Does the frost tolerance of *Chamaecrista fasciculata* seedlings differ along a regional gradient in the Midwest?

Ana I. Flores[1], Christopher Woolridge[2], Andrea Kramer[3], Jeremie Fant[2,3]

[1]Florida International University, [2]Northwestern University, [3]Chicago Botanic Garden

aflor039@fiu.edu; christopherwoolridge2017@u.northwestern.edu; akramer@chicagobotanic.org; jfant@chicagobotanic.org

INTRODUCTION

Land managers have traditionally aimed for obtaining locally-sourced seed for use in ecological restorations. The arguments for using only local sources, while valid, lack consideration of increasing climate variability and the difficulties that these abiotic factors pose to species' adaptive potential (2). Predictive provenancing combines climate projections and information from common garden experiments into models that can inform seed sourcing decisions (5). However, these models face challenges in anticipating increasingly unpredictable climate, including the damaging effects of extreme weather events on individuals with mistimed phenology.

A common garden study was conducted to compare the phenology and fitness of plants sourced from three potential regions differing by latitude. After observing low seedling establishment of southern-sourced plants in *Chamaecrista fasciculata*, we compared frost tolerance across sources, generation, and development stage. We expect that if frost tolerance differs by seed source, southern-sourced seeds will be more susceptible to frost damage, or death. We expect to see differences between parent and offspring, and expect that seedlings with first true leaves will be more tolerant of the frost.

METHODS

GERMINATION

Seeds made available from the common garden study were cold stratified, then incubated until emergence of the radicle (5-10 days); upon germination they were planted in germinating soil trays, and allowed to grow in growth chamber under simulated sunlight at 20°C/10°C day/night (optimum growth condition) for 2 weeks

FROST

Seedlings were later arranged randomly on trays, and subjected to 9h of cold acclimation before & after frost; frost period was 6h: incubator initially set to 0°C, actually reached -1.5°C (Group 1), -2.7°C (Group 2) according to sensors inside incubator; Group 1 was put through a colder frost at -3°C; all groups had 1 day in growth chamber at optimum conditions following frost

ANALYSIS

Seedlings were visually inspected for frost damage determined by discoloration and wilting, then scored as survived, damaged, or dead; A chi-square test was used to analyze the differences in survival across treatment, sources, generation, and development stage. Another analysis was conducted using a generalized linear model with a binomial distribution after converting damaged counts to counts of survival

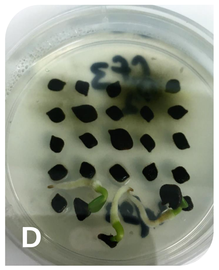
RESULTS

Our best fit model found northern-sourced seedlings to survive at a higher proportion (0.65) than those from the local (0.43) and southern (0.55) sources (GLM, $p < 0.05$).

Study Species

Chamaecrista fasciculata (partridge pea)

- Annual forb of the Fabaceae (leguminous) family
- Found in Midwestern and Eastern United States
- Grows in a broad range of soil conditions, & a Nitrogen fixer
- Controls overgrowth of neighboring species and weeds



Images. Post frost seedling with first leaf survived (A), one that was damaged (B), and one that was killed (C); Seeds germinating on agar plate (D); Post frost seedling cotyledon survived (E)

LITERATURE CITED

(1) Bischoff, A., Vonlanthen, B., Steinger, T., & Müller-Schärer, H. (2006), Basic and Applied Ecology, 7(4), 347-359. doi:10.1016/j.baae.2005.07.009
(2) Breed, M. F., Stead, M. G., Ottewell, K. M., Gardner, M. G., & Lowe, A. J. (2013). Conservation Genetics, 14(1), 1-10.
(3) Leiblein-Wild, M., Kaviani, R., & Tackenberg, O. (2014). Oecologia, 174(3), 739-750. doi:10.1007/s00442-013-2813-6
(4) Inouye, D. W. (2000). Ecology Letters, 3(5), 457-463.
(5) Sgro, C. M., Lowe, A. J., & Hoffmann, A. A. (2011). Evolutionary Applications, 4(2), 326-337.

RESULTS (cont.)

