Title
Conservation of Rare Serpentine Plants in the San Francisco Bay Area: Current Status and Future Prospects

Permalink
https://escholarship.org/uc/item/31d2n2t1

Author
Sauceda, Angelica

Publication Date
2021

Peer reviewed|Thesis/dissertation

Conservation of Rare Serpentine Plants in the San Francisco Bay Area: Current Status and Future Prospects

By

ANGELICA MARIA SAUCEDA
THESIS

Submitted in partial satisfaction of the requirements for the degree of

MASTER OF SCIENCE

in

Horticulture and Agronomy

in the

OFFICE OF GRADUATE STUDIES

of the

UNIVERSITY OF CALIFORNIA

DAVIS

Approved:

Daniel Potter, Chair

Valerie Eviner

Robert Robichaux

Committee in Charge

2021

This page intentionally left blank

Acknowledgments

I would like to express my deepest appreciation to Susan Frankel and Mia Ingolia for their encouragement and support throughout this project. I am deeply indebted to them for this opportunity to further my understanding of plant conservation and develop my research and writing skills.

Funding was provided by the San Francisco Public Utilities Commission. Cost-share funding was provided by the UC Davis Arboretum and Public Garden. Support was also provided by the US Forest Service, Pacific Southwest Research Station agreement 2020-JV-11272139-004.

I am grateful to Daniel Potter, Mary Burke, and Emily Griswold for their support of this work. Sincerest thanks to Robert Robichaux, Jake Schweitzer, Peggy Fiedler, Bruce Baldwin, and Valerie Eviner for their invaluable insights, advice, and contributions. I gratefully acknowledge Sonya Foree, Scott Simono, and Toni Corelli for providing field-based knowledge for this project. Many thanks to Kent Bradford, Riad Baalbaki, and David Burger for their technical support in the design and execution of the experimental trials. Additional thanks to Timothy Blank and Adrian Sbodio for ensuring safe access to lab equipment and space during a global pandemic. And special thanks to Mei Chau, Kylie Crisostomo-Rickman, Elena Rich, and Ayla Norris for their help implementing and monitoring the experimental trials.

Finally, a nod to my four-legged, loyal friend Tombola’s Delilah Jane and a heartfelt thanks to Sean Glantz for his love and support through this journey.

This page intentionally left blank

Abstract

The conservation of rare plants prioritizes *in situ* conservation by way of maintaining healthy and stable populations via monitoring and threat management. However, when populations appear to diminish additional effort must be taken to maintain their contribution to biodiversity in their natural habitat via *ex situ* conservation. This is done through the harvesting of seed for propagation for later augmentation and reintroduction. However, harvesting seed from threatened rare plant populations poses a challenge to land managers by potentially diminishing or disrupting regeneration. Therefore, germination and propagation protocols must be developed carefully to ensure that impacts are minimal and that the results can better support the long-term conservation of species.

The United States Fish and Wildlife Services' *Recovery Plan for the Serpentine Soil Species of the San Francisco Bay Area* identified several serpentine endemics proximal to the Crystal Springs Reservoir in San Mateo County whose conservation needs further study. The San Francisco Public Utility Commission Natural Resources and Lands Management division is responsible for managing ~63,000 acres of watershed lands that contain populations of three endemic, rare plant species: *Fritillaria biflora* var. *ineziana*, *Pentachaeta bellidiflora*, and *Eriophyllum latilobum*. Here, I present an analysis of the current conservation status and prospects for these species. I review the literature concerning endemism, rarity, serpentine ecology, climate modeling, species biology, and threats to provide the foundation for the conservation planning of these rare species. Additionally, the reintroductions of the rare species *Argyroxiphium kauense* and *Amsinckia grandiflora* and important frameworks are reviewed to identify approaches to rare plant recovery efforts. Finally, a supplemental research chapter concerning *Eriophyllum latilobum* germination trial results demonstrates the early approaches to identifying appropriate seed propagation methods for conservation planning.

This page intentionally left blank

CONTENTS

Introduction 1

Plant Endemism and Rarity 3

Serpentine Soil and Plants 6

- Serpentine Ecosystem of the San Francisco Peninsula 9
- Climate Trends of the San Francisco Peninsula 9

Hillsborough Chocolate Lily (*Fritillaria biflora* var. *ineziana)* 13

- Conservation Status 14
- Plant Description 14
- Occurrence Records 14
- Phylogeny 16
- Distribution 17
- Breeding System 18
- Seed Ecology 18
  - Dispersal. 18
  - Dormancy. 18
  - Germination. 19
- Demography 20
  - Sexual Reproduction by Seed. 20
  - Asexual Reproduction by Bulbs. 20
- Mutualists 21
  - Human Interactions. 21
  - Insect Interactions. 22
- Threats 23
- SFPUC Management 24
- Data Gaps 25

Whiteray Pygmydaisy (*Pentachaeta bellidiflora*) 26

- Conservation Status 26
- Plant Description 26
- Occurrence Records 27
- Phylogeny 28
- Distribution 29
- Breeding System 29
- Seed Ecology 30
  - Dispersal. 30
  - Germination. 30
- Demography 31
  - Seedling Recruitment. 31
- Mutualists 32
- Threats 33
- SFPUC Management 34
- Data Gaps 34

San Mateo Woolly Sunflower (*Eriophyllum latilobum*) 35

- Conservation Status 35
- Plant Description 36
- Occurrence Records 37
- Phylogeny 37

Distribution 38
Breeding System 39
Seed Ecology 40
    Dispersal. 40
    Dormancy. 40
    Germination. 41
Demography 41
    Seedling Recruitment. 42
    Asexual Reproduction. 42
Mutualists 42
Threats 43
SFPUC Management 43
Data Gaps 44

Strategies for rare plant recovery 45

Frameworks and Approaches 45
Case Studies 48
    Ka'ū silversword (*Argyroxiphium kauense*). 49
    Large-flowered fiddleneck (*Amsinckia grandiflora*). 53

Conclusion 60

Bibliography 61

Appendix A 76

Supplemental Research Chapter: *Eriophyllum latilobum* germination trial results 83

Abstract 84

Introduction 84

Materials and Methods 86

Seed Collection 86
    Initial Germination Test and Seed Viability Score. 86
    Disinfection, Preparation, and Imbibition of Seed. 87
Afterripening Experiment 88
Cold-moist Stratification Experiment 90
Scarification and Gibberellic Acid Experiment 91

Results 92

Effect of Afterripening on Germination 92
Effect of Cold-moist Stratification on Germination 92
Effect of Scarification and GA_3 on Germination 93

Discussion 94

Summary 96

Supplemental Research Bibliography 98

Appendix B 101

Conserving SFPUC’s Rarest Serpentine Plants

A SURVEY OF PRACTICAL APPROACHES & SCIENTIFIC PRINCIPLES

Introduction

The San Francisco Public Utilities Commission (SFPUC) provides power to San Francisco public services, wastewater treatment to San Francisco residents, and water via the Hetch Hetchy Regional Water System for 2.6 million customers in four Bay Area counties. The Natural Resources and Lands Management (NRLM) division is responsible for the management of ~63,000 acres of watershed lands and upholding the SFPUC’s Water Enterprise Environmental Stewardship Policy to ensure “responsible natural resources management that protects and restores viable populations of native species and maintains the integrity of the ecosystems that support them for current and future generations” (San Francisco Public Utilities Commission, 2006). Since 2008, SFPUC’s stewardship efforts have included the SFPUC-United States Department of Agriculture Forest Service (USDA FS) Research Partnership, a collaboration that funds projects aimed at improving the success of restoration activities and sustaining the health of vegetation in watershed lands managed by the SFPUC. This report has been fully funded via the SFPUC-USDA FS research partnership. It meets the goals of the SFPUC-USDA FS grant, by equipping San Francisco Bay Area land managers with the knowledge to prepare a conservation strategy for three endemic, rare plant species: *Fritillaria biflora* var. *ineziana*, *Pentachaeta bellidiflora*, and *Eriophyllum latilobum*.

These species are rare as they live in a narrow geographical range, occupy only one or few specialized habitats, and are found in small populations (Harrison *et al.*, 2008; Rabinowitz, 1981). Species most vulnerable to extinction share many defining characteristics of rare species (Grouios & Manne, 2009; Hockey & Curtis, 2009). Species with narrow geographical range, one or few populations (Cardillo *et al.*, 2008; Lawler *et al.*, 2010), small populations (Bulman *et al.*, 2007), and/or whose population size is declining (Peery *et al.*, 2004) are most vulnerable to extinction and require conservation planning.

Since 1974, the California Native Plant Society's *Inventory of Rare and Endangered Plants of California* has been the primary accepted authority regarding California's native plant rarity. The CNPS Rare Plant Program Committee created the CNPS Status Review Process, which ranks rare plants according to their extirpated and extant occurrences within and beyond California's border. Additionally, surrounding threats to the population(s) aid in determining a species CNPS Rare Plant Rank. The list is periodically reviewed as new data become available. The CNPS Rare Plant Ranks are found in Appendix A (Figure 1).

The species of this review, *F. biflora* var. *ineziana, P. bellidiflora*, and *E. latilobum*, are ranked as 1B.1, meaning that they are rare, threatened, or endangered in California, where over 80% of occurrences are threatened or have a high degree and immediacy of threat (Schmid & Tibor, 2002). They are threatened, endemic, and rare species part of, or adjacent to the serpentine ecosystem of the San Francisco Peninsula. Photos of these species are provided on pages 81-83. By reviewing the literature on endemism, rarity, serpentine ecology, climate modeling, species biology, and threats, biologists will be positioned to evaluate extinction risk and appropriate steps for species conservation. Two case studies, reintroductions of *Argyroxiphium kauense* and *Amsinckia grandiflora*, demonstrate the organizing principles and feasibility of recovery efforts for at-risk rare plant populations. These strategies are introduced in the section *Strategies for Rare Plant Recovery* to demonstrate how the Center for Plant Conservation's (CPC) *Best Reintroduction Practice Guidelines* and findings from Lessage *et al.* (2020) can be applied to plant mitigation and recovery efforts or strategies in California.

Plant Endemism and Rarity

Endemism refers to species found naturally in a single geographic area. Species that have recently evolved from closely related species are neoendemics, while paleoendemics are species whose nearest relatives have become extinct. The latter may have once been widespread, but due to unfavorable conditions may persist only in locations where climatic conditions favor them (Harrison & Noss, 2017).

Spatial phylogeneticists employ the concept of relative endemism which is a value on a continuous scale that is assigned according to the taxon's range. They also count evolutionary branches rather than the taxa at the tips of the phylogeny to measure endemism for the study area. This phylogenetic approach adds an evolutionary dimension to our understanding of species endemism. In an analysis of almost 1,110 lineages representing over 5,000 plant species, Thornhill *et al.* (2017) used existing sequence data to map the measure of relative endemism across California. Taxa were scored according to a weighted value which is the inverse of its range size as measured by the number of grid cells in which the taxa occur. These scores are summed for a given region, using different models, providing a comparison of endemism across the landscape. Statewide phylogenetic endemism analysis suggests that areas containing significantly high phylogenetic endemism receive annual precipitation less than 500mm annual precipitation. Furthermore, the Maxent model suggests the San Francisco Peninsula is a niche habitat able to support both paleo and neoendemics. This type of analysis may support further investigations of the potential evolutionary and ecological causes of endemism (Thornhill *et al.*, 2017).

While endemism and rarity are related, a key difference is that endemic species can occur with greater frequency. However, both rare and endemic plants have a high potential for extinction. Rabinowitz (1981) developed a theoretical framework to classify plant rarity (Table 1). It defines eight categories of commonness and rarity based on three traits: geographic range, habitat specificity, and local abundance. Seven of the eight combinations constitute a form of rarity, with one combination classified as commonness: wide range, broad habitat specificity, and large local abundance. While Rabinowitz's framework helps to classify species rarity, it is unable to identify the cause of rarity.

Table 1

A scheme for describing eight categories of commonness and rarity based on three traits

Geographic range		**Wide**		**Narrow**	
Habitat specificity		**Broad**	**Restricted**	**Broad**	**Restricted**
Local Abundance	**Somewhere large**	Common	Habitat Specialists	Unlikely	Endemic
	Everywhere small	Sparse		Unlikely	Rare

(Rabinowitz, 1981)

Fiedler (1995) reviewed the history of scientific inquiry on the topic of plant rarity and discussed five levels of investigation: age, ecology, genetics, evolutionary events, and pluralistic approaches. The analysis makes clear that complex factors contribute to plant rarity, and that there is no one cause for rarity.

Many botanists have debated whether rare species are simply "old" species (Darwin, 1872; Fernald, 1918, 1925, 1942, 1950, 1929; Lyell, 1830) or "new" species (Rafinesque, 1836; Willis, 1922). However, indeed, paleoendemic or nonendemic species may also be categorized as rare. There are a great number of rare California neoendemics, including many species of mariposa lilies (*Calochortus spp.)* (Fiedler *et al.*, 1998). For example, the evolutionarily young Tiburon mariposa-lily, *Calochortus tiburonensis*, is endemic to Ring Mountain on the Tiburon Peninsula of Marin County. Conversely, the rare *Carpenteria californica* is an evolutionarily old, paleoendemic species with only 13 occurrences in the wild, near the border of Madera and Fresno counties in the central Sierra Nevada foothills (Freeman, 2020; Raven & Axelrod, 1978).

Ecologically, rare species were believed to be habitat specialists (Watson, 1845), poor competitors (Griggs, 1940; Kruckeberg, 1951), or occurring infrequently as a result of niche climatic and edaphic requirements (Mason, 1946a, 1946b). While California's wide range of climatic conditions and varied geography has fostered great diversity, no generalizations can be made about rare plants being habitat specialists or poor competitors. However, in the case of serpentine plants, empirical evidence suggests that serpentine species are edaphically restricted and rare due to their inability to compete with other species on non-serpentine soil (Anacker, 2014; Kay *et al.*, 2011; Kruckeberg, 1950, 1954, 1984).

Through the emergent view of the Modern Synthesis[1], Stanley Cain (1940) suggested that low genetic heterozygosity limits a rare plant's ability to colonize new habitat, and G. Ledyard Stebbins (1942) hypothesized that rare species cannot respond to selective pressures and may have one or very few biotypes. However, Fielder maintains that not all rare share these characteristics (Fiedler *et al.*, 1998).

Wright (1956) and Huxley (2010) proposed that accidental fixation of deleterious mutation was the cause for rare species, but there are no empirical data to support this hypothesis (Fiedler, 1995). Evolutionary events causing catastrophic selection were demonstrated within the genus *Clarkia* (Raven, 1964). Mutations leading to new breeding systems have led to the speciation of rare taxa (Gottlieb, 1973), but this rare phenomenon cannot be applied to all cases of rarity. While rare species can

---

[1] In 1942, Julian Huxley popularized the term "Modern Synthesis", which combined Darwinian evolutionary theory and Mendelian genetics in an effort to disentangle how natural selection and hereditary variation cause adaptive change within species populations (Huxley, 2010). This approach emerged in the period spanning 1920 to 1950, and synthesized the studies of systematics, paleontology and botany with a populational view of evolutionary genetics.

originate by chromosomal rearrangement and the evolution of closely related taxa of different ploidy levels (Stebbins & Major, 1965), it is not universally the case.

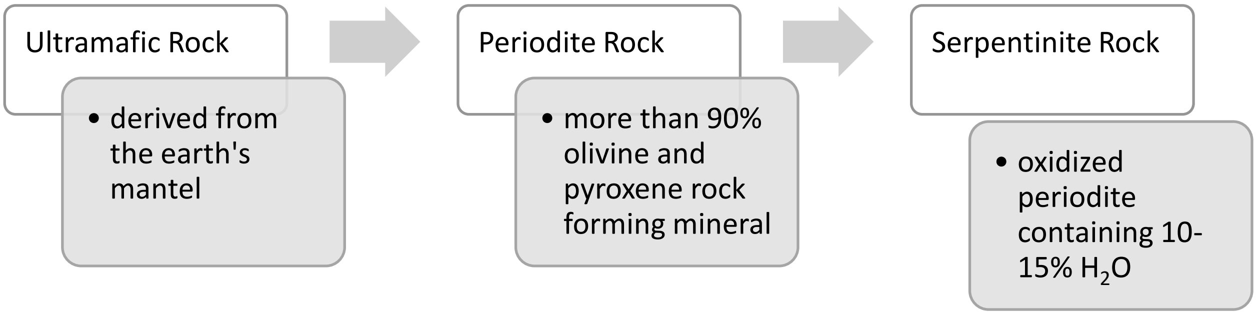
Finally, analysis of more than one factor, or pluralistic approaches, have been reviewed as causes for rarity. For example, Darwin (1872) proposed that rare species which are not resilient to environmental variation and are less competitive and thus less able to respond to selection pressure. Additionally, Stebbins’ (1980) Gene Pool-Niche Interaction theory proposed that a localized environment, specific population genetic structure, and past evolutionary history interact to form rare species (Stebbins, 1980). Since Darwin’s and Stebbins’s pluralistic syntheses only represent three variables that can explain rarity, they do not lead to broad generalizations that are applicable across all rare plants. Fiedler (1998) concluded the possible causes of a species rarity are diverse, complicated, and cannot be generalized. Given the unique factors of species rarity, the investigation of evolutionary age, ecology, genetics, and/or evolutionary events can inform the possible causes of rarity.

Serpentine Soil and Plants

As a result of tectonic activity along the San Andreas Fault, ultramafic-derived soils are found in the San Francisco Peninsula (Kruckeberg, 1984). These soils are the foundation of the serpentine ecosystem upon which serpentine plant communities occur. Serpentine species are rare due to the limited availability of serpentine soil for the establishment and their inability to compete on non-serpentine soil (Anacker, 2014; Brady *et al.*, 2005). The term ultramafic describes peridotite rock composed of more than 90% olivine and pyroxene derived from the mantle. Oxidized peridotite where seawater has hydrated the rock with 10-15% H_2O constitutes serpentinite or serpentine rock (Kruckeberg, 1984; Mével, 2003).

Figure 2

Serpentinite Rock Genesis Flowchart



Note: This flow chart depicts the genesis and specific qualities of serpentinite rock which are derived from parental peridotite and ultramafic rock.

Serpentinite rock types have complex geologic history unique to their situational formation on the uplifted margins of former crustal plates (R. C. Graham, 2009). The weathering of serpentinite rock results in the formation of soil which is comprised of at least 70% ferromagnesian minerals (Kruckeberg, 2002). The soil and the associated vegetation are referred to colloquially as serpentine soil and plants. Clay colloids such as the more common montmorillonite are characteristic in serpentine soils around the world. These soils tend to be heavier in texture, retain moisture longer, and are more intractable to cultivation (Kruckeberg, 1984).

Serpentine mineral composition varies but is generally characterized by low calcium and other essential plant nutrients, high magnesium and iron, and a gamut of heavy metals like chromium, nickel, and cobalt (Kruckeberg, 1984; H. D. Safford *et al.*, 2005). The low nutrient and cation imbalances of serpentine result in sparse plant cover. High-temperature effects, moisture stress, and biotic effects alter plant growth and survival on serpentinite-derived soils (Tadros, 1957).

Serpentine plants, endemic or tolerant, have adapted to the harsh soil conditions by either exclusion, avid uptake, or conversion of toxic minerals (Kruckeberg, 1984). Additionally, plants that can tolerate serpentine soils have relatively large root systems, small stature, and xeromorphic foliage (Krause, 1958; Pichi-Sermolli, 1948). However, experiments have shown that serpentine plants are unable to compete in non-serpentine soil (Anacker, 2014; Kay *et al.*, 2011; Kruckeberg, 1950, 1954, 1984). The plant traits typical of serpentine vegetation create contrasting sites known as contact zones where species composition, density, and pattern of distribution are distinct.

Plant species can be categorized according to their serpentine tolerance. Serpentine endemic species, due to their inability to compete with non-endemic species on non-serpentine soil, are those found exclusively on serpentine soil. Tolerant species are those that may occur on serpentine and non-serpentine soil. Intolerant species are taxa not observed on serpentine soil, which may be due to the absence of the soil type within their range (Anacker, 2011; Harrison & Inouye, 2001; H. Safford & Miller, 2020).

Anacker (2011) used sequence data to build the phylogeny of 23 serpentine-containing genera of the California Flora. The species used in the analysis were then categorized according to their serpentine tolerance. He found that transitions to or from serpentine tolerance were significantly more common than transitions to or from serpentine endemism. The analysis implies that serpentine tolerance is more easily gained or lost than serpentine endemism. Additionally, a majority of serpentine endemic-containing genera included serpentine tolerant species, but transitions to serpentine endemism were equally likely to occur from serpentine tolerant and intolerant ancestors. When a species transitions from serpentine intolerant to serpentine endemic, adaptation to serpentine is only implicated as the cause of speciation (Anacker, 2011, 2014). Conversely, other instances indicate tolerance may be a preadaptation to serpentine (P. L. Fiedler, 1992; Patterson & Givnish, 2002). In sister clade comparisons, Anacker (2011) concluded that serpentine endemism is generally an evolutionary dead-end for most California flora. However, Anacker (2011, 2014) believes that since serpentine soil has allowed adaptive evolution in plants and the soil type limits plant migration for serpentine endemics, future studies may detect early signs of climate change-induced evolution for these soil dependent species (Anacker, 2011, 2014).

Serpentine Ecosystem of the San Francisco Peninsula

California ranks third in the world for serpentine plant diversity and the count for rare plants on unusual soil types is greatest on serpentine soil. An estimated 285 species and varieties in California are serpentine endemics (Faber, 1997; Kruckeberg, 1984) There are over 2,848 square kilometers of serpentinite bedrock in California occurring on two north-south axes: the western Sierra Nevada from Plumas County south to Tulare County and the Coast Ranges-Klamath Mountains ranges from Del Norte County south to Santa Barbara County. Land-use changes due to urbanization, agriculture, or grazing impose threats to this ecosystem by habitat fragmentation, invasive species introductions, nitrogen deposition, altered fire regimes, and pollution (Grace *et al.*, 2007; Harrison *et al.*, 2020; Rajakaruna & Boyd, 2014; Hugh D. Safford & Harrison, 2004; Spasojevic *et al.*, 2014; Thorne *et al.*, 2011; Vallano *et al.*, 2012; Wolf, 2001).

The three species of interest, *F. biflora* var. *ineziana*, *P. bellidiflora,* and *E. latilobum,* occur within or near the serpentine ecosystem of the San Francisco Peninsula, within San Mateo County near the Crystal Springs Reservoir which forms a part of the Coast Ranges-Klamath Mountain serpentine axis.

Climate Trends of the San Francisco Peninsula

The diversity of serpentine flora of California is closely linked to latitude, proximity to the coast, and precipitation (Harrison *et al.*, 2000). The species discussed in this report are located within the San Francisco Peninsula, bordered by the Pacific Ocean to the west and the San Francisco Bay to the east. The climate is Mediterranean with warm, dry summers and mild, damp winters. Two corridors funnel an ocean breeze into the area: Crystal Springs Gap and San Bruno Gap. The San Andreas Fault travels through the westward hills creating a low-lying area known as the Crystal Springs Gap. This gap permits the ocean breeze to transport ocean cooling and fog, dampening the summer heat. Similarly, a gap between Montara Mountain and San Bruno Mountain, allows coastal weather to pass and cool surrounding Millbrae, Burlingame, and San Mateo areas (Gilliam, 2002).

The nearest weather station is located at the San Francisco International Airport (KSFO) which is approximately 20 kilometers from the SFPUC *F. biflora* var. *ineziana*, *P. bellidiflora,* and *E. latilobum* populations. The KSFO weather station calculates the National Oceanic and Atmospheric Administration (NOAA) 1981-2010 U.S. Climate Normals (Appendix A Figure 2). The NOAA Climate Normals represent the 30-year average of a particular climate variable such as temperature or precipitation across the period 1981-2010 (National Oceanic and Atmospheric Administration (NOAA), 2021). These data provide the basis for statements about regional climate trends and will be described as “Normal” throughout this text.

Normal high and low temperatures at the KSFO weather station illustrate a moderately cool-temperate climate where decreased temperatures and increased precipitation occur during December, January, and February. Normal annual precipitation at the KSFO weather station is 524.51 mm with December through February experiencing an average of 101.6 mm of precipitation per month (National Oceanic and Atmospheric Administration (NOAA), 2021).

The Palmer Drought Severity Index (PDSI) is a measure of the combined precipitation and temperature conditions. It has been successful at quantifying long-term drought (Dai & National Center for Atmospheric Research Staff, 2019). The index can capture the basic effect of climate change on drought through changes in potential evapotranspiration. The year 2014 marked the most severe drought in the Central Coast Drainage climate division in the 126-year history of NOAA’s records (Appendix A Figure 3). Conversely, the wettest year as reported by the PDSI occurred in 1983 (NOAA National Centers for Environmental Information, 2021). Overall, the frequency of wet years has diminished since 1920 illustrating a trend towards a warmer and drier climate. According to the PDSI values for the Central Coast Drainage climate division, drought conditions have persisted since 2012.

The uncertainty in projected climate change presents one of the greatest challenges in natural resource management. Climate models under all greenhouse gas emission scenarios suggest an increase in annual average minimum and maximum annual temperature as illustrated in Appendix A Figure 4. The observed average annual minimum and maximum temperature for the 30 years between 1961-1990 were 47.2 °F and 66.4 °F, respectively. This historical baseline represents the period in which the majority of California’s critical infrastructure was developed and when anthropogenic climate change signals were beginning to be felt (Cal-Adapt, 2020). The years between 2035 and 2064 represent the mid-century and the years between 2070 and 2099 represent the end of the century. Table 2 summarizes the modeled forecasted changes from baseline according to data derived from Pierce *et al.* 2018 which was produced for California's Fourth Climate Change Assessment.

Table 2

San Mateo Creek – Frontal San Francisco Bay Estuaries Watershed Annual Average Temperature Forecasts according to High and Medium Emission Table

		Models	Change from Baseline	30-year average	30-year range
Minimum Temperature	Observed (1961-1990)	-	-	47.2°F	-
	Baseline (1961-1990)	Historical	-	47.4°F	47.1 - 47.7°F
	Mid-Century (2035-2064)	Medium emissions (RCP 4.5)	+2.7°F	50.1°F	48.6 - 51.5°F
		High emissions (RCP 8.5)	+3.6°F	51.0°F	49.3 - 52.4°F
	End-Century (2070-2099)	Medium emissions (RCP 4.5)	+3.8°F	51.2°F	49.1 – 53.3°F
		High emissions (RCP 8.5)	+6.8°F	54. °F	51.6 – 57.3°F
Maximum Temperature	Observed (1961-1990)	-	-	66.4°F	-
	Baseline (1961-1990)	Historical	-	66.4°F	66.2 - 66.7°F
	Mid-Century (2035-2064)	Medium emissions (RCP 4.5)	+2.9°F	69.3°F	67.8 – 70.7°F
		High emissions (RCP 8.5)	+3.7°F	70.1°F	68.3 – 71.8°F
	End-Century (2070-2099)	Medium emissions (RCP 4.5)	+3.9°F	70.3°F	68.4 – 72.7°F
		High emissions (RCP 8.5)	+6.8°F	73.2 °F	70.5 – 77.0°F

Note: Summary table of annual average temperature forecasts according to medium and high emission models (Cal-Adapt, 2020; Pierce *et al.*, 2018)

In addition to changes in mean annual temperatures, the models also suggest that wet years will become wetter and dry years will become drier. Additionally, dry years are also likely to be followed by dry years, increasing the risk of drought. As a result of increased temperature and drought, there is increased wildfire risk. In recent years, California has experienced an earlier and longer-lasting fire season with extreme fire events (Ackerly *et al*., 2018; Cal-Adapt, 2020).

Warmer and wetter climates support greater plant diversity (Cowles *et al*., 2018; DeMalach *et al.*, 2017; Hawkins *et al.*, 2003) while, decreased water availability has produced a negative effect on plant diversity (Gornish & Tylianakis, 2013; Kreft & Jetz, 2007; Sommer *et al.*, 2010). Anacker *et al.* (2011), Cornelissen *et al*., (2003) Cornwell & Ackerly (2010), Damschen *et al*., (2012a) use surface leaf area (SLA) changes to compare the effects of increased warming and drought on plant community dynamics over time. Since serpentine species present functional traits such as low SLA, relatively large root systems, small stature, and xeromorphic foliage (Krause, 1958; Pichi-Sermolli, 1948), they are less sensitive to increased temperatures and limited water availability (Damschen *et al.*, 2012). Serpentine species, especially those occurring near or beneath canopy cover, are found to be less sensitive to climatic warming than serpentine species occurring in open understories (Harrison *et al.*, 2020). In the face of a changing and unpredictable climate, the persistence of serpentine communities will depend on the management of nutrient deposition, the encroachment of invasive species, and habitat loss over time (Damschen *et al.*, 2012). The study of a rare species evolutionary history, population, and biological ecology, and threats will assist land managers in stewarding populations into the volatile future these predicted by climate models.

Hillsborough Chocolate Lily (*Fritillaria biflora* var. *ineziana)*

Fritillaria biflora var. *ineziana* is a perennial bulbiferous herb that occurs on serpentinite soil in cismontane woodland, valley, and foothill grasslands at elevations ranging from 295 to 525 feet (90-160m)(California Native Plant Society, 2021b; McNeal & Ness, 2012). The type specimen is a 1914 Inez Smith collection in Hillsborough in San Mateo County, California (Missouri Botanical Garden, 2021). The variety is represented by a narrowly distributed population occurring within a quarter-mile radius, making it extremely at risk for extinction due to stochastic events. The population has reportedly declined in recent years, triggering habitat protection and enhancement measures, seed banking, and the development of an *ex situ* breeding program at the SFPUC Sunol nursery. The California Plant Rescue (CaPR) database lists two accessioned seed collections in the UC Botanic Garden (UCBG) and California Botanic Garden (CalBG).

