

Effects of Population and Temperature on Seed Germination of *Garberia*: A Florida Native with Ornamental and Ecological Value

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Summary

Seed propagation is the primary means of reproducing many native and endemic species, including garberia [*Garberia heterophylla* (W. Bartram) Merrill & F. Harper]. This sandhill species, belonging to a monotypic genus of Asteraceae, boasts an attractive display of purple inflorescences favored by an array of diverse pollinators. Yet it is scarcely found in nursery production and largely unknown to the gardening community. To better understand the seed biology of garberia, a series of experiments were conducted to evaluate the effects of population (North and Central Florida) on

seed viability and storability as well as germination response to four seasonal temperatures that included 11/22 °C (winter), 15/27 °C (fall), 19/29 °C (spring), and 24/33 °C (summer). Initial seed viability was 49% and 60% for Central and North Florida populations, respectively. Seeds germinated readily across populations and temperatures (reaching 50% of final germination within 3-10 days), revealing a lack of physical dormancy. After 28 days meaningful germination responses were observed for temperature and population effects.

When placed in different seasonal temperatures, seeds collected from the northern population had higher maximum germination than seeds collected from the central population, except in the winter temperature where no difference was observed. For the central population, maximum seed germination was greatest in winter (53.4%), followed by spring (53.0%), fall (48.2%), and summer (35.8%). For the northern population, maximum seed germination was similar among summer and fall temperature

treatments (55.4-55.6%) compared to spring and winter (53.4-54.0%). Further, it was observed that garberia seeds are intolerant of long-term, conventional dry storage, revealing a 9.8 and 36.3% reduction in germination after 3- and 6-months post storage, respectively. These findings contribute to the overall understanding of seed biology of underutilized species such as garberia, key to the development of efficient and reliable propagation systems for the nursery industry.

INTRODUCTION

Florida is the second largest producer of ornamental plants in the United States. Of total plant sales in 2019, approximately 15% were of species native to Florida (Khachatryan et al., 2021), meaning they occurred within the state boundaries prior to European contact, according to the best available scientific and historical documentation. In response to the many positive attributes native species can bring to urban gardens, growth in consumer interest and demand for residential and commercial native landscaping is expected to continue on an upward trajectory, with younger homeowners more likely to incorporate native plants in their landscapes (Gillis and Swim, 2020, Torres et al., 2024). To address this need, efforts are underway to support a widely available and diverse palette of native plants for commercial scale production (Rupp et al., 2018) and to develop efficient propagation systems for novel species (Wilson, 2020).

One such species with potential for widened use in the landscape is garberia (*Garberia heterophylla*). Garberia is a perennial shrub which blooms in Fall, boasting showy purple ray flowers as well as year-round foliar interest (**Fig. 1**). A pollinator plant, this species attracts a range of butterflies, bees and moths and naturally occurs in xeric plant communities. Vouchered as far north as Clay County and as far south as Highlands County in Florida (Wunderlin et al. 2024), garberia spans across cold hardiness zones 9A, 9B and 10A. Despite its many desirable traits, this species is carried by less than a handful of nurseries in the state (FANN, 2024) and its seed viability and germination requirements are largely unknown. Thus, the overall goal of this study was to develop optimal seed propagation practices for this underutilized species. Specific objectives were to: 1) determine the influence of population (seed origin) and temperature on seed viability and germination; and 2) assess seed storage longevity over time.



Figure 1. Images of garberia featuring: (A) upright habit and general form, (B) flower buds, (C) gray green foliage and two-ranked leaf arrangement, (D) purple inflorescence, (E) Achenes showing persistent pappi.

MATERIALS AND METHODS

Seed collection and initial viability. Seeds were collected from two natural populations situated on the periphery of Chiappini Farm Native Nursery (North Florida, USDA cold hardiness zone 9a) and The Natives Nursery (Central Florida, USDA cold hardiness zone 9b) within 2 weeks of each other in mid- to late- December 2023. A representative subsample of seeds from each population was sent to an independent seed testing facility to confirm initial pre-germination seed viability and embryo fill (US Forest Service National Seed Laboratory, Dry Branch, GA). First, seeds were non-destructively x-rayed using an Ultra Focus x-ray system with embryo fill calculated using Faxitron Vision software. Then, two replicates of 100 seeds per population were cut laterally and stained overnight at 37 C (98.6 F) in a 1.0% TZ (2,3,5-triphenyl-

2H-tetrazolium chloride) solution in accordance with the Association of Official Seeds Analysts (AOSA) rules for TZ testing (AOSA, 2010). Seeds were considered viable when firm embryos stained evenly red under 10× magnification.

