Management Strategic Plan
Seed Collection, Banking, and Bulking Plan
for
Conserved Lands in Western San Diego County

Prepared for

San Diego Association of Governments

Prepared by

Conservation Biology Institute and AECOM

in collaboration with

San Diego Management and Monitoring Program

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EXECUTIVE SUMMARY

The Conservation Biology Institute (CBI) and AECOM Technical Services, Inc. (AECOM) worked with the San Diego Management and Monitoring Program (SDMMP) and other regional partners to prepare a Management Strategic Plan (MSP) Seed Collection, Banking, and Bulking Plan (SCBBP) for MSP rare plants in the Management Strategic Planning Area in San Diego County, California. The Rare Plant Management Group Steering Committee guided development of the plan, while species Working Groups provided technical expertise (Appendix A). The plan was funded by the San Diego Association of Governments (SANDAG). The SCBBP is a living document that will be updated over time.

The SCBBP provides a strategic approach to managing seed resources for MSP rare plants on conserved lands in western San Diego County. This document does not replace existing NCCP obligations or requirements, and recommendations in the plan are advisory and meant to be implemented voluntarily if land owners and managers so desire. Recommendations are consistent with the intent of regional NCCP plans. The SCBBP fulfills an objective in the regional *Management and Monitoring Strategic Plan for Conserved Lands in Western San Diego County: A Strategic Habitat Conservation Roadmap* (MSP Roadmap), provides guidelines to implement selected management actions in the MSP Framework Rare Plant Management Plan (F-RPMP) related to seed resources, and is informed by regional and preserve-specific monitoring data and studies.

The SCBBP includes a general section and species-specific sections or chapters. In the general section, we discuss (1) the relationship of this plan to the MSP Roadmap and other regional plans, (2) the overall approach to seed management in the region, and (3) key factors for managing seed of rare plants, including regional monitoring, research, seed collection, banking, and bulking priorities and strategies, Best Management Practices, and potential sources of funding for seed-related management. Guidelines or recommendations in the general section are widely applicable to all MSP rare plants.

The species-specific section includes chapters for the MSP rare plants (*MSP target plants*) that have been evaluated to date, as summarized in the table below:

Year Prepared	Scientific Name	Common Name
2019	*Acanthomintha ilicifolia*	San Diego thornmint
	Acmispon prostratus	Nuttall's acmispon
	Chloropyron maritimum ssp. *maritimum*	Salt marsh bird's-beak
	Deinandra conjugens	Otay tarplant
2020	*Chorizanthe orcuttiana*	Orcutt's spineflower
	Dudleya brevifolia	Short-leaved dudleya
	Monardella viminea	Willowy monardella

The species chapters summarize relevant information specific to each MSP target plant, and identify species-specific strategies, management actions, and BMPs for seed collection, banking, and bulking. The SDMMP intends to prepare chapters for additional MSP priority plants in the future.

ACKNOWLEDGEMENTS

The San Diego Rare Plant Management Group Steering Committee provided guidance on the structure and content of the Seed Collection, Banking, and Bulking Plan (SCBBP), while many biologists and land managers provided expertise through species-specific working groups, interviews, and rare plant data collection (per the San Diego Management and Monitoring Program's Inspect and Manage rare plant program). All of these efforts were critical to plan development. Refer to Appendix A for a list of all participants.

In addition, the following people prepared, reviewed and/or commented on the SCBBP, or provided key information on costing, seed collections, and/or Best Management Practices:

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Dr. Kristine Preston of the San Diego Management and Monitoring Program (SDMMP) provided overall direction and resources to develop the SCBBP, and also reviewed and commented on the plan. Emily Perkins and Annabelle Bernabe of SDMMP prepared rare plant data and graphics. Finally, the San Diego Association of Governments (SANDAG) funded the SCBBP.

PHOTO CREDITS

Photos by Conservation Biology Institute (CBI; Patricia Gordon-Reedy, Jessie Vinje) except as credited in document. Other contributors: Earth Discovery Institute (Mary Duffy, Cathy Chadwick, Colin Richard), California Botanic Garden (RSA; John MacDonald), Recon Native Plant Nursery, San Diego Zoo Global (Stacy Anderson), Santa Barbara Botanic Garden (SBBG; Heather Schneider), Cal Photos (Ron Vanderhoff).

Cover Photos (clockwise from upper left): *Acanthomintha ilicifolia* (Patricia Gordon-Reedy, CBI), *A. ilicifolia* seed (John MacDonald, RSA), *Deinandra conjugens* (Patricia Gordon-Reedy, CBI), *D. conjugens* seed (John MacDonald, RSA), *Chloropyron maritimum* ssp. *maritimum* seed (Heather Schneider, SBBG), *C. maritimum* ssp. *maritimum* (Patricia Gordon-Reedy, CBI), *Acmispon prostratus* seed (John MacDonald, RSA), *A. prostratus* (Patricia Gordon-Reedy, CBI).

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ABBREVIATIONS

BLM	Bureau of Land Management
BMP	Best Management Practice
CaPR	California Plant Rescue
CBG	California Botanic Garden (formerly Rancho Santa Ana Botanic Garden)
ChBG	Chicago Botanic Garden
CBI	Conservation Biology Institute
CDFW	California Department of Fish and Wildlife
CMSP	Connectivity Monitoring Specific Plan
CNDDB	California Natural Diversity Database
CNLM	Center for Natural Lands Management
CPC	Center for Plant Conservation
EMP	Environmental Mitigation Program
GPS	Global Positioning System
IMG	Inspect and Manage
IPSP	Invasive Plant Strategic Plan
MOM	Master Occurrence Matrix
MSBP	The Millennium Seed Bank Partnership
MSCP	San Diego Multiple Species Conservation Plan
MSP	Management Strategic Plan for Conserved Lands in Western San Diego County
MSP F-RPMP	MSP Framework-Rare Plant Management Plan for Conserved Lands in Western San Diego County
MSP priority plant	MSP rare plant in the Species Management Focus Group
MSP rare plant	Rare plant addressed in MSP Roadmap
MSP Roadmap	Management and Monitoring Strategic Plan for Conserved Lands in Western San Diego County: *A Strategic Habitat Conservation Roadmap*
MSP SCBBP	MSP Rare Plant Seed Collection, Banking, and Bulking Plan for Conserved Lands in Western San Diego County
MSP target plant	MSP rare and priority plant addressed in species chapters in the SCBBP
MSPA	MSP Roadmap Area
MU	Management Unit
NCCP	Natural Community Conservation Plan
NLGRP	National Laboratory for Genetic Resources Preservation
NW	Native West Nursery (formerly Recon Native Plant Nursery)
RBG Kew	Royal Botanic Garden Kew
RSA	Rancho Santa Ana Botanic Garden
SANDAG	San Diego Association of Governments

SBBG	Santa Barbara Botanic Garden
SDBG	San Diego Botanic Garden
SDMMP	San Diego Management and Monitoring Program
SDZG	San Diego Zoo Global
TNC	The Nature Conservancy
TZ	Tetrazolium Test
USDA	U.S. Department of Agriculture
USFWS	U.S. Fish and Wildlife Service
USGS	U.S. Geological Survey

1.0 INTRODUCTION

The Conservation Biology Institute (CBI) and AECOM Technical Services, Inc. (AECOM), in coordination with the San Diego Management and Monitoring Program (SDMMP) and other regional partners, developed a Management Strategic Plan (MSP) Rare Plant Seed Collection, Banking, and Bulking Plan (SCBBP) for conserved lands in western San Diego County. This plan was funded by the San Diego Association of Governments (SANDAG), and is a living document that will be updated over time.

The SCBBP fulfills an objective in the regional Management and Monitoring Strategic Plan for Conserved Lands in Western San Diego County: A Strategic Habitat Conservation Roadmap (MSP Roadmap, SDMMP and The Nature Conservancy [TNC] 2017) and an achievement milestone in the TransNet Environmental Mitigation Program (EMP) Regional Management and Monitoring FY 2019-2020 Work Plan (Strategic Goal 1.1).