Conservation Status

The California Native Plant Society has designated *F. biflora* var. *ineziana* a California Rare Plant Rank of 1B.1, meaning it is rare in California and elsewhere and is seriously threatened throughout its range (California Native Plant Society, 2021b). It currently is not listed as endangered by the state or federal government, though its current decline may warrant such protection.

Plant Description

F. b. var. *ineziana* ranges in height from 114.3 to 431.8 mm and bears a slender stem with one or two nodding 30.4 to 61-mm-wide flowers varying from yellow-green to brown or purple. They are difficult to identify when not flowering in their grassland habitat. The flowers are 1.5 to 2 cm long and ovoid-shaped. The bloom period is typically March and April. Alternate leaves are often crowded above ground level and are 5 to 19 cm long. The Jepson Manual key distinguishes *F. biflora* var. *ineziana* from *F. biflora* var. *biflora* by a slight difference in leaf blade shape, where the former is linear to narrowly lanceolate and the latter is widely lanceolate to oblanceolate (McNeal & Ness, 2012, 2015). The ultramafic affinity index of both varieties is 5.4, meaning 85-94% of their occurrences have been recorded on ultramafic soil (H. D. Safford *et al.*, 2005).

Occurrence Records

At the time of this report, there are two California Natural Diversity Database (CNDDB) occurrence reports for *F. biflora* var. *ineziana*. The first observation was documented on March 12, 1914, based on the 1914 collection by Smith for the type specimen. Centered in the town of Hillsborough, the exact location is unknown but it is likely extirpated due to urbanization. The type locality is a valley and foothill grassland, or cismontane woodland.

The second observation was first documented by K. Himes on March 12, 1988, and has since been updated by subsequent surveys. This population is described as having three distinct colonies. The

CNDDB record describes the habitat as serpentine grassland. In 1995, Dean W. Taylor collected an herbarium specimen and noted the species occurred in sites dominated by *Perideridia kelloggii* and preferred humic clay soils. The latest CNDDB 2016 source described the habitat as serpentine bunchgrass with *Stipa pulchra, Chlorogalum pomeridianum, Muilla maritima,* and *Sisyrinchium bellum* forming part of the vegetation community. SFPUC reports *Hesperolinon congestum* ssp. *congesta* (a federally and state endangered and CNPS List 1B.1 species), *Erysimum franciscanum* (a CNPS List 4 species), *Lessingia arachnoidea* (a CNPS List 1B.2 species), *Brodiaea terrestris*, and *Delphinium variegatum* as associated species. *F. biflora* var. *ineziana* were reported in CNDD records to occur in deeper soil while *H. congestum* preferred thinner soil. In 2021, *H. congestum* ssp. *congesta* occurred in densities which may negatively impact one of the colonies. Additionally, the colony experiences increased seasonal moisture causing soils to become waterlogged. In 2015, this colony was the largest but has been in decline in recent years. Inundation may impact the regeneration and/or persistence of this colony. A separate colony may benefit from the removal of nearby tree species in late 2020. This will increase light availability and soil moisture which may benefit the population. Careful monitoring during the transition period will help determine whether the increased soil moisture is a limitation, at least initially.

Differences between the colonies are available to water storage (AWS) and shallowest depth to bedrock (SDB). AWS is the volume of water available to plants that the top 150cm of soil can store. Two colonies occur on Obispo clay with water storage ranging from 0-150 cm with an average of 4.2cm. The minimum depth to bedrock is 30 cm. The third colony occurs on Fagan loam whose water storage ranges from 0-150 cm with an average of 18.07 cm. The minimum depth to bedrock at this site is 109 cm. Both sites experience an average frost-free period of 303 days (USDA *et al.*, 2020).

Phylogeny

Molecular phylogeny identifies *Fritillaria* as a part of the tribe Lilieae within the Liliaceae family (Patterson & Givnish, 2002). This tribe appears to have evolved in the Himalayas and initial radiation occurred in montane and alpine habitats. Members of the core Liliales began diverging 36 million years ago; with members of Lileae diverging 12 million years ago (Patterson & Givnish, 2002). Liliales evolved in closed and shaded habitats but later shifted to open habitats and microsites. *Fritillaria meleagris*, type species for the genus, is distributed throughout most of Europe and western Asia (Roskov *et al.*, 2019). The genus holds about 140 species and 165 taxa of geophytic perennials found in temperate climatic regions of the Northern Hemisphere (Rønsted *et al.*, 2005).

Fritillaria colonized North America at least twice (Patterson & Givnish, 2002). There are a large number of *Fritillaria* species in Eurasia and North America implying several dispersal events. Intercontinental exchanges may have been possible due to the Bering land bridge during the Miocene, Pliocene, and Pleistocene periods (A. Graham, 1999; Patterson & Givnish, 2002; Tiffney, 1985; Wen, 1999).

There are twenty-one species of *Fritillaria* in North America. Nineteen species occur in California and eleven are state endemics (Calscape California Native Plant Society, 2021; McNeal & Ness, 2015). Molecular analysis suggests the North American Clade belongs to three subsections: *Affines, Liliorhiza,* and *Pudicae* (Ryan, 2014). *Fritillaria biflora* is identified as part of the monophyletic subsection *Liliorhiza* (Ryan, 2014). The Flora of North America and Jepson Manual identifies two varieties for this species: *F. biflora* var. *biflora* and *F. biflora var. ineziana*.

The *International Code of Nomenclature for Algae, Fungi, and Plants* designates the taxonomic rank of a variety below that of a species and subspecies, but above that of form (Turland *et al.*, 2018). According to the USDA's National Plant Materials Program Manual, "a variety consists of more or less recognizable entities within species that are not genetically isolated from each other, below the level of subspecies (USDA Natural Resource Conservation Service, 2010)." The literature review did not yield results showing that *F. biflora* var. *ineziana* DNA has been sequenced nor compared with *F. biflora* var. *biflora* or other sister taxa.

The base haploid chromosome number for the genus and the haploid number for the species are not described in the Flora of North America. However, several closely related species in the genus have a diploid number of 24 (2n=24) (Ness, 2020).

Of the 23 California *Fritillaria* species, the following are protected by state or federal listing:

Fritillaria gentneri	Federally Endangered
Fritillaria roderickii	State Endangered
Fritillaria striata	State Threatened

Additionally, the following species have CNPS Rare Plant Ranks:

Fritillaria agrestis	4.2	*Fritillaria liliaceae*	1B.2
Fritillaria biflora var. *ineziana*	1B.1	*Fritillaria ojaiensis*	1B.2
Fritillaria brandegeei	1B.3	*Fritillaria pinetorum*	4.3
Fritillaria eastwoodiae	3.2	*Fritillaria plauriflora*	1B.2
Fritillaria falcata	1B.2	*Fritillaria purdyi*	4.3
Fritillaria gentneri	1B.1	*Fritillaria roderickii*	1B.1
Fritillaria glauca	4.2	*Fritillaria striata*	1B.1
Fritillaria lanceolata var. *tristulis*	1B.1	*Fritillaria viridea*	1B.2

Distribution

Since the species *F. biflora* contains two infraspecific taxa: *F. biflora* var. *biflora* and *F. biflora* var. *ineziana,* it is important to understand their respective ranges. The Jepson treatment for *F. biflora* var. *biflora* identifies the variety as belonging to the North Coast Subregion (Mendocino County), North Coast Ranges Subregion (Napa County), Central Western California Region, and the Southwestern California Region. However, *F. biflora* var. *biflora* occurrence data shows that the species is observed with greater densities in its southern range (Calflora, 2020a). In contrast, *F. biflora* var. *ineziana* is known to occur exclusively in the Hillsborough area of San Mateo County (Baldwin *et al.*, 2020).

Breeding System

Liliaceae is regarded as an out-crossing family (Tamura, 1998). The type for the genus, *F. meleagris,* has been studied by Zych & Stpiczynska (2012). They demonstrated that the species can self and seed set was shown to be 0-5.5% of that set by out-crossing. Insects were able to induce selfing and increased seed set to 9.6-58.5%. However, the seeds were not tested for viability and were returned to the original population. Zych & Stpiczynska (2012) do not explain the decline in the species being attributed to pollination biology or the breeding system.

In March 2021, pollinator exclusion bags were installed on two *F. biflora* var. *ineziana* plants before the flowers opened. One individual set seed while the other did not (Mia Ingolia, *in litt.* 2021). The viability of the seed has not been tested.

Seed Ecology

Dispersal. *F. biflora* var. *ineziana* seed harvested from all SFPUC sites in 2020 produced an average of 96 seeds per sampled plant. The range of production varied between 3 to 224 seeds per plant, with some plants bearing up to three fruit capsules. The 4mm wide, 6mm long, flat, and deltoid-shaped seed is dispersed when the fruit capsule is dry. A small papery wing may aid in wind dispersal. Seeds are buoyant.

Dormancy. Morphophysiological dormancy is typical for members of the Liliaceae family (Carol C. Baskin & Baskin, 2014). This dormancy type is a combination of innate and enforced dormancy. At the time of seed dispersal, the embryo is not fully developed and is the cause of innate dormancy. Environmental factors such as moisture and temperature are the cause of enforced dormancy. Without a hard coat or additional tissue surrounding the embryo, these seeds are subject to desiccation.

For most species of *Fritillaria,* the optimum temperature for post-dispersal embryo development and germination is thought to be 4-5°C. In field conditions, it is believed that radicle emergence starts several months after root growth in adult bulbs. Seedling emergence occurs at or just before the emergence of adult bulbs (Laurence Hill, *in litt.* 2020). Field observations must be made to determine if this is the case for *F. biflora* var. *ineziana.*

Germination. In 2016, germination trials of *F. biflora* var. *ineziana* at CalBG documented an 87% germination rate after one month of cold-moist stratification on agar. After one year of frozen storage, a subsequent trial yielded an increased germination rate of 96% (Cheryl Birker, *in litt.* 2020).

In laboratory experiments with *F. meleagris,* a constant temperature of 12°C and moist conditions for three months were identified as the optimal conditions for germination (Zhang, 1983). Once the chilling requirements were met, the seeds could germinate at 4°C with or without light. Increased temperatures greater than 20°C produced an induced dormancy, which could be reversed by a rechilling treatment. This demonstrates that in addition to innate and enforced dormancy, *F. meleagris* is also regulated by induced dormancy (Zhang, 1983). Changes to the innate dormancy of freshly-harvested seed were not detected after five months of dry storage at 4°C and 20°C and moist storage at 20°C. Additionally, germinability after pre-chilling (moist storage at 4°C) was not impacted after one year of dry storage at 4°C.

The germination behavior of seeds stored in porous containers in the field was observed for 21 months. Zhang (1983) removed germinated seed from containers leaving dormant seed for further tests. After nine months, about 15% of sampled seeds could germinate in the laboratory at 12°C. After one year, the surviving proportion of seed was less than 1.5%. After two years, seeds that did not rot in the field and remained dormant were tested with 2,3,4-triphenyl tetrazolium chloride and found to be non-viable (Zhang, 1983).

Demography

To date, permanent demographic monitoring plots of *F. biflora* var. *ineziana* have not been established. A monitoring study must be developed and implemented to describe *in situ* seed ecology, seedling recruitment, life-stage transition, and survival rates for this species.

Sexual Reproduction by Seed. *The Gardener's Guide to Growing Fritillaries* (GGGF) broadly discusses Fritillary propagation and cultivation. If treated as mature bulbs in a nursery setting, plants grown from seed may bloom in three years. But a majority take additional two years to flower (Jefferson-Brown & Pratt, 1997). Many growers in the California native plant community echo these sentiments, but no other published reports specific to *F. biflora* were found in this review.

Asexual Reproduction by Bulbs. The GGGF section on bulb propagation discusses how rice grain bulblets of American Fritillaries can be brushed off and sown like a seed. They assert that plants resulting from rice grain bulblets bloom a season sooner than plants raised from seed during the same period. Still, they have found three to five years as the typical waiting period for blooms (Jefferson-Brown & Pratt, 1997). Despite this claim, other researchers have found that not all-American fritillary species possess rice grain bulblets (Laurence Hill, *in litt.* 2020).

Baranova *et al.* (2008) described the bulb development in several species of the genus *Fritillaria*, including the North American species: *F. biflora, F. agrestis, F. roderickii, F. lanceolata, F. liliacea,* and *F. pluriflora.* These species are grouped as having tiled bulbs with no stolon, consisting of a few rounded or lanceolate scales with narrow bases that are loosely distributed at the bottom. The scales develop into first-year shoots and leaves which protect the next year's emerging growth shoot (Baranova, 2008). While basal leaf scales do not possess a growing shoot if they are detached from the bulb they may produce a new scale with a growing shoot (Laurence Hill, *in litt.* 2020). However, as noted by Baranova *et al*. (2008), this North American group of *Fritillaria* possesses contractile roots which can position the bulb 15-25cm below the soil surface potentially barring the scales from successful vegetative reproduction (Baranova, 2008). Horticultural efforts should strive to test the asexual reproduction potential of *F. biflora* var. *ineziana* scales as these structures are likely to reduce the time to flower interval as compared to sexually reproduced plants.

Mutualists

Human Interactions. The mutualism between plants and humans is ancient, and it is possible that *F. biflora* var. *ineziana* was a food crop for the Ohlone people of the San Francisco Peninsula. The Ohlone are one of several distinct Native American groups belonging to the Bay-Delta area. They originally occupied the southern and central portion of the San Francisco Bay and traded with adjacent groups (Far Western Anthropological Research Group, 2010). These groups were impacted by European exploration and colonization and as result, they are not as well-known ethnographically compared to other California Native peoples (Byrd *et al.*, 2017). While the literature review did not yield documented ethnobotanical use of *F. biflora* var. *ineziana,* geophytes have a history of Native American management for use as a food crop. For example, the use of *F. affinis, F. camschatcensis F. micrantha, F. pudica, and F. recurva* as food has been documented among other Liliaceae species (Anderson & Lake, 2016; Kuhnlein & Turner, 1991; Ståhlberg & Svanberg, 2006).

The edible underground parts of geophytes (bulb, corms, taproots, tuber, or rhizomes) which remain after the above-ground stems die back, are generally of high nutritional value and contain fiber, complex carbohydrates, and essential vitamins and minerals (Kuhnlein & Turner, 1991). Stewarded by families and passed down intergenerationally, geophyte sites are sustainably harvested to encourage increased subsequent yields. While the large storage organs are used for consumption, smaller fragments such as scales or bulblets, are returned to the tilled soil to facilitate regeneration. In addition to tilling the patches to unearth the crop for harvest, the practice of weeding sites of non-desired vegetation and spreading mature seed in freshly tilled soil enhances the long-term regeneration of the crop. Furthermore, California Native peoples, like others in North America, use fire to eliminate unwanted vegetation and litter, improving nutrient cycling and water retention of the site. It is hypothesized that the heat from a fire stimulates contractile roots to pull the bulb further into the soil, thus forcing scales to shed from the mother bulb (Anderson & Lake, 2016). Furthermore, maintaining prairie habitat with few shrubs or closely growing trees enhances geophyte growth(Boyd, 1999).

California has approximately 285 species of bulbiferous or cormaceous monocots, of which 25% are listed as CNPS 1B ranked plants (Wilken, 2016). The management of rare geophytes would do well to incorporate traditional ecological knowledge to bolster declining populations. While evidence of *F. biflora* var. *ineziana* as a food crop may be lost to time, the site is shared with *Perideridia kelloggii* – a traditional staple food of the nearby Miwok and Pomo people. This occurrence suggests that the site may have endured a history of traditional resource management that may have been of benefit to *F. biflora* var. *ineziana*.

Insect Interactions. As primary out-crossers, *Fritillaria* rely on insect interactions that facilitate pollen transfer and sexual reproduction. While no formal studies have been conducted on the pollination biology of *F. biflora* var. *ineziana*, the species likely favor pollination by emergence time and nectar production. As a late-winter flowering species, *F. biflora* var. *ineziana* is one of few available food sources for pollinators. Additionally, the placement of nectar in the groves at the bases of tepals ensures increased visitation times that may increase the chance of pollen grain transmission. Pollination biology studies of *F. biflora* var. *ineziana* could help determine whether the species is pollinator-limited and whether this could be a reason for the populations' decline.

Zych and Stpiczyńska's 2012 study of the pollination biology and breeding system of *F. meleagris* suggests the species relies on generally rare pollinators. The four-year investigation attributed pollination to two taxonomic orders: Diptera and Hymenoptera. In two of the four study years, solitary bees belonging to the Andrenidae family and honey bees (*Apis mellifera)* were observed to perform 15-18% and 2-12% of visits, respectively. However, the genus *Bombus* was credited with 100% of visits in two of the four study years. The researchers credit the bumblebee's tolerance of unfavorable weather, and their seasonal and floral reliability as a reason for being the key pollinator for *F. meleagris* (Zych & Stpiczyńska, 2012). The study measured pollen loads of captured insects and grouped species into *Bombus, Apis melidera,* and solitary bee groups. For the study period, the highest average *Fritillaria* pollen load was carried by a solitary bee, and loads carried by bumblebees were highly variable. Pooled data from all study years showed that there were no differences between mean pollen loads through the study period for the three groups, however, solitary bees and honeybees were recorded in only two of the four study years.

F. biflora var. *ineziana* will attract its own suite of pollinators. Efforts to improve the habitat for observed pollinators may translate into increased reproductive success for *F. biflora* var. *ineziana* and should be an area of future study for this species.

Threats

Herbivory, non-native species competition, seasonal flooding or drought, nearby urban pollution, scrub encroachment, and potential for hybridization are the main threats posed to the remaining population.

Observations made in 2015 documented deer herbivory on approximately 60% of flowering plants in two of the three colonies (Ingolia, 2018). In the following three years, deer continued to impact reproductive structures at the site (Ingolia, 2018). Herbivore exclusion fencing will be installed in 2021 to eliminate deer herbivory. However, the site is also impacted by below-ground predators (gophers) that may disturb or consume the underground storage organs of *F. biflora* var. *ineziana.* Lastly, insectivore herbivory of the fruit capsule and seeds has been observed. An adult Spotted Cucumber Beetle (*Diabrotica undecimpunctata)* was reported in a CNDDB observation and noted to feed on foliage, pollen, and flowers.

The presence of locally non-native and invasive species is known to affect ecosystem function via altered water and nutrient cycling, and altered disturbance regimes caused by accumulated litter. Additionally, the buildup of thatch from non-native grasses has the potential to interfere with seedling establishment(Nomad Ecology, 2019).

The proximity of *F. biflora* var. *ineziana* to *F. liliacea* colonies increases their chases of hybridization. While some conservation goals welcome the continued diversification of plants, a colony of hybridized fritillaries within a non-hybridized population can potentially alter the genetics of the population.

Nomad Ecology's 2019 *Botanical Resources Survey Report,* prepared for the SFPUC and San Francisco Estuary Institute, identified invasive weeds/exotic annual grasses and shrub/tree encroachment as threats to the species. Increasing cover provided by coyote brush, Douglas fir, and coast live oak were mentioned as specific threats by changing the structure and sun exposure of communities, and potentially altering the fire ecology of *F. biflora* var. *ineziana* habitat (Nomad Ecology, 2019).

SFPUC Management

After conducting the 2015 census that documented the decline of the population since 1988, SFPUC Natural Resources staff started conducting plant protection measures to support *F. biflora* var. *ineziana.* In 2016 and 2017, several plants were caged to prevent herbivory, and seed heads were bagged for seed collection. No more than 5% of the total seed set was collected and almost 2000 seeds were sent to the long-term seed bank at the CalBG in Claremont, CA.

A SFPUC-USDA FS grant has funded the University of California, Davis Arboretum and Public Garden to conduct germination and propagation trials for the species. Progeny resulting from those trials are managed as an *ex situ* collection by the SFPUC Sunol nursery for further study.

In 2020, numerous non-native *Pinus radiata* and *Pinus coulteri* trees that were shading the plants were removed. The sites are monitored and managed to eliminate non-native species invasions (namely *Centaurea solstitialis*, yellow star-thistle). In Fall 2021, SFPUC installed fencing to discourage possible horticultural collecting by trespassers and browsing by ungulates.

SFPUC contracted Nomad Ecology to survey the watershed for new populations of *F. biflora* var. *ineziana.* Surveys were conducted during March, April, May, June, July, September, and October 2018. The upland acres of the watershed measure approximately 7.3 square kilometers. Surveys were conducted one day per month and covered approximately 8.7% of the watershed. No new populations were found.

Management considerations offered by Nomad Ecology (2019) included recommendations of flash grazing, mechanical removal, or prescribed fire to reduce the growth of exotic annual grasses and the resulting build-up of thatch. Nomad Ecology cautioned that management should be scheduled before germination and after seed set to support the long-term persistence of target species like *F. biflora* var. *ineziana*. SFPUC is planning to string trim the NW occurrence in fall 2021 to reduce the thatch build-up.

Data Gaps

High priority research needs for this species include the development of seed germination, greenhouse propagation techniques, and investigations of reproductive biology (mating system, dispersal, pollination). Demography studies help determine the potential for reintroduction success by reviewing/investigating the diverse factors that influence seed germination, life-history stages, and interspecific competition.

Whiteray Pygmydaisy (*Pentachaeta bellidiflora*)

Pentachaeta bellidiflora is an annual species found in cismontane woodland, valley and foothill grassland often on rocky serpentinite (California Native Plant Society Rare Plant Program, 2020). The type specimen is an 1874 Edward Lee Green collection from the foot of Mount Tamalpais (Consortium of California Herbaria, 2021). Today, the species range falls within a 5.6-km radius and is at high risk for extinction due to stochastic events. According to the California Plant Rescue database, a seed collection of *P. bellidiflora* is yet to be accessioned for conservation purposes. Holly Forbes, curator of the UC Botanical Garden at Berkeley, has a 2004 accession pending germination and viability testing before it can be made available for any project (Holly Forbes, email communication 2021).

Conservation Status

The California Native Plant Society has designated *B. bellidiflora* a California Rare Plant Rank of 1B.1, meaning it is rare in California and elsewhere and is seriously threatened throughout its range (California Native Plant Society Rare Plant Program, 2020). In June 1992, the California Department of Fish and Wildlife Service (CDFWS) classified the species as endangered. In February 1995, the United States Fish and Wildlife Service (USFWS) classified *P. bellidiflora* as endangered. Species biology and threats were documented in the associated USFWS 1998 *Recovery Plan for Serpentine Soil Species of the San Francisco Bay Area*. The most recent USFWS review for the species was published in September 2011.

Plant Description

P. bellidiflora generally flowers from March to May. Reaching 6 to 17 cm in height, the plant features small glabrous and narrow leaves less than 4.5cm long and 1mm wide that whorl up the stem.

Up to four composite blooms may occur on the short plants, with each inflorescence displaying both ray and disk flowers. Approximately 7 to 16 white ray flowers measure 3-6 mm. The underside of the flower is often but not always reddish. Approximately 16 to 38 yellow disc flowers radiate from the center of the bloom with 5-lobed corollas. The fruiting heads produce pappus with 5 bristles, rarely none, which are slightly expanded at the bases (Keil & Lane, 2021).

Based on herbaria, CNDDB records, and personal observations, *P. bellidiflora* occupies open, dry rocky, slopes and grassy areas, often on soils derived from serpentine bedrock. While several extirpated occurrences may have been misidentified, *P. bellidiflora* is reported to have an ultramafic affinity index of 2.4. This value means 55-64% of occurrences have been recorded on ultramafic soil (Calflora, 2020b). The only extant occurrence is found on ultramafic soil.

Occurrence Records

In CNDDB reports, *P. bellidiflora* is documented as having one remaining occurrence with the majority of the population occurring on SFPUC managed land. First reported by Thomas Libby Lindenmeyer in 1981, the Triangle colony was described as having one million plants in 1982. Thousands of plants were reported in 1987. Millions were documented in 1989, 1991, 1992, and 2000. The population is bisected by the San Junipero highway forming two colonies, one of which resides in San Mateo County's Edgewood Park. The Edgewood Park colony is much smaller with 100 individuals reported in 1989, 950 individuals in 1992, and 43 individuals in 2004. It occurs in association with *Plantago erecta, Nassella pulchra, Layia platyglossa, Brodiaea terrestris, Gilia tricolor, Dichelostemma capitatum, Sisyrinchium bellum, Ranunculus californicus, Eschscholzia californica, Castilleja densflora, Delphinium variegatum, Lasthenia glabrata, Hesperolinon congestum*, Acanthomintha duttonii** and *Cirsium fontinale* var. *fontinale** (U.S. Fish and Wildlife, 2019; U.S. Fish and Wildlife Service, 1998). Species marked with an asterisk represent federally and state endangered and CNPS List 1B.1 species.

The soil at the site is composed of Obispo clay. The water storage of this soil type ranges from 0-59 inches (0-150 cm) with an average of 1.6 inches (4.2cm). The minimum depth to bedrock is 11.8 inches (30 cm). The slope is fairly flat, approximately 5-7°, with a slight southern exposure(ESRI, 2019). Adjacent to the colony is a *Quercus durata* woodland and several *Umbellularia californica.*

In 2010, a small colony of *P. bellidiflora* was reported about 3.5 miles from the main population by Scott Simono [2]and Mike Vasey[3]. The site is described as a small remnant grassland that is not obviously serpentine but may be Franciscan, with some ultramafic component (S. Simono, *in litt.* 2021). This may be the original site described by an extant occurrence record, which is based on an 1867 Bolander collection near the vicinity of Crystal Springs Reservoir. A third occurrence record on SFPUC land is derived from a 1948 Lewis S. Rose herbarium voucher. The Skyline Boulevard locality above San Andreas Lake was visited in 1991 by Toni Corelli and she noted that the habitat has been replaced by a SFPUC managed road. These reports suggest that *P. bellidiflora* occurred with greater frequency on what are now SFPUC managed lands. The remaining 12 extirpated occurrences not on SFPUC managed land reflect possible misidentifications or have been extirpated due to man-made impacts.

Phylogeny

P. bellidiflora belongs to Astereae, the most diverse and abundant tribe in the Asteraceae family. Several centers of radiation for this tribe include the Americas, Africa, and Australia, with several genera occurring on all contents (Noyes & Rieseberg, 1999). Phylogenetic investigations suggest that the *Pentachaeta/Gigiopappus/Tracyina* clade is closely related to the North American genus *Ericameria* and represents another clade that arose from the major American radiation of tribe Astereae (Roberts &

---

[2] Biologist, SFPUC
[3] Director, San Francisco Bay National Estuarine Research Reserve

Urbatsch, 2003, 2004). Morphological characteristics had united *Pentachaeta* with *Chaetopappa*, but further genetic work has placed *Chaetopappa* more closely related to *Erigeron* (Roberts & Urbatsch, 2003, 2004).

There are six *Pentachaeta* species. Five *Pentachaeta* species are endemic to California, and the sixth occurs in southern California and Baja California (Nesom, 2020a). For the genus, the base haploid chromosome number is 9. *P. bellidiflora* is a diploid (Keil & Lane, 2021). According to Calflora, *P. bellidiflora* is the only species in the genus with an affinity to serpentine soil. *P. bellidiflora* overlaps with the species ranges of *P. alsinoides* and *P. exilis.* All species are small tap-rooted annuals. Of these, *P. bellidiflora* and *P. lyonii* are both State and Federally listed as endangered.

Distribution

Herbaria records and CNDDB occurrences indicate a range from Mount Tamalpais down to Santa Cruz County, but the records are frequently regarded as unreliable due to unlikely habitat and possible misidentifications near the southern range. However, the Jepson Manual treatment defines the bioregional distribution of *P. bellidiflora* as the California Central Coast and San Francisco Bay Area. Furthermore, the Flora of North America provides a narrower description limited to the San Francisco Bay region. Further investigation of the accuracy of herbaria vouchers is needed to verify the historic range of this species.

Breeding System

Van Horn (1973) conducted investigations of self-incompatibility or self-compatibility by preventing cross-pollination and manually pollinating from the same plant. As a result, Van Horn described *P. bellidiflora* as self-incompatible which is a very common breeding system for California annuals (Raven, 1973).

In addition, the breeding system of close relative *P. lyonii* was determined to be self-incompatible through additional study (Fotheringham & Keeley, 1998). Researchers tested self-compatibility with the following treatments (1) control with no manipulation, (2) bagged with fine nylon mesh and no further manipulation, (3) bagged with mesh and later 10 flowers were manually self-pollinated and marked with a thin colored wire, and (4) bagged with mesh and later 10 flowers manually cross-pollinated with pollen grains from a distant plant and marked with a colored wire. Treatments 1 and 2 showed marked differences in seed set with bagged inflorescences aborting. While the author mentions a p-value less than 0.01, suggesting a statistically significant difference, only four inflorescences per treatment were measured (Fotheringham & Keeley, 1998).