Seed germination. Seeds from each location were visually inspected and surface sterilized with 10% bleach solution (0.75% a.i. NaClO) for 5 min., then triple rinsed with sterile deionized (DI) water. Four replicates of 100 seeds were placed in 10.9 x 10.9 cm transparent polystyrene germination boxes with lids (Hoffman Manufacturing, Albany, OR) containing two sheets of white blotter paper underneath one sheet of unbleached crepe germination paper (Hoffman Manufacturing, Albany, OR). The germination boxes were moistened with 20 mL of sterile DI water, then placed in light and temperature-controlled incubators (Percival

I30VL, Percival Scientific, Perry, IA) set at 11/22 °C (winter), 15/27 °C (fall), 19/29 °C (spring), and 24/33 °C (summer) to mimic seasonal temperatures of Florida. Light was provided by cool white, fluorescent bulbs providing at an average of 40 $\mu\text{mol}\cdot\text{m}^{-2}\cdot\text{s}^{-1}$ at each shelf level for 12 hr, followed by 12 hr of darkness. Germination boxes were opened only as needed to provide moisture and prevent seeds from desiccating. Germination progress was recorded at the first sign of radical emergence every three days. At the end of a 28-day period, final germination percentage (FGP) and T50_{FG} (days to 50% of FGP) were determined for each treatment.

To determine how long seeds could be stored prior to losing viability, a subsample of the same seed lot collected from the central Florida population was placed in paper bags at room temperature (RT, 22-25°C) or in a refrigerator set to 4°C. Seeds were germinated after 0- 3- and 6-month storage times in an incubator set to 19/29°C (spring) using the same methods as previously described. Final germination was recorded after 28 days.

Experimental design and statistical analysis. Experiments utilized a modified randomized block design, with each of the four shelves of each incubator considered as a pseudo-block. Seed germination data was analyzed using generalized linear mixed model procedures through a 3-parameter logistic model in SAS (version 14.1, SAS Institute, Cary, NC).

RESULTS

Initial pre-germination tetrazolium tests of garberia revealed seed viability was 49% and 60% for Central and North Florida populations, respectively (data not presented). Significant effects of population, temperature and their interaction were observed for seed germination that ranged from 37.8 to 62.8% after 28 days. Population responses to temperature differed from each other among all temperature treatments except for winter (**Fig. 2 A and B**). For seeds collected from Central Florida, the greatest final germination occurred in the winter temperature (53.4%), followed by spring (53.0%), fall (48.2%) and then summer (37.8%). For seeds collected from North Florida, the greatest final germination occurred in the summer and fall temperatures (62.8-63.8%) compared to the spring and winter temperatures (60.0-60.8%).

In addition to germination percentage, the rate of seed germination was also influenced by population and temperature (**Fig. 2 A and C**). For the inflection point (T50_{FGP}) of spring and summer treatments, a population effect was not observed. Population effects were however observed for fall and winter treatments. Significant responses in germination time were also observed between each temperature treatment. For both populations, seeds germinated earlier in the summer temperature (day 3) than the spring (day 4). In the fall, seeds collected from Central Florida germinated slightly earlier (5 d inflection point) than seeds collected from North Florida (5.5 d inflection point). Likewise, in the winter, Central Florida seeds had an earlier inflection point (8.9 d) than North Florida seeds (10.2 d).

