, given the increasing calls for assisted gene flow and assisted migration, greater consideration of translocating populations in specific directions across climatic gradients is warranted. To the extent that local adaptation proceeds primarily in response to climatic conditions, assisted gene flow across climatic gradients is likely to promote species persistence in the face of climate change. However, if species are adapted to other abiotic and biotic factors, translocating populations across climatic gradients may have unintended and potentially maladaptive consequences. Here, we used extensive collections of seed materials from across the state of Minnesota, a field planting that established common conditions at a location that was near the southern extreme of all the source locations, and subsequent aster modeling of fitness data to examine the overall fitness consequences of translocating populations across the landscape. We found that populations from cooler, northern sources tended to have higher fitness than those from warmer, more southern locations. In addition, populations from more northern locations tended to have earlier flowering phenology relative to populations from more southern sources, perhaps conferring a fitness advantage. Taken together, our results suggest that latitude of origin may be an important factor to take into consideration during seed source selection for restoration work, and that the direction of the effects can be at odds with expectations based on climatic considerations.

Key words: **assisted gene flow, assisted migration, fitness, flowering phenology, latitude, seed source**

Implications for Practice

- Use of geographic distance to guide seed sourcing decisions can be problematic as it ignores other environmental and spatial variables that do not covary cleanly with geographic distance. Latitude of origin may also impact success of translocated populations.
- There is limited evidence supporting an inference that poleward assisted gene flow/migration is generally adaptive.
- Future studies investigating the relative importance to different species of the many environmental factors affected by climate change would provide valuable insight into assisted gene flow and assisted migration practices.
- When using assisted gene flow to augment populations' size and genetic variation, practitioners are advised to limit latitudinal distance between source populations and target populations to ensure sufficient overlap of flowering phenologies and maintain potential for admixture.

Introduction

Grasslands are diverse ecosystems that provide valuable ecosystem services. They are home to thousands of plant and animal species and aid in water infiltration, carbon storage, erosion control, and nutrient retention, as well as offering opportunities for hunting and other recreational activities (Tester 1995; Schulte et al. 2017; Bengtsson et al. 2019). However, grassland habitats are dwindling. Globally, over 45% of grasslands have been lost to agriculture and other uses (Hoekstra et al. 2005), while in Minnesota, U.S.A., less than 1% of original prairie habitat remains relative to pre-European settlement (Samson & Knopf 1994). The drastic loss of native prairie poses concerns about not only the resulting impairment of ecosystem services, but also the persistence of native species. Ecological restoration has become a critically important approach to mitigate habitat loss.

One of the keys to successful habitat restoration is choosing seeds that are likely to thrive in conditions at the restoration site.

Author contributions: NSR designed the study in consultation with RGS; RGS designed the Healthy Prairies Project experimental framework; SAF oversaw and implemented HPP seed collections, common garden planting, and maintenance; NSR conducted the field work for this study; NR analyzed the data with help from RGS; NSR wrote the manuscript with editing from RGS, SAF.

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A common practice is the preferential use of local seed, assuming a similar environment between the seed source and the restoration site (McKay et al. 2005; Broadhurst et al. 2008; Herman et al. 2014). According to evolutionary theory, if divergent selection outweighs gene flow, populations are expected to adapt to their local environments (Kawecki & Ebert 2004). As a result, the fitness of local populations in their home environment is expected to exceed that of foreign populations, and the fitness of populations planted at home sites is expected to exceed their fitness when planted at away sites. Local adaptation has been demonstrated to be widespread among plant species, although not ubiquitous (Leimu & Fischer 2008; Hereford 2009). However, the geographic scale of local adaptation is less clear, as is the magnitude of local adaptation (i.e. size of the fitness difference between local vs. foreign populations and fitness growing at home vs. at an away site).

The success of chosen seed sources may also be influenced by ongoing environmental change such as climate change. If current site conditions no longer match historical conditions, local seeds may no longer thrive. There is concern that local populations may not evolve quickly enough to keep pace with environmental changes, resulting in adaptational lag (Aitken et al. 2008). In response, it has been suggested that restoration practitioners engage in assisted migration (Aitken & Bemmels 2016), translocating seed from locations whose historical conditions more closely match the restoration site's current and predicted future conditions. This generally implies moving populations poleward, or in montane areas to higher elevations, with the idea that they are already adapted to the warmer temperatures predicted for these locations. Assisted gene flow is a related concept where populations are translocated along a climate gradient for the purpose of augmenting a resident population rather than initiating a new one. Assisted gene flow and assisted migration have been widely discussed (Aitken & Whitlock 2013; Breed et al. 2013; Vitt et al. 2016) and considerable work has been done with tree species (Williams & Dumroese 2013; Aitken & Bemmels 2016). However, there is a need for additional research into the logistics and effectiveness of this approach, particularly for herbaceous species (Hewitt et al. 2011; Bucharova 2017).

One potential consequence of translocating plant populations latitudinally is altered phenology, i.e. changes to timing of key life history events. Many species cue on photoperiod to initiate different life history stages, such as flowering. Moving populations latitudinally can cause their timing of flowering to differ from that of resident populations. For example, in a recent study Wadgymar and Weis (2017) translocated *Chamaecrista fasciculata* from Minnesota, Missouri, North Carolina, and Pennsylvania, to a common garden in Ontario. They found that in this extreme northerly location, plants from higher latitudes tended to begin flowering earlier than those from more southern latitudes, as Etterson (2004) also found for this species. Scalone et al. (2016) found a similar trend for *Ambrosia artemisiifolia*. As the shift in flowering timing increases, so do possible repercussions, such as an inability to complete fruit production before the onset of cold weather in the fall.