The MSP Roadmap applies to conserved lands (excluding military lands) within the MSP Roadmap Area (MSPA; Figure 1-1). The MSPA is divided into 11 Management Units (MUs) to facilitate coordinated management (Figure 1-2). The SDMMP delineated MUs by geography, vegetation, and threats and stressors, and MU size varies significantly, with smaller MUs found near the coast and larger MUs found inland (SDMMP and TNC 2017).

The MSP Roadmap addresses 57 rare plant species (*MSP rare plants*) within the MSPA (Figure 1-3). All of these species are covered under one or more Natural Community Conservation Plans (NCCPs). The SDMMP placed the 57 MSP rare plants into two management groups depending on the potential level of management needed for their long-term persistence: the Species Management Focus Group (32 species) and the Vegetation Management Focus Group (25 species). Species in the former category will likely require specific management measures, while species in the latter category are expected to persist by managing the vegetation community (SDMMP and TNC 2017).

The 32 MSP rare plants in the Species Management Focus Group are priorities for monitoring and management (*MSP priority plants*). These species are further categorized by potential risk of loss of either the species or significant occurrences[1] from the MSPA.

In this document, we develop species-specific seed collection, banking, and bulking guidelines for the MSP priority plants *(MSP target plants*) that have been evaluated to date. Refer to Table 1-1 for a list of all 57 MSP rare plants, many of which will be included in this living document over time. Table 1-1 also indicates which of these species are MSP priority and/or MSP target plants. Table 1-2 defines management categories for Species and Vegetation Management Focus Groups.

[1] A rare plant occurrence is similar to a 'population' without regard to whether individuals interbreed. The SDMMP follows California Natural Diversity Database (CNDDB) guidelines on defining unique occurrences based on distance (SDMMP 2019).

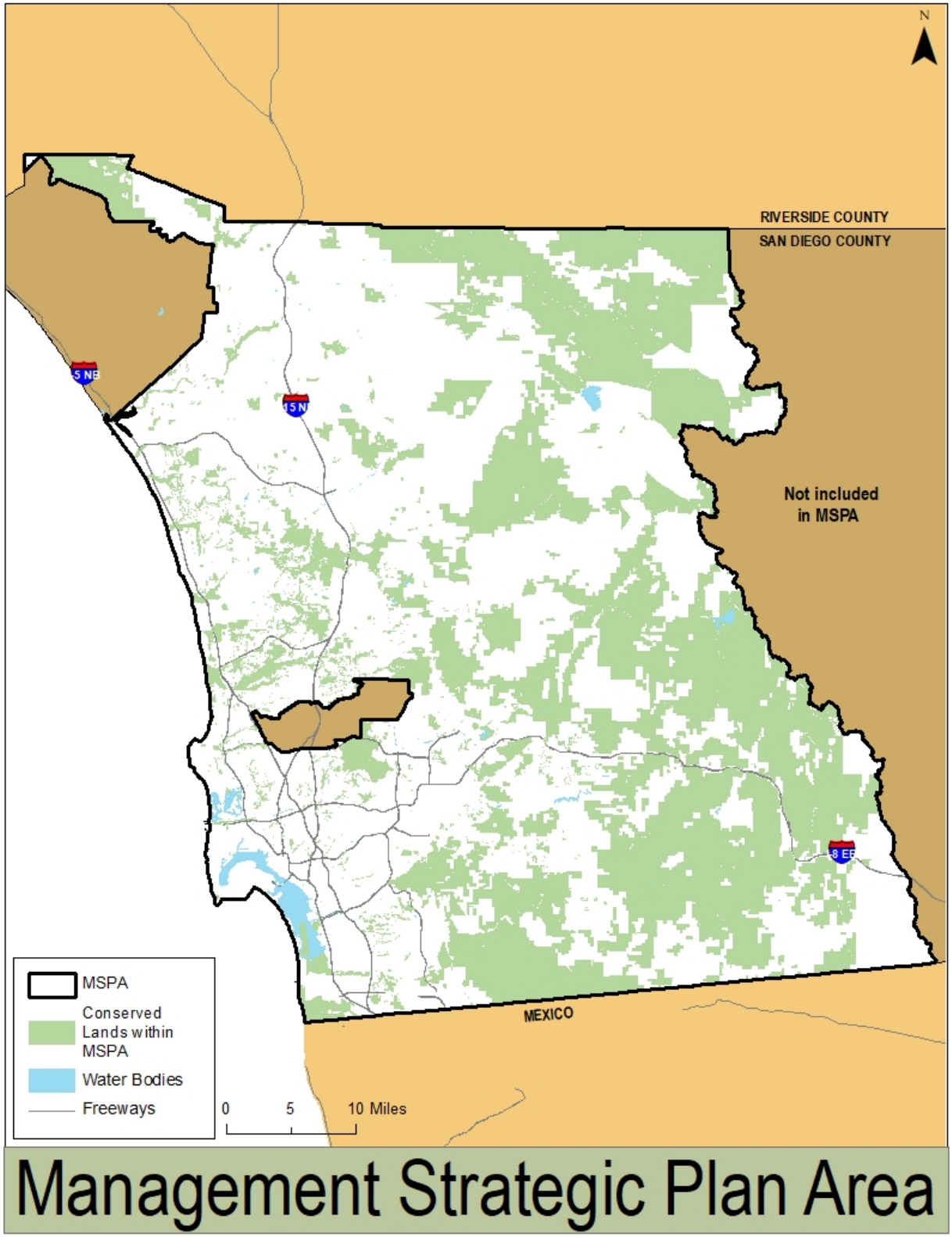

Figure 1-1. MSP Roadmap Area (MSPA) in Western San Diego County.