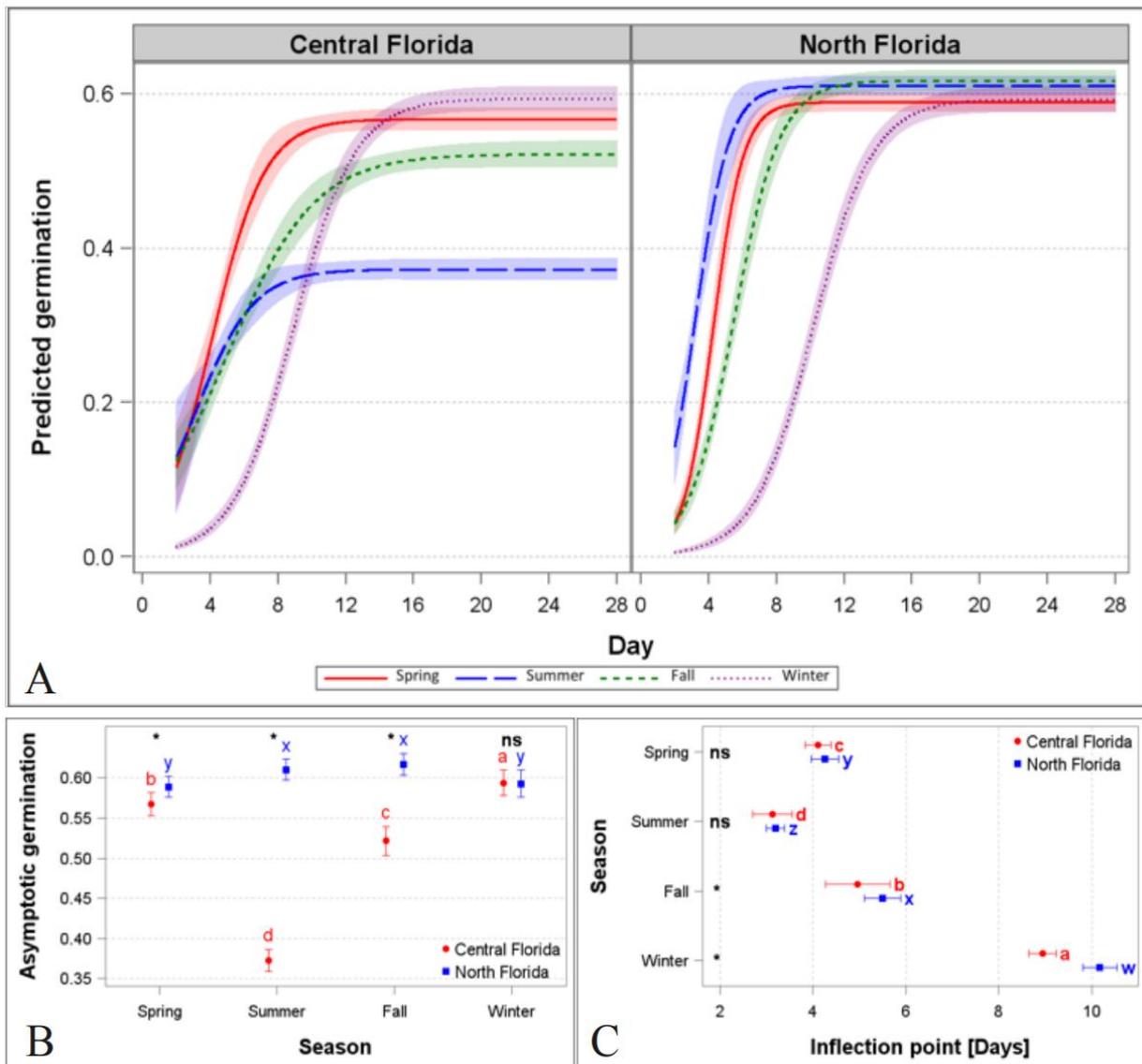


Figure 2. Seasonal germination response of garberia seed collected from North and Central Florida populations. **A.** Predicted germination proportion (symbols), fitted non-linear regression line (line) and 95% CI for the predicted response (colored areas). **B.** Asymptotic germination plus 95% CI. **C.** Inflection point (days to half-maximum germination) plus 95% CI. The colored letters indicate statistically meaningful differences among seasons within location ($\alpha = 0.05$). The * symbol indicates statistically meaningful differences among locations within season ($\alpha = 0.05$); ns=nonsignificant.

In seed longevity experiments, a significant effect of storage time and temperature was observed for seed germination of garberia. After three months of storage, a 9.8% decrease in seed germination was observed, regardless of whether seeds were

stored at room temperature or refrigerated conditions (**Fig. 3**). After six months, seed germination decreased by 38.3% and 34.3% when stored at room temperature or refrigeration, respectively.