Common Garden Study Findings

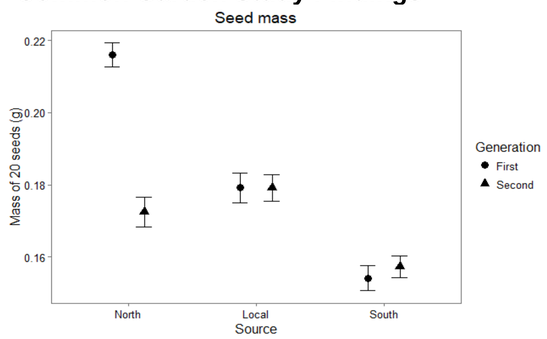


Figure 1. Average masses of 20 seeds for the first and second generations across the regional sources

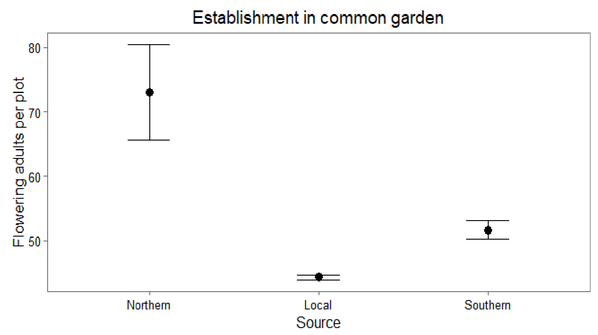


Figure 2. Number of established seedlings per plot across sources in common garden experiment conducted in 2016

Frost Tolerance Study Findings

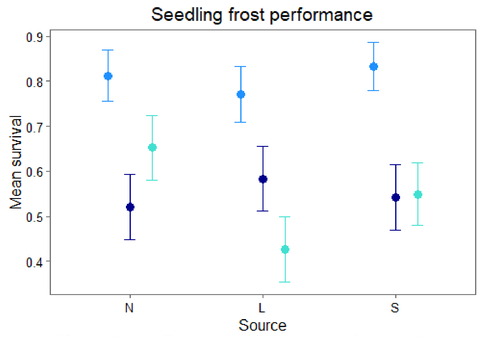


Figure 3. Seedling survival by source and group. Group 1 Test frost was unexpectedly mild, so a second Group 1 Test was done at lower temperature.

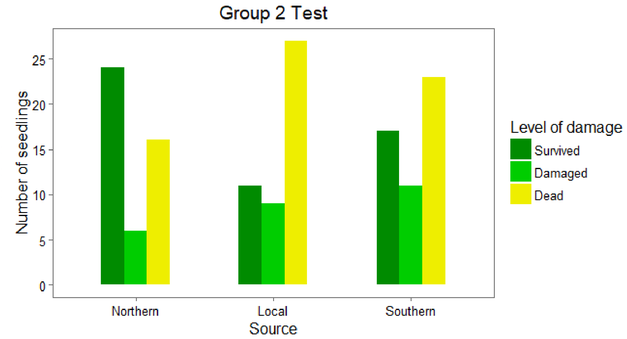


Figure 4. Seedling damage by source for Group 2 Test which temperature regime was 0.4°C for 9h, -1.5°C for 6h, -0.4°C for 9h. Damaged was based on amount of discoloration and wilting.

DISCUSSION

Northern-sourced seedlings survived at a higher proportion presumably as a function of local adaptation to more frequent frosts in their native region. Seeds from the northern-source used in the common garden experiment also had higher mass, which has shown to contribute to greater seedling frost tolerance (3). We expected that southern-sourced seedlings would have lower frost tolerance based on low seedling establishment observed in the common garden and in contrast, higher germination rates seen in lab trials. If southern-sourced seeds germinated at this high of a proportion in the field, a frost event may have occurred, resulting in high seedling mortality. However, the southern-sourced seedlings performed better than we predicted under frost exposure, experiencing only slightly more seedling deaths than locally-sourced seedlings. Southern-sourced seedlings experienced more damage after the colder frost simulation on Group 2, implying that southern-sourced seedlings may still be sensitive to frost. We did not expect the locally-sourced seedlings to experience so much mortality when exposed to frost. However, there are other findings of locally-sourced plants not performing the best when compared to others. Bischoff et al. (2010) found no evidence for fitness superiority of local provenance. The 'local is best' policy for seed provenance may not always be the case for every species and situation. With the frequency of late spring frosts expected to increase with longer growing seasons in temperate regions (4), understanding the extent to which climate variability is affecting our ability to successfully restore lost habitats has never been more pressing.

FUTURE DIRECTIONS

More studies like this are necessary to inform seed sourcing decisions that can lead to successful restoration projects. We recommend a bigger sample, with multiple species, and controlled various temperature regimes for future research. We suggest that the seeds be weighed, and seedlings be subjected to more than one frost event.

ACKNOWLEDGMENTS

We would like to thank the NSF-REU grant DBI-1461007 for financial support, and to the Chicago Botanic Garden for practical and logistical support. This project could not have been possible without the leadership and guidance of Christopher Woolridge, Andrea Kramer, and Jeremie Fant. Special thanks to Chris for major contributions to the data analysis, and for his patient mentorship; and to Kyrel McDavid from the CBG College First program.