

## Introduction

- Brighamia* is a genus of Hawaiian lobeliads; *B. rockii* is endemic to Moloka'i, while *B. insignis* is endemic to Ni'i'hau and Kauai'i, and is extinct in the wild (communication, Seana Walsh)
- Several metapopulations maintained in collections, mostly as living collections
- High rates of inbreeding depression and genetic drift within collections, lack of coordinated breeding system between institutions (Fant et al., 2016)
- Impacts to genetic diversity and fitness, complications for future re-introductions (Walsh 2015; Wood et al., 2020)
- The purpose of this project is to understand the impacts of ex situ conservation on the genetic diversity of the species**



**Figure 1:** Myself holding *B. insignis* from the Chicago Botanic Garden's living collection (left) and a picture of *B. insignis* in the wild, taken by Kenneth R. Wood, 2005 (right).

## Methods and Materials

### DNA extractions

- Dry leaf tissue from living collections represent modern populations; extracted using DNeasy Plant Mini Kit
- Herbarium samples represented the extinct wild populations; extracted using CTAB

### DNA amplification

- 6 microsatellites were amplified using standard PCR protocol

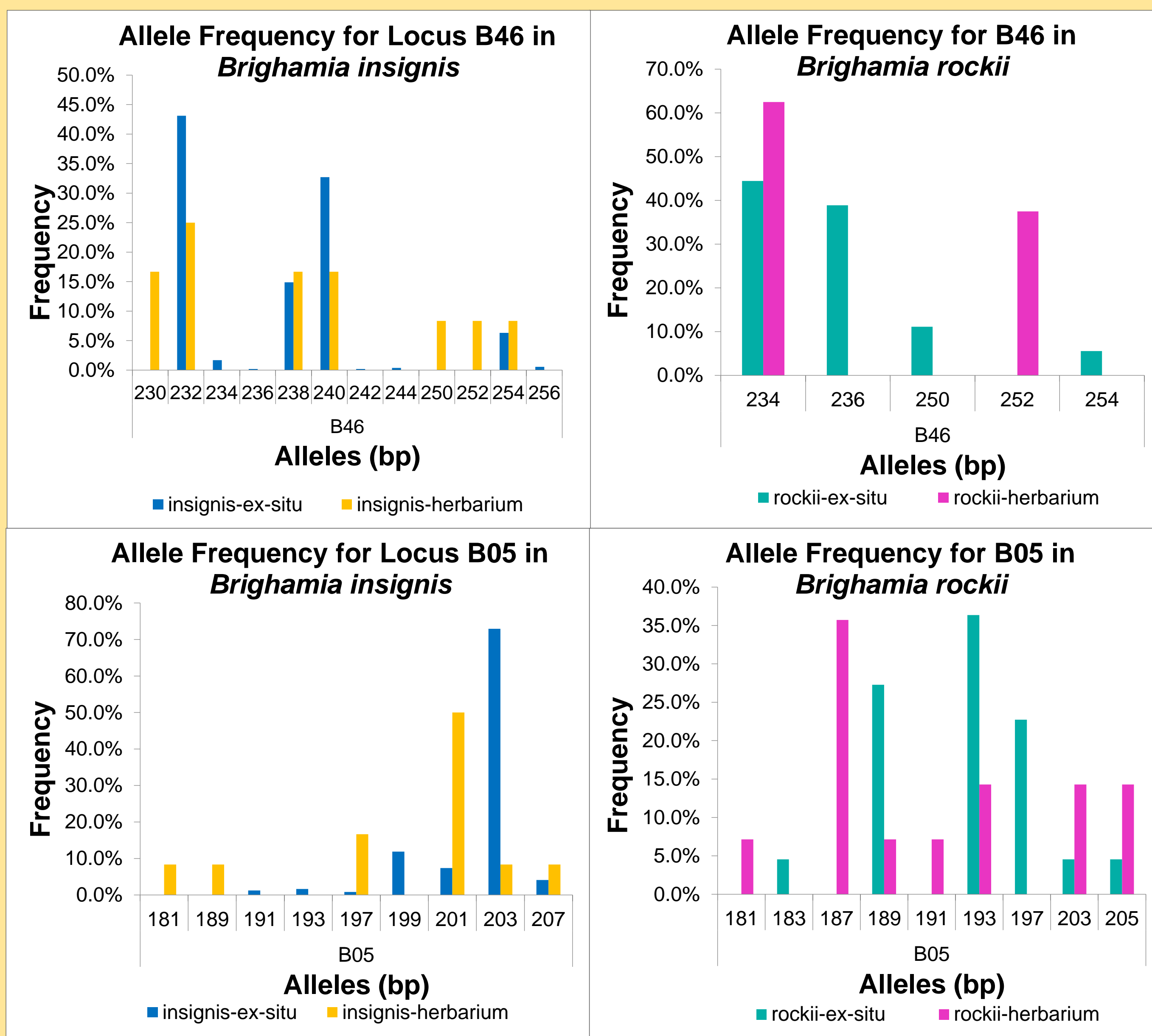
### DNA sequencing

- Amplified microsatellites loci were sequenced using the Beckman-Coulter CEQ-8000 electrophoretic sequencing
- Data were scored with Fant et al. 2019 as a reference for the allele size ranges

### Data analysis

- GenAlEx program was used to analyze data
- Ran frequency by population, private allele, and pairwise fst analyses
- Compared ex situ and herbarium specimen of *B. rockii* and *B. insignis*

## Results



**Figure 2:** Comparing the frequency and number of alleles in two target loci (B46 and B05) across two populations of *Brighamia insignis* and *Brighamia rockii*.

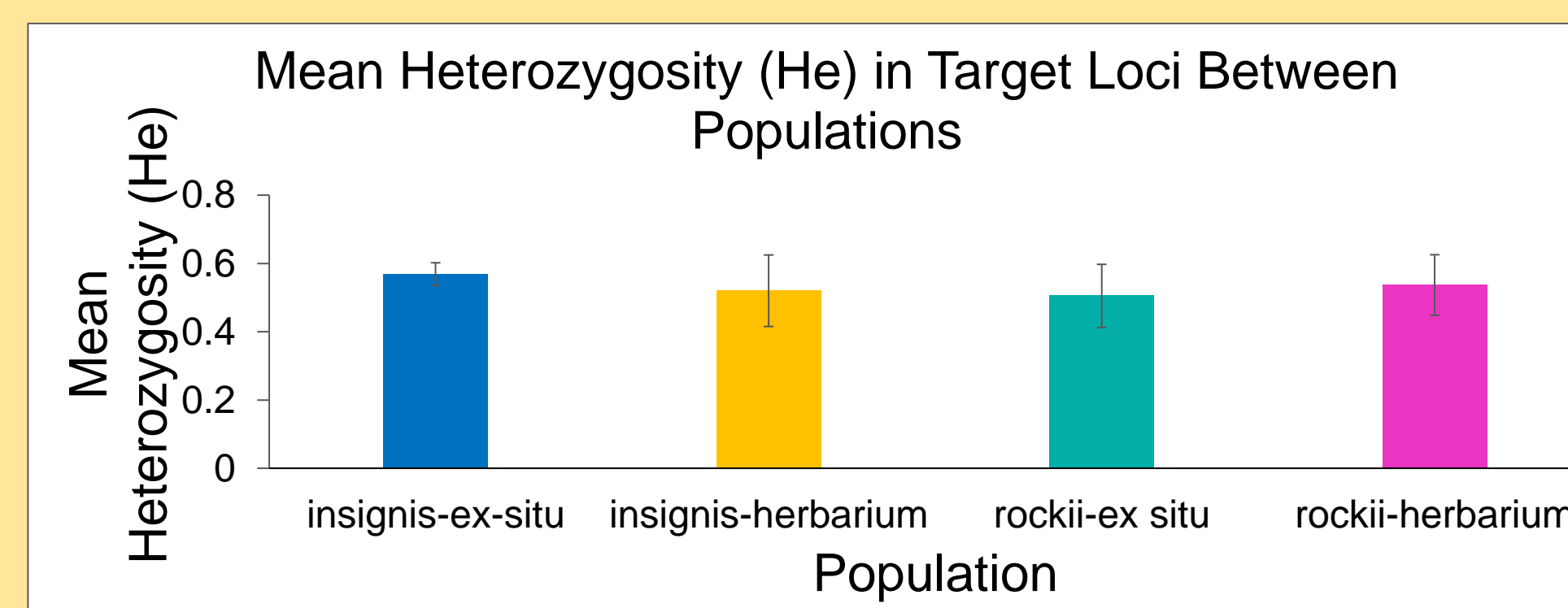
In *B. insignis*, there are several alleles in each loci found in the herbarium population that are absent from the modern ex-situ population (B46 alleles 230, 250, 252; B05 alleles 181, 189). Additionally, B05 alleles 197 and 201 are found in much higher frequency in the herbarium population.