With the goal of informing choice of seed sources, the objective of this research was to address the following questions: (1) How does translocation along a latitudinal gradient affect flowering phenology? (2) How does translocation across latitudes and geographic distances impact fitness? (3) How does the degree of adaptation vary among populations sampled across an extensive geographic scale? A common garden approach was used to address these questions. Seed of three perennial prairie species, *Anemone cylindrica*, *Dalea candida*, and *Dalea purpurea*, were each collected from 12 remnant prairies in Minnesota and then grown in a field planting in southeastern Minnesota. Date of first and last flower were recorded in order to assess the impact on phenology of translocating populations along a latitudinal gradient. Survival and seed head production were used to estimate fitness. Local adaptation was detected if populations from nearby sites had higher fitness in the common garden than those from more distant sites, or if populations from sites having similar climatic conditions to the common garden had higher fitness in the common garden than populations from dissimilar sites. A better understanding of the impacts on fitness of translocating populations will contribute to improving seed sourcing practices such as assisted gene flow and assisted migration, aiding future restoration and conservation work.

Methods

Study Species

As part of a larger study assessing the geographic scale of local adaptation of six species of prairie plants in Minnesota (U.S.A.), three perennial forb species were chosen for this study: *Anemone cylindrica* A. Gray (thimbleweed), *Dalea candida* Michx. ex Willd. (white prairie clover), and *Dalea purpurea* Vent. (purple prairie clover). These three species are all native to North American grasslands and are commonly used in prairie restoration.

D. candida and *D. purpurea* are both members of the legume family, Fabaceae, which form symbiotic relationships with nitrogen fixing rhizobia. Both *Dalea* species are found throughout the central United States, ranging from Texas to Minnesota, and from Indiana to Colorado (*D. purpurea*) and Utah (*D. candida*). Within Minnesota, *D. candida* and *D. purpurea* are present throughout the southern, central, and northwestern portions of the state (USDA n.d.). Both *Dalea* species attract numerous pollinator species, which gather both nectar and pollen (Cane 2006; Applegate et al. 2007; Pearce et al. 2012). *D. purpurea* is known to be mainly xenogamous (Cane 2006); breeding system for *D. candida* is not definitively known but is generally described as cross-pollinating (Wynia 2008; Molano-Flores et al. 2011).

A. cylindrica is a member of the buttercup family, Ranunculaceae. *A. cylindrica* is native throughout Minnesota and can be found from New England to the Rocky Mountains (USDA n.d.). Unlike *D. candida* and *D. purpurea*, *A. cylindrica* is primarily autogamous (Molano-Flores & Hendrix 1998) and receives far fewer pollinator visits.

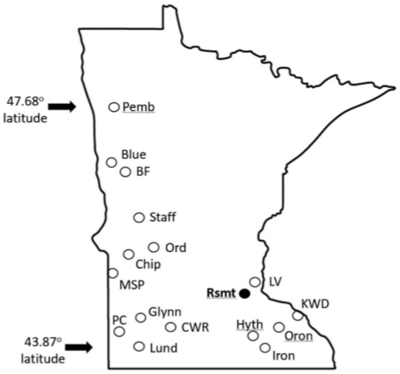

Figure 1. Map of seed sources for *Anemone cylindrica*, *Dalea candida*, and *Dalea purpurea* (open circles) and common garden site (filled circle) in Minnesota, U.S.A.

Seed Collection

A. cylindrica, *D. candida*, and *D. purpurea* seed were each collected from 12 native remnant prairies (16 prairies total) in Minnesota in 2014, under permit from the Minnesota Department of Natural Resources and The Nature Conservancy. Source sites were chosen to span much of Minnesota's native prairie, with three in southeastern Minnesota, three in southwestern Minnesota, three in west central Minnesota, and three in northwestern Minnesota (Fig. 1). At each collection site, seed was collected from at least 60 individuals per species. Seeds were collected from plants at least 3 m apart in order to minimize the chance of collecting seed from closely related individuals (Fenster 1991).

Plant Propagation

For each species and each of its 12 collection sites, 50 seeds were randomly chosen from each of 40 individuals. These seed were then pooled by site before cleaning for *A. cylindrica* and *D. candida* and after cleaning for *D. purpurea*. For *D. candida* and *D. purpurea* seed, hulls were removed and seeds were triple scarified using a sandpaper-lined drum. Seed for all three species was then stratified as follows: All seed from one species at one site was placed in a labeled, one gallon Ziploc bag containing a wet paper towel. Bags were placed in a cold room at 4°C for 30 days (*D. candida* and *D. purpurea*) or 60 days (*A. cylindrica*).

After stratification, seeds were planted in flats containing MVP Sungro Professional Growing Mix in January 2015. Each flat contained seed from one species at one site. Flats were then placed in the growth chamber in a randomized array. Growth chamber settings alternated between 12 hours at 16°C with lights off and 12 hours at 25°C with lights on. Relative humidity was maintained at 40%. In January–June 2015, seedlings were transplanted into D19 Deepot Cells containing MVP Sungro Professional Grow Mix and moved to the greenhouse. Seedling arrangement was randomized in the greenhouse. Once seedlings had at least two true leaves, they were transplanted into field plots in June–July 2015.