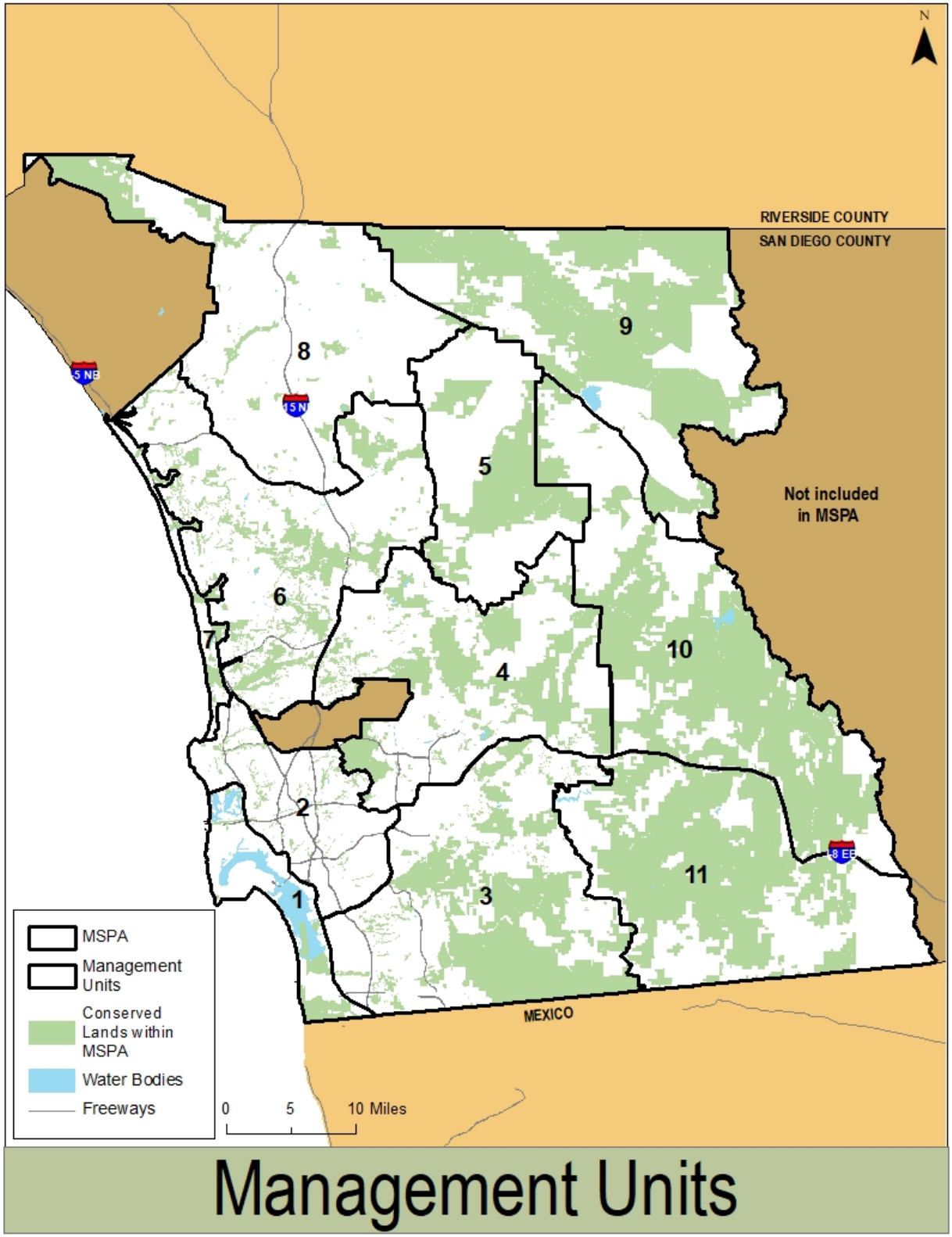

Figure 1-2. Management Units (MUs) within the MSPA.

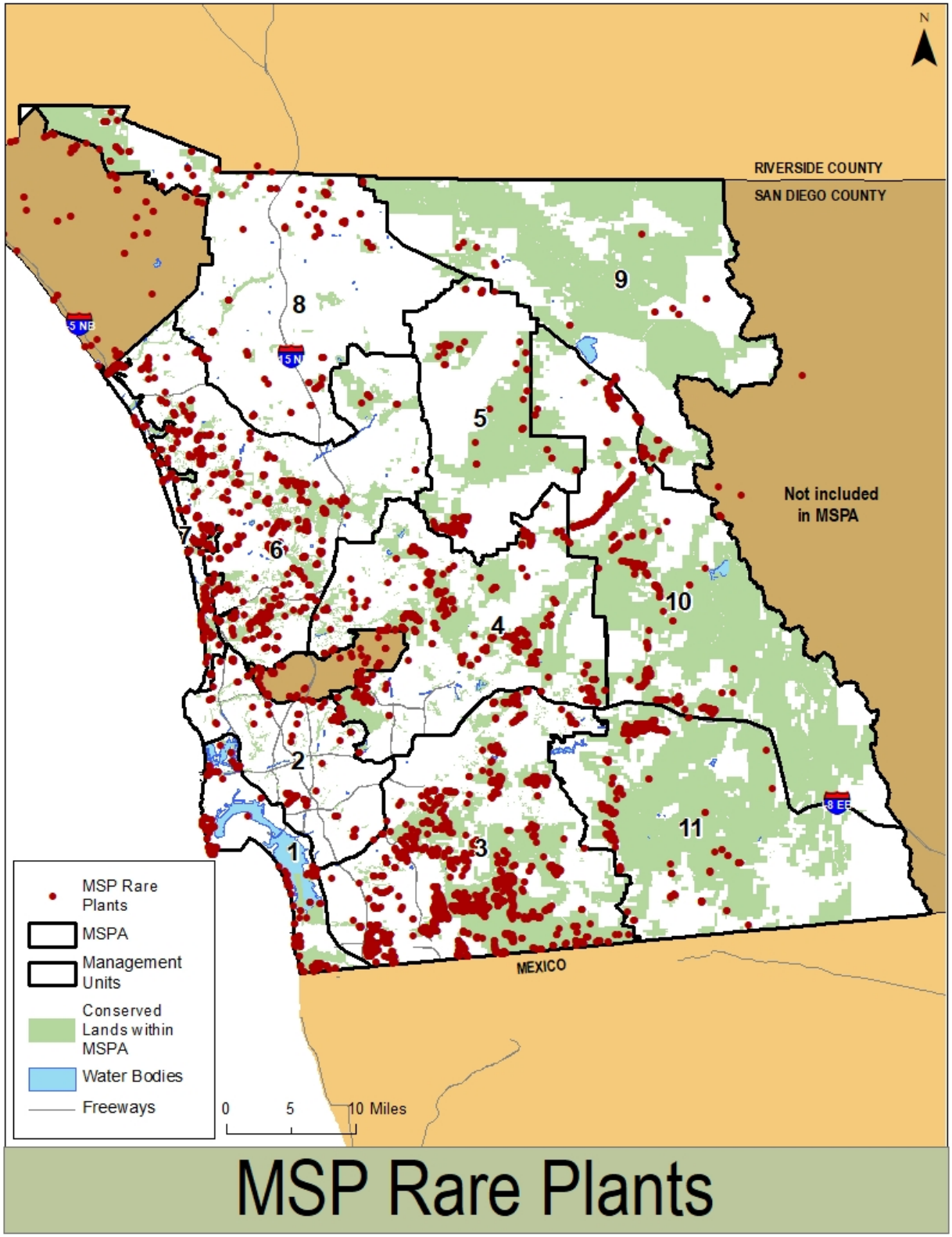

Figure 1-3. MSP Rare Plants Detected since 2000 within the MSPA.

Table 1-1. MSP Rare Plant Species.[1]

Scientific Name[2]	Common Name	Management Category[3]	MSP Rare Plants[4]	MSP Priority Plants[5]	MSP Target Plants[6]
Acanthomintha ilicifolia	San Diego thornmint	SO	✓	✓	✓
Acmispon prostratus	Nuttall's acmispon	SO	✓	✓	✓
Adolphia californica	California adolphia	VG	✓		
Agave shawii var. *shawii*	Shaw's agave	SL	✓	✓	
Ambrosia pumila	San Diego ambrosia	SO	✓	✓	
Aphanisma blitoides	Aphanisma	SL	✓	✓	
Arctostaphylos glandulosa ssp. *crassifolia*	Del Mar manzanita	VF	✓		
Arctostaphylos otayensis	Otay manzanita	VF	✓		
Arctostaphylos rainbowensis	Rainbow manzanita	VF	✓		
Atriplex coulteri	Coulter's saltbush	VF	✓		
Atriplex parishii	Parish brittlescale	VF	✓		
Baccharis vanessae	Encinitas baccharis	SO	✓	✓	
Bloomeria clevelandii	San Diego goldenstar	SS	✓	✓	
Brodiaea filifolia	Thread-leaved brodiaea	SS	✓	✓	
Brodiaea orcuttii	Orcutt's brodiaea	SO	✓	✓	
Brodiaea santarosae	Santa Rosa brodiaea	SS	✓	✓	
Calochortus dunnii	Dunn's mariposa lily	VG	✓		
Ceanothus cyaneus	Lakeside ceanothus	VF	✓		
Cenaothus verrucosus	Wart-stemmed ceanothus	VF	✓		
Centromadia parryi ssp. *australis*	Southern tarplant	VF	✓		
Chloropyron maritimum ssp. *maritimum*	Salt marsh bird's-beak	SL	✓	✓	✓
Chorizanthe orcuttiana	Orcutt's spineflower	SL	✓	✓	
Clinopodium chandleri	San Miguel savory	SL	✓	✓	
Comarostaphylis diversifolia ssp. *diversifolia*	Summer-holly	VG	✓		
Cylindropuntia californica var. *californica*	Snake cholla	VF	✓		
Deinandra conjugens	Otay tarplant	SS	✓	✓	✓
Dicranostegia orcuttiana	Orcutt's bird's-beak	SL	✓	✓	
Dudleya blochmaniae	Blochman's dudleya	SL	✓	✓	
Dudleya brevifolia	Short-leaved dudleya	SL	✓	✓	
Dudleya variegata	Variegated dudleya	SS	✓	✓	
Dudleya viscida	Sticky dudleya	SS	✓	✓	
Ericameria palmeri ssp. *palmeri*	Palmer's goldenbush	VF	✓		