Seed Ecology

Dispersal. *P. bellidiflora* achenes are about 1-1.5 mm long and can have up to 5 pappus bristles, allowing this species to travel a short distance by wind in updrafts or by secondary dispersal over the substrate. *P. bellidiflora* achenes readily dehisce from involucres when ripe. Wildlife can potentially aid in long-distance dispersal. In the family of Asteraceae, small-seed eating mammals such as ground squirrels (*Citellus* sp.), pocket mice (*Perognathus* sp.), kangaroo rats (*Dipodomys* sp.), and birds, including quail (*Lophortyx* sp.) have been reported to facilitate dispersal (Martin *et al.*, 1961; U.S Fish and Wildlife Service, 2008).

Germination. Van Horn (1973) stated that fresh seeds will not germinate unless placed at 10°C (50°F) and that all achenes of *Pentachaeta* germinate in four to seven days following moist conditions at 28°C (82°F). Additionally, Van Horn (1973) noted that seeds which endure winter rain are unlikely to germinate the next season, however, no evidence is provided to support the claim.

In 2004, Holly Forbes of the UC Botanical Garden collected *P. bellidiflora* as part of a conservation seed banking project with the Center for Plant Conservation and National Park Service.

While a formal greenhouse study was not conducted, Forbes reported that their garden grew the species in their annual boxes (wooden boxes about 2 ft wide and long and 1.5 ft high) without treatment suggesting overcoming dormancy is not a barrier to establishment (Holly Forbes *in litt.,* 2020).

CalBG has grown *P. lyonii* as part of their viability testing for stored seed. They note that germination rates for this species vary slightly by accession number and pretreatment, but the average germination rate is 64-94% for trials whose storage year was not far from the treatment start date (tested on filter paper and agar). One accession, having been stored for 20 years was documented with a 29% germination rate with no pretreatment on agar. Further investigation into *P. bellidiflora* germination will reveal whether without treatment, it will have similar germination as *P. lyonii* over time.

Demography

At the time of this report, permanent demographic monitoring plots of *P. bellidiflora* have not been established. A monitoring study must be developed and implemented to describe *in situ* seed ecology, seedling recruitment, life-stage transition, and survival rates for this species.

Seedling Recruitment. No studies were found that examine the demographic processes that may describe seedling recruitment for either *P. bellidiflora* or other closely related species. However, Fotheringham (1998) conducted a community characterization study for *P. lyonia* which found a positive correlation between soil depth and species density. While the species can establish on shallow substrates, it is more likely to grow at higher densities on deeper soils. Additionally, an increased soil depth increased the growing season for the species (Fotheringham & Keeley, 1998).

Population growth and survivorship can depend on population density (Taylor & Hastings, 2005). An Allee effect is defined to occur when species decline at low population size or density and is often caused by mate or pollen limitation. Holt (2011) investigated potential Allee effects caused by population densities and presumed limitations in pollinator visitation for *P. lyonii*. Observations were carried out at three sites over two flowering seasons. A total of 5,720 insects were observed from the *Apidae*, *Bombyliidae, Lepidoptera, Megachilidae, Melyridae*, and *Syrphidae* families. Sparsely populated areas of the three populations were visited by pollinators, and the species was not reported to experience Allee effects (Holt, 2011).

Mutualists

The self-incompatible breeding system of *P. bellidiflora* requires a pollinator for seed production. The species flowers March to May and may be pollinated by the bay checkerspot butterfly (*Euphydryas editha bayensis*), however, the primary pollinators are unknown (Robison & Morey, 1992). While the literature search did not reveal any studies of pollinator interactions for *P*. *bellidiflora,* the pollinator interactions of *P. lyonii* have been investigated.

Fotheringham and Keeley's (1998) recorded pollinator visitation rates for *P. lyonii*. Their study focused on six randomly selected plants for 20-minute observation periods at mid-day for about two hours on three occasions in late spring. After insects were collected by sweep net, they were transferred to vials so their pollen sacs could be examined. The anthers of nearby plants were collected and prepared on a glass slide for examination under 100x magnification. Pollination behavior was observed in nine species of visitors mostly belonging to *Hymenoptera* and *Diptera*. Four bee species, three wasp species, and two fly species were observed. The highest visitation was attributed to the digger bee (Fotheringham & Keeley, 1998). A second study found 29 species from 7 orders with hoverfly (*Mesograpta marginata*), deerfly (*Lepidanthrax* sp.), andrenid bee (Andrenidae), and megachiuid bee (*Ashmeadiella caifornica californica*) visiting most frequently and throughout the season (U.S. Fish and Wildlife Service, 1999). Pollen analysis revealed that the top three pollinator species rely on nearby *Adenostoma fasciculatum, Brassica nigra* (invasive non-native), *Centaurea melitensis* (invasive non-native)*, Clarkia purpurea, Dichelostemma capitatum, Deinandra fasciculata,* and *Salvia leucophylla* for forage. These findings suggest that the protection of associated forage plants may support the pollination of *P. lyonii.* A similar study for *P. bellidiflora* can inform habitat protection and/or restoration measures to support pollinators necessary for seed set.

Threats

A nearby county park trail can be an avenue for non-native introductions to the *P. bellidiflora* population. Additionally, nearby nitrogen deposition from the San Junipero Freeway (I-280) may accelerate the establishment and encroachment of non-native grasses in *P. bellidiflora* habitat. While annual grasses are not a current threat to *P. bellidiflora,* their negative impact on the sister taxon *P. lyonii* is documented as involving direct competition and an increase in fire frequency and severity in *P. lyonii* habitat(U.S Fish and Wildlife Service, 2008). Nearby threats to *P. bellidiflora* are teasel (*Dipsacus* sp.), jubata grass (*Cortaderia jubata*), fennel (*Foeniculum vulgare*), Harding grass (*Phalaris aquatica*), hoary mustard (*Hirschfeldia incana*), bull thistle (*Cirsium vulgare*), and Spanish broom(*Spartium junceum*), but none is documented to occur within the population (U.S. Fish and Wildlife Service, 2011). However, the recently reported colony is comparatively remote in comparison, making the risk of invasion by non-native species and impact from nitrogen deposition much lower.

As an annual, *P. bellidiflora* depends on seasonal rainfall and environmental conditions for regeneration. The effects of a poor growing season can bring the population below the minimum viable population size. A reduction in the population size may change gene frequencies due to the founder effects, random fixation, or inbreeding thus leaving the population with less genetic material to adapt to changes in the environment (Shaffer, 1981). With the two remaining populations of *P. bellidiflora* occurring within a 6.4-km distance from each other, the populations are extremely vulnerable to weather patterns or stochastic events that may lead to extirpation or extinction.

Nomad Ecology's 2019 *Botanical Resources Survey Report,* prepared for the SFPUC and San Francisco Estuary Institute, identified invasive weeds and exotic annual grasses as threats to the species. These threats may alter the fire ecology of the habitat, interfere with seedling establishment by increased thatch build-up, and alter nutrient and water availability to the detriment of *P. bellidiflora*.

SFPUC Management

The SFPUC colony of *P. bellidiflora* occurs in association with several other state and federally listed rare plants. The site is guarded with fencing. The site is monitored to prevent and eliminate invasion by non-native species.

A SFPUC-USDA FS grant has funded the University of California, Davis Arboretum and Public Garden to conduct germination trials permitted by the California Department of Fish and Wildlife (CDFW permit No. 2081(a)-20-006-RP).

Management considerations offered by Nomad Ecology's *Botanical Resources Survey Report* included recommendations of flash grazing, mechanical removal, or prescribed fire to reduce the growth of exotic annual grasses and the resulting build-up of thatch. Nomad Ecology cautioned that management should be scheduled before germination and after seed set to support the long-term persistence of target species like *P. bellidiflora*.

SFPUC contracted Nomad Ecology to survey the watershed for new populations of *P. bellidiflora*. Surveys were conducted between March and October of 2018 and did not report new populations of the species.

Data Gaps

The USFWS and CDFWS have gathered knowledge of the species which is limited to historic occurrences, species description, habitat, and known threats. According to the USFWS, no research or grant-supported activities are known to have been conducted on this species since their 5-year review (U.S. Fish and Wildlife Service, 2010). The literature review did not yield published findings of *P. bellidiflora,* therefore congener data have been presented.

Research needs identified by the 1998 *Recovery Plan for the Serpentine Soil Species of the San Francisco Bay Area* are: investigations of soil affinity, seed germination, and greenhouse propagation techniques, demography (soil seed bank, limiting life history stages), and reproductive biology (mating system, dispersal, pollination).

San Mateo Woolly Sunflower (*Eriophyllum latilobum*)

Eriophyllum latilobum is a perennial species found in oak woodland habitats at altitudes between 100 and 150 meters (California Native Plant Society, 2021a). The type specimen was collected on Half Moon Bay Road by Amos Arthur Heller in 1907. Thought to range from Napa, San Mateo, San Benito, Mariposa, and Riverside Counties, CNDDB records suggest many occurrences are misidentified or extirpated. Today, verified occurrences are within San Mateo County with the greatest densities on SFPUC managed land. The species tends to occur along steep roadsides and is impacted by road maintenance and erosion. The California Plant Rescue database reports one accession collected in 2009 stored at CalBG.

Conservation Status

In 1992, *E. latilobum* was State listed as endangered. The CNPS California Rare Plant Rank is 1B.1 meaning it is rare or seriously endangered in California. *E. latilobum* is a perennial herb native and endemic to the San Francisco Peninsula. The U.S. Government designated it as Federally endangered on February 3, 1995. Additionally, within the genus, *E. congdonii* is State-listed as rare.

Plant Description

It is described as a perennial subshrub that can reach between 18.3- 48.8 cm in height. The deeply triangular-lobed leaves are diamond-shaped to more or less narrow at the base (obovate) and are 2-6 cm in size. The alternately arranged leaves are smooth (glabrous) on their upper surface. At maturity, the plant is generally glabrous or smooth. Flowering generally occurs from May to June, with one to ten flowering heads occurring on 1-8 cm length peduncles (stems). They are enveloped in a 4-7mm, widely bell-shaped involucre with 6-10 sharply pointed, free, and barely overlapping phyllaries. A cross-section of the inflorescence will reveal a flat receptacle that is conic in the center. Six to thirteen yellow ray flowers occur with the ray generally ranging 6 -10 mm. Forty to seventy yellow disk flowers are 3-4 mm in size and glandular. Anther tips are ovate. Mature fruit are 3-4 mm and covered with short stiff adpressed hairs. The small mature fruit is topped with very short pappus measuring 0.3-1 mm in length (Keil & Lane, 2021; Nesom, 2020b).

E. latilobum may be easily confused with sister taxa, especially those of its purported parentage: *E. confertiflorum* and *E. lanatum* var. *arachnoideum.* The *Jepson Manual: Vascular Plants of California* key differences relate to the number of ray flowers and inflorescence size and shape (Baldwin *et al.*, 2012). *E. latilobum* is identified as having larger flower heads and inflorescences that are more open with eight ray flowers while *E. confertiflorum* has five. *E. lanatum* var. *arachnoideum* is defined as having 13 ray flowers and shallowly cleft leaves.

SFPUC measurements of over 180 plants in 2021 documented *E. latilobum* as having anywhere between 5-14 ray flowers (M. Ingolia, *in litt.* 2021). The features used to distinguish the plants in the field relate to location and growth form. Mature plants of *E. latilobum* on the peninsula watershed are mostly subshrubs found on shady, often disturbed slopes, while *E. confertiflorum* is a larger shrub that is found in full sun areas. *E. lanatum* var. *arachnoideum* has only been identified in one location on the

Peninsula Watershed (Montara Mountain) and is entirely herbaceous. Beginning in 2021, the SFPUC is surveying all known individuals of *E. latilobum* to capture morphology data to create a more well-defined description of the species since many of the currently defined diagnostic features overlap with the parental species.

Occurrence Records

E. latilobum is documented as having several occurrences on SFPUC and nearby San Mateo County managed land. Six occurrences occur on SFPUC managed land. An additional two are not on SFPUC managed land and require field work to determine if they are extant and correctly identified. For example, in 1988 Toni Corelli visited one of such occurrences and only found parent species *E. confertiflorum* and *E. lanatum* var. *arachnoideum* (T. Corelli, *in litt.* 2021). A review of CNDDB occurrence records identifies *E. latilobum's* associated species as: *Pseudotsuga menziesii, Aesculus californica, Umbellularia californica, Arbutus menziesii , Ceanothus thyrsiflorus, Quercus agrifolia, Pinus radiata (introduced), Rubus ursinus, Toxicodendron diversilobum, Artemisia douglasiana, Lotus scoparius, Mimulus aurantiacus, Eriogonum sp., Brassica (tournefortii)*, *Juncus patens, Lonicera sp., Madia sp., Polystichum sp , Stachys bullata, Iris douglasii, Brassica (tournefortii)., Cirsium vulgare,* and *Lactuca serriola.*

Phylogeny

A part of the Asteroideae subfamily in Asteraceae, *Eriophyllum* belongs to the supertribe Heliantheae or sunflower tribe which was reorganized in 2002 according to results from nuclear rDNA evidence (Baldwin *et al.*, 2002). It is the third-largest tribe in Asteraceae. A few genera are pantropical, with most being found in North and South America, particularly centered in Mexico (Johnson & Mooring, 2021). *E. latilobum* forms a part of the Helenieae tribe and Baeriinae subtribe, which is sometimes combined with other subtribes within Heliantheae (Robinson 1981, Baldwin 2002, Bruce

Baldwin, *in litt.* 2020). Baeriinae occur mostly in western North America, with a few occurring in western South America (Baldwin *et al.*, 2002). The subtribe is closely related to Madieae (Baldwin, *in litt.* 2020).

Currently, there are 13 *Eriophyllum* species and 14 subspecies described in the Flora of North America. The genus *Eriophyllum* features an array of annuals, perennials, subshrubs, and shrubs. They are present in a range of habitats such as seashore, chaparral, grassland, desert, forest, and alpine communities.

Current unpublished data from the Bruce Baldwin lab at UC Berkeley, confirms *E. latilobum* as a true species of *Eriophyllum*. Ongoing phylogenetic work will help determine whether species belonging to *Pseudobahia* and *Syntrichopappus* should be treated within *Eriophyllum* or not (Baldwin, *in litt.* 2020). Constance (1937) postulated *E. confertiflorum* var. *confertiflorum* and *E. lanatum* var. *arachnoideum* as likely parental lineages for *E. latilobum* (Mooring, 1994; Munz, 1959). This hybridization likely occurred many thousands of years ago, and subsequent natural changes in the environment and plant populations are likely why the geographic proximity to possible parents is no longer observed. *E. latilobum* is likely an allotetraploid; a product of two diploid species that hybridized and lead to the genomic doubling that resulted in a genomically stable and true-breeding, distinct species (Baldwin, *in litt.* 2020). As a tetraploid, it has four sets of chromosomes (Carlquist 1956, Mooring 1973).

Distribution

Following anatomical and cytotaxonomic study of closely related *Eriophyllum, Pseudobahia, Syntrichopappus,* and *Monolopia,* Constance (1937) identified the center of dispersal for *Eriophyllum* as the California Inner Coast Ranges between the San Francisco Bay and the junction of the Los Angeles Ranges with the Sierra Nevada (Constance, 1937).

Despite having been rated with the highest affinity towards serpentine (H. Safford & Miller, 2020) and having been included in the *Serpentine Recovery Plan,* the remaining populations occur on soil that does not appear to be serpentine. Further research is warranted regarding the species endemism to serpentine-derived soil.

Breeding System

Mooring (1994) carried out several hybridization experiments for the genus and identified probable parent species *E. confertiflorum* and *E. lanatum* as self-incompatible(Mooring, 1994)(Mooring, 1994)(Mooring, 1994)(Mooring, 1994)(Mooring, 1994). Mooring (1994) suspected that *E. latilobum* is likely self-incompatible.

Seed Ecology

Dispersal. Seeds sit in an involucre when ripe, which may limit dispersal. Heavy wind, rain, and/or disturbance may be necessary to disperse the seed. Mature fruit are 3-4 mm and covered with short stiff adpressed hairs. The small mature fruit is topped with very short pappus measuring 0.3 to 1 mm in length. These features suggest that the fruit is unlikely to travel very far by adhesion to a dispersing animal or by wind.

Dormancy. No studies have been published describing the dormancy of *E. latilobum*. While some species in the family are nondormant, physiological dormancy (PD) is the only dormancy class known for the family; with non-deep PD as the most common type (Carol C. Baskin & Baskin, 2014). Non-deep PD can be tested by the successful development of embryos excised from seeds, resulting in normal seedlings. Cold stratification treatments ranging from 5 to 90 days, as well as storage of dry seed at temperatures equal to or greater than room temperature, known as afterripening, can overcome this dormancy type. The duration of afterripening is typically much longer than that required for dormancy loss during cold stratification. Non-deep PD can be broken by chemicals. Other potential types of dormancy could be intermediate or deep PD, with excised embryos of deep PD producing abnormal seedlings (Nikolaeva, 1977).

Germination. In unpublished data, Mooring described the germination rate for *E. latilobum* in a greenhouse setting as less than 10% (U.S. Fish and Wildlife Service, 2011). However, CalBG incubated seed in a germination chamber set to 13 hours of light at 20°C and 11 hours of dark at 12°C. This first trial reported a 28% germination on a 0.5% agar substrate. During their ten-year follow-up test in 2010, seeds were imbibed and set in a 5°C environment for 19 days, then transferred to their germination chamber for a five-week observation, resulting in a final germination percentage of 48% (Cheryl Birker, *in litt.* 2021). These findings suggest that a chilling period is an important factor to release seed from dormancy.

Closely related *E. confertiflorum* demonstrated improved germination with the use of a water-soluble extract of charred wood (charate extract) and heated chaparral soil extract(Keeley & Nitzberg, 1984). Control sample germination rates on chaparral soil and potting soil are 8% and 12%, respectively. A control sample on filter paper yielded a 62% germination rate. Charate extract alone increased germination on filter paper to 75%. However, the highest germination rate of 77% was achieved by using sand as a growing medium and applying heated chaparral soil extract for germination (Keeley & Nitzberg, 1984). Although the fire-return-interval for the San Francisco Peninsula is far greater than that of *E. confertiflorum* chaparral, a charate treatment may be administered and tested for effects on germination for *E. latilobum.*

Demography

At the time of this report, permanent demographic monitoring plots of *E. latilobum* have not been established. A monitoring study must be developed and implemented to describe *in situ* seed ecology, seedling recruitment, life-stage transition, and survival rates for this species.

Seedling Recruitment. No demography studies have been established. In unpublished reports, Mooring noted that competing species such as *Carduus* sp. may affect *E. latilobum* germination and seedling establishment (U.S. Fish and Wildlife Service, 2011).

Asexual Reproduction. This literature review did not yield any published studies on asexual reproduction by layering or cuttings. However, *E. lanatum* is common in the nursery trade and easily propagated by root cuttings or crown divisions in the late winter (Hebda, 2017; Lady Bird Johnson Wildflower Center, 2016; Norton *et al.*, 1985).

Mutualists

Although *E. latilobum* pollination ecology has not been formally studied, syrphid flies and bees are identified as pollinators in the *Recovery Plan for the Serpentine Soil Species of the San Francisco Bay Area.* Additionally, Calscape identifies *E. latilobum* as being a likely host and/or forage plant for *Amblyptilia pica* (geranium plume moth)*, Argyrotaenia franciscana* (orange tortix moth)*, Phymatopus californicus* (lupine ghost moth)*, Phalonidia latipunctana,* and *Platyptilia williamsii* (CalScape, 2020).

As with *F. biflora* var. *ineziana,* evidence of ethnobotanical use of *E. latilobum* has been lost during colonization of the San Francisco Peninsula (see *F. biflora* var. *ineziana* mutualists section). The Native American Ethnobotany database identifies parent species *E. confertiflorum* and *E. lanatum* as being used by indigenous peoples. The seeds of *E. confertiflorum* can be ground into flour and is regarded as a staple food for the Cahuilla people. *E. lanatum* is a documented dermatological aid of the Skagit people and has documented use as a love medicine by the Chehalis people. Nearby Miwok applied *E. lanatum* var. *leucophyllum* leaves in a poultice as an antirheumatic (Moerman & Best, 2003). The documented use of parent species by several Californian native peoples suggests that *E. latilobum* may have been utilized by the Ohlone of the San Francisco Peninsula.

Threats

Insects, invasive plant species, landslides, climate change, roadside maintenance, and fuel reduction activities all pose a threat to *E. latilobum.* In a report to the California Department of Fish and Wildlife, S. Morey and T. McGuire shared observation of beetle larvae in seed heads but the extent of the predation was not known (U.S. Fish and Wildlife, 2019).

Carduus sp., *Cirsium vulgare, Toxicodendron diversilobum,* and *Rubus ursinus* appear to compete with *E. latilobum* (U.S. Fish and Wildlife Service, 2011). The dense shade of *Umbellularia californica* seems to impact the vigor and reproductive success of *E. latilobum* as compared to individuals situated under the dappled shade of *Quercus agrifolia.*

As *E. latilobum* occurs primarily on roadsides, road maintenance such as mowing, herbicide application, and grading are documented threats to the species. The threat of climate change coupled with *E. latilobum'*s few occurrences of small population size compounds the effects of the aforementioned threats.

Nomad Ecology's 2019 *Botanical Resources Survey Report,* prepared for the SFPUC and San Francisco Estuary Institute, identified invasive weeds/exotic annual grasses, road, trail, or firebreak maintenance, and fire suppression as threats to the species. While mastication is a short-term fire surrogate, controlled burns can create post-fire conditions that may improve the long-term persistence of *E. latilobum in situ* (Nomad Ecology, 2019).

SFPUC Management

E. latilobum occurs on roads essential to SFPUC Crystal Springs Reservoir operations. These roads are maintained to prevent weed invasion, reduce fire risk, and allow safe access for SFPUC and emergency vehicles. Each year, plants are flagged for avoidance in advance of roadside mowing activities. Since the species tends to colonize unstable soils, road-blocking debris from landslides that might contain seeds is spread thinly in appropriate habitats to encourage germination after landslides.

In 2019, a SFPUC-USDA FS agreement funded germination and propagation trials at the University of California, Davis Arboretum and Public Garden (CDFW permit No. 2081(a)-20-006-RP). As a result of those trials, an *ex situ* collection is currently managed for further study.

Management considerations offered by Nomad Ecology's *Botanical Resources Survey Report* include recommendations of flash grazing, mechanical removal, or prescribed fire to reduce the growth of non-natives and/or exotic annual grasses. Nomad Ecology cautioned that management should be scheduled, before germination and after seed set, to support the long-term persistence of *E. latilobum*.

In 2009, a quantity of 7,788 seeds from 254 individuals was collected by SFPUC Biologist, Sonya Foree, and submitted to the CalBG germplasm repository for conservation. Given the sizeable collection, a portion of the seed was sent to the USDA National Center for Genetic Resource Preservation in Fort Collins, Colorado(Wall & Arnold, 2009).

SFPUC contracted Nomad Ecology to survey the watershed for new populations of *E. latilobum.* Surveys were conducted during the months of March, April, May, June, July, September and October 2018. No new populations were found.

Data Gaps

The *Recovery Plan for the serpentine soil species of the San Francisco Bay Area* published in 1998 by the California Department of Fish and Wildlife highlights research needs for the species. High priority research areas are: greenhouse propagation techniques (including propagation from cuttings), factors influencing seed germination (including the possible importance of disturbance and competing species), the possible impact of beetle predation of seeds, and affinity to serpentine soils (U.S. Fish and Wildlife Service, 1998). Of lower priority is research on genetics, phenotypic plasticity, demography, and reproduction (U.S. Fish and Wildlife Service, 1998). The five-year review published in 2011 emphasizes the need for understanding germination and propagation to determine the potential for reintroduction success (U.S. Fish and Wildlife Service, 2011). The literature review did not yield any new publications that fill these data gaps as identified by the California Department of Fish and Wildlife.

Strategies for rare plant recovery

Frameworks and Approaches

Strategies for rare plant recovery require a combination of efforts such as habitat conservation, restoration, seed-banking, genetic material preservation (e.g. pollen), and *ex situ* collections in botanical gardens to preserve biodiversity and ecosystem services (Center for Plant Conservation, 2019; Maschinski & Haskins, 2012).

There are three primary strategies for rare plant recovery, restoration, reintroduction, and relocation. Restoration is the process of assisting the recovery of an ecosystem that has been degraded, damaged, or destroyed (Society for Ecological Restoration International Science & Policy Working Group, 2004). However, conservationists have managed plant recovery more directly via reintroduction practices.

Reintroduction, or re-establishment, is specific to the release of individuals into a formerly occupied area after the native population has been lost or become extinct (IUCN/SSC, 2013). The addition of individuals to an existing population to increase population size or diversity and thereby improve viability is considered as a type of reintroduction (Maschinski & Haskins, 2012). Reinforcement, augmentation, enhancement, or restocking are synonymous, with this latter category of reintroduction. Finally, introductions, managed relocation, or translocation are the deliberate introduction of an organism outside of their native ranges to counteract the effects of climate change (Hellmann *et al.*, 2008).

The Center for Plant Conservation has developed the *CPC Best Reintroduction Practice Guidelines* (CPCBRPG) which provides a framework (See Appendix A Figure 5) that can be applied to all three types of recovery efforts aiming to establish self-sustaining populations in natural habitats (Maschinski *et al.*, 2012). Additionally, the framework could be applied to mitigation efforts which may be required if the population of the species in this report decline. Rare plants may occupy sites that are ephemeral or face major threats that are not subject to regulatory control. Mitigation efforts that compensate elsewhere for the loss of species at these sites, if designed and implemented experimentally, can inform future augmentation, reintroduction, or introduction efforts (referred to as reintroductions).

The CPCBRPG offers a strategic response to help combat the rapid loss of plant diversity and rare plants from land use, invasive species, climate change, and other threats (Maschinski & Haskins, 2012). When a species is at risk for extinction, the CPCBRPG emphasizes first conserving the species *in situ* and preserving wild populations in natural habitats in as many locations as possible, before reintroduction can be considered. The species should be handled for *ex situ* collection, threats must be minimized, and the habitat managed to avoid extirpation (Guerrant *et al.*, 2004). These steps would allow for advancements in horticultural, genetic, and ecological knowledge that may make it feasible to conduct a reintroduction if deemed necessary and appropriate for the species.

Researchers compiled responses from practitioners involved in reintroduction projects for 14 Californian species (Lesage *et al.*, 2020). The paper distilled key advice regarding the reintroduction of rare species and identified critical resources which were lacking that would have improved reintroduction outcomes. The highest-ranked advice was for practitioners to study organisms in the field and to use experiments as a part of the reintroduction process. The authors included a schematic illustrating practitioners' common advice and lessons learned to improve reintroduction outcomes (Appendix A Figure 6).

Interviewees revealed that more and longer-term funding, coupled with long-term monitoring and active management was missing from their projects and would result in improved outcomes (Lesage *et al.*, 2020). Additionally, as the average tenure of an individual involved in a project can be short, practitioners advised for projects to begin with clearly defined record-keeping protocols and well-described management methodologies so that high-quality data can be stored and shared to inform future decisions. A final key insight was to invoke the use of establishing a reintroduction under the 1982 Sect. 10(j) amendment of the Endangered Species Act. Doing so would designate the reintroduction project as an experimental population allowing practitioners to gain knowledge about techniques and best practices without concern for creating land-use restrictions or incidental take limitations for the experimental reintroduction (Lesage *et al.*, 2020).

A reintroduction has two components for success: biological success and project success. Biological success is specific to the condition and performance of individuals, the size and performance of a population, the number and distribution of populations, and the persistence of populations. By establishing taxon-specific objectives that meet the goals of abundance, extent, resilience and persistence, practitioners can design studies that measure and assess biological success accordingly. Project success is much broader, where an experimental design coupled with monitoring can contribute to our knowledge of the species biology and ecology as well as inform the development of new ecosystem management techniques. So then, even if a reintroduction *biologically* fails, a reintroduction can still result in project success that informs future rare plant recovery efforts (Pavlik, 1996a).

The CPCBRPG and analysis of practitioner surveys emphasize the need for carefully thought-out planning, implementation, and monitoring of projects. Success in rare plant recovery efforts is possible when species ecology and biology are understood, and a predefined set of objectives and measurable desired outcomes are defined. This allows project managers to identify and adapt project steps while also defining biological, ecological, and site-specific variables that may affect the outcome of reintroduction.

Case Studies

The following cases are presented as they provide insight into elements that lead to rare plant recovery project successes. The efforts to reestablish the Ka‘ū silversword (*Argyroxiphium kauense*) and the rare large-flowered fiddleneck (*Amsinckia grandiflora*) will be discussed. Both cases reveal that addressing key research questions concerning the species biology and site ecology, and the development of *ex situ* collections, propagation and cultivation methods, experimental reintroduction design, data-informed adaptive management, and interagency collaboration are required to optimize reintroduction project success.

Kaʻū silversword (*Argyroxiphium kauense*). Like serpentine endemics which are restricted to their soil type, the Kaʻū silversword (*A. kauense*) is restricted to the island of Hawaiʻi and threatened by the impacts of non-native species and ungulates which have degraded their habitat. In 1993, the endemic species became federally listed as endangered. Two natural populations occupy distinct habitats in the moister Kahuku and drier Kapāpala region. The species is a rosette perennial shrub that is typically single-stemmed and monocarpic. Vegetative stems are 1 to 24 inches long and reproductive stems can be 2 to 8 feet long. After many years of vegetative growth, the self-incompatible plant dies after flowering and fruit set (U.S. Fish and Wildlife Service, 1995). The multi-agency partnerships that were formed through this project's 20-year span are central to the project's success (R. H. Robichaux et al., 2017). The efforts to mitigate threats, restore habitat and develop a breeding program make this project of notable importance.