Field Planting Design

The field planting was located in Rosemount, MN at the Rosemount Research and Outreach Center. Within the common garden there were 12 plots, each separated by at least 100 m in order to minimize cross pollination between plots (Figs. 2 & S1). Before planting, plots were sprayed with Roundup at a rate of 3.5 L per hectare and then tilled to kill agricultural weeds. The soil in each plot was tested for phosphorus (P) and potassium (K). Plots were then fertilized with P_2O_5 and K_2O as needed based on soil tests (application rates ranged from 0 to 45 kg/ha). Plots were covered with weed barrier to minimize competition with agricultural weeds at the site. In addition, plots were weeded as needed throughout the growing season and in subsequent years to reduce weed pressure, and fenced to minimize herbivory by deer and rabbits.

Each plot was divided into subplots, each of which contained 100 individuals from one of the three study species: *A. cylindrica*, *D. candida*, or *D. purpurea*. Seed sources for each species were randomly assigned to the 12 plots such that each plot had a random combination of seed sources for the three species, while each subplot comprised individuals of a single species from a single source. Seedlings were transplanted into holes cut in the weed barrier and watered as needed after transplanting. Seedlings were arranged in alternating rows of 12 and 13 plants in hexagonal spacing. Seedlings were 0.3 m apart from each other within each row and also from their two nearest neighbors in both flanking rows. Seedling spacing was consistent across all three study species.

Climate Data

Climate data were gathered from publicly available sources for each of the source sites. Mean July maximum temperatures and mean January minimum temperatures are from the 1981–2010 Normals Map Tool on the Minnesota Department of Natural Resources website (https://www.dnr.state.mn.us/climate/summaries_and_publications/normalsportal.html). Mean annual precipitation was calculated as the mean of annual observed precipitation totals for the years 1995–2014 from the National Weather Service's database (https://water.weather.gov/precip/). The coefficient of variation (CV) for mean annual precipitation was calculated as σ/μ, the standard deviation divided by the mean. Mean annual evapotranspiration was calculated as the mean of annual evapotranspiration for the years 2000–2017. Evapotranspiration data was acquired from the Simplified Surface Energy Balance Actual Evapotranspiration data for the Conterminous United States available at the USGS Geo Data Portal webpage (https://cida.usgs.gov/gdp/). Climate data are displayed in Table 1.

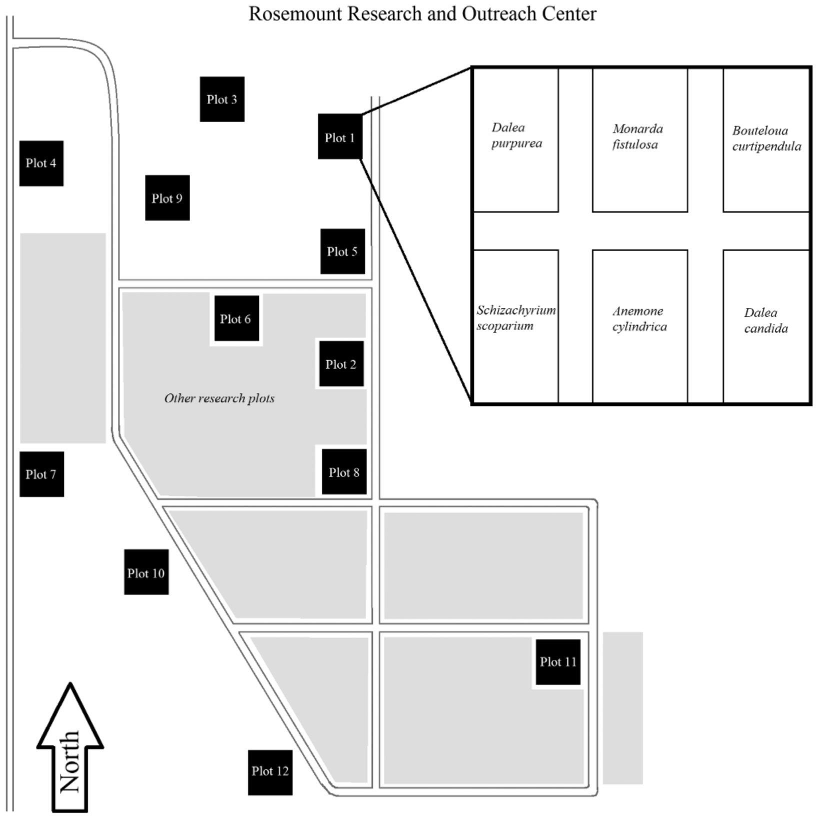

Figure 2. The field planting was located at the Rosemount Research and Outreach Center in Rosemount, MN, U.S.A. The field planting consisted of 12 plots. All plots were separated by at least 100 m to minimize cross pollination between plots. Each plot was divided into six subplots, each of which contained 100 individuals from one species. Seed sources for each species were randomly assigned to the 12 plots such that each plot had a random combination of seed sources for the six species, while each subplot was comprised of individuals of a single species from a single source. *Anemone cylindrica*, *Dalea candida*, and *Dalea purpurea* were the focal species for this project.

Phenology Data

Phenological data were collected in 2017 for *A. cylindrica*, *D. candida* and *D. purpurea*. Plots were visited one to two times a week beginning on 27 April 2017, around the time of emergence for *A. cylindrica*, and ending on 6 September 2017, at the conclusion of flowering for *D. purpurea*. The following data were recorded for all three species: date of emergence in spring and presence or absence of a living plant during each census. In addition, for each plant, the number of flower heads with open flowers was recorded at each census for *A. cylindrica* and *D. candida*. Length of flowering period was calculated as date of last flower – date of first flower +1. For *D. purpurea*, only

Table 1. Location and climate aspects of source locations. Distance given is distance between source site and common garden location in Rosemount, MN, U.S.A. (rsmt*).