Similarly, various alleles of both loci in *B. rockii* are found in the herbarium population that are not present in ex-situ (B46 allele 252; B05 alleles 181, 187, 191), while B05 alleles 203 and 205 have a much greater frequency in herbarium populations.

**Figure 3:** Comparing the mean heterozygosity for all target loci per population, as a measure of diversity among populations.

The genetic diversity of *B. insignis* herbarium population is not significantly different from that in the ex-situ population.

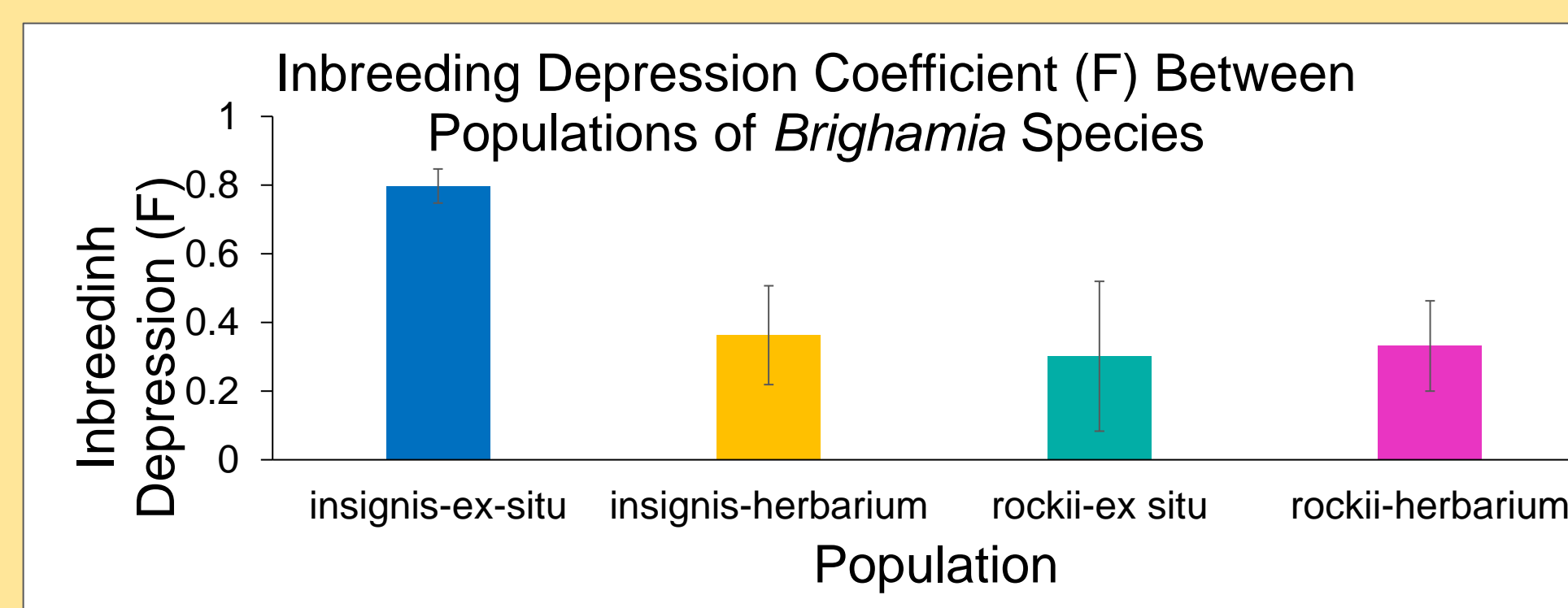
Similarly, *B. rockii* does not have significant difference in diversity between the two populations.