Table 1-1. MSP Rare Plant Species.[1]

Scientific Name[2]	Common Name	Management Category[3]	MSP Rare Plants[4]	MSP Priority Plants[5]	MSP Target Plants[6]
Eryngium aristulatum var. *parishii*	San Diego button-celery	VF	✓		
Erysimum ammophilum	Coast wallflower	SL	✓	✓	
Euphorbia misera	Cliff spurge	VF	✓		
Ferocactus viridescens	San Diego barrel cactus	VF	✓		
Fremontodendron mexicanum	Mexican flannelbush	SL	✓	✓	
Hazardia orcuttii	Orcutt's hazardia	SL	✓	✓	
Hesperocyparis forbesii	Tecate cypress	VF	✓		
Iva hayesiana	San Diego marsh-elder	VG	✓		
Lepechinia cardiophylla	Heart-leaved pitcher sage	SL	✓	✓	
Lepechinia ganderi	Gander's pitcher sage	VG	✓		
Monardella hypoleuca ssp. *lanata*	Felt-leaved monardella	VF	✓		
Monardella stoneana	Jennifer's monardella	SL	✓	✓	
Monardella viminea	Willowy monardella	SL	✓	✓	
Navarretia fossalis	Spreading navarretia	VF	✓		
Nolina cismontana	Chaparral nolina	SL	✓	✓	
Nolina interrata	Dehesa nolina	SO	✓	✓	
*Orcuttia californica**	California Orcutt grass	SL	✓	✓	
Packera ganderi	Gander's ragwort	SO	✓	✓	
Pinus torreyana ssp. *torreyana*	Torrey pine	VF	✓		
Pogogyne abramsii	San Diego mesa mint	VF	✓		
*Pogogyne nudiuscula**	Otay mesa mint	SL	✓	✓	
Quercus dumosa	Nuttall's scrub oak	VF	✓		
Quercus engelmannii	Engelmann oak	VF	✓		
Rosa minutifolia	Small-leaved rose	SS	✓	✓	
Tetracoccus dioicus	Parry's tetracoccus	SS	✓	✓	

[1] MSP plant species as defined in the MSP Roadmap (SDMMP and TNC 2017).
[2] Plant species nomenclature generally follows Baldwin et al. 2012.
[3] Management Category: **SL** = at risk of loss from MSPA, **SO** = significant occurrences at risk of loss from MSPA, **SS** = stable and persistent, but require species-specific management; **VF** = limited distribution or require vegetation management, **VG** = may benefit from management for VF species. See Table 1-2 for full definitions.
[4] MSP rare plants = all plant species in the MSP Roadmap, which are covered under one or more NCCPs.
[5] MSP priority plants = all MSP rare plants in the Species Management Focus Group. MSP priority plants with an asterisk (*) are monitored per the Vernal Pool Management and Monitoring Plan (City of San Diego 2017) rather than the Inspect and Manage (IMG) program. All MSP priority plants are also MSP rare plants.
[6] MSP target plants = species included in the species chapters of this document. MSP target plants are also MSP rare and priority plants.

Table 1-2. Management Focus Groups and Categories.

Management Category[1]	Definition
Species Management Focus Group	
SL	Species at high risk of loss from MSP Roadmap Area (MSPA) without immediate management action above and beyond daily maintenance activities.
SO	Species with significant occurrence(s) at high risk of loss from MSPA without immediate management action above and beyond daily maintenance activities.
SS	Species with occurrences stable and persistence at lower risk than SL and SO species, but still require species-specific management actions.
Vegetation Management Focus Group	
VF	Species with limited distribution in the MSPA or needing specific vegetation characteristics requiring management.
VG	Species is not managed specifically, but may benefit from vegetation management for VF species.

[1] Focus group/management category designations and definitions per the MSP Roadmap (SDMMP and TNC 2017).

1.1 OVERVIEW

The SCBBP includes a general section and species-specific sections or chapters that provide the framework to manage seed of MSP rare plants within the MSPA. In the general section, we discuss (1) the relationship of this plan to the MSP Roadmap and other regional plans, (2) the overall approach to seed management in the region, and (3) key factors for managing seed of rare plants, including:

- Regional monitoring to inform management
- Seed-oriented research
- Seed collection, banking, and bulking priorities and strategies
- Best management practices (BMPs)
- Potential funding sources

Information in the general section is broadly applicable to all MSP rare plants, with a focus on MSP priority plants. Information in the species chapters is specific to the MSP target plants that have been prepared to date:

Year Prepared	Scientific Name	Common Name
2019	*Acanthomintha ilicifolia*	San Diego thornmint
	Acmispon prostratus	Nuttall's acmispon
	Chloropyron maritimum ssp. *maritimum*	Salt marsh bird's-beak
	Deinandra conjugens	Otay tarplant
2020	*Chorizanthe orcuttiana*	Orcutt's spineflower
	Dudleya brevifolia	Short-leaved dudleya
	Monardella viminea	Willowy monardella

The species chapters summarize relevant information specific to each MSP target plant, and identify species-specific strategies, management actions, and BMPs for seed collection, banking, and bulking. The SDMMP intends to prepare chapters for additional MSP priority plants in the future.

Guidelines in the SCBBP incorporate recommendations from the western San Diego County Regional Rare Plant Management Group Steering Committee (Rare Plant Management Group Steering Committee) and species-specific Working Groups, and from established seed collection programs, monitoring, management, restoration, and research and experimental studies, among others. Refer to Appendix A for a list of Rare Plant Management Group Steering Committee and Working Group participants and the reference section for sources used to develop the SCBBP.

1.2 Purpose and Need

The SCBBP provides a strategic approach to managing seed resources for MSP rare plants that (1) identifies and prioritizes rare plant species and occurrences requiring seed conservation and/or restoration, (2) directs seed management actions and funding where they are most needed or will be most effective, and (3) provides land managers with information to manage seed resources for their occurrences effectively.

Developing the SCBBP is possible because of the efforts of many individuals and institutions over the years, including land managers, biologists, botanists, researchers, ecologists, government and non-governmental entities, seed banks or seed storage facilities, commercial nurseries or growers, private organizations, and others that contribute or provide:

- Rare plant monitoring data
- Research or experimental studies and data
- BMPs for seed collection, banking, and bulking
- Support for seed-related activities (e.g., regional seed banking)
- Supplemental funding for seed management activities

1.3 Relationship to MSP Roadmap and other Regional Plans

There are a number of regional strategic plans or documents for western San Diego County that relate directly or indirectly to MSP rare plants. The MSP Roadmap (SDMMP and TNC 2017) is the overarching document that guides monitoring and management in the region and incorporates elements of many earlier plans. We summarize plans most relevant to this document below; refer to Table 1.3-1 for sources and links to these and other regional documents related to rare plant management.

Management Strategic Plan for Conserved Lands in Western San Diego County (MSP)

The MSP provides a comprehensive approach for managing multiple species within the region by establishing biological goals and measurable objectives to implement management actions (SDMMP 2013). The MSP categorizes and prioritizes species and vegetation communities, identifies geographic locations for management actions, provides specific timelines for implementation, and establishes a process for coordination and implementation. For MSP priority species, the document summarizes status, identifies management threats, develops management approaches, and outlines regional and MU goals and objectives.

The SCBBP updates general information in the MSP on seed biology and seed collection, banking, and bulking needs based on IMG monitoring data, research, or other studies.