Seed Source	*Latitude*	*Distance (km)*	*Mean July High (°C)*	*Mean Jan. Low (°C)*	*Mean Annual Precip. (cm)*	*CV of Mean Annual Precip.*	*Mean Annual Evapotranspiration (mm)*
Pemb	47.68	417	26.39	−20.00	54.61	0.14	423.21
Blue	46.85	356	27.22	−18.33	66.04	0.19	470.90
BF	46.69	330	26.67	−18.89	68.58	0.19	456.79
Staff	45.82	243	27.22	−17.78	40.64	0.26	503.05
Ord	45.45	192	27.22	−16.67	67.95	0.19	536.47
Chip	45.15	237	27.78	−16.67	48.90	0.16	552.84
LV	44.80	24	27.78	−14.44	75.57	0.15	503.74
MSP	44.77	269	28.06	−15.56	77.47	0.69	487.05
Rsmt*	44.70	0	27.78	−15.00	101.60	0.13	555.95
Glynn	44.26	214	27.78	−15.56	66.04	0.27	537.47
KWD	44.26	103	27.78	−14.44	81.28	0.19	488.00
CWR	44.20	171	27.78	−15.56	72.39	0.23	509.16
Oron	44.14	77	27.22	−15.00	82.55	0.14	498.11
PC	44.12	253	27.78	−15.56	77.47	0.22	503.11
Hyth	44.02	74	26.94	−14.44	87.00	0.17	496.05
Lund	43.93	227	26.67	−15.00	67.31	0.18	604.16
Iron	43.87	93	26.67	−14.44	88.90	0.15	487.11

the presence/absence of flowers on each plant was recorded due to time constraints and the very large number of flowerheads on *D. purpurea*.

Fitness Data

Fitness data were collected in 2017 and 2018 for *A. cylindrica* and *D. candida*. Fitness data were not collected for *D. purpurea* in either year due to time limitations. Number of seed heads per plant was used as an approximation of reproductive fitness. Seed head number has been shown to approximate an individual's contribution of seed to the next generation (Clark & Watkins 2010; Mahajan et al. 2020), while avoiding the laborious process of counting the very small seeds on each head individually and determining viability of each seed. Seed heads were counted at the end of the growing season for each species when the majority of plants had finished flowering. This occurred in July for *A. cylindrica* and August for *D. candida*.

Analysis of Phenology Data

Phenology data for *A. cylindrica*, *D. candida* and *D. purpurea* were analyzed using linear models in R (R Core Team 2019). Date of first flower, date of last flower, and length of flowering period were regressed on latitude of seed source in order to assess the relationship between latitude and flowering phenology. Adjusted R^2 values are reported. In addition, least squares means for date of first flower, date of last flower, and length of flowering period were estimated for each population using linear models including population as the sole predictor. Plants that did not emerge in spring, or that did not flower after emergence were excluded from these analyses.

Analysis of Fitness Data

Aster analysis (Shaw et al. 2008) was used to model mean fitness of *A. cylindrica* and *D. candida* populations in the field plantings. Individual fitness comprises multiple components—germination, survival, flowering, and fruiting—each characterized by a particular statistical distribution (e.g. Bernoulli, Poisson, normal). This situation is further complicated in perennial species because these components are expressed over the course of multiple growing seasons and include multiple rounds of seed production. Aster models use appropriate statistical distributions for each component of fitness, with later elements of fitness dependent upon earlier elements, thereby allowing analysis of overall fitness in a single model that encompasses all fitness components. Aster is available as a package in R (Shaw et al. 2008; R Core Team 2019).

A. cylindrica and *D. candida* are perennial species. Their fitness is dependent on survival and seed head production in multiple years, as depicted in the graphical model (Fig. 3). Bernoulli distributions were used to model survival from 2015 to 2017 and from 2017 to 2018. Poisson distributions were used to model number of seed heads per plant in 2017 and 2018 for analyses (1), (2), and (3) (described further below). Negative binomial distributions were used for analysis (4) (described further below) as they were a better fit for these relationships. Fitness of individual plants, comprising survival and seed head production, was modeled using subsets of the predictors: seed source, latitude of source site, geographic distance between source site and common garden site, mean July maximum temperature of source site, mean January minimum temperature of source site, mean annual precipitation of source site, CV of mean annual precipitation at source site, mean annual evapotranspiration of source site, and date of first flower. Statistical significance of factors was tested using R's ANOVA procedure to compare nested models. Separate analyses were conducted for (1) source site, (2) latitude of source site and geographic distance between source site and common garden, (3) mean July maximum

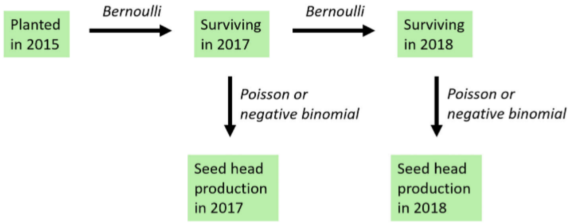

Figure 3. Graphical aster model depicting dependence of later fitness components (seed head production in 2017, survival in 2018, seed head production in 2018) on earlier fitness components (planting in 2015 and survival in 2017), for the study species *Anemone cylindrica* and *Dalea candida*. Bernoulli distributions were used to model survival from 2015 to 2017 and from 2017 to 2018. Poisson distributions were used to model seed head production for aster analyses involving latitude of source, distance between source and common garden site, and source climate conditions. Negative binomial distributions were used to model seed head production for aster analyses involving date of first flower.

temperature, mean January minimum temperature, mean annual precipitation, CV of mean annual precipitation, and mean annual evapotranspiration, and (4) date of first flower.