***Threat Mitigation and Habitat Restoration*.** Post-colonial Hawaii features a gamut of non-native ungulates that have run feral across natural areas to the detriment of landscape ecology and native species biodiversity (Nogueira-Filho *et al.*, 2009; Pratt & Stone, 1990; Robichaux *et al.*, 1998). The native vegetation has no preadaptation to the mammalian herbivores as the only non-human terrestrial mammals native to the region are bats. Because of this threat, fencing was installed to protect the Kahuku population in 1982 and the Kapāpala population in 1999. Recent improvements to the Kahuku fence in 2004 used 2-meter tall fencing to better exclude mouflon sheep. Hawaii Volcanoes National Park (HVNP) has since expanded its boundaries, installing improved fencing suitable for rugged volcanic terrain. The reintroduction sites were further protected by two exclosures protecting 95% of the outplanted seedlings.

As in California, the invasion of Hawaii by non-native plants has demonstrated the ability to transform native ecosystems by altering fire regimes and nutrient cycling (Antonio *et al.*, 2015; Loh &

Daehler, 2008; Vitousek & Walker, 1989). In 2015, HVNP implemented a monitoring and sanitation program for field vehicles, equipment, and gear to prevent the further introduction of non-native plants, insects, and other organisms to the habitat (David Benitez, *in litt.* 2021). Additionally, the Park has implemented ground-based and aerial-assisted monitoring and management protocols to control the impacts of ungulates and non-native plants.

Additional threats to Hawaiian ecology include the loss of native insect pollinators by competition with non-native social insects, loss of native bird pollinators and seed dispersers by non-native avian diseases, and the predation of seeds by non-native rodents and slugs (Atkinson & LaPointe, 2009; Hartley *et al.*, 2010; Joe & Daehler, 2008; Pender *et al.*, 2013; Pratt & Stone, 1990; Wilson *et al.*, 2009). The impacts of these non-native species have transformed the landscape and the range or persistence of native plants. Additionally, land-use changes associated with agriculture, logging, and ranching further reduce the resilience of the Hawaiian biota (Pratt & Stone, 1990).

To mitigate these compounded impacts, reintroduction efforts were coupled to large-scale landscape restoration in the Park and adjacent state and private lands. Land managers controlled the occurrence and spread of non-native plants while also augmenting canopy and understory species for improved habitat connectivity across large tracts of the landscape. Plants for managed breeding and seedlings for reintroduction were grown at the Volcano Rare Plant Facility (VRPF) of the University of Hawaii at Mānoa. The Three Mountain Alliance, a federal, state, and private watershed partnership, implements landscape restoration efforts in state and private lands adjacent to the Park. These efforts engage the local community with the recovery of Hawaiian rare plants and the recovery of ecologically functional habitats and wildlife corridors (Three Mountain Alliance, 2007).

Breeding Program, Field Management, and Monitoring. Before developing an *ex situ* collection, efforts were made to rediscover *A. kauense* individuals throughout their native range. In 2004, the Kahuku population totaled 381 individuals. In 1998, the Kapāpala population totaled 127 individuals, which were diminutive because of prior ungulate browsing. The Kahuku population served as a source for the reintroduction of the species at the Kahuku area of the Park. The Kapāpala population was the source for the Kipuka Kulalio area. These new reintroduction sites were in areas where concentrated restoration and habitat improvement measures had taken place. Having found all known individuals, biologists developed a managed breeding program.

Whole plants for propagation were retrieved from the field from both sites, without roots. VRPF's *ex situ* propagation efforts were able to produce flowering plants much quicker, as compared to *in situ* plants. These plants were used in combination with wild-collected seeds to develop an *ex situ* breeding program. When plants reached flowering maturity, their pollen was collected, mixed, and applied to increase the genetic diversity offered by the 37 maternal founders from the Kahuku source population and 96 founders from the Kapāpala source population. Thirty-six plants that flowered *in situ* in the Kahuku population were crossed in the same manner.

After seed maturity, they were harvested, stored, and viability tested according to maternal lines. This informed the propagation of the seedlings to be used for out-planting. Each maternal founder represented no more than 2.5% of the total number of seedlings used in the reintroduction at each site. Between 2004 and 2009, VRPF was able to produce and reintroduce 10,212 seedlings for the Kahuku site. In 2014, surveys found 5,894 remaining individuals from the reintroduction. In contrast, an assembly of 11,060 seedlings were introduced to Kipuka Kulalio. In the summer of 2010, only 165 of the 11,060 seedlings were alive. Once propagules were reintroduced to a site they were watered manually at planting, and 1 to 3 times in the following 2 to 4 weeks if rainfall was not adequate.

Facilitated *in situ* assisted achene dispersal within each reintroduction site is an interesting feature of the *A. kauense* recovery effort. Following out-planting, a portion of mature fruit from remnant plants were collected and dispersed in the reintroduced population. In late 2016, the Kahuku site reported 2,845 seedlings as progeny resulting from assisted achene dispersal. In contrast, 578 seedlings were established from 69 fruiting plants at the Kipuka Kalalio site. This difference in survival and recruitment suggests that the latter site may be challenging for establishment due to its soil depth and precipitation.

The VRPF continues to produce propagules for the outplanting of the Kaʻū silversword and other endangered plants. The Kaʻū silversword recovery effort is part of a larger effort to restore the possibility of adaptive radiation for the silversword lineage and other lineages on the island of Hawai’i. This multi-agency, multi-year project will continue to work towards this end by minimizing threats to endangered plants of Hawai’i and increasing their resiliency through carefully planned reintroductions.

Large-flowered fiddleneck (*Amsinckia grandiflora*). In 1982, the large-flowered fiddleneck was listed as endangered by the California Department of Fish and Wildlife, and federally endangered by the US Fish and Wildlife Service in 1985. The large-flowered fiddleneck *(A. grandiflora)* is an annual species of fiddleneck endemic to California preferring slopes with mesic grassland habitat. The species is heterostylous, which is a genetic polymorphism where style and stigma lengths differ between "pin" morphs (wherein the style is taller than the stamen) and "thrum" morphs (wherein the stamen are taller than the style). This genetic difference causes like morph phenotypes to be incompatible and reproduction less likely. The species historically ranged from northern Contra Costa County at the hills above the San Joaquin River Delta, south to Corral Hollow in the hills of southwestern San Joaquin County. Populations of the large-flowered fiddleneck have been reduced by development and non-specific grazing regimes, and the species is further imperiled by the invasion of non-native plants, erosion, ungulate trampling, and altered fire frequency.

In the earliest reintroduction year, 1988, two populations existed within the Lawrence Livermore Laboratory's Site 300 Drop Tower and Draney Canyon and were represented by yearly fluctuations of 50 to 400 individuals. In 1991, a second naturally occurring population was located nearby in Corral Hollow. To safeguard extinction, the USFWS called for the establishment of four new populations within its historic range. The recovery of *A. grandiflora* can be divided into two periods. The first period was headed largely by the efforts of Bruce M. Pavlik (1988, 1990, 1991, 1993, 1994) and the second by Vollmar Natural Lands Consulting (VNLC), whose reintroduction efforts are still underway.

A. grandiflora recovery efforts have spanned beyond extant populations across its historic range. To do this, both Pavlik and VNLC developed site criteria that were informed by known natural occurrences and habitat observations, sampling, and scoring (Holland, 1998; Pavlik, 1990; 1991; Pavlik *et al.*, 1993; Pavlik & Heisler, 1988; Vollmar Natural Lands Consulting, 2016). However, VNLC has leveraged the latest geographic information systems technology to develop a model that incorporates aerial photography, soil and geology data layers, climate isobars and interpolated grids, and a 10-meter elevation model to analyze the percent slope, aspect, and elevation values for occurrence sites. Additionally, solar radiation values were extracted from the extant natural population to narrow the thousands of sites the analysis provided, based on geomorphology and climate. Buffer values were set to allow more sites to be identified based on the upper and minimum topographic and climatic limits of known occurrences. Finally, habitat values were included in the model to classify each targeted area concerning the species' preferred habitat of grassland, woodland, or scrub. Field surveys of extirpated or extant, natural, and reintroduced sites were used to further refine the model.

Modeled target habitats and adjacent areas required field surveys and evaluation during the *A. grandiflora* blooming season (March to April). Site-specific parameters such as ecological (soil, habitat, and disturbance) and logistical (land ownership and proximity to road) characteristics required scoring. Field surveyors found that the lower-ranked habitat areas were consistently too dry to support the species. A second survey was conducted after the blooming season (May). Interestingly, VNLC's final identified sites were steeper than those of Pavlik's previous reintroductions.

Before field surveying could begin, VNLC ecologists entered a process of negotiating access with landowners. Land trust organizations and Resource Conservation Districts' efforts enabled VNLC to network and build trust with land owners and managers of potential recipient sites. VNLC produced Safe Harbor Agreements for private land owners, which exonerated them if the reintroduced species becomes extirpated on their property following introduction. This, as well as liability release forms, insurance forms, letters of understanding, documentation of environmental laws and regulations, and detailed project descriptions, required upfront effort but proved worthwhile to secure access to reintroduce *A. grandiflora* to these sites (Jake Schweitzer*, in litt. 2021)*.

Threat Mitigation and Habitat Management. In place of fire disturbance, judicious grazing is believed to reduce competition from annual grasses and confer a greater benefit to *A. grandiflora* vigor and fecundity (Pavlik, 1991). While Lawrence Livermore Laboratory's Site 300 has never been graded or plowed, the landscape has a history of prescribed fire as a method of reducing grass cover and thatch buildup. This practice may have encouraged the persistence of *A. grandiflora* and other native flora, particularly bunchgrasses at this site. Additionally, the Corral Hollow site has a history of managed grazing.

Pavlik's reintroductions (1991) experimentally tested clipping, burning, and herbicidal treatment on annual grasses and their effect on reintroduced *A. grandiflora* fitness. Demographic monitoring revealed competitive dynamics with annual grass cover that reduced survivorship from seed. Future efforts led Pavlik to test grazing effects. The results led to his recommendation of fire or herbicide application a few weeks after grass emergence following near- or above normal rain in late fall. In contrast, fire or herbicide treatments would not be necessary for years with below-normal rainfall in October, November, December, and January (Pavlik, 1991). Finally, grazing treatments revealed the need for conservation easements near reintroduction sites or the development of post-dispersal stocking schedules to minimize threats to reintroduced populations.

While grazing can reduce exotic annual grass cover, the negative risks of erosion and trampling remain (Pavlik, 1994; Pavlik, 1996b; VNLC, 2016). However, researchers noted the long integral component of grazing at the remaining natural population[4] as possibly conferring some benefit to *A. grandiflora*. So, VNLC reintroductions continue to assess this management technique. They found gated

---

[4] Vollmar identifies the natural populations at Site 300 Drop Tower and Draney Canyon as possibly extirpated. However, reintroduced plants adjacent to the once extant natural Site 300 remain.

enclosures as more likely to impede access, so barbed-wire enclosures were installed around 3 of 6 plots in the 9 out of 10 reintroduction sites. The barbed wire fence was selected over gated enclosures as it could be coiled away to allow wider, controlled access for experimental grazing. However, two sites suffered severe damage from livestock trampling outside of the fencing. This was later mitigated by extending fencing to protect the still relatively small, vulnerable plots. However, it was found that some amount of grazing was critical for maintaining the populations, as ungrazed areas became overrun by annual grasses and other non-native and native plants.

VNLC's ongoing management includes the weeding and irrigation of plots. To minimize soil disturbance within plots, invasive and weedy species are cut at their base or pulled at their roots to reduce competition. However, all native grasses and less weedy forbs were left undisturbed. Plots were irrigated with 2 to 3 gallons of water using a 4-gallon pump-action backpack sprayer. Each plant receives approximately 10 to 20 seconds of irrigation. During the 2014-2015 drought, all plots were irrigated at the time of planting and twice in January. With contingency-based irrigation practices, February rains made irrigation unnecessary for reintroduced plots.

Lastly, while bird netting was used initially to protect plants from predation, they proved logistically difficult to manage and also posed a risk for wildlife, which were found tangled in the nets. Mylar flashing ribbon was used experimentally as well, but it was found to be ineffective. The benefits of nets and ribbons did not outweigh the costs (Jake Schweitzer, *in litt.* 2020).

Breeding Program, Field Management, and Monitoring. Building on formal investigations of the species' reproductive biology. In 1976 the UC Botanical Garden (UCBG) in Berkeley developed well-established propagation and cultivation protocols for the species. Pavlik (1988) researched nutlet production and germination using garden-derived seeds. Demographic findings resulting from greenhouse-grown plants and demography measurements formed the basis for Pavlik's reintroduction plans (1990, 1991, 1993).

The VNLC reintroduction is informed by a solid understanding of population genetics, as UC Botanic Garden assigned a single source material to each site. Two sites near the extant natural population in Corral Hollow used propagules from the nearby natural site. Likewise, the reintroduction site nearest to the extirpated Site 300 populations used Site 300 propagules. All other recipient locations contain both Site 300 and Corral Hollow propagules in separate but proximal sites to maximize the potential for genetic exchange. The VNLC reintroductions relied on propagules produced according to CPC guidelines (i.e. along matrilineal lines).

VNLC reintroductions included a mix of seed and seedlings. Due to budget constraints, a compromise was made favoring out-plant timing for seedlings. A ratio of 2,000 seeds to 200 seedlings was assigned to each reintroduction site. Plants were clustered into smaller groups, as per CPC guidelines, to reduce impacts to the plot during planting, management, and monitoring. Plots were replanted on a biannual basis as to not interfere with multi-year dynamics and monitoring. Second-year plantings focused on the most successful microhabitats at each site (including areas of natural recruitment outside of the plots).

Two plot designs tested densities and ratios of seeds to seedlings, while also allowing for the easy monitoring between propagule types. While greater spacing between seedlings was required due to the disturbance caused by planting, early monitoring suggested that seeds may do better with increased spacing as well. Initial findings have led VNLC to favor seedlings, over seed, in subsequent and ongoing *A. grandiflora* reintroductions as they are more likely to survive to reproduction in the field (Jake Schweitzer, email communication). The germination rate of *A. grandiflora* seeds is naturally low, such that seedlings may confer more fitness than seeds. The establishment of seedlings, while more expensive, was found to be worth the added costs. The VNLC goal is approximately 2,000 to 3,000 individuals per site, which has been suggested as the minimum viable population for a self-sustaining occurrence(Maschinski & Haskins, 2012; VNLC, 2016). This is also the average population size per year of the lone natural occurrence.

VNLC's six plots per reintroduction site were divided into fenced and un-fenced plots. Plots were designed with monitoring and maintenance accessibility in mind. Plots occupied various microhabitats within a given site. Plots were divided into nine cells, with each cell divided into quadrants for random sampling. Quadrant level sampling included plant height, morph (pin or thrum flower), number of flowering branches, and phenology for each individual in the quadrant. This sampling method allowed for the estimation of nutlet productivity among plots. Additionally, each quadrant sample was further characterized by plot type (seed or mixed), topography (concave, convex, flat, undulating), and presence of fencing or bird netting. The corner of each plot was marked with a rebar post, given a plot ID, and mapped using a handheld GPS unit.

As surveying was limited to spring, the measurement of residual dry matter (RMD) which is the standard for measuring dead annual grasses in the fall was not possible. Instead, a photo monitoring method was used to approximate annual grass and vegetation cover (Guenther, 2008). The method involved the use of a Robel pole and golf balls positioned to allow a photograph to capture vegetation density and height. Additional observations of disturbance from burrowing mammals, livestock, and erosion were classified per site as well. Four randomly selected locations within each site, divided among fenced and unfenced plots, gave some indication of *A. grandiflora* competition within a site.

Efforts to construct pathways and remove vegetation increased the ease of monitoring. Pathways allowed for easy access to plants on very steep terrain without damage to plots. Clearing allowed for reduced competition, but too much clearing of vegetation increased competition. Early metrics of each site allowed VNLC to reallocate resources to monitor and maintain the most successful sites.

Monitoring revealed microhabitats and site characteristics that may positively influence *A. grandiflora* survival and reproductive success. Generally, successful plots were steeper and moister with north-facing slopes with fertile, relatively moist, clay to clay loam soil dominated by patchy perennial bunchgrass cover. Plots near trees were determined to be unfavorable at the drip-line, however, some near oak trees were highly successful despite an increased chance of disturbance and predation from associated species. Frequent surveying throughout the year aided in identifying species that may reveal subtle plot differences favoring success. Finally, the previous land management regime became an obvious variable affecting the habitat at each site and is something to consider when establishing a new population.

UCBG has played a key role in conserving *A. grandiflora* for decades via *ex situ* collection and the nursery augmentation of wild collections resulting in over 100,000 seeds stored along maternal lines. Since 2016, on a biannual basis, VNLC's four top-performing sites have received one thousand out-plantings each. This was made possible through partnerships between UCBG, the U.S. Bureau of Reclamation, VNLC's and community volunteers. Preliminary data suggest that judicious grazing abates competition from annual grasses and out-planting of plugs yields better *A. grandiflora* survival and reproductive outcomes. It was found that protecting the plots within fenced enclosures for the duration of germination and flowering of *A. grandiflora*, but then grazing outside of these phases of the life cycle, allowed for the establishment of robust pioneer populations. Once populations were large enough, they appeared to be able to withstand the vagaries of grazing and weather abnormalities. It is believed that a contiguous population will increase the chance for long-term persistence at each site. A Phase 2 report is due in Fall 2021 and will include a detailed site and plot-level analysis including survivorship and vigor as a function of the site, microhabitat, vegetation cover, and fence and unfenced plots.

Conclusion

The serpentine endemics of the San Francisco Bay Area, *Fritillaria biflora* var. *ineziana, Pentachaeta bellidiflora,* and *Eriophyllum latilobum* are ranked as rare, threatened, or endangered in California, where over 80% of occurrences are threatened or have a high degree and immediacy of threat (Schmid & Tibor, 2002). Their high extinction potential warrants the development of a conservation strategy to ensure a planned response should populations decline due to stochastic events (e.g. drought or fire) or prolonged stressors (e.g. climate change, nitrogen deposition, disturbance). SFPUC's conservation efforts include managing threats, seed banking, surveying for additional populations, and funding the development of germination and propagation methods. The Center for Plant Conservation's (CPC) *Best Reintroduction Practice Guidelines* and Lesage *et al.*, 2020 offer a framework and considerations which support managers in developing a plan that is customized to each species unique biology and site ecology. Species biology, site ecology, and threats described in this review serve as a foundation for the development of such a plan. The reintroductions of *Argyroxiphium kauense* and *Amsinckia grandiflora* highlight the importance of threat mitigation, habitat restoration, breeding programs, field management, and monitoring in conservation planning.

Bibliography

Ackerly, D., Jones, A., Stacey, M., Riordan, B., & (University of California Berkeley). (2018). San Francisco Bay Area Summary Report. *California's Fourth Climate Change Assessment*, *CCCA4-SUM-2018–005*. https://www.climateassessment.ca.gov/regions/

Anacker, B. L. (2011). Phylogenetic Patterns of Endemism and Diversity. In *SerpentineThe Evolution and Ecology of a Model System* (pp. 49–70). University of California Press. https://doi.org/10.1525/california/9780520268357.003.0003

Anacker, B. L. (2014). The nature of serpentine endemism. *American Journal of Botany*, *101*(2), 219–224. https://doi.org/10.3732/ajb.1300349

Anderson, M. K., & Lake, F. K. (2016). Beauty, Bounty, and Biodiversity: The story of California Indians' Relationship with Edible Native Geophytes. *Fremontia*, *44*(3), 44–51.

Antonio, C. M. D., Hughes, R. F., Tunison, J. T., & Antonio, D. (2015). Long-term impacts of invasive grasses and subsequent fire in seasonally dry Hawaiian woodlands. *Ecological Applications*, *21*(5), 1617–1628.

Atkinson, C. T., & LaPointe, D. A. (2009). Introduced avian diseases, climate change, and the future of Hawaiian honeycreepers. *Journal of Avian Medicine and Surgery*, *23*(1), 53–63. https://doi.org/10.1647/2008-059.1

Baldwin, B. G., Keil, D. J., Markos, S., Mishler, B. D., Patterson, R., Rosatti, T. J., & Wilken, D. H. (2020). *Jepson eFlora: Geographic Subdivisions*. Jepson Flora Project (Eds.) Jepson EFlora. https://ucjeps.berkeley.edu/eflora/geography.html

Baldwin, B. G., Keil, D. J., Patterson, R., Rosatti, T. J., & Goldman, D. (2012). *The Jepson Manual: Vascular Plants of California* (2nd ed.). University of California Press.

Baldwin, B. G., Wessa, B. L., & Panero, J. L. (2002). Nuclear rDNA evidence for major lineages of helenioid Heliantheae (Compositae). *Systematic Botany*, *27*(1), 161–198. https://doi.org/10.1043/0363-6445-27.1.161

Baranova, M. V. (2008). *The ecologo-morphological peculiarities of the underground organs of the representatives of the genus Fritillaria (Liliaceae)*. www.fritillariaicones.com

Baskin, C. C., & Baskin, J. M. (2003). When Breaking Seed Dormancy Is a Problem: Try a Move-along Experiment. *Native Plants Journal*, *4*(1), 17–21. https://doi.org/10.3368/npj.4.1.17

Baskin, Carol C., & Baskin, J. M. (2014). *Seeds : ecology, biogeography, and evolution of dormancy and germination*. Elsevier Science.

Baskin, Carol C., & Baskin, J. M. (2020). Breaking seed dormancy during dry storage: A useful tool or major problem for successful restoration via direct seeding? *Plants*, *9*(5). https://doi.org/10.3390/plants9050636

Baskin, J. M., & Baskin, C. C. (1985). The Annual Dormancy Cycle in Buried Weed Seeds: A Continuum. *BioScience*, *35*(8), 492–498. https://doi.org/10.2307/1309817

Batlla, D., & Benech-Arnold, R. L. (2004). A predictive model for dormancy loss in Polygonum aviculare L. seeds based on changes in population hydrotime parameters. *Seed Science Research*, *14*(3), 277–286. https://doi.org/10.1079/ssr2004177

Batlla, D., & Benech-Arnold, R. L. (2010). Predicting changes in dormancy level in natural seed soil banks. *Plant Molecular Biology*, *73*(1–2), 3–13. https://doi.org/10.1007/s11103-010-9601-z

Batlla, D., & Benech-Arnold, R. L. (2015). A framework for the interpretation of temperature effects on dormancy and germination in seed populations showing dormancy. *Seed Science Research*, *25*(2), 147–158. https://doi.org/10.1017/S0960258514000452

Benech-Arnold, R. L., Sánchez, R. A., Forcella, F., Kruk, B. C., & Ghersa, C. M. (2000). Environmental control of dormancy in weed seed banks in soil. *Field Crops Research*, *67*(2), 105–122. https://doi.org/10.1016/S0378-4290(00)00087-3

Bewley, J., Bradford, K., Hilhorst, H., & Nonogaki, H. (2013). *Seeds: Physiology of Development, Germination and Dormancy.* (Third). Springer.

Bhatla, S. C., & A. Lal, M. (2018). Plant Physiology, Development and Metabolism. In *Plant Physiology, Development and Metabolism*. Springer.

Boyd, R. (1999). *Indians, Fire, and the Land in the Pacific Northwest*. Oregon State University Press.

Brady, K. U., Kruckeberg, A. R., & Bradshaw, H. D. (2005). Evolutionary ecology of plant adaptation to serpentine soils. In *Annual Review of Ecology, Evolution, and Systematics* (Vol. 36, Issue 1, pp. 243–266). https://doi.org/10.1146/annurev.ecolsys.35.021103.105730

Bulman, C. R., Wilson, R. J., Holt, A. R., Bravo, L. G., Early, R. I., Warren, M. S., & Thomas, C. D. (2007). Minimum viable metapopulation size, extinction debt, and the conservation of a declining species. *Ecological Applications*, *17*(5), 1460–1473. https://doi.org/10.1890/06-1032.1

Byrd, B. F., Whitaker, A. R., Mikkelsen, P., & Rosenthal, J. S. (2017). *San Francisco Bay-Delta Regional Context and Research Design for Native American Archaeological Resources , Caltrans District 4*. *June*.

Cal-Adapt. (2020). *Local Climate Change Snapshot for San Mateo Creek-Frontal San Francisco Bay Estuaries Watershed, California*. https://cal-adapt.org/tools/local-climate-change-snapshot/

Calflora. (2020a). *Fritillaria biflora var. biflora. Calflora: Information on California plants for education, research and conservation, with data contributed by public and private institutions and individuals, including the Consortium of California Herbaria.* The Calflora Database Taxon Report. https://www.calflora.org/app/taxon?crn=3629

Calflora. (2020b). *Pentachaeta bellidiflora. Calflora: Information on California plants for education, research and conservation, with data contributed by public and private institutions and individuals, including the Consortium of California Herbaria.* The Calflora Database Taxon Report. https://www.calflora.org/app/taxon?crn=6227

California Native Plant Society, R. P. P. (2021a). *Eriophyllum latilobum*. Inventory of Rare and Endangered Plants of California. https://rareplants.cnps.org/Plants/Details/779

California Native Plant Society, R. P. P. (2021b). *Fritillaria biflora var. ineziana*. Inventory of Rare and Endangered Plants of California (Online Edition, v9-01 0.0). https://rareplants.cnps.org/Plants/Details/1682

California Native Plant Society Rare Plant Program. (2020). *Pentachaeta bellidiflora. Inventory of Rare and Endangered Plants of California.* https://rareplants.cnps.org/Plants/Details/1241

CalScape. (2020). *San Mateo woolly sunflower (Eriophyllum latilobum)*. https://calscape.org/Eriophyllum-latilobum-(San-Mateo-Woolly-Sunflower)?srchcr=sc59322f3da740f

Calscape California Native Plant Society. (2021). *Fritillaria native to California*. https://calscape.org/loc-california/Fritillaria(all)/vw-list?view_style=list

Cardillo, M., MacE, G. M., Gittleman, J. L., Jones, K. E., Bielby, J., & Purvis, A. (2008). The predictability of extinction: Biological and external correlates of decline in mammals. *Proceedings of the Royal Society B: Biological Sciences*, *275*(1641), 1441–1448. https://doi.org/10.1098/rspb.2008.0179

Center for Plant Conservation. (2019). *CPC Best Plant Conservation Practices to Support Species Survival in the Wild*. Center for Plant Conservation.

Commander, L. E., Rokich, D. P., Renton, M., Dixon, K. W., & Merritt, D. J. (2013). Optimising seed broadcasting and greenstock planting for restoration in the Australian arid zone. *Journal of Arid Environments*, *88*, 226–235. https://doi.org/10.1016/j.jaridenv.2012.08.012

Consortium of California Herbaria. (2021). *Type Specimen Accession Details: Pentachaeta bellidflora*. https://ucjeps.berkeley.edu/cgi-bin/new_detail.pl?GH11000&YF=0

Constance, L. (1937). A Systematic Study of the Genus Eriophyllum lag. In *University of California Publications in Botany* (pp. 69–136).

Cornea-Cipcigan, M., Pamfil, D., Sisea, C. R., & Mărgăoan, R. (2020). Gibberellic acid can improve seed germination and ornamental quality of selected cyclamen species grown under short and long days. *Agronomy*, *10*(4). https://doi.org/10.3390/agronomy10040516

Cowles, J., Boldgiv, B., Liancourt, P., Petraitis, P., & Casper, B. (2018). Effects of increased temperature on plant communities depend on landscape location and precipitation. *Ecology and Evolution*, *8*, 5267–5278. https://doi.org/10.1002/ece3.3995

Dai, A., & National Center for Atmospheric Research Staff, (Eds.). (2019). *The Climate Data Guide: Palmer Drought Severity Index (PDSI)*. https://climatedataguide.ucar.edu/climate-data/palmer-drought-severity-index-pdsi

Damschen, E. I., Harrison, S., Ackerly, D. D., Fernandez-Going, B. M., & Anacker, B. L. (2012). Endemic plant communities on special soils: early victims or hardy survivors of climate change? *Journal of Ecology*, *100*(5), 1122–1130. https://doi.org/10.1111/j.1365-2745.2012.01986.x

Darwin, C. (1872). *On the origin of species* (6th ed.).

DeMalach, N., Zaady, E., & Kadmon, R. (2017). Contrasting effects of water and nutrient additions on grassland communities: A global meta-analysis. *Global Ecology and Biogeography*, *26*(8), 983–992. https://doi.org/10.1111/geb.12603

Dhillon, B. S., Kumar, V., Sagwal, P., Kaur, N., Mangat, G. S., & Singh, S. (2021). Seed Priming with Potassium Nitrate and Gibberellic Acid Enhances the Performance of Dry Direct Seeded Rice (Oryza sativa L.) in North-Western India. *Agronomy*, *11*(5).

Elzenga, J. T. M., Bekker, R. M., & Pritchard, H. W. (2019). Maximising the use of native seeds in restoration projects. *Plant Biology*, *21*(3), 377–379. https://doi.org/10.1111/plb.12984

ESRI. (2019). *USA Soils Map Units*. USDA Natural Resources Conservation Service. https://landscape11.arcgis.com/arcgis/rest/services/USA_Soils_Map_Units/featureserver

Faber, P. M. (1997). California's wild gardens : A living legacy. In *California Native Plant Society for the California Department of Fish and Game*.