Management and Monitoring Strategic Plan for Conserved Lands in Western San Diego County: A Strategic Habitat Conservation Roadmap (MSP Roadmap)

The MSP Roadmap expands on the 2013 MSP by including monitoring, adaptive management, additional species, vegetation communities, and threats derived, in part, from other planning documents in the region (e.g., Connectivity Monitoring Strategic Plan [SDMMP 2011], Invasive Plant Strategic Plan [CBI et al. 2012]). The MSP Roadmap also includes a Wildfire Element that addresses plant fire risk and management actions, as well as databases and mapping tools ("MSP Portal") that are available on the SDMMP interactive website: https://sdmmp.com/portal.php.

Preparing the SCBBP is an objective in the MSP Roadmap (MGT-PRP-SBPL).[2] The SCBBP addresses specific action items under this objective, including:

- Consult the Rare Plant Working Group Steering Committee and species-specific Working Groups for input and recommendations.

[2] MGT-PRP-SBPL indicates that this is a Management (MGT) objective to prepare (PRP) a seed banking plan (SBPL).

Table 1.3-1. Regional Plans or Documents Related to MSP Rare Plant Management in the MSPA.[1]

Regional Plan	Source[2]	Link(s) to Document or Relevant Sections
Management Strategic Plan for Conserved Lands in Western San Diego County (MSP)	SDMMP 2013	Volume 1: https://sdmmp.com/view_article.php?cid=CID_eperkins%40usgs.gov_588f7c6408184 Volume 2: https://sdmmp.com/view_article.php?cid=CID_eperkins%40usgs.gov_588f7ce9c0f68 Volume 3: https://sdmmp.com/view_article.php?cid=CID_eperkins%40usgs.gov_588f7d49c7d8b
Management and Monitoring Strategic Plan for Conserved Lands in Western San Diego County: *A Strategic Habitat Conservation Roadmap* (MSP Roadmap)	SDMMP and TNC 2017	Volume 2A: https://sdmmp.com/view_article.php?cid=CID_eperkins%40usgs.gov_590233783f742 Volume 2B: https://sdmmp.com/view_article.php?cid=CID_eperkins%40usgs.gov_59024838d1636 Volume 2C: https://sdmmp.com/view_article.php?cid=CID_eperkins%40usgs.gov_590233f2e2c53
Connectivity Monitoring Strategic Plan for the San Diego Preserve System (CMSP)	SDMMP 2011 SDMMP 2014	https://sdmmp.com/view_article.php?cid=CID_tedgarian%40usgs.gov_57acfb763b9ff https://sdmmp.com/view_article.php?cid=CID_tedgarian%40usgs.gov_57acf913214cc
Management Priorities for Invasive Non-native Plants: A Strategy for Regional Implementation, San Diego County, California (IPSP)	CBI et al. 2012	https://sdmmp.com/view_article.php?cid=CID_201604011922_38
Vernal Pool Management and Monitoring plan	City of San Diego 2017	https://www.sandiego.gov/sites/default/files/vp-mmp.pdf
Framework Management Plan: Guidelines for Best Practices with Examples of Effective Monitoring and Management	Lewison and Deutschman 2014	https://sdmmp.com/view_article.php?cid=CID_201604011922_110
Adaptive Management Framework for the Endangered San Diego Thornmint (*Acanthomintha ilicifolia*), San Diego County, California	CBI 2014a	https://sdmmp.com/view_article.php?cid=CiteID_1603251358356080
Otay Tarplant Management Vision	CBI 2012	https://databasin.org/groups/92c7bce8d88d43b3a800dd686195007e/ (see Supporting documents/South County grasslands/Project documents/OTP Goals and Objectives 10-29-12)

[1] Table includes only regional plans related to MSP priority plants, with a focus on the target plants covered by this document evaluated to date.
[2] Source: CBI = Conservation Biology Institute, SDMMP = San Diego Management and Monitoring Program, TNC = The Nature Conservancy. Refer to reference section for full citations.

- Prepare a SCBBP for MSP priority species with species-specific chapters that include guidelines and protocols for seed collection, testing, bulking, and storage.
- Prepare protocols and guidelines for collecting and submitting voucher specimens.
- Submit project metadata and the SCBBP to the MSP Web Portal.

San Diego Thornmint Adaptive Management Framework Plan

This regional framework plan reviewed status and threats, developed conceptual models for management, identified potential environmental correlates and opportunity areas for restoration, developed detailed goals and objectives, and compiled or developed seed banking and other BMPs and monitoring metrics for San Diego thornmint in San Diego County (CBI 2014).

The SDMMP incorporated key elements of this plan into the MSP Roadmap. The SCBBP will build on both the Adaptive Management Framework Plan and the MSP Roadmap by updating seed biology information, and refining BMPs for seed banking for San Diego thornmint.

Otay Tarplant Management Vision

CBI, in partnership with TNC and with input from other biologists and land managers, prepared a framework for coordinated management of Otay tarplant in MU 3 of the MSPA (CBI 2012). The Otay Tarplant Management Vision identified key areas to manage or restore Otay tarplant occurrences and to improve connectivity for pollinators, and developed both landscape-level and preserve-specific goals and objectives for this species, including guidelines for seed banking (e.g., seed collection, testing, storage, bulking, and outplanting).

The SDMMP incorporated elements of the Management Vision into both the MSP and MSP Roadmap. The SCBBP will build on both the Management Vision and the MSP Roadmap by updating seed biology information, and refining BMPs for seed banking for Otay tarplant.

1.4 Relationship to Framework Rare Plant Management Plan

The MSP Roadmap includes objectives to develop two closely related framework plans for rare plants: a MSP Framework Rare Plant Management Plan for Conserved Lands in Western San Diego County (F-RPMP) and the SCBBP. The F-RPMP identifies priorities, locations, and actions to manage rare plant occurrences, while the SCBBP provides guidelines to implement selected management actions related to seed resources. For example, where the F-RPMP calls for restoring occurrences of a target species, the SCBBP details seed collecting, banking, and bulking practices to maximize both genetic diversity and restoration success.

1.5 Relationship to Preserve Management

The SCBBP provides guidelines for managing seed resources for MSP rare plants on conserved lands in western San Diego County. This document does not replace existing NCCP obligations or requirements. Further, recommendations in this plan are advisory and not required. Rather, they are to be implemented voluntarily if land owners and managers so desire. Plan recommendations are also meant to be consistent with the intent of regional NCCP plans. The SCBBP aligns directly with goals, objectives, and management actions in the MSP Roadmap, and is informed by regional and preserve-specific monitoring data and studies, as detailed in the F-RPMP.

This document provides land managers with general and species-specific guidelines and BMPs to conserve and restore MSP rare plant occurrences using seed resources, and provides links and references to more detailed sources of information. In addition, rare plant occurrences prioritized in the F-RPMP for restoration (e.g., reintroduction using seed) may be eligible for funding assistance for seed-related management actions through SANDAG's *Transnet* EMP land management grants, depending on grant cycle priorities.

2.0 REGIONAL SEED COLLECTION PROGRAMS

Seed collection programs that include California native plants (including MSP rare plants) are housed within various institutions or seed banks within and beyond the state. These programs acquire and maintain living seeds for conservation and/or research purposes. In addition, seeds may be distributed for restoration or horticultural activities, depending upon seed availability and policies of the holding institution (California Plant Rescue [CaPR] 2019, Meyer et al. 2014).

In this section, we highlight key programs or institutions that collect, store, or bank seed, and/or conduct research for MSP rare plants *ex situ*. Some institutions also support living plant collections. We limit our discussion of seed banks to those that are widely available to land managers (directly or indirectly), but note that other groups operate seed banks for specific purposes or for their own use (e.g., California Department of Fish and Wildlife [Rancho Jamul Ecological Reserve], Center for Natural Lands Management [CNLM]).