Assessment of Local Adaptation

In a common garden experiment, the strength of local adaptation of the study species is determined by examining fitness in the common garden along with attributes of the sources, such as geographic distance or environmental characteristics. Here, we focused on geographic distance between seed source and common garden site and five climate measures (described previously in Climate Data section). We detect the presence of local adaptation in a particular species if populations that have higher fitness in the common garden come from sources having similar climatic conditions and/or come from a shorter geographic distance, while populations that have lower fitness in the common garden come from sources having dissimilar climatic conditions and/or are from a more distant source. During the years of this study, our common garden location tended to have climatic conditions that were extreme relative to historic conditions at our source sites: high mean July maximum temperature (Fig. S2), high mean January minimum temperature (Fig. S3), extremely high mean annual precipitation (Fig. S4), low CV of mean annual precipitation (Fig. S5), and high mean annual evapotranspiration (Fig. S6). Therefore, in the comparisons of aster models, evidence of local adaptation for a given species was indicated by the following relationships with fitness: positive relationship with mean July high temperature, positive relationship with mean January low temperature, positive relationship with mean annual precipitation, negative relationship with mean CV of annual precipitation, and positive relationship with mean annual evapotranspiration.

Results

Phenology

The regression analysis showed a significant negative relationship between date of first flower and source latitude, with northern populations tending to start flowering earlier than southern populations in all three species (*A. cylindrica*, $p = 0.0033$, $R^2 = 0.025$, *D. candida*, $p = 0.0099$, $R^2 = 0.015$, *D. purpurea*, $p < 0.0001$, $R^2 = 0.10$) (Fig. 4). (Detailed p-values for all instances where $p < 0.0001$ are given in Table S1.) The estimated difference in average date of first flower between the northernmost and southernmost populations is 2 days for *A. cylindrica*, 4 days for *D. candida*, and 7 days for *D. purpurea*. The mean date of last flower was also earlier in plants from northern sources for *D. candida* (11-day difference, $p < 0.0001$, $R^2 = 0.20$) and *D. purpurea* (7-day difference, $p < 0.0001$, $R^2 = 0.076$) but not for *A. cylindrica* ($p = 0.37$) (Fig. S7). Mean length of flowering period was shorter for plants from northern sources for *D. candida* (8-day difference, $p < 0.0001$, $R^2 = 0.078$) but was not significantly different by source latitude for *A. cylindrica* ($p = 0.52$) or *D. purpurea* ($p = 0.62$) (Fig. 5). There was substantial variation around the linear prediction of population means for all flower phenology data (Figs. 4, 5, & Fig. S7). Three populations had sample sizes of less than five for phenology data as few plants survived and/or flowered in these populations (*D. candida* populations from Ord and KWD, *D. purpurea* population from Glynn).

Fitness and Local Adaptation

In aster models with latitude as the sole predictor, plants from northern sources had higher fitness in the common garden than those from southern sources for *A. cylindrica* ($p < 0.0001$) and marginally so for *D. candida* ($p = 0.084$). With distance as the sole predictor, plants from more distant sources had higher fitness for *A. cylindrica* ($p < 0.0001$) while there was no relationship between distance and fitness for *D. candida* ($p = 0.47$). Aster models containing both latitude and distance had a better fit than either the distance-only model ($p < 0.0001$, $p < 0.0001$) or the latitude-only model ($p < 0.0001$, $p = 0.00033$) for *A. cylindrica* and *D. candida*, respectively. These models showed that fitness depended positively on latitude of origin and negatively on distance from source site for both species. For a given latitude of

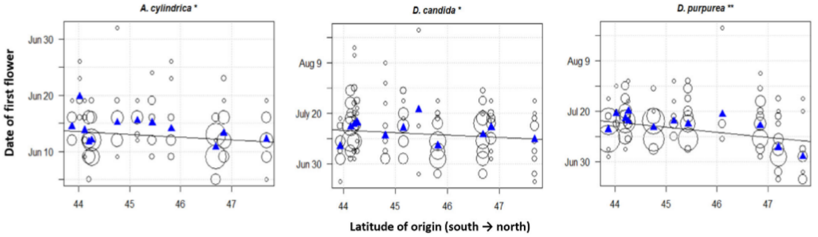

Figure 4. Date of first flower of *Anemone cylindrica* ($n = 645$), *Dalea candida* ($n = 569$), and *D. purpurea* ($n = 453$) overlaid with regression of first date of flowering by latitude ($*p < 0.01$, $**p < 0.0001$). Blue triangles indicate predicted population means based on linear models using source as the sole predictor.

origin, closer plants tended to have higher fitness, as can be seen with the *D. candida* populations from sources around 44° of latitude in Figure 6. Similarly, for a given distance from the source, those from higher latitudes expressed higher mean fitness, such as the *A. cylindrica* populations from sources in the 200–300 km range in Figure 6. For *A. cylindrica*, the direction of the effect for distance differs depending on whether latitude is included in the model. In the joint model, fitness has a negative relationship with distance, while in the model with distance as the sole predictor, fitness has a positive relationship with distance. The change in the effect of distance is likely due to the strong impact of latitude of origin on fitness and the fact that the more distant sources in this study also tended to be from more northerly locations.