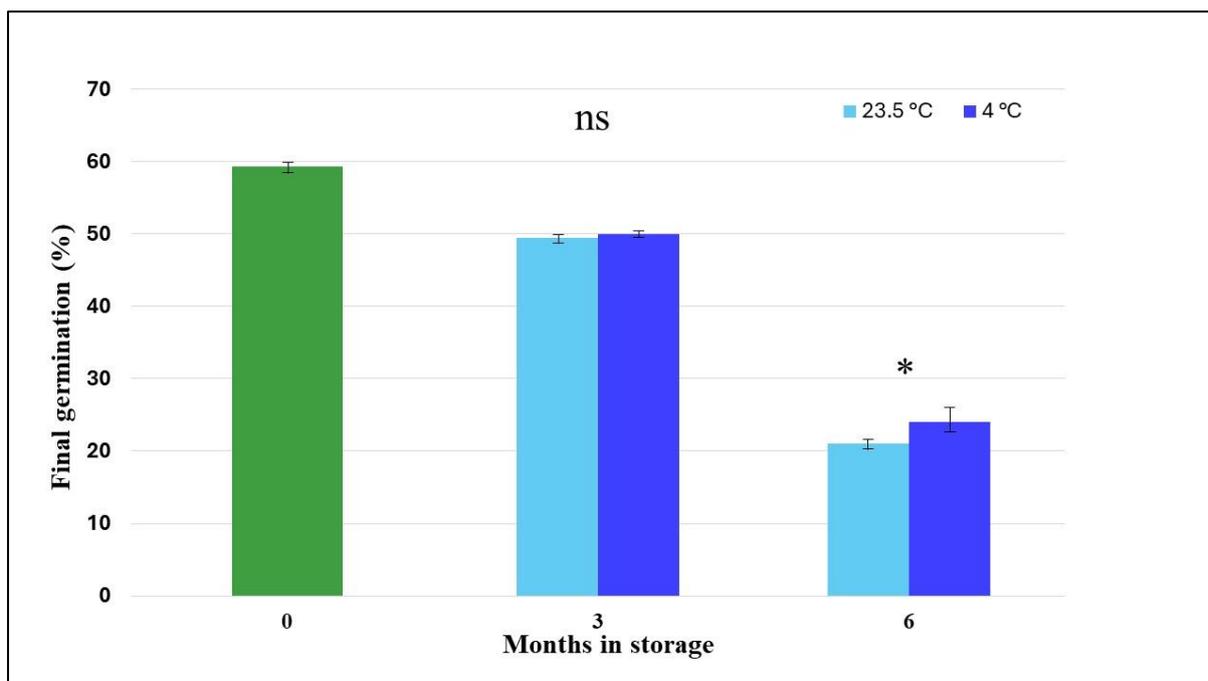


Figure 3. Final germination percentage of garberia seed that was freshly collected (Central Florida population) and dry stored at room temperature (23.5 °C) or refrigerated conditions (4.0 °C) for a period of 0-, 3-, and 6-months. After each storage interval, a subset of seeds was removed and placed in germination incubators set at 19/29 °C for 28 days. The vertical bars above the columns represent \pm standard error of the mean. The * symbol indicates statistically meaningful differences among storage conditions within each storage interval ($\alpha = 0.05$); ns= nonsignificant.

DISCUSSION

Understanding the seed biology of native species allows nursery growers to anticipate the effect of seed viability for determining the sowing rate and to predict the varying degree of germination that may be influenced by seed provenance and season of planting. Likewise, knowledge of a species' seed storability index relative to deterioration can help inform decisions of collection times and production scheduling. Our results showed that garberia produces a considerable proportion of seeds that lack viability ($\leq 60\%$) even when freshly collected from robust plant communities. This is not uncommon for wildflower species, particularly those of Asteraceae (Baskin and Baskin, 2014). Producers can account for

this by using specialized seed separation equipment or simply by increasing the seed planting density per cell (Davies et al., 2018).

Results also revealed that germination responses of garberia varied not only by the geographic location (population) in which the seeds were collected but also by the temperature they were subjected to. It is of interest to note the lower germination of the Central Florida seed when germinated in the summer temperature (24/33 °C). Dell et al. (2021) also observed this effect when germinating a similar Asteraceae species, Eggert's sunflower (*Helianthus eggertii*), under warm temperatures (20/35°C), as did Wilson (2020) for other wildflower species. However, it remains unclear why the North

Florida seeds did not have the same response to the warmer temperature. This underscores the importance of considering the role of population effects when evaluating the seed propagation of native species for nursery production. Lastly, this study provided important insight into the storability of garberia. Unlike some species of Asteraceae that can survive at least a year of dry storage (Jiménez-Vázquez et al., 2021), garberia seed began to lose viability within 3 months of storage. Additional studies are warranted to determine if seeds are tolerant of longer-term storage options such as cryopreservation. Given the narrow seed collection window and lack of seed longevity for garberia, further studies exploring cutting propagation of this species may be worthwhile to ensure year-round nursery availability.

CONCLUSION

Research exploring germination responses of underutilized species is vital in developing propagation protocols and establishing best practices in the collection and storage of native seeds. Results presented herein suggest garberia is an excellent candidate for nursery production by seed. Using freshly collected seed from known populations, sowing at 3-4 seeds per cell, and planting in late winter or early spring will ensure best germination responses.

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