Far Western Anthropological Research Group, I. (2010). *The Contact-Period Native California Community Distribution Model: A Dynamic Digital Atlas and Wiki Encyclopedia*.

Favier, J. F. (1995). A Model for Germination Rate during Dormancy Loss in Hordeum vulgare. *Annals of Botany*, *76*(6), 631–638. https://doi.org/10.1006/anbo.1995.1141

Fernald, M. L. (1918). The geographic affinities of the vascular flora of New England, the Maritime Provinces, and Newfoundland. *American Journal of Botany*, *5*, 219–247.

Fernald, M. L. (1925). Persistence of Plants in Unglaciated Areas of Boreal America. *Memoirs of the American Academy of Arts and Sciences*, *15*(3), 239–342. https://doi.org/10.2307/25058128

Fernald, M. L. (1942). Misinterpretation of Atlantic coastal plain species. *Rhodora*, *44*, 238–246.

Fernald, M. L. (1950). *Gray's Manual of Botany*. D. Van Nostrand Co.

Fernald, M. L. (1929). Some relationships of the floras of the Northern Hemisphere. *Proceedings of the International Congress of Plant Sciences*, 1487–1507.

Fiedler, P. L. (1992). Cladistic test of the adaptational hypothesis for serpentine endemism. In A. J. M. Baker, J. Proctor, & R. D. Reeves (Eds.), *The Vegetation of Ultramafic (Serpentine) Soils: Proceedings of the First International Conference on Serpentine Ecology* (pp. 421–432). Intercept.

Fiedler, P. (1995). Rarity in the California Flora: New thoughts on old ideas. *Madroño*, *42*(2), 127–141. https://www.jstor.org/stable/41425060

Fiedler, Peggy, Knapp, B. E., & Fredricks, N. (1998). *Rare Plant Demography: Lessons from the Mariposa Lilies (Calochortus: Liliaceae)*.

Finch-Savage, W. E., & Footitt, S. (2012). To germinate or not to germinate: A question of dormancy relief not germination stimulation. *Seed Science Research*, *22*(4), 243–248. https://doi.org/10.1017/S0960258512000165

Finch-Savage, W. E., & Leubner-Metzger, G. (2006). Seed dormancy and the control of germination. *New Phytologist*, *171*(3), 501–523. https://doi.org/10.1111/j.1469-8137.2006.01787.x

Foley, M. E. (2016). Germination of leafy spurge (Euphorbia esula L.) seeds under alternating temperatures: The effects of amplitude, midpoint, and imbibition in gibberellic acid. *Canadian Journal of Plant Science*, *96*(3), 321–328. https://doi.org/10.1139/cjps-2015-0269

Fotheringham, C. J., & Keeley, J. E. (1998). *Ecology and Distribution of Braunton's milkvetch (Astragalus brauntonii) and Lyon's pentachaeta (Pentachaeta lyonii)*.

Freeman, C. C. (2020). *Carpenteria californica. In: Flora of North America Editorial Committee*. Flora of North America North of Mexico [Online]. http://www.efloras.org/florataxon.aspx?flora_id=1&taxon_id=220002379

Gilliam, H. (2002). *Weather of the San Francisco Bay Region*. University of California Press.

Gornish, E. S., & Tylianakis, J. M. (2013). Community shifts under climate change: Mechanisms at multiple scales. In *American Journal of Botany* (Vol. 100, Issue 7, pp. 1422–1434). https://doi.org/10.3732/ajb.1300046

Gottlieb, L. D. (1973). Genetic Differentiation, Sympatric Speciation, and the Origin of a Diploid Species of Stephanomeria. *American Journal of Botany*, *60*(6), 545. https://doi.org/10.2307/2441378

Grace, J. B., Safford, H. D., & Harrison, S. (2007). Large-scale causes of variation in the serpentine vegetation of California. *Plant and Soil*, *293*(1–2), 121–132. https://doi.org/10.1007/s11104-007-9196-6

Graham, A. (1999). *Late Cretaceous and Cenozoic history of North American vegetation, north of Mexico*.

Graham, R. C. (2009). Serpentine Geoecology of Western North America. *Soil Science*, *174*(3), 193. https://doi.org/10.1097/SS.0b013e318199f342

Griggs, R. F. (1940). The Ecology of Rare Plants. *Bulletin of the Torrey Botanical Club*, *67*(7), 575. https://doi.org/10.2307/2481578

Grouios, C. P., & Manne, L. L. (2009). Utility of measuring abundance versus consistent occupancy in predicting biodiversity persistence. *Conservation Biology*, *23*(5), 1260–1269. https://doi.org/10.1111/j.1523-1739.2009.01253.x

Guenther, K. (2008). *Monitoring Annual Grassland Residual Dry Matter. A Mulch Manger's Guide for Monitoring Success*. Wildlands Solutions.

Guerrant, E. O., Fiedler, P. L., Havens, K., & Maunder, M. (2004). Revised Genetic Sampling Guidelines for Conservation Collections of Rare and Endnagered Plants. *Ex Situ Plant Conservation: Supporting Species Survival in the Wild*, *May 2017*, 419–438.

Harrington, J. (1972). Seed storage and longevity. In T. T. Kozlowski (Ed.), *Seed Biology Volume III Insects, and Seed Collection, Storage, Testing, and Certification.* (pp. 145–245).

Harrison, S., & Inouye, B. D. (2001). High beta diversity in the flora of Californian serpentine 'islands.' *Biodiversity and Conservation*, *11*, 1869–1876.

Harrison, S., & Noss, R. (2017). Endemism hotspots are linked to stable climatic refugia. *Annals of Botany*, *119*(2), 207–214. https://doi.org/10.1093/aob/mcw248

Harrison, S., Spasojevic, M. J., & Li, D. (2020). Climate and plant community diversity in space and time. *Proceedings of the National Academy of Sciences of the United States of America*, *117*(9), 4464–4470. https://doi.org/10.1073/pnas.1921724117

Harrison, S., Viers, J. H., & Quinn, J. F. (2000). *Climatic and Spatial Patterns of Diversity in the Serpentine Plants of California*. *6*(3), 153–161.

Harrison, S., Viers, J. H., Thorne, J. H., & Grace, J. B. (2008). Favorable environments and the persistence of naturally rare species. *Conservation Letters*, *1*(2), 65–74. https://doi.org/10.1111/J.1755-263X.2008.00010.X

Hartley, S., Krushelnycky, P. D., & Lester, P. J. (2010). Integrating physiology, population dynamics and climate to make multi-scale predictions for the spread of an invasive insect: The Argentine ant at Haleakala National Park, Hawaii. *Ecography*, *33*(1), 83–94. https://doi.org/10.1111/j.1600-0587.2009.06037.x

Hawkins, B. A., Field, R., Cornell, H. V., Currie, D. J., Guégan, J. F., Kaufman, D. M., Kerr, J. T., Mittelbach, G. G., Oberdorff, T., O'Brien, E. M., Porter, E. E., & Turner, J. R. G. (2003). Energy, water, and broad-scale geographic patterns of species richness. *Ecology*, *84*(12), 3105–3117. https://doi.org/10.1890/03-8006

Hebda, R. (2017). *Royal BC Museum Curator Report on Eriophyllum lanatum*. Royal BC Museum Blog. https://staff.royalbcmuseum.bc.ca/2017/01/09/common-woolly-sunflower/

Hellmann, J., McLachlan, J., Sax, D., & Schwartz, M. (2008). *Managed relocation*. https://www3.nd.edu/~hellmann/MRWorkingGroup/Managed_relocation.html

Hilhorst, H. W. M. (1995). A critical update on seed dormancy. I. Primary dormancy. *Seed Science Research*, *5*(2), 61–73. https://doi.org/10.1017/S0960258500002634

Hockey, P. A. R., & Curtis, O. E. (2009). Use of basic biological information for rapid prediction of the response of species to habitat loss. *Conservation Biology*, *23*(1), 64–71. https://doi.org/10.1111/j.1523-1739.2008.01028.x

Holland, R. F. (1998). Great Valley Vernal Pool Distribution, Photorevised 1996. *Ecology, Conservation, and Management of Vernal Pool Ecosystems – Proceedings from a 1996 Conference*, 71–75.

Holt, J. R. (2011). No allee effects in Lyon's Pentachaeta, A Federally Listed Endangered Sunflower. In *California State University, Northridge*. https://doi.org/10.1128/AAC.03728-14

Ingolia, M. (2018). *Habitat Protection Measures Fritillaria biflora var . ineziana (Hillsborough chocolate lily)*.

IUCN/SSC. (2013). Guidelines for Reintroductions and Other Conservation Translocations. In

IUCN Species Survival Commission (Ed.), *Ecologial Applications*.

James, J. J., Sheley, R. L., Erickson, T., Rollins, K. S., Taylor, M. H., & Dixon, K. W. (2013). A systems approach to restoring degraded drylands. In *Journal of Applied Ecology* (Vol. 50, Issue 3, pp. 730–739). https://doi.org/10.1111/1365-2664.12090

James, J. J., Svejcar, T. J., & Rinella, M. J. (2011). Demographic processes limiting seedling recruitment in arid grassland restoration. *Journal of Applied Ecology*, *48*(4), 961–969. https://doi.org/10.1111/j.1365-2664.2011.02009.x

Jefferson-Brown, M. J., & Pratt, K. (1997). *The gardener's guide to growing fritillaries*. David & Charles.

Joe, S. M., & Daehler, C. C. (2008). Invasive slugs as under-appreciated obstacles to rare plant restoration: Evidence from the Hawaiian Islands. *Biological Invasions*, *10*(2), 245–255. https://doi.org/10.1007/s10530-007-9126-9

Johnson, D. E., & Mooring, J. S. (2021). *Eriophyllum. In: Flora of North America Editorial Committee, eds. 1993+*. Flora of North America North of Mexico [Online]. http://floranorthamerica.org/Eriophyllum

Kay, K. M., Ward, K. L., Watt, L. R., & Schemske, D. W. (2011). Plant Speciation. In S. Harrison & N. Rajaaruna (Eds.), *Serpentine: The Evolution and Ecology of a Model System* (pp. 71–95). University of California Press.

Keeley, J. E., & Nitzberg, M. E. (1984). Role of Charred Wood in the Germination of the Chaparral Herbs Emmenanthe penduliflora (Hydrophyllaceae) and Eriophyllum confertiflorum (Asteraceae). In *Madroño* (Vol. 31, Issue 4). https://about.jstor.org/terms

Keil, D. J., & Lane, M. A. (2021). *Pentachaeta bellidiflora*. Jepson Flora Project (Eds.) Jepson EFlora. https://ucjeps.berkeley.edu/eflora/eflora_display.php?tid=4279

Kildisheva, O. A., Dixon, K. W., Silveira, F. A. O., Chapman, T., Di Sacco, A., Mondoni, A., Turner, S. R., & Cross, A. T. (2020). Dormancy and germination: making every seed count in restoration. *Restoration Ecology*, *28*(S3), S256–S265. https://doi.org/10.1111/rec.13140

Krause, W. (1958). Andere Bodenspezialisten. In *Die Mineralische Ernährung der Pflanze / Mineral Nutrition of Plants* (p. 756). Springer Berlin Heidelberg. https://doi.org/http://doi-org-443.webvpn.fjmu.edu.cn/10.1007/978-3-642-94729-2_26

Kreft, H., & Jetz, W. (2007). Global patterns and determinants of vascular plant diversity. *Proceedings of the National Academy of Sciences of the United States of America*, *104*(14), 5925–5930. https://doi.org/10.1073/pnas.0608361104

Kruckeberg, A. R. (1950). *An experimental inquiry into the nature of endemism on serpentine soils*. University of California, Berkeley.

Kruckeberg, A. R. (1951). Intraspecific Variability in the Response of Certain Native Plant Species to Serpentine Soil. *American Journal of Botany*, *38*(6), 408. https://doi.org/10.2307/2438248

Kruckeberg, A. R. (1954). The Ecology of Serpentine Soils. III. Plant Species in Relation to

Serpentine Soils. *BMC Ecology*, *35*(2), 267–274.

Kruckeberg, A. R. (1984). *California serpentines : Flora, vegetation, geology, soils, and management problems* (University). University of California Press.

Kruckeberg, A. R. (2002). The influences of lithology on plant life. In *Geology and Plant Life: The Effects of Landforms and Rock Type on Plants* (pp. 160–181). University of Washington Press.

Kuhnlein, H. V., & Turner, N. J. (1991). *Traditional Plant Foods of Canadian Indigenous Peoples : Nutrition, Botany, and Use*. Gordon and Breach.

Ladouceur, E., Jiménez-Alfaro, B., Marin, M., De Vitis, M., Abbandonato, H., Iannetta, P. P. M., Bonomi, C., & Pritchard, H. W. (2018). Native Seed Supply and the Restoration Species Pool. *Conservation Letters*, *11*(2), 1–9. https://doi.org/10.1111/conl.12381

Lady Bird Johnson Wildflower Center. (2016). *Eriophyllum lanatum (Common woolly sunflower) | Native Plants of North America*. Lady Bird Johnson Wildflower Center Plant Database. https://www.wildflower.org/plants/result.php?id_plant=erla6

Lawler, J. J., Tear, T. H., Pyke, C., Shaw, R. M., Gonzalez, P., Kareiva, P., Hansen, L., Hannah, L., Klausmeyer, K., Aldous, A., Bienz, C., & Pearsall, S. (2010). Resource management in a changing and uncertain climate. *Frontiers in Ecology and the Environment*, *8*(1), 35–43. https://doi.org/10.1890/070146

Lesage, J. C., Press, D., & Holl, K. D. (2020). Lessons from the reintroduction of listed plant species in California. *Biodiversity and Conservation*, *29*(13), 3703–3716. https://doi.org/10.1007/s10531-020-02045-y

Loh, R. K., & Daehler, C. C. (2008). Influence of woody invader control methods and seed availability on native and invasive species establishment in a Hawaiian forest. *Biological Invasions*, *10*(6), 805–819. https://doi.org/10.1007/s10530-008-9237-y

Lyell, C. (1830). *Principles of geology* (1st ed.).

Martin, A. C., Zim, H. S., & Nelson, A. L. (1961). Upland Weed and Herbs. In *American wildlife and plants - a guide to wildlife food habits* (pp. 400–406). Dover Publications.

Maschinski, J., Albrecht, M. A., Monks, L., & Haskins, K. E. (2012). *Reintroduction Practice Guidelines*. https://doi.org/10.5822/978-1-

Maschinski, J., & Haskins, K. E. (2012). Plant Reintroduction in a Changing Climate. In *Plant Reintroduction in a Changing Climate*. https://doi.org/10.5822/978-1-61091-183-2

Mason, H. L. (1946a). The Edaphic Factor in Narrow Endemism. I. The nature of environmental influences. *Madroño*, *8*(7), 209–226. https://www.jstor.org/stable/41431623

Mason, H. L. (1946b). The Edaphic Factor in Narrow Endemism. II. The geographic occurrence of plants of highly restricted patterns of distribution. *Madroño*, *8*(8), 241–257.

McNeal, D. W., & Ness, B. D. (2012). *Fritillaria biflora var. ineziana*. Jepson Flora Project (Eds.) Jepson EFlora. https://ucjeps.berkeley.edu/eflora/eflora_display.php?tid=59252

McNeal, D. W., & Ness, B. D. (2015). *Fritillaria*. Jepson Flora Project (Eds.) Jepson EFlora. https://ucjeps.berkeley.edu/eflora/eflora_display.php?tid=9513

Merritt, D. J., & Dixon, K. W. (2011). Restoration Seed Banks— A Matter of Scale. *Science*, *332*, 424–425. https://doi.org/10.1126/science.1203083

Mével, C. (2003). Serpentinization of abyssal peridotites at mid-ocean ridges. *Geoscience*, *335*(10–11), 825–852. https://doi.org/10.1016/j.crte.2003.08.006

Missouri Botanical Garden. (2021). *Fritillaria biflora var. ineziana*. Tropicos.Org. https://www.tropicos.org/name/18406288

Moerman, D., & Best, J. (2003). *Native American Ethnobotany Database*. http://naeb.brit.org/uses/search/?string=eriophyllum

Mooring, J. S. (1994). A cytogenetic study of Eriophyllum confertiflorum (Compositae, Helenieae). *American Journal of Botany*, *81*(7), 919–926. https://doi.org/10.2307/2445773

Munz, P. A. (1959). *A California Flora*. University of California Press.

Nagel, R., Durka, W., Bossdorf, O., & Bucharova, A. (2019). Rapid evolution in native plants cultivated for ecological restoration: not a general pattern. *Plant Biology*, *21*(3), 551–558. https://doi.org/10.1111/plb.12901

National Oceanic and Atmospheric Administration (NOAA). (2021). *National Climatic Data Center, Data Tools: 1981-2010 Normals, San Francisco International Airport, CA US*. Data Tools: 1981 - 2010 Normals. https://www.ncdc.noaa.gov/cdo-web/datatools/normals

Nesom, G. L. (2020a). *Pentachaeta. In: Flora of North America Editorial Committee*. Flora of North America North of Mexico [Online]. http://beta.floranorthamerica.org/Pentachaeta

Nesom, G. L. (2020b, November 5). *Pentachaeta bellidiflora. In: Flora of North America Editorial Committee, eds. 1993+*. Flora of North America North of Mexico [Online]. http://floranorthamerica.org/Pentachaeta_bellidiflora

Ness, B. (2020, November 5). *Fritillaria. In: Flora of North America Editorial Committee, eds.* Flora of North America. http://floranorthamerica.org/Fritillaria

Nikolaeva, M. G. (1977). Factors controlling the seed dormancy pattern. In ed. A. A. Khan (Ed.), *The Physiological and Biochemistry of Seed Dormancy and Germination* (pp. 51–74).

NOAA National Centers for Environmental Information. (2021). *Climate at a Glance: Divisional Time Series California, Climate Division 4. Central Coast Drainage Palmer Modified Drought Index (PMDI) Rankings*. https://www.ncdc.noaa.gov/cag/divisional/rankings/0404/pmdi/189501

Nogueira-Filho, S. L. G., Nogueira, S. S. C., & Fragoso, J. M. V. (2009). Ecological impacts of feral pigs in the Hawaiian Islands. In *Biodiversity and Conservation* (Vol. 18, Issue 14, pp. 3677–3683). https://doi.org/10.1007/s10531-009-9680-9

Nomad Ecology. (2019). *Botanical Resources Survey Report. Peninsula Watershed. San Mateo County, California [Draft]*.

Norton, R. A., Finlayson, A. J., & Towers, G. H. N. (1985). Two dithiacyclohexadiene polyacetylenes from Chaenactis douglasii and Eriophyllum lanatum. *Phytochemistry*, *24*(2), 356–357. https://doi.org/10.1016/S0031-9422(00)83554-2

Noyes, R. D., & Rieseberg, L. H. (1999). Its sequence data support a single origin for North American astereae (asteraceae) and reflect deep geographic divisions in Aster s.l. *American Journal of Botany*, *86*(3), 398–412. https://doi.org/10.2307/2656761

Oldfield, S. (2019). The US National Seed Strategy for Rehabilitation and Restoration: progress and prospects. *Plant Biology*, *21*(3), 380–382. https://doi.org/10.1111/plb.12851

Palmerlee, A. P., & Young, T. P. (2010). Direct seeding is more cost effective than container stock across ten woody species in California. *Native Plants Journal*, *11*(2), 89–102. https://doi.org/10.2979/npj.2010.11.2.89

Patterson, T. B., & Givnish, T. J. (2002). Phylogeny, Concerted Convergence, and Phylogenetic Niche Conservatism in the Core Liliales: Insights from rbcL and ndhF Sequence D. In *Evolution* (Vol. 56, Issue 2).

Pavlik, B. M. (1990). Reintroduction of Amsinckia grandiflora to Stewartville. In *Endangered Plant Program*. https://doi.org/10.19009/jjacg.17.1_19

Pavlik, B. M. (1991). *Reintroduction of Amsinckia grandiflora to Three Sites Across its Historic Range* (Issue June).

Pavlik, B. M. (1994). *Tales of Success and Failure: Trends in Natural and Reintroduced Populations of Amsinckia grandiflora Under Different Management Regimes*.

Pavlik, B. M. (1996a). Defining and Measuring Success. In D. A. Falk, C. I. Miller, & M. Olwell (Eds.), *Restoring Diversity: stategies for reintroduction of endangered plants* (pp. 127–156). Island Press.

Pavlik, B. M. (1996b). *Populations of Amsinckia grandiflora (Large-flowered fiddleneck): Growth and decline under different management regimes* (Issue December 1996).

Pavlik, B. M., & Heisler, K. (1988). *Habitat Characterization Establishment and Selection of Potential Sites for Establihment of New Populations of Amsinckia grandiflora* (Issue December 1988).

Pavlik, B. M., Nickrent, D. L., & Howald, A. M. (1993). The Recovery of an Endangered Plant. I. Creating a New Population of Amsinckia grandiflora. *Biology*, *7*(3), 510–526.

Peery, M. Z., Beissinger, S. R., Newman, S. H., Burkett, E. B., & Williams, T. D. (2004). Applying the declining population paradigm: Diagnosing causes of poor reproduction in the Marbled Murrelet. *Conservation Biology*, *18*(4), 1088–1098. https://doi.org/10.1111/j.1523-1739.2004.00134.x

Pender, R. J., Shiels, A. B., Bialic-Murphy, L., & Mosher, S. M. (2013). Large-scale rodent control reduces pre- and post-dispersal seed predation of the endangered Hawaiian lobeliad, Cyanea superba subsp. superba (Campanulaceae). *Biological Invasions*, *15*(1), 213–223. https://doi.org/10.1007/s10530-012-0280-3

Pichi-Sermolli, R. E. (1948). An Index for Establishing the Degree of Maturity in Plant Communities. *The Journal of Ecology*, *36*(1), 85. https://doi.org/10.2307/2256647

Pierce, D. W., Kalansky, J. F., Cayan, D. R., & Brown, E. G. (2018). *Climate, Drought, and Sea Level Rise Scenarios for California's Fourth Climate Change Assessment*. www.climateassessment.ca.gov.

Pratt, L. W., & Stone, C. P. (1990). *Alteration of Native Hawaiian Vegetation : Effects of Humans, Their Activities and Introductions*. University of Hawaii, Cooperative National Park Resources Studies Unit.

Qureshi, U. S., Shakeel, M., Qureshi, K. M., Saleem, A., Mir, A. R., & Qureshi, A. A. (2016). Effect of seed priming through gibberellic acid (GA3) and imidacloprid on bismarckia palm seed. *International Journal of Biosciences (IJB)*, *8*(4), 18–27. https://doi.org/10.12692/ijb/8.4.18-27

Rabinowitz, D. (1981). Seven forms of rarity. In *The Biological Aspects of Rare Plant Conservation* (pp. 205–217). https://www.esf.edu/efb/parry/Invert_Cons_14_Readings/Rabinowitz_1981.pdf

Rafinesque, C. S. (1836). *New flora of North America*. Publisher unknown.

Rajakaruna, N., & Boyd, R. S. (2014). Serpentine Soils. *Ecology*. https://doi.org/10.1093/obo/9780199830060-0055

Raven, P. H. (1964). Catastrophic Selection and Edaphic Endemism. *Evolution*, *18*(2), 336–338. https://www.jstor.org/stable/2406409

Raven, P. H. (1973). The Evolution of Mediterranean Floras. In F. di Castri & H. A. Mooney (Eds.), *Mediterranean type ecosystems; origin and structure.* (p. 223). Springer-Verlag.

Raven, P. H., & Axelrod, D. (1978). *Origin and Relationships of the California Flora*. University of California Press.

Roberts, R. P., & Urbatsch, L. E. (2003). Molecular phylogeny of Ericameria (Asteraceae, Astereae) based on nuclear ribosomal 3′ ETS and ITS sequence data. *Taxon*, *52*(2), 209–228. https://doi.org/10.2307/3647390

Roberts, R. P., & Urbatsch, L. E. (2004). Molecular Phylogeny of Chrysothamnus and Related Genera (Asteraceae, Astereae) Based on Nuclear Ribosomal 3′ ETS and ITS Sequence Data. *Systematic Botany*, *29*(1), 199–215. https://doi.org/10.1600/036364404772973410

Robichaux, R., Canfield, J., Warshauer, F., Bruegmann, M., & Friar, E. (1998). Restoring Mauna Kea's Crown Jewel. *Endangered Species Bulletin*, 21.

Robichaux, R. H., Moriyasu, P. Y., Enoka, J. H., McDaniel, S., Loh, R. K., Bio, K. F., Bakutis, A., Tunison, J. T., Bergfeld, S. T., Perry, J. L., Warshauer, F. R., Wasser, M., Cole, T. C., Agorastos, N. R., Cole, I. W., Camara, J. K., Rubenstein, T., Whitehead, A. N., VanDeMark, J. R., … Bruegmann, M. M. (2017). Silversword and lobeliad reintroduction linked to landscape restoration on Mauna Loa and Kīlauea, and its implications for plant adaptive radiation in Hawaiʻi. *Biological Conservation*, *213*(July), 59–69.

https://doi.org/10.1016/j.biocon.2017.07.001

Robison, R. A., & Morey, S. (1992). *Report to the Fish and Game Commission on the status of white- rayed pentachaeta (Pentachaeta bellidiflora)*.

Rønsted, N., Law, S., Thornton, H., Fay, M. F., & Chase, M. W. (2005). Molecular phylogenetic evidence for the monophyly of Fritillaria and Lilium (Liliaceae; Liliales) and the infrageneric classification of Fritillaria. *Molecular Phylogenetics and Evolution*, *35*(3), 509–527. https://doi.org/10.1016/j.ympev.2004.12.023

Roskov, Y., Ower, G., Orrell, T., Nicolson, D., Bailly, N., Kirk, P. M., Bourgoin, T., DeWalt, R. E., Decock, W., Nieukerken, E., Zarucchi, J., & L., P. (2019). *Fritillaria meleagris*. Species 2000 & ITIS Catalogue of Life, 2019 Annual Checklist. http://www.catalogueoflife.org/annual-checklist/2019/details/species/id/7414c88fa3fb96179f4021d3963f207e

Ryan, S. P. (2014). *Molecular phylogeny and character evolution of Fritillaria subgenus Liliorhiza (Liliaceae)*.

Safford, H. D., Viers, J. H., & Harrison, S. P. (2005). Serpentine Endemism in the California Flora: A Database of Serpentine Affinity. *Madroño*, *52*(4), 222–257.

Safford, H., & Miller, J. E. D. (2020). an Updated Database of Serpentine Endemism in the California Flora. *Madroño*, *67*(2), 85–104. https://doi.org/10.3120/0024-9637-67.2.85

Safford, Hugh D., & Harrison, S. (2004). Fire effects on plant diversity in serpentine vs. sandstone chaparral. *Ecology*, *85*(2), 539–548. https://doi.org/10.1890/03-0039

San Francisco Public Utilities Comission. (2006). *SFPUC Water Enterprise Environmental Stewardship Policy FINAL DRAFT*.

Schmid, R., & Tibor, D. P. (2002). California Native Plant Society's Inventory of Rare and Endangered Plants of California. In *Taxon* (Sixth, Vol. 51, Issue 1). https://doi.org/10.2307/1555013

Shaffer, M. L. (1981). Minimum Population Sizes for Species Conservation. *BioScience*, *31*(2), 131–134. https://doi.org/10.2307/1308256

Society for Ecological Restoration International Science & Policy Working Group. (2004). *The SER International Primer on Ecological Restoration*. Society for Ecological Restoration International. www.ser.org

Soltani, E., Baskin, C. C., & Baskin, J. M. (2017). A graphical method for identifying the six types of non-deep physiological dormancy in seeds. *Plant Biology*, *19*(5), 673–682. https://doi.org/10.1111/plb.12590

Sommer, J. H., Kreft, H., Kier, G., Jetz, W., Mutke, J., & Barthlott, W. (2010). Projected impacts of Climate change on regional capacities for global plant species richness. *Proceedings of the Royal Society B: Biological Sciences*, *277*(1692), 2271–2280. https://doi.org/10.1098/rspb.2010.0120

Spasojevic, M. J., Damschen, E. I., & Harrison, S. (2014). Patterns of seed dispersal syndromes on serpentine soils: Examining the roles of habitat patchiness, soil infertility and correlated functional traits. *Plant Ecology and Diversity*, *7*(3), 401–410. https://doi.org/10.1080/17550874.2012.678506

Ståhlberg, S., & Svanberg, I. (2006). Sarana in Eurasian folk botany. *Suomalais-Ugrilaisen Seuran Aikakauskirja*, *2006*(91). https://doi.org/10.33340/SUSA.81952

Stebbins, G. L. (1980). Rarity of Plant Species: A synthetic viewpoint. *Rhodora*, *82*(829), 77–86. https://www.jstor.org/stable/23311891

Stebbins, G. L., & Major, J. (1965). Endemism and Speciation in the California Flora. *Ecological Monographs*, *1*(1), 1–35.

Tadros, T. M. (1957). Evidence of the Presence of an Edapho-Biotic Factor in the Problem of Serpentine Tolerance. *Ecology*, *38*(1), 14–23. https://doi.org/10.2307/1932121

Tamura, M. N. (1998). Liliaceae. In K. K. (Ed.), *The families and genera of vascular plants. III. Flowering plants. Monocotyledons, Lilianae (except Orchidaceae)* (pp. 343–353). Springer.