Aster analysis of the relationship between fitness and climatic factors yielded contrasting results for *A. cylindrica* and *D. candida*. For *A. cylindrica*, seeds from sources having a higher mean July maximum temperature, lower mean January minimum temperature, higher mean annual precipitation, and higher mean annual precipitation CV tended to have higher fitness at the common garden site ($p < 0.0001$) (Table S2). (Mean annual evapotranspiration did not improve model fit for *A. cylindrica*, $p = 0.077$.) For *D. candida*, on the other hand, seeds from sources having a lower mean July maximum temperature, higher mean January minimum temperature, lower mean annual precipitation, higher mean annual precipitation CV, and higher mean annual evapotranspiration tended to have higher fitness at the common garden site ($p < 0.0001$) (Table S2). In comparison to source sites, environmental conditions at the common garden site during the study period were characterized by a high mean July maximum temperature, high mean January minimum temperature, very high mean annual precipitation, low mean annual precipitation CV, and high mean annual evapotranspiration. For *A. cylindrica*, seeds from sources having climatic conditions more similar to the common garden site tended to have higher fitness (with the exception of January minimum temperatures), evidence of local adaptation of this species to climatic factors within the geographic area covered by this study. For *D. candida*, seeds from sources with contrasting climatic conditions to the common garden site tended to have higher fitness (again, with the exception of January minimum temperatures), evidence not consistent with local adaptation to climatic factors for this species. Predicted mean population fitness values for climate and latitude/distance analyses are displayed in Table 2.

Aster analysis of fitness in relation to date of first flower indicates that fitness decreases as date of first flower gets later in

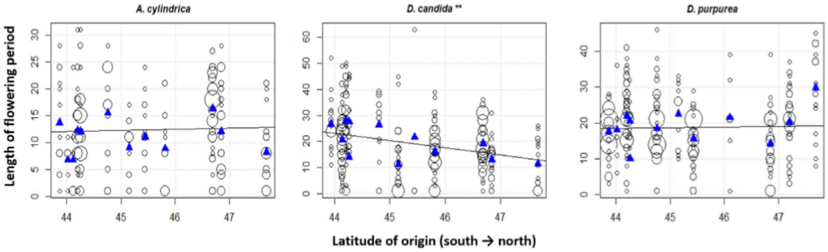

Figure 5. Length of flowering period of *Anemone cylindrica* ($n = 645$), *Dalea candida* ($n = 569$), and *D. purpurea* ($n = 453$) overlaid with regression of length of flowering period by latitude ($**p < 0.0001$). Blue triangles indicate predicted population means based on linear models using source as the sole predictor.

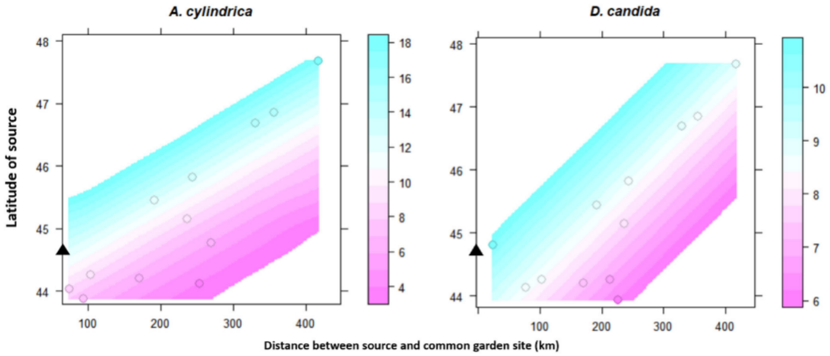

Figure 6. Predicted mean fitness of *Anemone cylindrica* ($n = 645$) and *Dalea candida* ($n = 569$) populations based on latitude of their source sites and distance between common garden and seed source. The colored bar to right of each figure is the fitness scale for each species, with higher numbers indicating higher fitness levels. Black triangles indicate latitude of common garden site.

both *A. cylindrica* and *D. candida* (Fig. 7). A quadratic model provided a better fit than the linear model for both species (*A. cylindrica*: $p = 0.00024$, *D. candida*: $p < 0.0001$).

Discussion

Seed sourcing strategies have received considerable attention in the restoration literature and are a key component of effective management for restoration and conservation of natural areas. Research and discussion tend to focus on optimal distances and environmental similarities between seed sources and planting sites (e.g. McKay et al. 2005; Broadhurst et al. 2008; Herman et al. 2014). However, given the increasing discussion of assisted gene flow and assisted migration (Whitely et al. 2015; Vitt et al. 2016; Bell et al. 2019), greater consideration of translocating populations in specific directions across climatic

Table 2. Mean population fitness is given for each source based on aster models. Sources are listed by latitude, with the most northern latitude at the top of the table. Empty cells indicate locations where seed was not collected for a given species. Columns 2 and 5 use source as the sole predictor. Columns 3 and 6 use latitude of source and distance between source and common garden site as the predictors. Column 4 uses the following environmental factors of source sites as the predictors: mean July maximum temperature, mean January minimum temperature, mean annual precipitation, and CV of mean annual precipitation. Column 7 uses mean annual evapotranspiration, in addition to the previous four environmental factors, as the predictors.