Conservation Terms

Ex Situ refers to the offsite conservation of genetic resources (e.g., plants, seeds), often in a botanic garden, seed bank, or seed storage facility.

In Situ refers to the onsite conservation of genetic resources (e.g., plants, seed) in the species' natural habitat.

California Plant Rescue

The California Plant Rescue (CaPR) is not a seed banking facility per se, but a consortium of institutions that conserve native plants in California and the California floristic province through field work and long-term seed or living plant collections (CaPR 2019). The goal of the CaPR Initiative is to obtain representative samples of all populations of a species to conserve its genetic diversity. The program addresses all California native plant taxa; however, the current focus is on collecting and storing seeds of sensitive plant species. Toward this end, partner institutions work collaboratively to establish priorities, identify target species to collect, share data, and fundraise (CaPR 2019). Some of these institutions also research seed biology and propagation methods. Table 2-1 lists institutions or groups that participate in the CaPR Initiative.

The CaPR also has an online tool to search collections from partner institutions: https://www.caplantrescue.org/caseedbanks.html#publicsearch. We include relevant seed information from the CaPR database in this document.

California Botanic Garden

CBG in Claremont, California maintains the largest seed bank for California native plants. CBG currently stores over 5,000 seed accessions representing more than 2,000 California native plant species and cultivars, with a focus on rare plant species from southern California (RSA 2019).

Table 2-1. California Plant Rescue Initiative: Participating Institutions and Organizations.[1]

Institution or Agency	Regional Seed Bank[2]	Website
CaPR Institutions		
California Botanic Garden	✓	https://www.rsabg.org/
California Native Plant Society		https://www.cnps.org/
Center for Plant Conservation		https://saveplants.org/
Institute for Conservation Research, San Diego Zoo Global	✓	https://institute.sandiegozoo.org/plant-conservation
Regional Parks Botanic Garden (Tilden Park)		https://nativeplants.org/
San Diego Botanic Garden		http://www.sdbgarden.org/
Santa Barbara Botanic Garden	✓	https://www.sbbg.org/
University of California Botanical Garden at Berkeley	✓	https://botanicalgarden.berkeley.edu/
University of California, Davis Arboretum and Public Garden		https://arboretum.ucdavis.edu/
University of California, Los Angeles, Mildred E. Mathias Botanical Garden		https://www.botgard.ucla.edu/
University of California, Santa Cruz Arboretum and Botanic Garden	✓	https://arboretum.ucsc.edu/
Supporting Partners		
California Department of Fish and Wildlife Natural Diversity Database		https://www.wildlife.ca.gov/Data/CNDDB
Consortium of California Herbaria		http://ucjeps.berkeley.edu/consortium/ http://www.cch2.org/portal/
Jepson Herbarium		http://ucjeps.berkeley.edu/
National Laboratory for Genetic Resources Preservation (NLGRP)	✓[3]	https://www.ars.usda.gov/plains-area/fort-collins-co/center-for-agricultural-resources-research/paagrpru/

[1] Information from CaPR 2019.
[2] Regional Seed Bank: indicates facility functions as a regional seed bank.
[3] The NLGRP serves as backup seed storage facility for regional seed collections.

CBG has long been involved with preserving seeds, and processes and stores seed for both long-term conservation and shorter-term restoration efforts. Seeds are also used for research, and may be distributed to other seed bank institutions for backup storage and/or sharing.

The Seed Conservation Program at CBG has developed policies and guidelines for collecting and storing seed, and data forms to document seed collection in the field. The program maintains lists of seed accessions and germination data that are accessible on the CBG website (https://www.rsabg.org/conservation/seed-conservation). In addition, they have produced an extensive collection of seed images (http://www.hazmac.biz/rsabghome.html).

CBG currently stores seed of 37 MSP plant species (Table 2-2). Some of these accessions are from the 1980s and 1990s and represent occurrences that have been lost due to development.

San Diego Zoo Global Native Plant Seed Bank

The Institute for Conservation Research, San Diego Zoo Global (SDZG) maintains a native plant seed bank at the Beckmann Center for Conservation Research at the San Diego Zoo Safari Park in Escondido, California. The native seed bank is dedicated to preserving the floristic diversity of San Diego County by collecting and storing seed of both common and sensitive plant species. The bank currently includes seed accessions for over 375 plant taxa. Accessions in long-term storage act as a hedge against catastrophic loss or extirpation, and provide seed for research and restoration. The seed bank started in the early 2000s in collaboration with the Millennium Seed Bank (Royal Botanic Gardens [RBG] Kew), and more recently joined the CaPR Initiative, which focuses on rare and endangered species.

The seed bank team collects seed of sensitive plants in San Diego County for *ex situ* conservation. They also develop or refine protocols for germination and propagation, and work with land managers and others on projects to restore or conserve sensitive plant species. The seed bank currently stores seed for 44 MSP plant species and maintains an additional MSP species as a living collection (Table 2-2). While SDZG contains many of the same MSP plants as CBG, their accessions are generally more recent in age (5 years old or less). The seed bank is also in the process of creating high quality images of seeds.

The SDZG was awarded a SANDAG *Transnet* Environmental Mitigation Program (EMP) land management grant in 2017 to establish genetically diverse, high quality seed collections for six MSP rare plant species: San Diego thornmint, salt marsh bird's-beak, Orcutt's bird's-beak, short-leaved dudleya, Mexican flannelbush, and willowy monardella. Under this grant, the SDZG has also bulked seed of selected MSP plants for specific restoration projects in the region. For additional information on the SDZG's native seed bank, refer to https://institute.sandiegozoo.org/resources/native-plant-seed-bank.

Santa Barbara Botanic Garden

The Santa Barbara Botanic Garden (SBBG) in Santa Barbara, California houses a conservation seed bank for California native species, with a focus on sensitive and disjunct species. The seed bank currently stores over 175,000 seeds representing 106 taxa. The collections may be available for authorized research or species restoration. Although the seed bank does not store any MSP rare plants that are endemic to San Diego County, it does include six more wide-ranging MSP plants (Table 2-2). For additional information on the SBBG seed bank, refer to https://www.sbbg.org/explore-garden/pritzlaff-conservation-center/conservation-seed-bank.

Table 2-2. MSP Plant Species: Existing Seed Collections.[1]

Scientific Name[2]	Common Name	CBG[3]	SBBG[3]	SDBG[3]	SDZG[3]
Acanthomintha ilicifolia	San Diego thornmint	✓			✓
Acmispon prostratus	Nuttall's acmispon	✓			✓
Adolphia californica	California adolphia	✓			✓
Agave shawii var. *shawii*	Shaw's agave	✓	✓	---[4]	
Ambrosia pumila	San Diego ambrosia	---[4]	---[4]		✓
Aphanisma blitoides	Aphanisma		✓		
Arctostaphylos glandulosa ssp. *crassifolia*	Del Mar manzanita	---[4]	---[4]	✓	✓
Arctostaphylos otayensis	Otay manzanita	---[4]	---[4]		✓
Arctostaphylos rainbowensis	Rainbow manzanita	---[4]			
Atriplex coulteri	Coulter's saltbush	✓			✓
Atriplex parishii	Parish's brittlescale	✓			✓
Baccharis vanessae	Encinitas baccharis	---[4]		✓	✓
Bloomeria clevelandii	San Diego goldenstar				✓
Brodiaea filifolia	Thread-leaved brodiaea	✓			✓
Brodiaea orcuttii	Orcutt's brodiaea				✓
Brodiaea santarosae	Santa Rosa brodiaea	---[4]			
Calochortus dunnii	Dunn's mariposa lily	✓			✓
Ceanothus cyaneus	Lakeside ceanothus	✓			✓
Cenaothus verrucosus	Wart-stemmed ceanothus	✓		✓	✓
Centromadia parryi ssp. *australis*	Southern tarplant	✓	✓		✓
Chloropyron maritimum ssp. *maritimum*	Salt marsh bird's-beak	✓	✓		✓
Chorizanthe orcuttiana	Orcutt's spineflower	✓			
Clinopodium chandleri	San Miguel savory				✓
Comarostaphylis diversifolia ssp. *diversifolia*	Summer-holly	✓			✓
Cylindropuntia californica var. *californica*	Snake cholla				---[4]
Deinandra conjugens	Otay tarplant	✓			✓
Dicranostegia orcuttiana	Orcutt's bird's-beak				✓
Dudleya blochmaniae	Blochman's dudleya	✓			✓
Dudleya brevifolia	Short-leaved dudleya	✓			✓
Dudleya variegata	Variegated dudleya	✓			✓
Dudleya viscida	Sticky dudleya	---[4]	---[4]		✓
Ericameria palmeri ssp. *palmeri*	Palmer's goldenbush				✓