Taylor, C. M., & Hastings, A. (2005). Allee effects in biological invasions. *Ecology Letters*, *8*(8), 895–908. https://doi.org/10.1111/j.1461-0248.2005.00787.x

Thorne, J. H., Huber, P. R., & Harrison, S. (2011). Systematic conservation planning: Protecting rarity, representation, and connectivity in regional landscapes. In S. Harrison & N. Rajakaruna (Eds.), *Serpentine: The evolution and ecology of a model system* (pp. 309–328). University of California Press.

Thornhill, A. H., Baldwin, B. G., Freyman, W. A., Nosratinia, S., Kling, M. M., Morueta-Holme, N., Madsen, T. P., Ackerly, D. D., & Mishler, B. D. (2017). Spatial phylogenetics of the native California flora. *BMC Biology*, *15*(1), 1–18. https://doi.org/10.1186/s12915-017-0435-x

Three Mountain Alliance. (2007). *Three Mountain Alliance Management Plan*.

Tiffney, B. H. (1985). Perspectives on the Origin of the Floristic Similarity between Eastern Asia and Eastern North America. *The Arnold Arboretum of Harvard University*, *66*(1), 73–94.

Turland, N. J., Wiersema, J. H., Barrie, F. R., Greuter, W., Hawksworth, D. L., Herendeen, P. S., Knapp, S., Kusber, W.-H., Li, D.-Z., Marhold, K., May, T. W., McNeill, J., Monro, A. M., Prado, J., Price, M. J., & Smith, G. F. (2018). *International Code of Nomenclature for algae, fungi, and plants* (Vol. 159). Koeltz Botanical Books. https://doi.org/10.12705/Code.2018

U.S. Fish and Wildlife. (2019). *Recovery Plan for Serpentine Soil Species of the San Francisco Bay Area: Recovery Plan Amendment*.

U.S. Fish and Wildlife Service. (1995). *Recovery plan for the Ka'u Silversword, Argyroxiphium kauense*. https://doi.org/10.1002/fsat.3501_4.x

U.S. Fish and Wildlife Service. (1998). *Recovery plan for the serpentine soil species of the San Francisco Bay Area*.

U.S. Fish and Wildlife Service. (1999). *Recovery Plan for Six Plants from the Mountains Surrounding the Los Angeles Basin*.

U.S. Fish and Wildlife Service. (2010). *U.S. Fish and Wildlife Service 5-Year Review:*

Acanthomintha obovata ssp. duttonii (San Mateo thornmint), Cirsium fontinale var. fontinale (fountain thistle), Pentachaeta bellidiflora (white-rayed pentachaeta).

U.S. Fish and Wildlife Service. (2011). *Eriophyllum latilobum (San Mateo woolly sunflower). 5-Year Review : Summary and Evaluation*.

U.S Fish and Wildlife Service. (2008). *Lyon's pentachaeta (Pentachaeta lyonii) 5-Year Review: Summary and Evaluatio*.

USDA Natural Resource Conservation Service. (2010). *National Plant Materials Manual - Fourth Edition* (Issue July). https://doi.org/https://www.nrcs.usda.gov/Internet/FSE_DOCUMENTS/stelprdb1042145.pdf

USDA, NRCS, & ESRI. (2020). *USA Soils Map Units*. ArcGIS Online. https://ucdavis.maps.arcgis.com/home/item.html?id=06e5fd61bdb6453fb16534c676e1c9b9

Vallano, D. M., Selmants, P. C., & Zavaleta, E. S. (2012). Simulated nitrogen deposition enhances the performance of an exotic grass relative to native serpentine grassland competitors. *Plant Ecology*, *213*(6), 1015–1026. https://doi.org/10.1007/s11258-012-0061-1

Vitousek, P. M., & Walker, L. (1989). Biological Invasion by Myrica Faya in Hawai'i : Plant Demography, Nitrogen Fixation, Ecosystem Effects. *Ecological Monographs*, *59*(3), 247–265. https://www.jstor.org/stable/1942601

Vollmar Natural Lands Consulting. (2016). *Phase 1 Implementation Final Report - Captive Propagation and Reintroduction of Large-flowered Fiddleneck (Amsinckia grandiflora) in Contra Coasta, Alameda, and San Joaquin Counties, California*.

Wall, M., & Arnold, G. (2009). *California Botanic Garden Seed Bank Collection Report*.

Watson, H. C. (1845). On the theory of 'progressive development,' applied in explanation of the origin and transmutation of species. *Phytologist*, *2*, 108–113, 140–147, 161–168, 225–228.

Wen, J. (1999). Evolution of Eastern Asian and Eastern North American Disjunct Distributions in Flowering Plants. *Annual Review of Ecology and Systematics*, *30*, 421–455.

Wilken, D. (2016). Rarity, Threats, and the Conservation Status of California Geophytes. *Fremontia*, *44*(3), 20–25.

Willis, J. C. (1922). *Age and area* (Cambridge).

Wilson, E. E., Mullen, L. M., & Holway, D. A. (2009). Life history plasticity magnifies the ecological effects of a social wasp invasion. *Proceedings of the National Academy of Sciences of the United States of America*, *106*(31), 12809–12813. https://doi.org/10.1073/pnas.0902979106

Wolf, A. (2001). Conservation of endemic plants in serpentine landscapes. *Biological Conservation*, *100*(1), 35–44. https://doi.org/10.1016/S0006-3207(00)00205-6

Zhang, L. (1983). Vegetation ecology and population biology of Fritillaria meleagris L. at the Kungsangen Nature Reserve, eastern Sweden. *Acta Phytogeographica Suecica*, *73*.

Zych, M., & Stpiczyńska, M. (2012). Neither protogynous nor obligatory out-crossed: Pollination biology and breeding system of the European Red List Fritillaria meleagris L. (Liliaceae). *Plant Biology*, *14*(2), 285–294. https://doi.org/10.1111/j.1438-8677.2011.00510.x

Appendix A

Figure 1. California Native Plant Society Rare Plant Ranks (Schmid & Tibor, 2002)

- 1A: Plants presumed extirpated in California and either rare or extinct elsewhere
- 1B: Plants rare, threatened, or endangered in California and elsewhere
- 2A: Plants presumed extirpated in California but common elsewhere
- 2B: Plants rare, threatened, or endangered in California but common elsewhere
- 3: Plants about which more information is needed, a review list
- 4: Plants of limited distribution, a watch list

Ranks at each level also include a threat rank (e.g. 1B.1) and are determined as follows:

- 0.1: Seriously threatened in California (over 80% of occurrences threatened / high degree and immediacy of threat)
- 0.2: Moderately threatened in California (20-80% occurrences threatened / moderate degree and immediacy of threat)
- 0.3: Not very threatened in California (less than 20% of occurrences threatened / low degree and immediacy of threat or no current threats known)

The category of Considered but Rejected is given to species that were previously listed or were considered for addition to the *Inventory* but rejected. Additionally, a category of Postponed exists for plants that were considered as possible additions, but were postponed due to due to significant taxonomic uncertainty and/or lack of information regarding distribution, abundance, rarity and/or endangerment.

Figure 2. KSFO Weather Station National Oceanic and Atmospheric Administration U.S. Climate Normals are graphed according to high and low temperature in degrees Fahrenheit along with monthly precipitation in inches derived from 1981-2010 averages

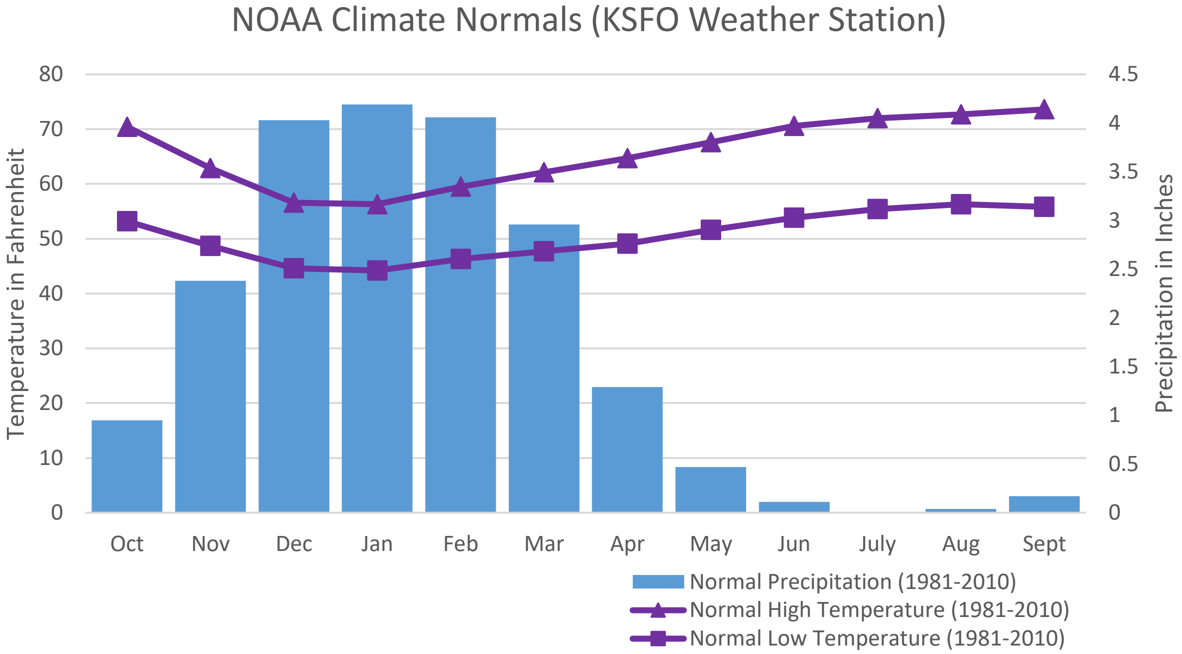


Figure 3. The graphed Palmer Drought Severity Index (PDSI) for California, Climate Division 4 illustrates wet and dry years with values below -3 representing severe to extreme drought (NOAA National Centers for Environmental Information, 2021).

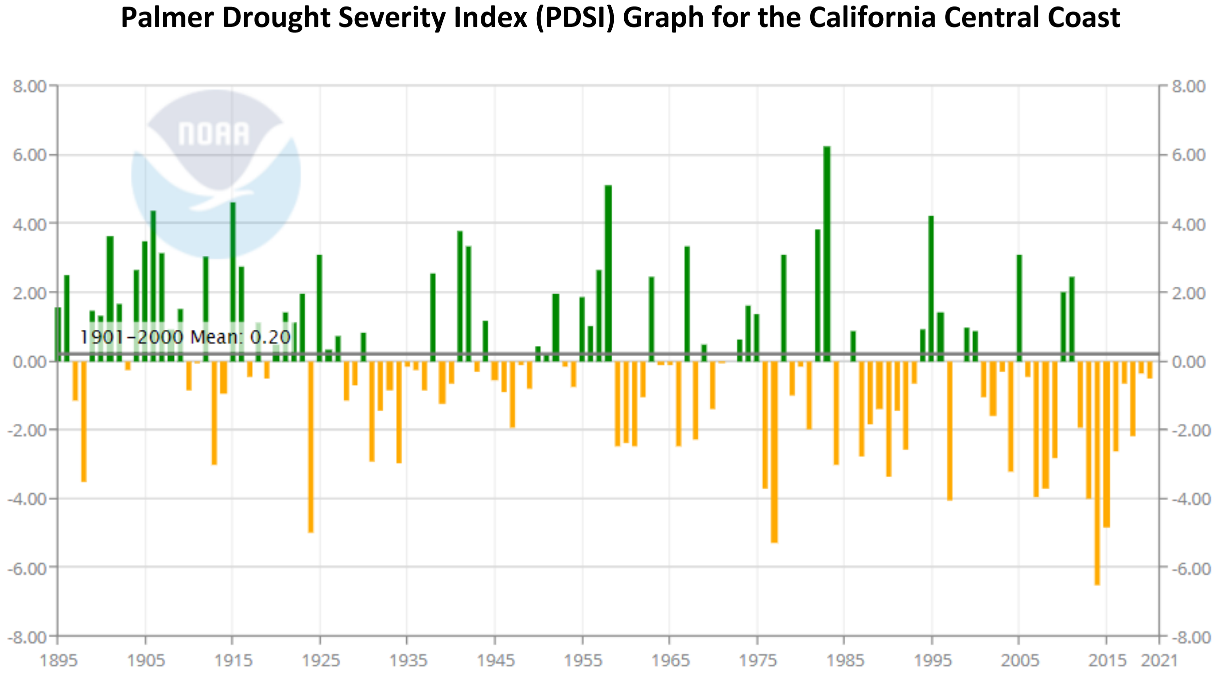


Figure 4. Annual average minimum and maximum temperature under modeled historical, medium and high emission scenarios for San Mateo Creek-Frontal San Francisco Bay Estuaries Watershed, California. (Cal-Adapt, 2020)

Annual Average Minimum and Maximum Temperature Forecasts according to

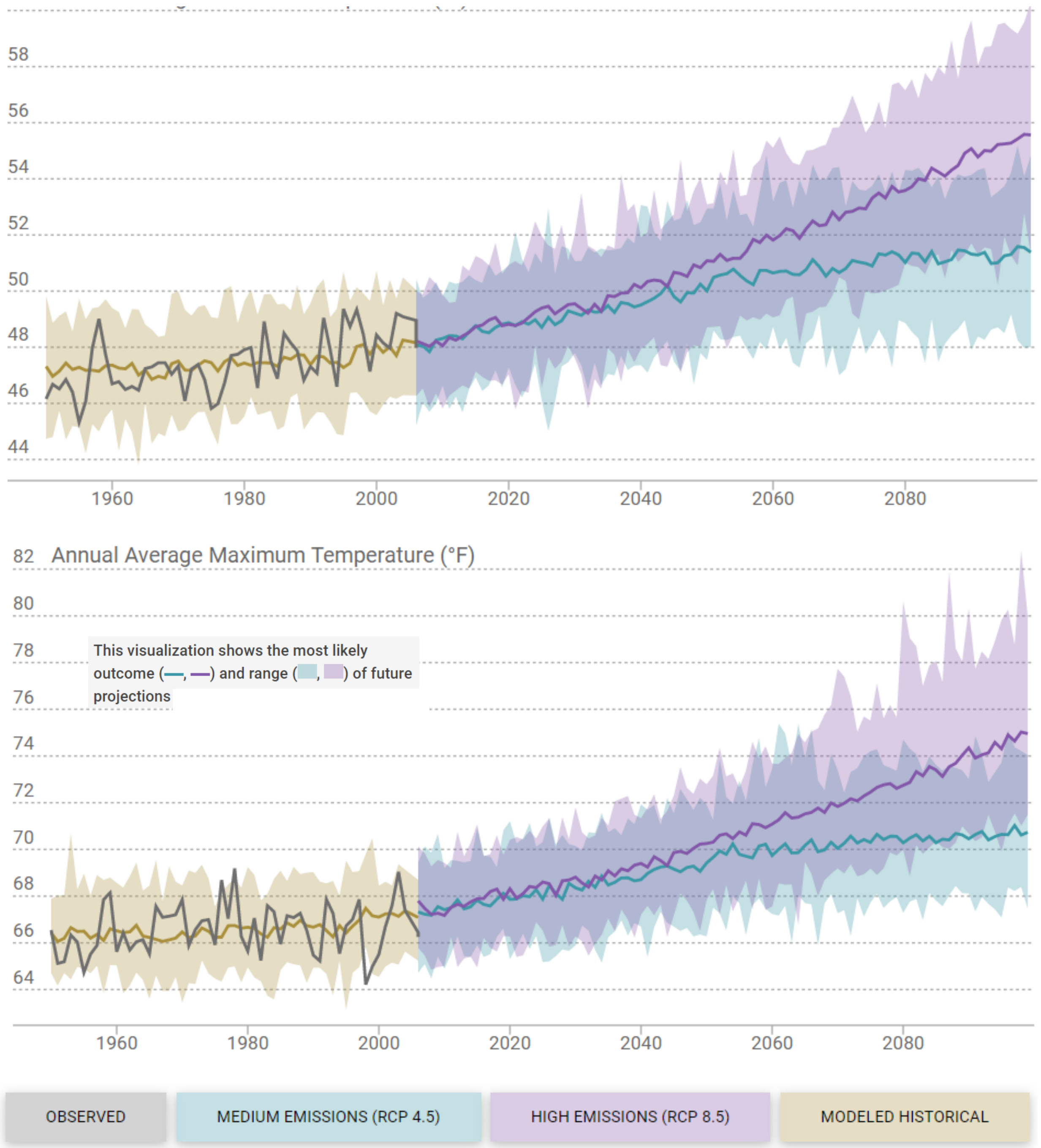


Figure 5.

Center for Plant Conservation Best Reintroduction Practice Guidelines Flow Diagram (Maschinski & Haskins, 2021)

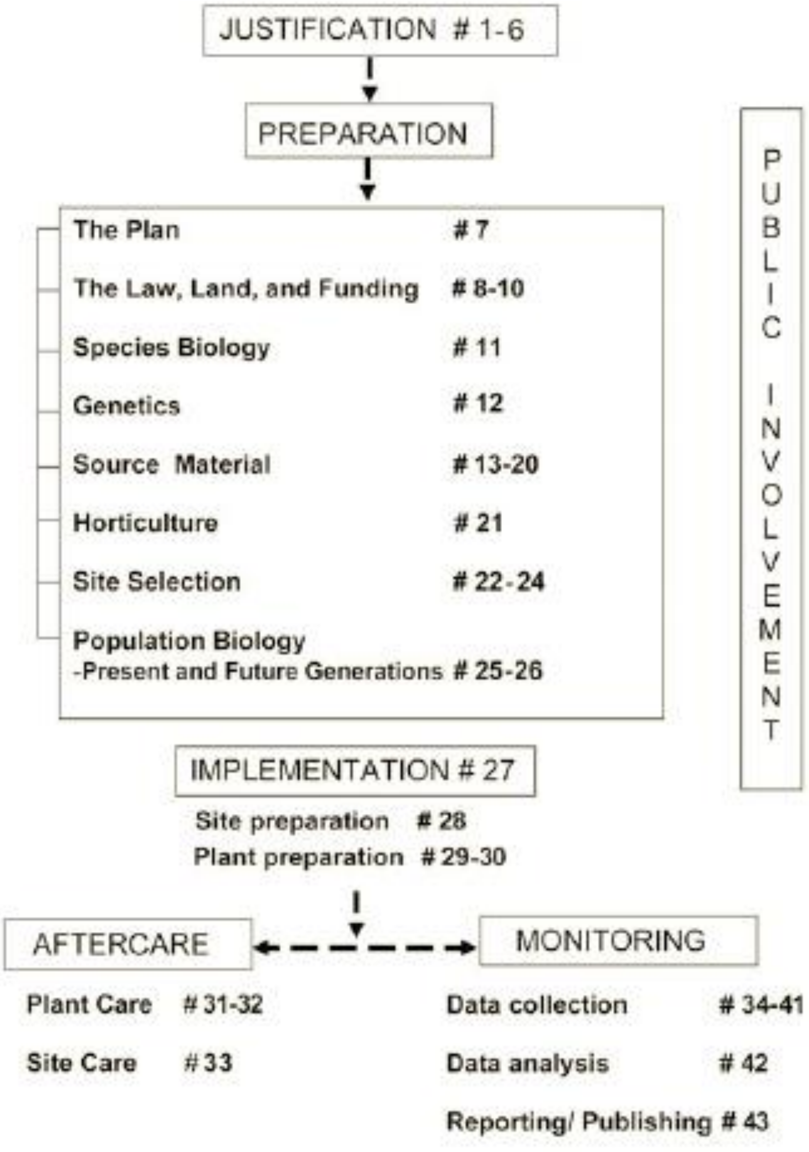
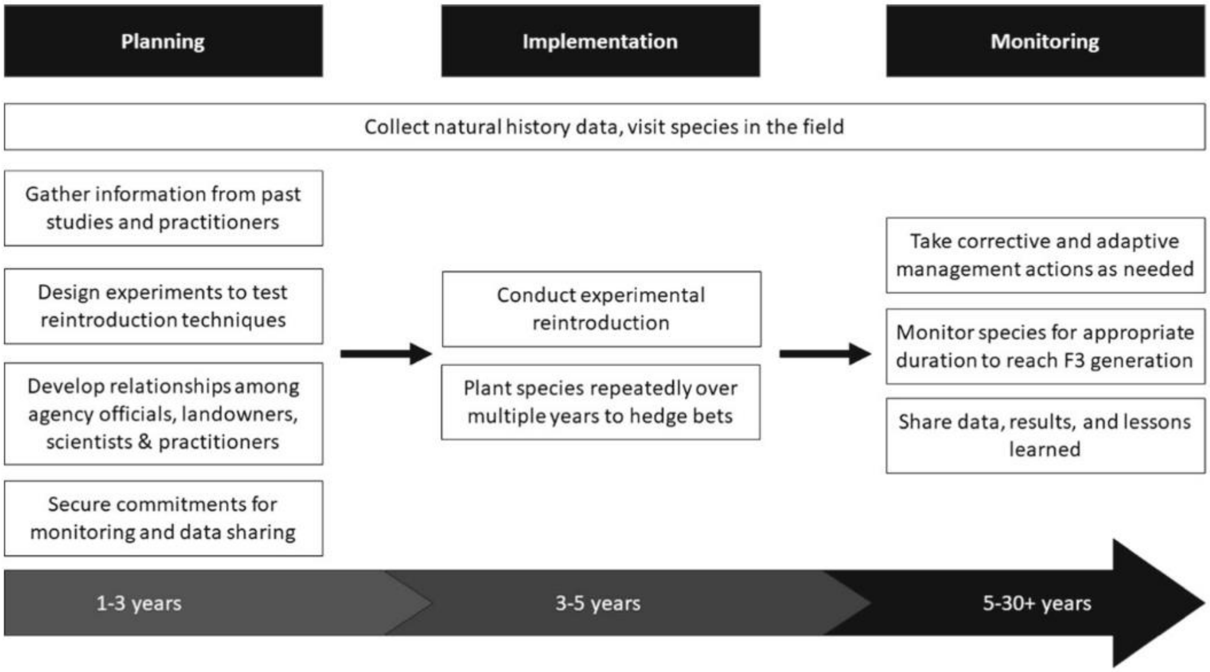


Figure 6.

Schematic of Practitioners Reintroduction Advice (Lesage, Press and Holl, 2020)



Fritillaria biflora **var.** ***ineziana*** **Photos**

a. *F. biflora* var. *ineziana* with a single flower 4/6/2018

b. *F. biflora* var. *ineziana* with a triple flower 4/6/2018

c. *F. biflora* var. *ineziana* triple fruit (note possible caterpillar web) 4/28/2020

d. *F. biflora* var. *ineziana* vegetative stage close up 1/22/2021

e. *F. biflora* var. *ineziana* vegetative stage emerging from surrounding vegetation 1/22/2021

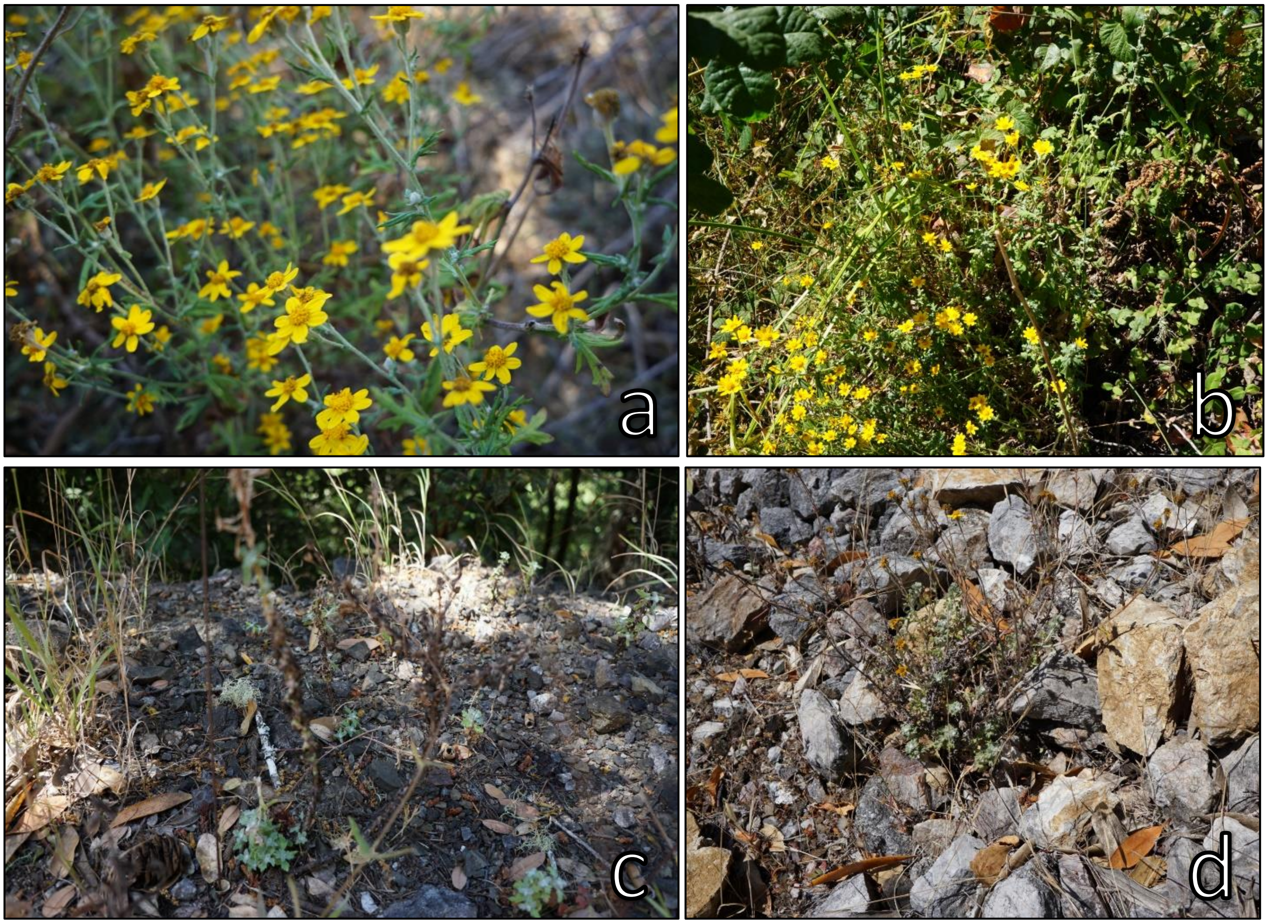
Pentachaeta bellidiflora **Photos**

a. *P. bellidiflora* close up with both young and senescing flowers 4/28/2020

b. *P. bellidiflora* senescing flowers beneath *Layia platyglossa* 4/28/2020

c. *P. bellidiflora* in dense bloom 3/9/2021

Eriophyllum latilobum Photos



a. *E. latilobum* at Occurrence 4 White Rock 7/17/2020

b. *E. latilobum* at Occurrence 7 beneath *Toxicodendron diversilobum* 7/17/2020

c. *E. latilobum* at Occurrence 4 San Mateo #2 along roadside 7/17/2020

d. *E. latilobum* at Occurrence 4 San Mateo #2 within abandon quarry site 7/17/2020

Abstract

Eriophyllum latilobum (Rydb.) is a Federal and State listed endangered plant belonging to the Asteraceae family. There are six extant occurrences in the San Francisco Peninsula in San Mateo County and two extirpated, possibly misidentified occurrences at the southern edge of the County. Invasive weeds, road, trail, or firebreak maintenance, and fire suppression are identified threats to the species (Nomad Ecology, 2019; U.S. Fish and Wildlife Service, 1998). In this study, the germination ecology of *E. latilobum* was investigated to identify treatments that may be used in the propagation of this species. Initial studies tested the effects of seed afterripening and cold-moist stratification on germination. Results from these preliminary trials did not demonstrate a significant effect on dormancy break or germination. A final trial tested the effects of scarification and gibberellic acid (GA_3) hormone on germination. Statistical analysis did not detect a significant effect of GA_3 on germination, however, scarification was found to improve the odds of germination by 45% as compared to the non-scarified group (p-value < 0.05; 95% confidence interval of odds ratio is 1.26, 1.68). However, viability was scored at 83.7% for the seeds used in this experiment. Therefore, a large proportion of the seeds did not receive adequate conditions for dormancy break. We conclude that further investigation should examine the effect of diurnal temperature on germination outcomes for the species.

Introduction

The greenhouse propagation of *Eriophyllum latilobum* is identified as a high priority research activity in the US Department of Fish and Wildlife's *Recovery plant for the Serpentine Soil Species of the San Francisco Bay Area* Report (U.S. Fish and Wildlife Service, 1998). In unpublished data, Mooring carried out several hybridization experiments for the genus and reported *E. latilobum* germination as less than 10 percent in a greenhouse setting (U.S. Fish and Wildlife Service, 2011). The California Botanic Garden (CalBG) maintains an *ex situ* accession which they have tested for germination percentages. Their 2009 accession was incubated in a germination chamber set to 13 hours of light at 20°C and 11 hours of dark at 12°C. The first trial reported 28% germination on a 5% agar substrate. Their follow up test in 2010, seeds were imbibed and set in a 5°C environment for 19 days, then transferred to their germination chamber for a five-week observation, resulting in 48% germination (Cheryl Birker, *in litt.* 2021).