	Mean Population Fitness					
	Source	*Lat. + Dist.*	*Environment*	*Source*	*Lat. + Dist.*	*Environment*
Seed Source	A. cylindrica	A. cylindrica	A. cylindrica	D. candida	D. candida	D. candida
Pemb	2.90	17.49	12.35	2.71	8.98	12.69
Blue	18.06	14.70	22.98	4.70	8.60	5.78
BF	41.76	14.79	22.90	18.18	8.75	8.35
Staff	2.18	12.83	0.32	22.46	8.72	16.74
Ord	5.76	12.63	6.95	0.51	8.93	5.17
Chip	3.62	8.89	1.02	7.76	7.78	4.74
LV				7.44	10.62	7.05
MSP	7.96	5.91	8.33			
Glynn				5.37	6.81	7.97
KWD	28.59	8.90	10.90	0.01	8.43	8.16
CWR	5.36	6.28	9.57	5.91	7.33	7.03
Oron				19.21	8.63	7.23
PC	3.04	3.95	18.24			
Hyth	1.11	8.64	4.84			
Lund				5.52	6.19	8.84
Iron	1.78	7.12	3.95			

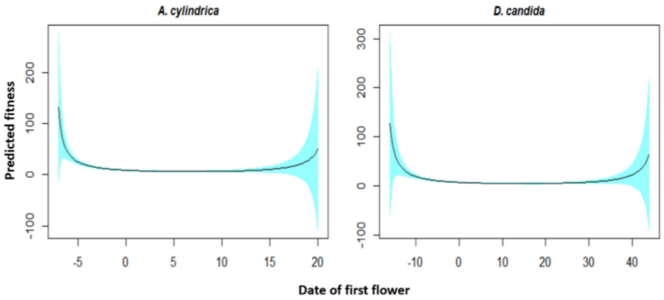

Figure 7. Predicted fitness of *Anemone cylindrica* ($n = 645$) and *Dalea candida* ($n = 569$) based on date of first flower. Initial flowering dates in figure are adjusted such that a zero date of first flower is the median date of flowering for that species. Light blue area indicates ± 95% CI.

gradients is warranted. To the extent that local adaptation proceeds primarily in response to climatic conditions, assisted gene flow across climatic gradients is likely to promote species persistence in the face of climate change. A growing body of literature has addressed this issue, particularly in tree species, using genecological models to inform translocation along climatic gradients (e.g. Hamann et al. 2011; Kilkenny 2015; Mahoney et al. 2020). However, if species are adapted to other abiotic and biotic factors, translocating populations across climatic gradients may have unintended and potentially maladaptive consequences (Bucharova 2017; Wadgymar & Weis 2017). Here, we used extensive collections of seed materials from across the state of Minnesota, a field planting that established common conditions, and subsequent aster modeling of fitness data to examine the fitness consequences of translocating populations across the landscape. We found that, in the common garden near the southern limit of the source locations, populations from cooler, northern source sites tended to have higher fitness than those from warmer, more southern locations. In addition, populations from more northern locations tended to have earlier flowering phenology relative to populations from more southern sources, perhaps conferring a fitness advantage. Evidence of local adaptation to climate was less clear, with *A. cylindrica* showing signs of local adaptation to three out of five climate factors, while *D. candida* showed signs of local adaptation to only one of five factors. Taken together, our results suggest that latitude of origin may be an important factor to take into consideration during seed source selection for restoration work.

Local adaptation is commonly found in plants, although it is not universal. In Hereford's (2009) quantitative survey, local adaptation was found in 65% of 892 estimates, while Leimu and Fischer (2008) found local adaption in 45% of 1,032 pairwise comparisons in their meta-analysis. Although it is useful to have an understanding of the general prevalence of local adaptation, in restoration ecology it is also helpful to understand the spatial extent to which specific species manifest local adaptation. In our study, evidence of local adaptation within Minnesota for *A. cylindrica* and *D. candida* was mixed. Latitude of origin had a positive effect for both species, with populations from more northern locations (which were also generally more distant) tending to outperform populations from more southern locations. Once latitude was accounted for, populations from more local sources tended to have higher fitness than those from more distant sources, again for both species. Our results provide evidence of local adaptation for both *A. cylindrica* and *D. candida* within the 75,000 km^2 area (approximately 420 km north to south and 220 km east to west) of our study. For these species there is likely to be a decrease in fitness when seed is translocated over greater distances, a finding which supports the use of more local seed during restoration work. These results align with the current seed sourcing guidelines of both the Minnesota Department of Natural Resources (Schulte & Westbrook 2013) and the Minnesota Board of Water and Soil Resources (Shaw 2019). Both organizations use a tiered approach to seed sourcing where collections are preferentially made in local areas and only from farther away when local sources are unavailable, with a maximum recommended seed transfer distance of 175 miles (282 km). Our work also demonstrates the importance of latitude of origin, which will be discussed further below.

Local adaptation to climatic factors differed between the two species. We detected evidence of local adaptation in *A. cylindrica* to July temperature highs and annual precipitation, but not to January low temperatures or annual evapotranspiration. *D. candida*, in contrast, showed evidence of local adaptation only to January low temperatures. Our results highlight the difficulty of trying to generalize about effects of environmental distance between sites. In addition, our results for *D. candida* indicate that environmental similarity as measured between source and planting site may not always aid in predicting transplant success, contrary to common assumptions. The differing responses of these two species underscore the difficulty and importance of developing species-specific restoration guidelines.

Several important environmental factors vary with changing latitudes, including photoperiod, temperature, growing season length, and biodiversity (De Frenne et al. 2013). As a result, translocating populations across latitudes may result in fitness differences based on latitude of origin. However, the direction of potential fitness differences is variable. In a study of two *Quercus* species in Minnesota, Etterson et al. (2020) found that seedlings from further south outperformed seedlings from more northern locations when planted in northern common gardens. Similarly, McGraw et al. (2015) found that the optimum temperature for populations of long-lived *Eriophorum vaginatum* in Alaska was at locations 140 km north of their home sites. In contrast, Torang et al. (2015) and Colautti and Barrett (2013) found strong evidence of local adaptation in *Arabis alpina* and *Lythrum salicaria*, respectively, as regional populations outperformed nonlocal populations in common gardens in both studies. Our findings, that northern populations tended to outperform southern populations, present yet a third scenario (although it is important to note that our study included only a limited range of sources from south of our common garden location). Our findings are similar to those of Wadgymar and Weis (2017) who found that northern populations of *Chamaecrista fasciculata* outperformed southern populations when grown in a common garden. A common assumption in the literature on assisted gene flow is that when southern populations are translocated northwards, they will outperform resident populations because they are already adapted to the warming temperatures that climate change is imposing on northern locations (Parmesan & Hanley 2015). The variability among the studies mentioned here indicates that poleward assisted gene flow may not always successfully address population declines due to climate change and may in some cases be maladaptive.