Table 2-2. MSP Plant Species: Existing Seed Collections.[1]

Scientific Name[2]	Common Name	CBG[3]	SBBG[3]	SDBG[3]	SDZG[3]
Eryngium aristulatum var. *parishii*	San Diego button-celery	✓			✓
Erysimum ammophilum	Coast wallflower		✓		✓
Euphorbia misera	Cliff spurge	✓		---[4]	
Ferocactus viridescens	San Diego barrel cactus	✓		---[4]	✓
Fremontodendron mexicanum	Mexican flannelbush	✓			✓
Hazardia orcuttii	Orcutt's hazardia	✓		✓	
Hesperocyparis forbesii	Tecate cypress	✓		---[4]	✓
Iva hayesiana	San Diego marsh-elder	✓		---[4]	
Lepechinia cardiophylla	Heart-leaved pitcher sage	✓			
Lepechinia ganderi	Gander's pitcher sage	✓			✓
Monardella hypoleuca ssp. *lanata*	Felt-leaved monardella				✓
Monardella stoneana	Jennifer's monardella				✓
Monardella viminea	Willowy monardella	✓		---[4]	✓
Navarretia fossalis	Spreading navarretia	✓			✓
Nolina cismontana	Chaparral nolina	✓	✓		
Nolina interrata	Dehesa nolina	✓			✓
Orcuttia californica	California Orcutt grass	✓			✓
Packera ganderi	Gander's ragwort				✓
Pinus torreyana ssp. *torreyana*	Torrey pine	✓		---[4]	✓
Pogogyne abramsii	San Diego mesa mint	✓			✓
Pogogyne nudiuscula	Otay mesa mint	✓			✓
Quercus dumosa	Nuttall's scrub oak	---[4]	---[4]	---[4]	✓
Quercus engelmannii	Engelmann oak	---[4]	---[4]	---[4]	✓
Rosa minutifolia	Small-leaved rose	✓		---[4]	✓
Tetracoccus dioicus	Parry's tetracoccus	✓			✓

[1] MSP plant species as defined in the MSP Roadmap (SDMMP and TNC 2017); information in table is obtained primarily from the California Plant Rescue (CaPR) conservation collection database and from seed bank staff (C. Birker [CBG], S. Anderson [SDZG], H. Schneider [SBBG]).

[2] Nomenclature generally follows Baldwin et al. 2012.

[3] Institutions housing seed collections or other plant material: **CBG** = California Botanic Garden, **SBBG** = Santa Barbara Botanic Garden, **SDBG** = San Diego Botanic Garden, **SDZG** = San Diego Zoo Global.

[4] --- indicates the presence of a living collection (e.g., plants growing in the botanic garden) and is noted only where no seed collection has been identified at any of the three institutions. Where seed collections have been identified, a living collection may or may not occur at CBG, SBBG, or SDBG.

San Diego Botanic Garden

The San Diego Botanic Garden (SDBG) in Encinitas, California has decades of experience working with sensitive species native to San Diego County. Currently, 12 MSP rare plants are represented at SDBG as seed, stock, or living collections. The SDBG is currently working with the California Department of Fish and Wildlife (CDFW) to propagate and establish Encinitas baccharis at the garden and in surrounding, natural areas. The SDBG also supports a population of Orcutt's hazardia that was planted in 1995 in cooperation with the CDFW and CNLM. The SDBG's new horticulture campus includes a seed bank dedicated to research and recovery projects. Collection materials and institutional expertise can be made available for conservation and research purposes.

National Laboratory for Genetic Resources Preservation

The National Laboratory for Genetic Resources Preservation (NLGRP) is located on the campus of Colorado State University in Fort Collins, Colorado, and is run by the U.S. Department of Agriculture (USDA). This facility houses more than 500,000 accessions that represent 12,000 plant species in the plant collection; accessions are stored in a vault-like setting. Like the Svalbard Global Seed Vault in Norway, the NLGRP protects genetic material from loss due to natural or man-made disasters.

In addition to preserving genetic material, seeds at the NLGRP are also used for research and to some degree, for conservation and commercial purposes. The NLGRP offers seed testing services, conducts international seed exchange for research, and develops technology to improve seed storage. Although originally focused on forestry and crop species, the NLGRP now includes native plants. It stores seed of endangered species for future reintroduction efforts and provides backup storage capabilities for some of the seed banks that participate in the CaPR Initiative.

Other Institutions

Although not likely to play a key role in seed conservation for the San Diego region, seed and/or living plant collections of a few MSP rare plants occur at the (1) Millennium Seed Bank at the RBG Kew, (2) University of California Botanical Garden at Berkeley, and (3) University of California, Santa Cruz. Additionally, commercial nurseries and a few private firms provide seed collection services and operate banking and bulking facilities.

3.0 SEED ZONES

A seed zone is defined as an area within which plant material can be moved from one location to another with minimal genetic risk to the population or species (Bower et al. 2014, Miller et al. 2011, McKay et al. 2005, Johnson et al. 2004, Rogers and Montalvo 2004, St. Clair and Johnson 2004, Hufford and Mazer 2003, Lesica & Allendorf 1999). As indicated in the F-RPMP, *seed for restoration purposes should be obtained from the target occurrence, if at all possible*. However, where seed is not available or the occurrence is small, declining, and/or genetically depauperate, then it may be necessary to source seed from another occurrence. Under these conditions, seed zones guide seed movement to minimize potential, adverse effects.

The concept of seed zones arises from forestry practice. More recently, seed zones have been used in restoration to ensure that outplanted material is adapted to current environmental conditions and ideally, contains enough genetic diversity to respond to changing conditions, as well. Introducing poorly adapted seed into a restoration site may affect the success of the effort and/or extant occurrences in proximity (McKay et al. 2005).

Seed zones are based traditionally on climate, topography, soils, and other broad factors (Bower et al. 2014, Johnson et al. 2010). Some practitioners use ecoregions to estimate seed zones for widespread species (Miller et al. 2011). Bower et al. (2014) combined climate and ecoregion to refine seed zones for selected grasses and forbs in the western U.S. (Bower et al. 2014). Common garden studies provide even more precise zones based on patterns of genetic variation (e.g., St. Clair et al. 2013, Wilson et al. 2008). However, the most precise seed zones are based on studies that examine genetic differentiation and diversity within a species.

Seed zones based on climatic or other broad factors are termed *provisional seed zones*, and they estimate suitable limits for moving seed in the absence of genetic data. Seed zones based on genetic data are referred to as *genetic seed zones*, and they provide more precise limits for moving seed. Seed zones are typically depicted on maps. Refer to Figure 3-1 for examples of both provisional and genetic seed zone maps.

In the species-specific chapters, we include provisional seed zone maps for species where genetic studies have not yet been conducted and genetic seed zone maps for species with genetic information. Although we show seed zone boundaries as discrete lines or polygons, species often occur along climatic, ecologic, or genetic gradients. Therefore, we provide the following guidelines to further direct seed movement within these zones (e.g., St. Clair and Johnson 2004):

- Move seed only within the designated seed zone (i.e., both the source occurrence and target occurrence should be in the same seed zone).