Direct sowing of seed in the field is the most economical approach used in restoration and reintroduction efforts (Baskin & Baskin, 2020; Elzenga et al., 2019; Ladouceur et al., 2018; Oldfield, 2019; Palmerlee & Young, 2010). However, the freshly-matured seed of many species may be dormant at maturity and therefore require treatments to break dormancy before sowing (Baskin & Baskin, 2020). *Eriophyllum latilobum* is a member of the Asteraceae family. Typically, seeds of this family have nondeep physiological dormancy (nondeep PD) which is a type of physiological dormancy (Baskin & Baskin, 2014; Bhatla & A. Lal, 2018). Nondeep PD can break during dry storage; this method is known as afterripening (Baskin & Baskin, 2020). Following afterripening treatment, germination percentages and rates (speed) increase and the window of favorable conditions for germination widens (Baskin & Baskin, 1985; Favier, 1995; Soltani *et al.*, 2017). Additionally, cold or warm stratification, scarification, and gibberellic acid (GA_3) treatment have demonstrated dormancy break on nondeep PD seed (Finch-Savage & Footitt, 2012).

Since CalBG germination data suggest that *E. latilobum* has nondeep PD, the effects of seed afterripening, cold-moist stratification, scarification, and gibberellic acid (GA_3) treatments on germination were measured in a controlled environment at the University of California, Davis. The research objective is to identify a treatment that would relieve a majority of the seeds from dormancy.

Materials and Methods

Seed Collection

Seeds were collected in bulk from California Natural Database (CNDDB) Occurrences 1, 4, 6 and 7 on July 17, 2020. The chaff was separated from seeds using a seed blower and sieve. Seeds were dehydrated to 10-15% relative humidity. The seeds were then stored in sealed zip-lock bags at 4°C.

Initial Germination Test and Seed Viability Score. Seed dormancy was confirmed following an initial germination test within 2 weeks of harvest. After sterilization in 10% bleach solution, a lot of 30 seed was set on moistened blotter paper in a Zip-lock at 20°C and observed for two weeks. However, seeds did not germinate during the two weeks. According to Baskin (2014) a two-week germination test is generally considered the appropriate period to observe germination outcomes in response to treatment. If germination percentages are increasing at the end of the 2 weeks, consideration should be given to extending the germination period to a maximum of 3 or 4 weeks (Baskin & Baskin, 2014; Bhatla & A. Lal, 2018). This protocol was observed for all trials described in this report.

A tetrazolium (TZ) viability test according to AOSA Rules was administered to a sample containing an equal portion of seed from occurrences 1, 4, 6, and 7 and a self-fertilized sample. Seeds were separated into two groups: clean and light seeds. Using a seed blower, heavy seeds were separated from the light, unfilled and inert matter present in the sample. The light seeds were picked after blowing. Light seed viability is expected to be low since these seeds, when present, will have underdeveloped or decayed embryos. The low number of light seeds in Table 1 reflects the fact that many were not found in the blown portion. The viability of the clean or heavy seeds is what is usually considered the true viability of the sample after all other material is separated. Seeds were cut laterally, removing the distal end of cotyledons, then stained with 1% TZ 32°C for 24 hours for testing. The test of the bulk sample of 200 treated clean seeds resulted in an 83.7% viability score. The test of 24 seeds from the light seeds fraction resulted in a 25% viability score. The test of the self-fertilized sample of 16 seeds resulted in a 12.5% viability score.

Table 1

Eriophyllum latilobum Tetrazolium Viability Test Scores

		Clean seeds	Light seeds
Bulk	Viability	83.7 %	25.0%
	Number of seeds tested	200	24
Self-fertilized	No separation into groups	Viability = 12.5%	Number of seeds tested: 16

Disinfection, Preparation, and Imbibition of Seed. In afterripening and CMS experiments, seeds were treated using a 10% bleach dilution for 15 min. Seeds were rinsed three times using DI water. Seeds were placed into MilliporeSigma Petri Dishes lined with sterile absorbent pads. To allow for the imbibition of water by seeds, the pads were periodically moistened with a 0.2% Plant Preservative Mixture (Plant Cell Technology, Inc., Washington, DC; contains 5-choloro-2-methyl-isothiazole), henceforth called “prepared petri dishes”. The UC Davis Seed Biotechnology Center has found this preparation to prevent fungal growth in longer-term experiments and has not found effects on germination. Petri dishes were placed in a propagation flat with no holes at the base and sealed with a clear dome lid. These flats were subjected to a photoperiod of 12 light hours and 12 dark hours in their respective treatments.

Seeds that filled the seed coat with firm tissue were selected for afterripening and cold-moist stratification trials. This was determined by pressing the seed with a needle, if the seed did not collapse under pressure this suggested living tissue was present and the seed was probably viable. In experiment

3, many seeds became soft after imbibition and therefore the selection criteria in experiments 1 and 2 were not reliable.

In the scarification and GA_3 treatment experiment, seeds were sorted into microcentrifuge tubes according to their assigned treatment. Following imbibition, solutions were removed from the microcentrifuge tube using a micropipette leaving behind the treated seeds. Seeds were then sterilized with a 20% bleach, 2% tween-20 solution for twenty minutes. In a laminar flow hood following sterilization, the bleach-tween solution was removed using a micropipette, and the treated seeds were rinsed three times with nanopore water. This method is an adapted high-throughput sterilization technique by Lindsey *et al* (2020).

In a laminar flow hood, autoclaved 7% phytoagar substrate (Bio-World PlantMedia™) and 0.2% Plant Preservative Mixture were poured in 10mm x 10mm square, polystyrene petri dishes and stored until the following day for plating. Sterilized and rinsed seeds were plated and petri dishes (trays) and were sealed with surgical tape. Trays were then placed in a 10°C environment and subjected to a photoperiod of 9 light hours and 15 dark hours.

Afterripening Experiment

The first experiment treated seeds via afterripening treatments, which required dry seeds to be subjected to hot and dry conditions. The seeds were sampled at varying intervals, imbibed, and incubated at 20°C to measure germination response to various treatments. An equal share of seeds from each occurrence were used per treatment.

Table 2

Afterripening Treatments and Exposure Time

Treatment	Exposure Time
4°C (39°F) treatment	4 days
37°C (99°F)	7, 14, and 24 days
50°C (122°F)	2, 4, 7, 9, 11 and 24 days
20°C (68°F) salt container	7 days

The rate of seed aging doubles with every 5°C increase in temperature(Bewley *et al.*, 2013; Harrington, 1972). Therefore, one day of 50°C is about equal to one week of 37°C. The 37°C and 50°C treatments will indicate whether afterripening will work and if so, how long and strong a treatment is required to get the majority of seeds out of dormancy. If neither works or if seeds appear to need several weeks of 37°C or 50°C treatment, a salt slurry can expedite aging. A salt slurry is produced by adding enough water to table salt to allow it to become viscose. Placed in a sealed container, the salt slurry creates a 75% relative humidity environment which aggressively ages the seeds (Bewley *et al.*, 2013; Harrington, 1972).

On November 16, 2020, dry seeds were placed in four controlled environments to mimic natural afterripening effects on seeds. As an indicator of baseline dormancy for the seed lot, a sample of 50 dry seeds were placed in a sealed container at 4°C for four days. A sample of 145 dry seeds were placed in a sealed container at 37°C, 130 dry seeds were placed in a sealed container at 50°C, and 48 dry seeds were placed in a sealed container containing a salt slurry at 20°C. Seeds were sampled from each environment at various time intervals and stored at 4°C until incubation. On January 11, 2021 seeds across all treatments were set to incubate in a 20°C environment with a 12-hour photoperiod. See Appendix B Table 1 for a summary of treatments, corresponding sample sizes, and percent germination outcomes.

On January 11, prepared seeds were transferred into petri dishes with blotter paper and set to incubate at 20°C for 15 days. The seeds were considered to have germinated when the size of their radicle measured half the seeds' length for all experiments. The first germination event occurred on January 19 (Day 7). Germination events were then recorded on days 7, 9, 11, and 15 ending on January 27, 2020 (Day 15). The germination percentages were compared across treatments using a line graph (See Appendix B Figure 1).

Cold-moist Stratification Experiment

The second experiment subjected seeds to CMS treatments, which required imbibed seeds to be set at 5°C and sampled at intervals to measure germination response to days at 5°C. An equal share of seeds from each occurrence were used per treatment.

Table 3

Cold-moist Stratification Treatments and Incubation Temperatures

Treatment: days in CMS at 4°C	Incubation temperature
8,12,16,20,24,36,44,48,52,56,60,64,68,72,75	10°C
8,12,16,20,24,36,	15°C

On November 21, 2020, 20 petri dishes containing 20 prepared seeds were placed in a 5°C CMS environment. These samples were transferred to an incubation environment of 10°C or 15°C at various time intervals. Once a sample was transferred, germination frequency was recorded daily for 2 weeks. See Appendix B Tables 4 and 5 for a summary of treatments, corresponding sample sizes, and percent germination outcomes.

Germinated seeds were removed from petri-dishes to reduce competition and the risk of contamination. Germination occurring in the CMS environment was noted as a daily frequency. Incubation temperatures of 10°C and 15°C were tested to determine whether the seeds had an upper-temperature limit that would inhibit germination. However, due to increased germination events at 5°C and slightly better germination at 10°C, the 15°C incubation temperature was dropped to allow more days in CMS for the remaining seed.

The germination percentages were compared across treatments using a line graph (See Appendix B Figure 3).

Scarification and Gibberellic Acid Experiment

The third experiment treated seeds with a combination of stratification and GA_3 treatments. On September 21st, seeds were sorted by occurrence, then divided by weight into scarified and non-scarified groups. Scarification was employed with seeds set between two sheets of 200 grit sandpaper. The top sheet was pressed down and drawn in a circular pattern, clockwise, then counter-clockwise for 1 minute in each direction. The scarified group and non-scarified group were then separated into four separate lots by equal weight, stored individually in a corresponding labeled 1.5 microcentrifuge tube, and stored at 5°C until imbibition the following day.

Table 4

Scarification Groups and GA_3 Hormone Concentration Treatments

	Scarified Group		Non-Scarified Group
Tube 1	Control - 0 PPM GA_3	Tube 5	Control - 0 PPM GA_3
Tube 2	10 PPM GA_3	Tube 6	10 PPM GA_3
Tube 3	100 PPM GA_3	Tube 7	100 PPM GA_3
Tube 4	1000 PPM GA_3	Tube 8	1000 PPM GA_3

Solutions of 0, 10, 100, and 1000 PPM GA_3 were prepared using nanopure water. On September 22nd, for each scarified and non-scarified group one of the four tubes were treated with 0, 10, 100, or 1000 PPM GA_3 solution. Tubes with presorted seeds were filled with solution, agitated, and sealed using parafilm. The seeds were set to imbibe for 24 hours on a mechanical agitator in a 5°C environment. The agitation ensured that all seeds were exposed to the solution throughout the 24-hour imbibition period. On September 23rd, treated seeds were plated onto agar in a 10 x 10 grid where each seed was placed in an approximately 5mm^2 cell. Due to the disproportionate amount of seeds available for each occurrence and the logistics of applying the treatments, the total number of seeds for each tray is variable. Most trays contain the full amount of 100 seeds, however, smaller amounts are possible (minimum 16).

As the counts of germinated seeds from a total number of planted seeds were collected, we modeled the proportion of germinated seeds using a generalized linear model, which reports odds ratios of germination under different treatments.

Results

Effect of Afterripening on Germination

Appendix B Table 1 describes the specific afterripening treatment per sample and corresponding percent germination per observation day. The graphed data illustrates that seeds subjected to 37°C for seven days had a 13% final germination percentage as compared to other treatments (Appendix B Figure 8). However, the control sample percent germination (10%) is similar. Statistical analysis was not possible given the small sample sizes.

Effect of Cold-moist Stratification on Germination

Appendix B Table 4 and 5 describe the specific cold-moist stratification treatments per sample and corresponding percent germination per observation day. The graphed data illustrates that for seeds incubated at 10°C, the highest final germination percentages 33%, 30%, and 27%, are associated 76, 48, and 72 days of CMS, respectively (Appendix B Figure 2). Additionally, daily percent germination appeared to be greater when incubated at 10°C as compared to 15°C (Appendix B Figure 3). No statistical analysis was used given the small sample sizes.

Effect of Scarification and GA_3 on Germination

Appendix B Figure 4 compares the proportion of germinated seeds per tray with and without scarification over incubation time in days. The visualization illustrates that scarified seeds were more likely to germinate than non-scarified seeds. Appendix B Figure 5 compares the germination of trays with 0, 10, 100, and 1000 ppm GA3 over incubation time in days. There is no indication that hormone treatment increased or decreased germination. Appendix B Table 6 contains the daily germination frequencies for all trays used in the analysis. This experiment contained many seeds per treatment combination allowing for statistical analysis for hypothesis testing.

The tested hypotheses are:

- Hypothesis 1:
    - Null hypothesis: scarification does not affect germination
    - Alternative hypothesis: scarification does affect germination
- Hypothesis 2:
    - Null hypothesis: hormone does not affect germination
    - Alternative hypothesis: hormone does affect germination

To see how the seeds in a tray germinated over time, we calculated the percentage of germinated seeds in a tray at each day and plotted the percentage as a function of days in incubation. To account for the possible differences among seeds collected from different occurrences, the occurrences are treated as a random variable in a generalized mixed model, while the scarification and hormone treatment are considered the fixed variables. The measured effects from the model are as follows:

Table 5

Regression of germination proportion per treatment

Variable	Estimate	p.value	Odds ratio (95% CI)
(intercept)	-1.8553452	0.0000	
Scarified1	0.3742454	0.0000003	1.45 (1.26 ,1.68)
hormone10	-0.1300592	0.2054426	0.88 (0.72 ,1.07)
hormone100	-0.1829169	0.0716438	0.83 (0.68 ,1.02)
hormone1000	-0.0704010	0.4738448	1.07 (0.88 ,1.3)

The result shows that treatment with scarification increased the odds of germination by 45%. The 95% confidence interval of odds ratio is (1.26, 1.68) and p-value is < 0.05. Therefore, we can reject the null hypothesis that scarification does not affect germination.

We cannot reject the null hypothesis that hormone treatment does not affect germination. The p-values of the treatment with 10, 100, and 1000 ppm GA_3 are all greater than 0.05, and the 95% confidence intervals of odds ratio of all hormones include 1. We do not have enough evidence to claim that hormone treatment affects germination.

Discussion

Baskin & Baskin (2014) report that an average of 80% of dryland flora produce seeds with some level of dormancy. While the direct sowing of seeds in the field is most cost-effective, it can lead to high levels of plant establishment failure and waste (Commander *et al.*, 2013; James *et al.*, 2011, 2013; Merritt & Dixon, 2011). Additionally, the horticultural propagation of wild species may result in unintended selection pressures and loss of traits(Nagel *et al.*, 2019). Our research objective is to identify treatments that would release a majority of seeds from dormancy, thus reducing or eliminating these risks. Other Asteraceae species commonly possess nondeep PD which can be relieved by afterripening, cold or warm stratification, scarification, and GA_3 (Baskin & Baskin, 2014; Bhatla & A. Lal, 2018). Given the rarity of *E. latilobum*, a conservative amount of seeds was used to incrementally test their response to afterripening and cold-moist stratification. A final experiment tested commonly used horticultural practices, scarification, and GA_3 treatments, on a larger lot of seeds. This experiment resulted in higher germination percentages, which allowed the use of statistical analysis to determine treatment effects.

The afterripening treatment sample sizes ranged from 10-50 seeds. The afterripening treatment did not produce a pronounced effect on breaking dormancy. A sample size of 20 seeds per treatment was allocated for the CMS experiment. However, due to unexpected germination at 5°C, fewer seeds were available per sample. The data suggests that 10°C provides a slight benefit to germination rates, and so 15°C was dropped from sampling to allow the remaining seed to receive up to 76 days in CMS. An interesting finding from this trial was the lower than anticipated temperature for germination at 5°C. Additionally, we can say that 15°C and 10°C are within the temperature limits for seed germination. Although statistical significance cannot be derived from the results of this experiment, it seems that these seeds are highly variable in their response to environmental signals for dormancy break. Due to the low germination results and sample sizes per treatment, statistical analysis would not bear meaningful comparisons for either afterripening or cold-moist stratification experiments. Afterripening or cold-moist stratification alone did not relieve a majority of the seeds from dormancy.

Since *E. latilobum* inhabits soil that is prone to slippage and erosion, we hypothesized that scarification may increase germination as compared to non-scarified seeds. Additionally, a large body of evidence demonstrates that an increase in the ratio of gibberellins to abscisic acid in a dormant seed is a mechanism for dormancy break(Bewley *et al.*, 2013; Finch-Savage & Leubner-Metzger, 2006; Hilhorst, 1995). Since treating seed with GA_3 has demonstrated favorable germination outcomes for horticultural and agronomic crops (Cornea-Cipcigan *et al.*, 2020; Dhillon *et al.*, 2021; Foley, 2016; Qureshi *et al.*, 2016), we applied four GA_3 (10, 100, 1000 ppm) concentrations to both scarified and non-scarified seeds. This experiment treated over 5,000 seeds in 60 separate trays. However, due to the logistics of applying the treatments, the number of seeds per tray ranged between 16 and 100, averaging 85 seeds.

This difference in the number of seeds per tray required the analysis to compare the proportions of germination for each treatment combination. The analysis did not find any concentration of GA_3 to have a significant effect on the proportion of germinated seeds, whereas scarification was found to increase the odds of germination by 45% (p-value < 0.05), suggesting that the species may require weathering or disturbance for dormancy release.

Seasonal changes in soil temperature and the hydration level of seeds have been identified as key factors influencing dormancy break (Baskin & Baskin, 2014; Batlla & Benech-Arnold, 2004; Benech-Arnold *et al.*, 2000). However, dormancy exists on a continuum where certain environmental conditions may induce dormancy (Batlla & Benech-Arnold, 2010, 2015; Kildisheva et al., 2020). Baskin & Baskin (2003) have offered a technique to identify a species' favored conditions without using thousands of seeds, called the "move-along experiment". This would require access to five controlled environments (i.e. growth chambers) for one year. Seeds are treated by either winter or summer progression diurnal temperatures, including four control temperatures. A total of 18 samples with 50 seeds per tray are recommended and allow for three replicates per treatment. Since seeds respond to specific environmental signals such as nitrate, ethylene, carbon dioxide, alternating temperature, and light (Finch-Savage & Footitt, 2012; Finch-Savage & Leubner-Metzger, 2006), it is possible to incorporate these variables in the move-along experiment as well (Baskin & Baskin, 2003).

Summary

In this study, dormancy-breaking treatments were tested for the propagation of *Eriophyllum latilobum* from seed. Nondeep physiological dormancy is typical for species in the family Asteraceae. Afterripening, cold-moist stratification, scarification, and gibberellic acid (GA_3) treatments have demonstrated efficacy in breaking nondeep PD. The effects of these treatments were measured in three separate experimental trials. Results from these experiments did not find a significant effect of afterripening, cold-moist stratification, or GA_3 treatments on seed germination. However, the effect of scarification was found to be significant, improving the odds of germination by 45% as compared to the non-scarified group (p-value < 0.05; 95% confidence interval of odds ratio is 1.26, 1.68). As the viability of a bulk sample of 200 treated clean seeds resulted in an 83.7% viability score, the results illustrate that scarification alone does not release all the seeds from dormancy. Other factors such as the effect of diurnal temperature on germination should be investigated for improved germination outcomes for *E. latilobum.*

Supplemental Research Bibliography

Baskin, C. C., & Baskin, J. M. (2003). When Breaking Seed Dormancy Is a Problem: Try a Move-along Experiment. *Native Plants Journal*, *4*(1), 17–21. https://doi.org/10.3368/npj.4.1.17

Baskin, Carol C., & Baskin, J. M. (2014). *Seeds : ecology, biogeography, and evolution of dormancy and germination*. Elsevier Science.

Baskin, Carol C., & Baskin, J. M. (2020). Breaking seed dormancy during dry storage: A useful tool or major problem for successful restoration via direct seeding? *Plants*, *9*(5). https://doi.org/10.3390/plants9050636

Baskin, J. M., & Baskin, C. C. (1985). The Annual Dormancy Cycle in Buried Weed Seeds: A Continuum. *BioScience*, *35*(8), 492–498. https://doi.org/10.2307/1309817

Batlla, D., & Benech-Arnold, R. L. (2004). A predictive model for dormancy loss in Polygonum aviculare L. seeds based on changes in population hydrotime parameters. *Seed Science Research*, *14*(3), 277–286. https://doi.org/10.1079/ssr2004177

Batlla, D., & Benech-Arnold, R. L. (2010). Predicting changes in dormancy level in natural seed soil banks. *Plant Molecular Biology*, *73*(1–2), 3–13. https://doi.org/10.1007/s11103-010-9601-z

Batlla, D., & Benech-Arnold, R. L. (2015). A framework for the interpretation of temperature effects on dormancy and germination in seed populations showing dormancy. *Seed Science Research*, *25*(2), 147–158. https://doi.org/10.1017/S0960258514000452

Benech-Arnold, R. L., Sánchez, R. A., Forcella, F., Kruk, B. C., & Ghersa, C. M. (2000). Environmental control of dormancy in weed seed banks in soil. *Field Crops Research*, *67*(2), 105–122. https://doi.org/10.1016/S0378-4290(00)00087-3

Bewley, J., Bradford, K., Hilhorst, H., & Nonogaki, H. (2013). *Seeds: Physiology of Development, Germination and Dormancy.* (Third). Springer.

Bhatla, S. C., & A. Lal, M. (2018). Plant Physiology, Development and Metabolism. In *Plant Physiology, Development and Metabolism*. Springer.

Commander, L. E., Rokich, D. P., Renton, M., Dixon, K. W., & Merritt, D. J. (2013). Optimising seed broadcasting and greenstock planting for restoration in the Australian arid zone. *Journal of Arid Environments*, *88*, 226–235. https://doi.org/10.1016/j.jaridenv.2012.08.012

Cornea-Cipcigan, M., Pamfil, D., Sisea, C. R., & Mărgăoan, R. (2020). Gibberellic acid can improve seed germination and ornamental quality of selected cyclamen species grown under short and long days. *Agronomy*, *10*(4). https://doi.org/10.3390/agronomy10040516

Dhillon, B. S., Kumar, V., Sagwal, P., Kaur, N., Mangat, G. S., & Singh, S. (2021). Seed Priming with Potassium Nitrate and Gibberellic Acid Enhances the Performance of Dry Direct Seeded Rice (Oryza sativa L.) in North-Western India. *Agronomy*, *11*(5).

Elzenga, J. T. M., Bekker, R. M., & Pritchard, H. W. (2019). Maximising the use of native seeds in restoration projects. *Plant Biology*, *21*(3), 377–379. https://doi.org/10.1111/plb.12984

Favier, J. F. (1995). A Model for Germination Rate during Dormancy Loss in Hordeum vulgare.

Annals of Botany, *76*(6), 631–638. https://doi.org/10.1006/anbo.1995.1141

Finch-Savage, W. E., & Footitt, S. (2012). To germinate or not to germinate: A question of dormancy relief not germination stimulation. *Seed Science Research*, *22*(4), 243–248. https://doi.org/10.1017/S0960258512000165

Finch-Savage, W. E., & Leubner-Metzger, G. (2006). Seed dormancy and the control of germination. *New Phytologist*, *171*(3), 501–523. https://doi.org/10.1111/j.1469-8137.2006.01787.x

Foley, M. E. (2016). Germination of leafy spurge (Euphorbia esula L.) seeds under alternating temperatures: The effects of amplitude, midpoint, and imbibition in gibberellic acid. *Canadian Journal of Plant Science*, *96*(3), 321–328. https://doi.org/10.1139/cjps-2015-0269

Harrington, J. (1972). Seed storage and longevity. In T. T. Kozlowski (Ed.), *Seed Biology Volume III Insects, and Seed Collection, Storage, Testing, and Certification.* (pp. 145–245).

Hilhorst, H. W. M. (1995). A critical update on seed dormancy. I. Primary dormancy. *Seed Science Research*, *5*(2), 61–73. https://doi.org/10.1017/S0960258500002634

James, J. J., Sheley, R. L., Erickson, T., Rollins, K. S., Taylor, M. H., & Dixon, K. W. (2013). A systems approach to restoring degraded drylands. In *Journal of Applied Ecology* (Vol. 50, Issue 3, pp. 730–739). https://doi.org/10.1111/1365-2664.12090

James, J. J., Svejcar, T. J., & Rinella, M. J. (2011). Demographic processes limiting seedling recruitment in arid grassland restoration. *Journal of Applied Ecology*, *48*(4), 961–969. https://doi.org/10.1111/j.1365-2664.2011.02009.x

Kildisheva, O. A., Dixon, K. W., Silveira, F. A. O., Chapman, T., Di Sacco, A., Mondoni, A., Turner, S. R., & Cross, A. T. (2020). Dormancy and germination: making every seed count in restoration. *Restoration Ecology*, *28*(S3), S256–S265. https://doi.org/10.1111/rec.13140

Ladouceur, E., Jiménez-Alfaro, B., Marin, M., De Vitis, M., Abbandonato, H., Iannetta, P. P. M., Bonomi, C., & Pritchard, H. W. (2018). Native Seed Supply and the Restoration Species Pool. *Conservation Letters*, *11*(2), 1–9. https://doi.org/10.1111/conl.12381

Merritt, D. J., & Dixon, K. W. (2011). Restoration Seed Banks— A Matter of Scale. *Science*, *332*, 424–425. https://doi.org/10.1126/science.1203083

Nagel, R., Durka, W., Bossdorf, O., & Bucharova, A. (2019). Rapid evolution in native plants cultivated for ecological restoration: not a general pattern. *Plant Biology*, *21*(3), 551–558. https://doi.org/10.1111/plb.12901

Nomad Ecology. (2019). *Botanical Resources Survey Report. Peninsula Watershed. San Mateo County, California [Draft]*.

Oldfield, S. (2019). The US National Seed Strategy for Rehabilitation and Restoration: progress and prospects. *Plant Biology*, *21*(3), 380–382. https://doi.org/10.1111/plb.12851

Palmerlee, A. P., & Young, T. P. (2010). Direct seeding is more cost effective than container stock across ten woody species in California. *Native Plants Journal*, *11*(2), 89–102. https://doi.org/10.2979/npj.2010.11.2.89

Qureshi, U. S., Shakeel, M., Qureshi, K. M., Saleem, A., Mir, A. R., & Qureshi, A. A. (2016). Effect of seed priming through gibberellic acid (GA3) and imidacloprid on bismarckia palm seed. *International Journal of Biosciences (IJB)*, *8*(4), 18–27. https://doi.org/10.12692/ijb/8.4.18-27

Soltani, E., Baskin, C. C., & Baskin, J. M. (2017). A graphical method for identifying the six types of non-deep physiological dormancy in seeds. *Plant Biology*, *19*(5), 673–682. https://doi.org/10.1111/plb.12590

U.S. Fish and Wildlife Service. (1998). *Recovery plan for the serpentine soil species of the San Francisco Bay Area*.

U.S. Fish and Wildlife Service. (2011). *Eriophyllum latilobum (San Mateo woolly sunflower). 5-Year Review : Summary and Evaluation*.

Appendix B

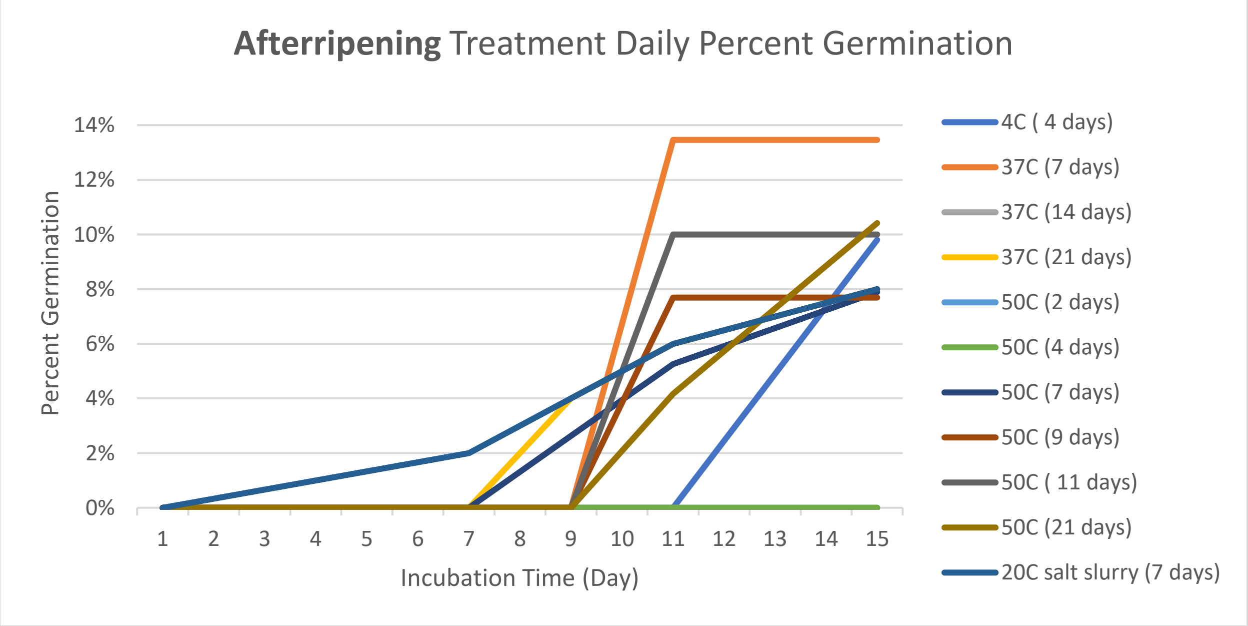
Table 1.