Latitude of origin can impact flowering phenology of translocated populations, either due to changes in photoperiod or changes in accumulation of growing degree days (Griffith & Watson 2006; Wadgymar et al. 2015). Flowering phenology, in turn, can impact fitness via temporal mismatch with key pollinators and other mutualists (Rafferty & Ives 2012), reproductive isolation from resident populations (Weis 2015; Wadgymar & Weis 2017), and inability to complete fruit production before the onset of cold weather in the fall (Griffith & Watson 2006). For *A. cylindrica*, *D. candida*, and *D. purpurea*, we found that populations from northern sources tended to flower earlier than populations from southern sources when grown in our field planting. This may be due to changes in the accumulation of growing degree days, where northern sources have a lower threshold of growing degree day accumulation required to initiate flowering relative to southern sources. Earlier flowering was associated with increased fitness for *A. cylindrica* and *D. candida*, the two species for which we collected fitness data. Some research has indicated that earlier flowering may be associated with increased fitness due to pollinator preferences (Elzinga et al. 2007). Alternatively, it may be that earlier flowering aligns the plant's life history with the earlier start to the growing season that is resulting from increased temperatures due to climate change. Parmesan and Yohe (2003) conducted a meta-analysis of recent phenological change, looking at 172 species including plants, birds, butterflies, and amphibians. They found a mean shift toward earlier spring timing of 2.3 days per decade, matching climate change predictions. Perhaps the northern populations in our study are better suited to the earlier spring occurring to their south, resulting in increased fitness relative to more southern populations which tend to flower later. These results suggest that when considering the impacts of climate change, it may be informative to look beyond increases in temperature per se and consider additional factors such as changes to the start and end of the growing season.

In addition to the fitness decrease and flowering delay associated with populations from more southern sources in this project, latitude of origin also determined the degree of flowering overlap between populations from different latitudes, particularly for *D. candida* and *D. purpurea*. The difference between the predicted initiation of flowering of the northernmost and southernmost populations was four and seven days, while predicted completion of flowering differed by eleven and seven days for *D. candida* and *D. purpurea*, respectively. This limits the degree of flowering overlap between the two populations, thereby limiting the potential for admixture. Although in this experiment there was still substantial overlap between populations, sourcing populations over greater distances could result in the complete separation of flowering. For example, Wadgymar et al. (2015) grew *C. fasciculata* from Minnesota, Missouri, North Carolina and Pennsylvania in a common garden in Ontario. They found that the North Carolina population was almost completely reproductively isolated from the Minnesota and Pennsylvania populations in the common garden due to its later flowering schedule. In similar work, Weis (2015) collected *Brassica rapa* from three sites along a flowering phenology cline in California and grew them in a common garden. Weis (2015) found that differences in flowering time limited admixture between the populations by up to half in the common garden. Such differences in flowering schedule could disrupt plans for assisted gene flow if immigrant populations do not hybridize with resident populations (Way & Montgomery 2015). Our research, along with that of Wadgymar et al. (2015) and Weis (2015) suggest that impacts of flowering phenology on the admixture of translocated populations should be taken into consideration in plans for assisted gene flow.

Assisted gene flow and assisted migration are two seed sourcing strategies that have been suggested to mitigate the adverse impacts of climate change that are affecting many native plant and animal populations. In this study, we used three native prairie plant species and a common garden approach to demonstrate that latitude of origin can impact the success of populations that are translocated across latitudes. In particular, we found that populations from northern sources tended to have earlier flowering schedules and higher fitness than those from southern sources. Our findings suggest that despite the current focus on northward assisted gene flow and migration, there may be situations where translocation in different directions would be beneficial. Climate change causes many environmental factors to differ from historic means, but these factors do not consistently covary across latitudes. Future studies that investigate the relative importance of these different factors to populations and species would provide valuable insight for assisted gene flow and assisted migration practices. In addition, one limitation of this study was the use of a single common garden. Further studies that incorporate multiple common gardens at different latitudes could provide a more nuanced understanding of the impacts of movement of populations across latitudes.

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Supporting Information

The following information may be found in the online version of this article:

Table S1. Detailed values for all *p*-values of less than 0.0001.
Table S2. Results of climate analysis using ANOVA to compare nested aster models.
Figure S1. One of the twelve plots in the field planting.
Figure S2. Mean July maximum temperature (°C) of seed sources, graphed by longitude and latitude of source location.
Figure S3. Mean January minimum temperature (°C) of seed sources, graphed by longitude and latitude of source location.
Figure S4. Mean annual precipitation (cm) of seed sources graphed by longitude and latitude of source location.
Figure S5. Mean annual precipitation CV of seed sources graphed by longitude and latitude of source location.
Figure S6. Mean annual evapotranspiration (mm) of seed sources graphed by longitude and latitude of source location.
Figure S7. Date of last flower of *A. cylindrica* ($n = 645$), *D. candida* ($n = 569$), and *D. purpurea* ($n = 453$) overlaid with regression of last date of flowering by latitude.

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