**Figure 4:** Comparing the inbreeding depression coefficient between populations of *B. insignis* populations and between *B. rockii* populations.

The ex-situ population of *B. insignis* has a significantly higher inbreeding depression coefficient than the herbarium population.

There is no significant difference between populations of *B. rockii*.



## Literature Cited

- Fant, J. B., Havens, K., Kramer, A. T., Walsh, S. K., Callicrate, T., Lacy, R. C., . . . Smith, P. P. (2016). What to do when we can't bank on seeds: What botanic gardens can learn from the zoo community about conserving plants in living collections. *American Journal of Botany*, 103(9), 1541-1543. doi:10.3732/ajb.1600247
- Fant, J. B., Fluckes, M., James, E., Noble, H., & Wood, J. (2019). Characterization of microsatellite loci IN Brighamia insignis and transferability to other genera in the Hawai'ian lobelioid group. *Applications in Plant Sciences*, 7(11). https://doi.org/10.1002/aps3.11303
- Wood, J., Ballou, J. D., Callicrate, T., Fant, J. B., Griffith, M. P., Kramer, A. T., . . . Havens, K. (2020). Applying the zoo model to conservation of threatened exceptional plant species. *Conservation Biology*, 34(6), 1416-1425. doi:10.1111/cobi.13503
- Walsh, S. K. (2015). *Floral biology, breeding system, pollination ecology, and ex situ genetic diversity of the endangered Hawaiian species, Brighamia insignis*. A. Gray (Campanulaceae) [Unpublished Master's thesis]. University of Hawai'i at Manoa
- Brueggemann, M. M. & Caraway, V. (2003). *Brighamia rockii*. The IUCN Red List of Threatened Species 2003: e.T44081A10849477. https://dx.doi.org/10.2305/IUCN.UK.2003.RLTS.T44081A10849477.en.

## Discussion

### *Brighamia insignis*

- Due to a relatively small number of *Brighamia insignis* herbarium samples, conclusions are considered preliminary
- Ex-situ populations extensively sampled; several alleles that have gone missing from modern populations (Figure 2), demonstrating loss of valuable genetic diversity
- Mean diversity not significantly different between wild (herbarium) and modern (ex-situ) (Figure 3)
- Large significant difference in inbreeding depression coefficient; ex-situ population has a much higher amount of inbreeding than herbarium population (Figure 4)
- Conservation and breeding programs should focus on preserving existing diversity, prevent further loss (Fant et al., 2016)

### *Brighamia rockii*

- Has also experienced loss in alleles in collections (Figure 2)
- No significant loss in the mean diversity between wild (herbarium) and modern (ex-situ) populations (figure 3)
- No significant difference in inbreeding coefficient (figure 4)
- Not considered extinct in the wild (Brueggemann, 2003), still possible to introduce alleles into collections for conservation
- Maintain existing diversity with proper breeding programs (Fant et al., 2016)
- Herbaria are valuable source of genetic material for extinct plants and populations, require specific protocols

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