Afterripening Treatment Percent Germination (20°C)

		Incubation Day				
Treatment	*n*	Day 1	Day 7	Day 9	Day 11	Day 15
5°C (4 days)	50	0%	0%	0%	0%	10%
37°C (7 days)	52	0%	0%	0%	13%	13%
37°C (14 days)	43	0%	0%	0%	0%	0%
37°C (21 days)	50	0%	0%	4%	6%	8%
50°C (2 days)	51	0%	0%	0%	0%	0%
50°C (4 days)	10	0%	0%	0%	0%	0%
50°C (7 days)	11	0%	0%	3%	5%	8%
50°C (9 days)	38	0%	0%	0%	8%	8%
50°C (11 days)	13	0%	0%	0%	10%	10%
50°C (21 days)	10	0%	0%	0%	4%	10%
20°C + salt slurry (7 days)	48	0%	2%	4%	6%	8%

Note: Summary table of afterripening treatments, corresponding sample size, and percent germination outcomes per observation day (days 1, 7, 9, 11, and 15).

Figure 1.



Note: A line graph illustrating percent germination per tray for each afterripening treatment for observation days 7, 9, 11 and 15.

Table 2.

Cold-moist Stratification Treatment Percent Germination (15°C Incubation Temperature)

CMS Treatment	Incubation Day														
	n	1	2	3	4	5	6	7	8	9	10	11	12	13	14
5°C (8 Days)	20	0%	0%	0%	0%	0%	5%	5%	5%	5%	10%	10%	10%	10%	10%
5°C (12 Days)	20	0%	0%	5%	5%	5%	5%	5%	10%	10%	10%	10%	10%	10%	10%
5°C (15 Days)	18	0%	0%	6%	6%	6%	6%	6%	6%	6%	6%	6%	6%	6%	6%
5°C (20 Days)	19	5%	11%	11%	11%	11%	16%	16%	16%	16%	16%	16%	16%	16%	16%
5°C (24 Days)	17	6%	12%	12%	12%	18%	18%	18%	18%	18%	18%	18%	18%	18%	18%
5°C (36 Days)	19	0%	5%	5%	5%	5%	5%	5%	5%	5%	5%	5%	5%	5%	5%

Note: Summary table of cold-moist stratification treatments, corresponding sample size, and percent germination outcomes per observation day (days 1-14) for seed incubated at 15°C*).*

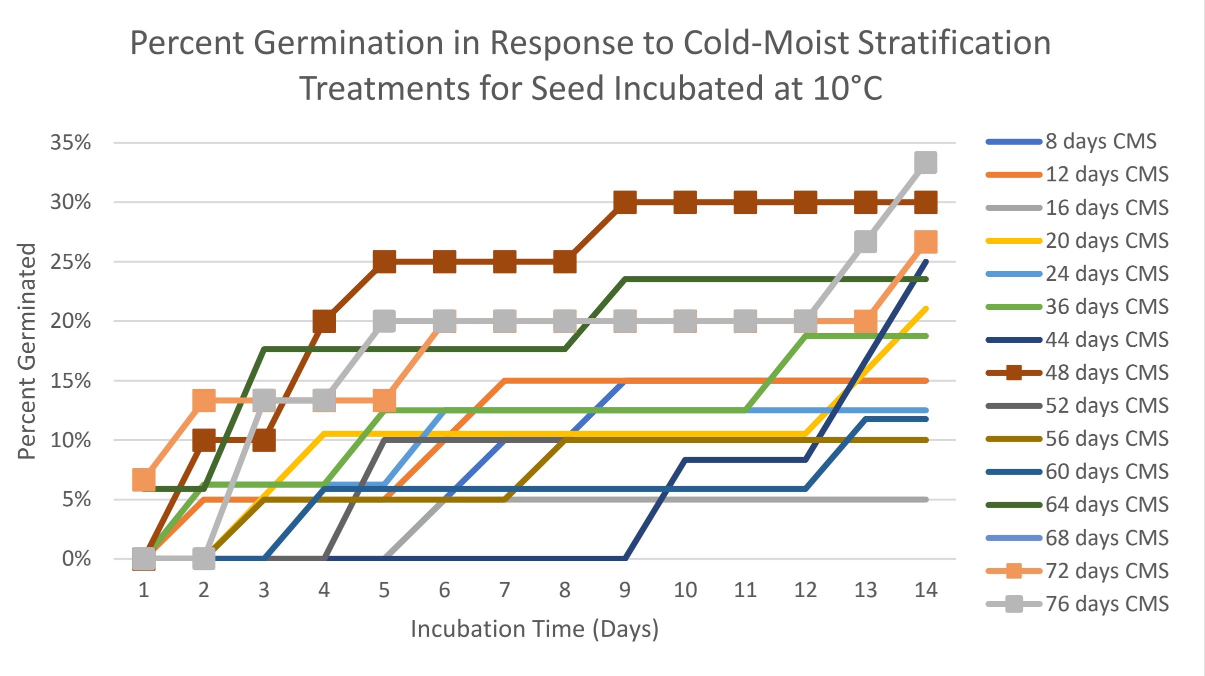
Table 3.

Cold-moist Stratification Treatment Percent Germination (10°C Incubation Temperature)

CMS Treatment	Incubation Day														
	n	1	2	3	4	5	6	7	8	9	10	11	12	13	14
5°C (8 Days)	20	0%	0%	0%	0%	0%	5%	10%	10%	15%	15%	15%	15%	15%	15%
5°C (12 Days)	20	0%	5%	5%	5%	5%	10%	15%	15%	15%	15%	15%	15%	15%	15%
5°C (15 Days)	18	0%	0%	0%	0%	0%	5%	5%	5%	5%	5%	5%	5%	5%	5%
5°C (20 Days)	19	0%	0%	5%	11%	11%	11%	11%	11%	11%	11%	11%	11%	16%	21%
5°C (24 Days)	16	0%	6%	6%	6%	6%	13%	13%	13%	13%	13%	13%	13%	13%	13%
5°C (36 Days)	16	0%	6%	6%	6%	13%	13%	13%	13%	13%	13%	13%	19%	19%	19%
5°C (44 Days)	12	0%	0%	0%	0%	0%	0%	0%	0%	0%	8%	8%	8%	17%	25%
5°C (48 Days)	20	0%	10%	10%	20%	25%	25%	25%	25%	30%	30%	30%	30%	30%	30%
5°C (52 Days)	20	0%	0%	0%	0%	10%	10%	10%	10%	10%	10%	10%	10%	10%	10%
5°C (56 Days)	20	0%	0%	5%	5%	5%	5%	5%	10%	10%	10%	10%	10%	10%	10%
5°C (60 Days)	17	0%	0%	0%	6%	6%	6%	6%	6%	6%	6%	6%	6%	12%	12%
5°C (64 Days)	17	6%	6%	18%	18%	18%	18%	18%	18%	24%	24%	24%	24%	24%	24%
5°C (68 Days)	17	6%	6%	6%	6%	6%	6%	6%	6%	6%	6%	12%	12%	12%	12%
5°C (72 Days)	15	7%	13%	13%	13%	13%	20%	20%	20%	20%	20%	20%	20%	20%	27%
5°C (76 Days)	15	0%	0%	13%	13%	20%	20%	20%	20%	20%	20%	20%	20%	27%	33%

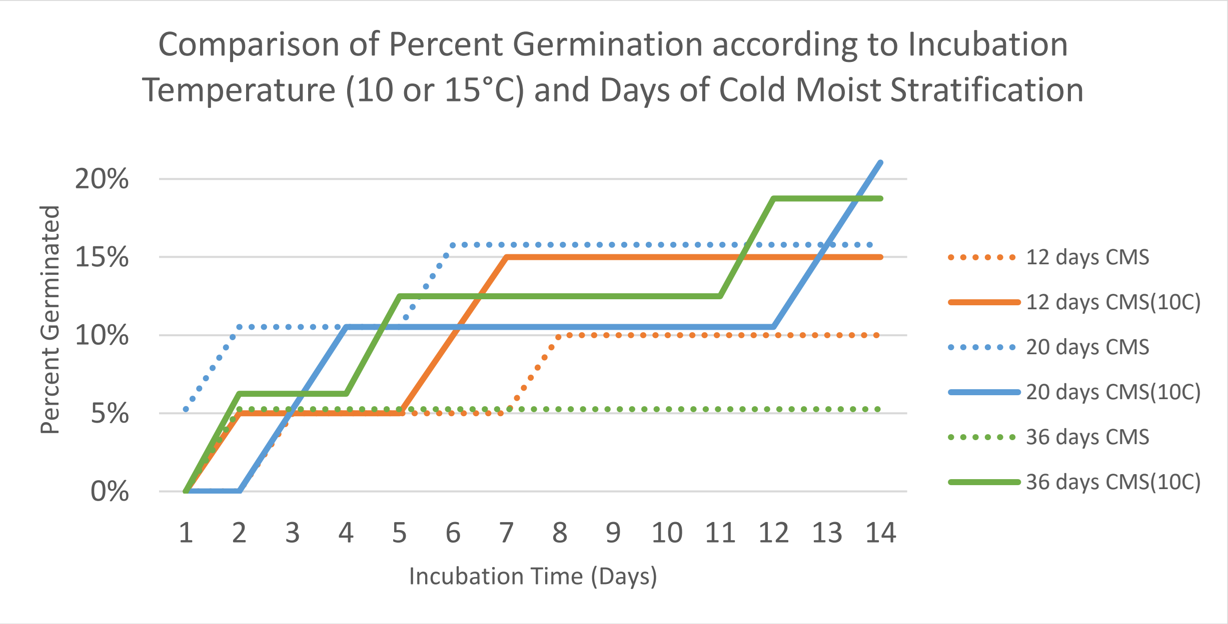
Note: Summary table of cold-moist stratification treatments, corresponding sample size, and percent germination outcomes per incubation day (days 1-14) for seed incubated at 10°C*).*

Figure 2.



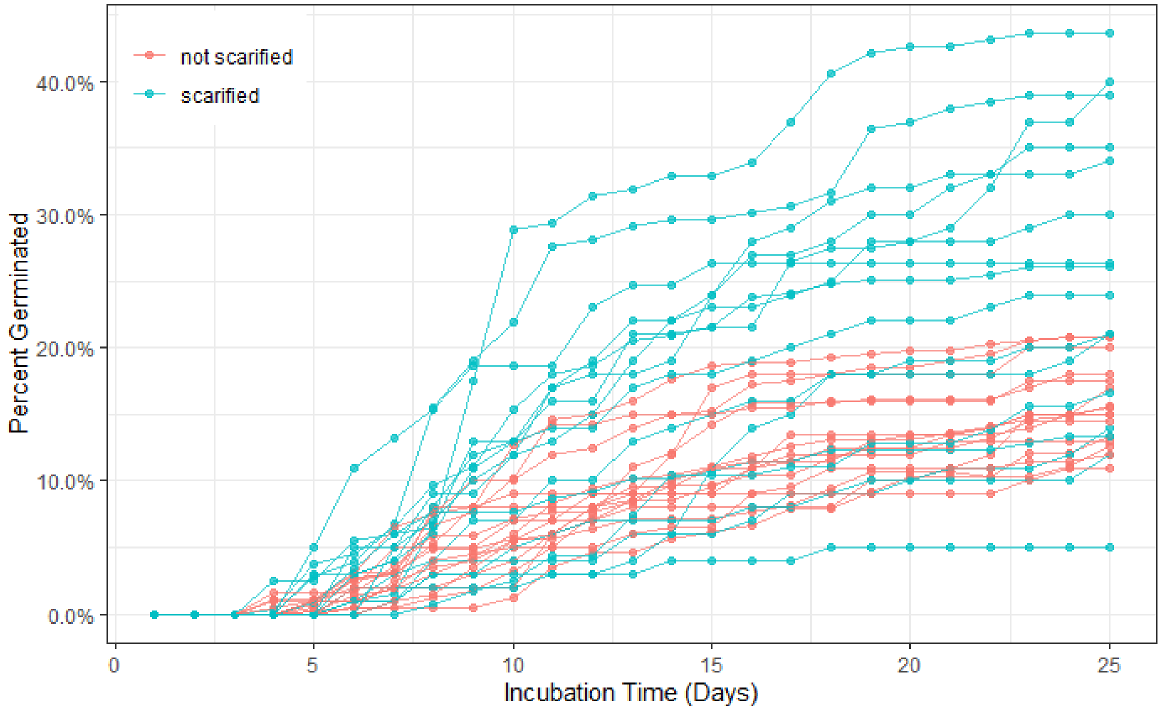
Note: A line graph illustrating percent germination per tray for each cold-moist stratification treatment incubated at 10°C. Percent germination was recorded daily for fourteen days of incubation.

Figure 3.



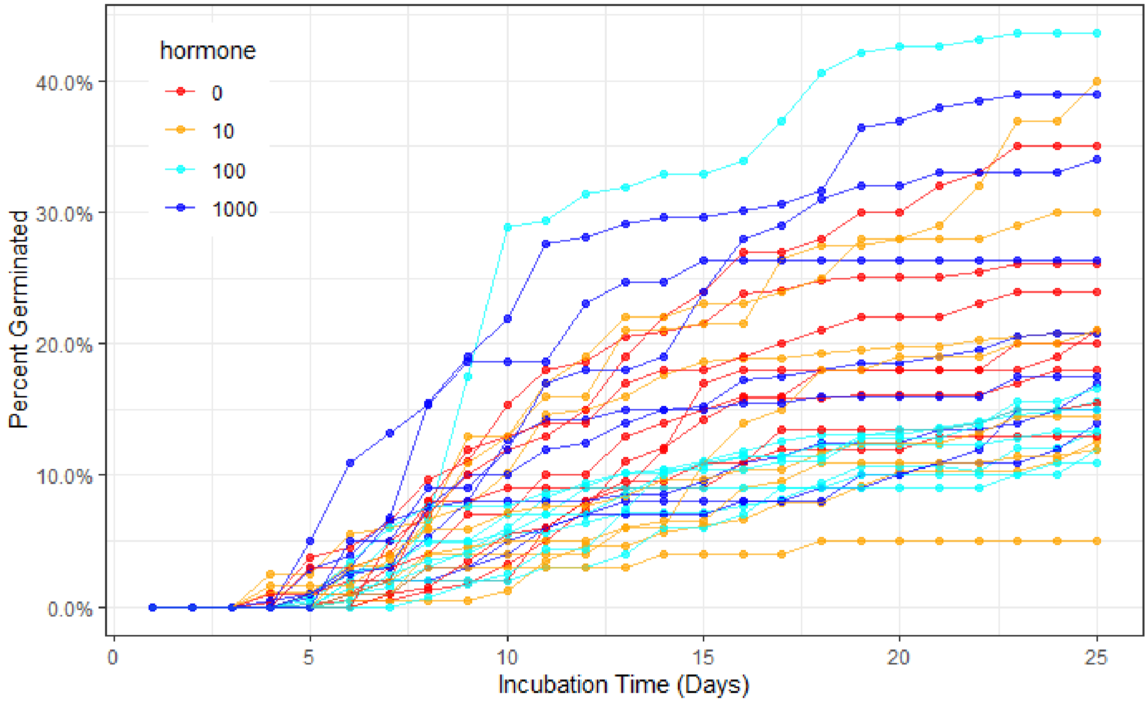
Note: A line graph illustrating differences in percent germination per tray incubated at 10°C and *15°C. Percent germination was recorded daily for fourteen days of incubation. Only cold-moist stratification treatment days 12, 20 and 26 are compared. Percent germination at 10°C is marginally greater.*

Figure 4.



Note: A line graph illustrating percent germination per tray for scarified and non-scarified groups.

Figure 5.



Note: A line graph illustrating percent germination per tray treated with 0, 10, 100, or 1000 ppm GA$_3$.

Table 4.

Scarification and GA_3 Treatment Daily Germination Frequencies per Incubation Day

				Incubation Day									
Occurrence	Scarified	Hormone	*n*	1	2	3	4	5	6	7	8	9	10
1	0	0	100	0	0	0	0	0	0	0	1	3	5
1	0	0	100	0	0	0	0	0	0	2	2	4	6
1	0	10	100	0	0	0	0	0	0	1	4	4	5
1	0	10	65	0	0	0	2	2	2	4	5	5	6
1	0	100	100	0	0	0	0	0	1	3	3	3	5
1	0	100	100	0	0	0	0	0	0	2	4	5	6
1	0	1000	100	0	0	0	0	2	2	2	8	10	10
1	0	1000	100	0	0	0	0	0	3	4	7	10	10
1	1	0	100	0	0	0	0	0	0	5	7	10	12
1	1	10	100	0	0	0	0	0	3	4	6	11	13
1	1	100	68	0	0	0	0	0	0	0	1	1	2
1	1	100	100	0	0	0	0	0	0	0	0	2	2
1	1	1000	100	0	0	0	0	0	5	5	9	9	12
3	0	0	100	0	0	0	1	1	1	2	2	2	2
3	0	10	100	0	0	0	0	0	1	1	1	1	1
3	0	10	69	0	0	0	0	0	0	0	0	0	1
3	0	100	100	0	0	0	0	1	2	3	5	5	5
3	0	100	64	0	0	0	0	0	2	2	3	3	4
3	0	1000	100	0	0	0	0	0	1	2	2	3	4
3	1	0	100	0	0	0	0	0	1	3	4	7	7
3	1	10	100	0	0	0	0	0	0	1	4	4	4
3	1	100	100	0	0	0	0	0	2	2	2	2	2
3	1	1000	100	0	0	0	0	0	1	1	3	3	5
4	0	0	100	0	0	0	0	1	2	2	8	8	9
4	0	10	100	0	0	0	0	1	1	3	5	6	7
4	0	10	100	0	0	0	0	1	1	1	3	3	3
4	0	100	100	0	0	0	0	0	2	5	5	5	7
4	0	1000	100	0	0	0	1	1	2	3	4	4	4
4	0	1000	100	0	0	0	0	1	4	10	11	12	12
4	1	0	100	0	0	0	0	3	3	4	8	12	13
4	1	10	100	0	0	0	0	0	1	1	3	3	3
4	1	100	100	0	0	0	0	0	0	2	2	2	2

Note: Summary table of scarification and GA_3 treatments, corresponding sample size, and daily germination frequencies per incubation day (days 1-10).

Table 4 continued.

Scarification and GA_3 Treatment Daily Germination Frequencies per Incubation Day

				Incubation Day									
Occurrence	Scarified	Hormone	*n*	1	2	3	4	5	6	7	8	9	10
4	1	100	60	0	0	0	0	1	4	6	8	8	8
4	1	1000	30	0	0	0	0	3	6	7	8	10	10
4	1	1000	100	0	0	0	0	0	2	3	4	4	4
5	0	0	27	0	0	0	0	0	0	0	0	0	0
5	0	10	24	0	0	0	1	1	2	2	4	4	5
5	0	100	24	0	0	0	0	0	1	1	1	1	1
5	0	1000	16	0	0	0	0	0	0	0	0	1	3
5	1	0	27	0	0	0	0	2	2	2	3	3	6
5	1	10	20	0	0	0	1	1	2	2	2	4	4
5	1	100	16	0	0	0	0	0	0	0	1	4	7
5	1	1000	21	0	0	0	0	1	1	2	4	4	5
6	0	0	100	0	0	0	0	1	2	2	3	4	5
6	0	0	100	0	0	0	0	0	0	0	0	0	3
6	0	0	100	0	0	0	0	0	0	0	2	3	5
6	0	10	100	0	0	0	0	0	1	1	2	2	3
6	0	10	100	0	0	0	0	0	2	2	5	5	7
6	0	10	100	0	0	0	0	0	1	1	3	7	10
6	0	100	100	0	0	0	0	0	0	1	2	2	5
6	0	100	100	0	0	0	0	1	1	2	2	4	6
6	0	100	100	0	0	0	0	0	0	0	4	7	9
6	0	1000	100	0	0	0	0	2	2	3	5	6	9
6	0	1000	100	0	0	0	0	1	6	6	11	14	16
6	0	1000	100	0	0	0	0	0	3	3	5	6	7
6	1	0	100	0	0	0	1	3	3	6	9	10	10
6	1	0	100	0	0	0	0	1	3	6	9	12	14
6	1	10	100	0	0	0	0	0	1	2	3	6	6
6	1	100	100	0	0	0	0	0	2	3	7	10	14
6	1	1000	100	0	0	0	0	1	3	4	12	19	20

Note: Summary table of scarification and GA_3 treatments, corresponding sample size, and daily germination frequencies per incubation day (days 1-10) (continued). Values 1 in the scarified field indicate scarification, values 0 indicate non-scarified. Values 0, 10, 100, and 1000 in the hormone field indicate parts per million of GA_3.

Table 4 continued.

Scarification and GA_3 Treatment Daily Germination Frequencies per Incubation Day

				Incubation Day									
Occurrence	Scarified	Hormone	*n*	11	12	13	14	15	16	17	18	19	20
1	0	0	100	6	8	9	9	10	10	15	15	15	15
1	0	0	100	6	8	10	10	12	12	12	12	12	12
1	0	10	100	6	6	6	7	7	7	7	8	8	8
1	0	10	65	6	6	7	8	8	9	9	10	11	11
1	0	100	100	6	6	8	9	9	10	10	10	12	13
1	0	100	100	8	8	9	11	13	13	13	13	14	14
1	0	1000	100	12	12	13	14	14	14	14	15	15	15
1	0	1000	100	12	13	15	16	16	17	17	17	17	17
1	1	0	100	13	15	19	22	24	27	27	28	30	30
1	1	10	100	17	19	22	22	23	23	24	25	28	28
1	1	100	68	4	4	6	8	8	8	9	9	10	10
1	1	100	100	3	3	6	9	9	9	9	9	11	11
1	1	1000	100	17	18	18	19	24	28	29	31	32	32
3	0	0	100	6	8	9	12	17	18	18	18	18	18
3	0	10	100	4	5	5	7	8	9	10	10	11	12
3	0	10	69	2	3	3	3	3	3	4	4	5	6
3	0	100	100	5	5	5	5	5	6	7	8	9	9
3	0	100	64	4	5	6	6	6	6	6	7	8	8
3	0	1000	100	6	7	8	8	8	8	8	8	10	10
3	1	0	100	10	10	13	14	15	16	16	18	18	18
3	1	10	100	4	4	6	6	11	14	15	18	18	19
3	1	100	100	3	3	4	6	6	7	9	9	10	10
3	1	1000	100	6	7	7	7	7	8	8	9	9	10
4	0	0	100	9	9	9	9	9	11	12	12	12	12
4	0	10	100	7	7	8	9	9	12	13	15	15	15
4	0	10	100	3	3	4	4	4	6	6	7	7	7
4	0	100	100	7	9	9	9	9	9	9	9	9	9
4	0	1000	100	4	4	4	4	5	7	8	9	9	9
4	0	1000	100	12	12	13	13	14	15	15	16	16	16
4	1	0	100	14	14	17	18	18	19	20	21	22	22
4	1	10	100	3	3	3	4	4	4	4	5	5	5
4	1	100	100	4	5	7	7	8	8	8	8	8	8

Note: Summary table of scarification and GA_3 treatments, corresponding sample size, and daily germination frequencies per incubation day (days 11-20). Values 1 in the scarified field indicate scarification, values 0 indicate non-scarified. Values 0, 10, 100, and 1000 in the hormone field indicate parts per million of GA_3.

Table 4 continued.

Scarification and GA$_3$ Treatment Daily Germination Frequencies per Incubation Day

				Incubation Day									
Occurrence	Scarified	Hormone	*n*	11	12	13	14	15	16	17	18	19	20
4	1	100	60	8	8	8	8	8	9	9	10	10	10
4	1	1000	30	10	12	13	13	14	14	14	14	14	14
4	1	1000	100	4	6	6	6	6	6	6	6	6	6
5	0	0	27	0	1	3	3	4	5	5	5	5	5
5	0	10	24	7	7	7	8	8	8	8	8	8	8
5	0	100	24	2	3	3	3	3	3	3	3	3	3
5	0	1000	16	3	3	3	3	3	4	4	4	4	4
5	1	0	27	7	7	8	8	8	9	9	9	9	9
5	1	10	20	5	5	7	7	7	7	9	9	9	9
5	1	100	16	7	7	7	7	7	7	8	9	9	9
5	1	1000	21	7	7	7	7	7	7	7	7	9	9
6	0	0	100	7	12	14	16	19	20	20	20	21	21
6	0	0	100	5	6	10	11	11	13	13	13	13	13
6	0	0	100	8	8	9	10	12	12	12	12	12	12
6	0	10	100	3	5	5	5	6	6	6	7	7	8
6	0	10	100	10	10	10	12	12	13	13	13	13	13
6	0	10	100	16	16	20	20	23	23	23	24	25	25
6	0	100	100	6	6	7	7	7	8	10	11	11	11
6	0	100	100	8	8	10	10	11	12	12	12	12	12
6	0	100	100	11	11	11	12	14	15	16	17	17	17
6	0	1000	100	10	10	11	11	11	12	13	13	15	15
6	0	1000	100	19	19	21	21	21	22	22	23	23	23
6	0	1000	100	9	9	9	9	10	10	10	11	11	11
6	1	0	100	12	12	12	12	14	16	16	17	18	18
6	1	0	100	16	18	20	21	21	22	23	24	24	24
6	1	10	100	7	7	7	7	8	8	8	10	10	11
6	1	100	100	15	19	20	22	22	24	24	25	28	29
6	1	1000	100	22	23	25	26	26	27	28	30	30	31

Note: Summary table of scarification and GA$_3$ treatments, corresponding sample size, and daily germination frequencies per incubation day (days 11-20)(continued). Values 1 in the scarified field indicate scarification, values 0 indicate non-scarified. Values 0, 10, 100, and 1000 in the hormone field indicate parts per million of GA$_3$.

Table 4 continued.

Scarification and GA_3 Treatment Daily Germination Frequencies per Incubation Day

				Incubation Day				
Occurrence	Scarified	Hormone	*n*	21	22	23	24	25
1	0	0	100	15	16	17	17	18
1	0	0	100	12	12	13	13	13
1	0	10	100	8	8	9	9	9
1	0	10	65	11	12	13	13	13
1	0	100	100	13	13	13	13	13
1	0	100	100	14	15	17	17	17
1	0	1000	100	15	15	16	16	16
1	0	1000	100	17	17	19	19	19
1	1	0	100	32	33	35	35	35
1	1	10	100	28	28	29	30	30
1	1	100	68	10	10	11	11	11
1	1	100	100	11	13	15	15	17
1	1	1000	100	33	33	33	33	34
3	0	0	100	18	18	20	20	20
3	0	10	100	12	12	12	12	12
3	0	10	69	6	6	6	7	9
3	0	100	100	9	9	10	10	11
3	0	100	64	8	8	9	9	10
3	0	1000	100	11	12	15	15	17
3	1	0	100	18	18	18	19	21
3	1	10	100	19	19	20	20	21
3	1	100	100	10	10	10	10	12
3	1	1000	100	11	11	11	12	14
4	0	0	100	13	13	13	13	13
4	0	10	100	15	15	16	16	16
4	0	10	100	7	7	7	7	8
4	0	100	100	9	9	10	11	11
4	0	1000	100	9	9	9	10	11
4	0	1000	100	18	18	19	19	19
4	1	0	100	22	23	24	24	24
4	1	10	100	5	5	5	5	5
4	1	100	100	8	8	9	10	10

Note: Summary table of scarification and GA_3 treatments, corresponding sample size, and daily germination frequencies per incubation day (days 21-25). Values 1 in the scarified field indicate scarification, values 0 indicate non-scarified. Values 0, 10, 100, and 1000 in the hormone field indicate parts per million of GA_3.

Table 6 end.

Scarification and GA_3 Treatment Daily Germination Frequencies per Incubation Day

				Incubation Day				
Occurrence	Scarified	Hormone	*n*	21	22	23	24	25
4	1	100	60	10	10	10	10	10
4	1	1000	30	14	14	14	14	14
4	1	1000	100	6	6	6	6	6
5	0	0	27	5	5	6	7	7
5	0	10	24	8	8	8	8	8
5	0	100	24	3	3	3	3	3
5	0	1000	16	4	4	4	4	4
5	1	0	27	9	9	9	9	9
5	1	10	20	9	10	12	12	13
5	1	100	16	9	9	9	9	9
5	1	1000	21	9	9	9	9	9
6	0	0	100	21	21	21	21	21
6	0	0	100	13	13	13	13	13
6	0	0	100	12	12	12	12	12
6	0	10	100	8	9	10	10	10
6	0	10	100	13	13	13	14	14
6	0	10	100	25	26	26	26	26
6	0	100	100	11	12	12	12	12
6	0	100	100	12	12	12	12	13
6	0	100	100	19	20	22	23	25
6	0	1000	100	16	17	18	18	18
6	0	1000	100	23	23	24	25	25
6	0	1000	100	12	13	15	15	15
6	1	0	100	18	19	21	21	21
6	1	0	100	24	24	24	24	24
6	1	10	100	13	14	14	14	15
6	1	100	100	29	30	31	31	31
6	1	1000	100	33	34	35	35	35

Note: Summary table of scarification and GA_3 treatments, corresponding sample size, and daily germination frequencies per incubation day (days 21-25)(continued). Values 1 in the scarified field indicate scarification, values 0 indicate non-scarified. Values 0, 10, 100, and 1000 in the hormone field indicate parts per million of GA_3.