- Within a seed zone, source seed from large occurrences (if available) to maximize genetic diversity and minimize introducing poorly adapted seed. Where seed zones are large, use source occurrences near the target occurrence, if possible.
- For formerly widespread species that have been fragmented recently, source seed from multiple occurrences within the seed zone (composite seed provenancing) to maximize diversity (Miller et al. 2011).
- Within the target occurrence, outplant seed into microsites that match the source occurrence, to the degree feasible.

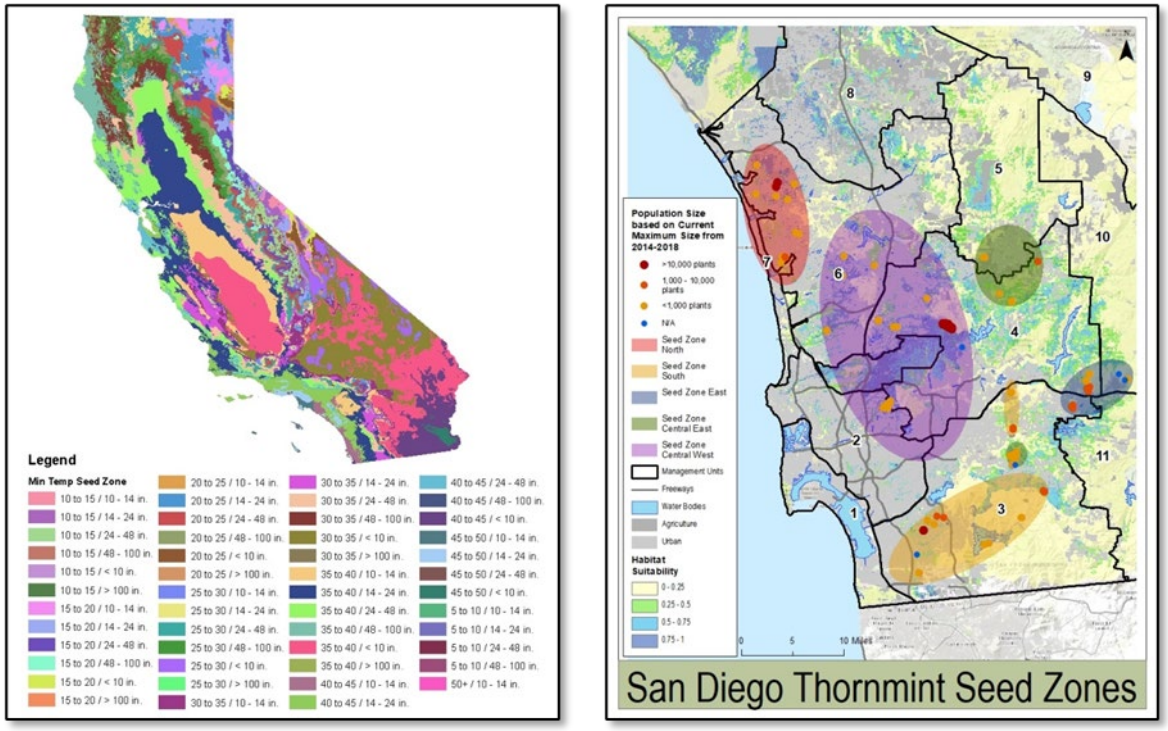

Figure 3-1. Seed Zone Maps
(left) Provisional Seed Zones for California Native Plants based on Temperature and Precipitation (http://www.forestseedlingnetwork.com), (right) Genetic Seed Zones for San Diego Thornmint in San Diego County.

Several spatial decision-support tools have been developed recently to help restoration practitioners' select appropriate seed for restoration (Table 3-1). These tools are generally specific to certain regions and/or a select suite of species. Although none of these tools has been adapted for use in the MSPA or currently includes any MSP rare plants, we provide this information for reference purposes in case these (or similar) tools become available in the future.

Table 3-1. Web-based Applications to Match Seed Source to Planting Site.

Application	Website	Description
Threat and Resource Mapping (TRM) Seed Zone Map	https://usfs.maps.arcgis.com/apps/webappviewer/index.html?id=cf1136a05dd84ec2bb4f51068b0336aa	Interactive 2D map that allows users to view/download seed zone to develop sampling and restoration strategies.
Climate Distance Mapper	https://www.usgs.gov/media/images/climate-distance-mapper-0	Spatial decision support tool to match seed source with restoration site based on current and future climate scenarios. Currently available for four regions in the desert southwest.
Seed Lot Selection Tool	https://seedlotselectiontool.org/sst/	Web-based mapping application to match seed lots with planting sites based on current or future climatic scenarios. Currently available for the western U.S, Canada, and Mexico; focuses on forest trees.

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4.0 GENERAL FRAMEWORK FOR SEED COLLECTION, BANKING, AND BULKING

Detailed guidelines for seed collection, banking, and bulking are available through a number of sources, including the Center for Plant Conservation (CPC 2018), Millennium Seed Bank Partnership (MSBP 2015), CBG (Wall 2009, Wall and McDonald 2009), and RBG Kew (RBG Kew 2001), among others. In Section 4.0, we summarize information from these and other sources that is relevant to the SCBBP; refer to the original documents for more detailed information on the elements discussed below (Table 4.1-1).

4.1 SEED COLLECTING

Qualifications for Seed Collectors

We include minimum requirements for seed collectors to ensure that collections are high quality, documented adequately, and represent the range of ecological and phenological conditions within the target species or occurrence. Thus, seed collectors should be able to:

- Develop or follow a seed sampling strategy
- Access the seed collection site safely
- Identify the target species in different phenological stages (e.g., flowering, fruiting)
- Differentiate between ripe and unripe seed
- Differentiate the target species from closely-related or visually similar species
- Monitor occurrences throughout the season to determine the optimal time for seed collection
- Conduct simple field tests for seed viability
- Collect seed using a variety of methods
- Document the seed collection digitally or on paper, using standard seed collection forms
- Record Global Positioning System (GPS) coordinates
- Prepare voucher specimens to deposit at herbaria
- Photograph the occurrence, individual plants, and fruit/seed in the field
- Transport seed to a storage facility immediately or store on a short-term basis.

Seed Collection Terms

Accession: A genetically unique plant sample from a specific location that is stored in a seed bank. Each accession has a unique identifier or number.

Bulk Collection: Seeds from all plants are collected and stored together within the same accession.

Conservation Collection: A seed collection that captures genetic diversity by collecting and storing seed along maternal lines for at least 50 plants per occurrence.

Maternal Line Collection: Seeds from each plant are collected and stored separately within the same accession.

Restoration Collection: A seed collection designed to restore a species or occurrence by bulking and/or outplanting seed back into the wild. Seed for restoration may be collected along maternal lines or in bulk.

Voucher Specimen: A pressed and dried plant specimen that documents plant identity and occurrence at a specific location.

Table 4.1-1. Detailed Sources for Seed Collection, Banking, and Bulking.

Document	Seed Collection	Seed Banking	Seed Bulking	Document Link
Bureau of Land Management: Technical protocol for the collection, study and conservation of seeds from native plant species for Seeds of Success (BLM 2016)	✓	✓		https://www.blm.gov/sites/blm.gov/files/uploads/SOS%20Protocol%206.20.18.pdf
Center for Plant Conservation: Best plant conservation practices to support species survival in the wild (CPC 2019)	✓	✓	✓	https://saveplants.org/wp-content/uploads/2019/05/CPC-Best-Practices-5.22.2019.pdf
Rancho Santa Ana Botanic Garden: Seed collection guidelines for California native plant species (Wall 2009)	✓			https://uploads-ssl.webflow.com/59613419dcbd9b40dceb57a6/5acd728da1daf8feb5427a0c_Seed%20Collecting%20and%20Storage%20Guidelines.pdf
Rancho Santa Ana Botanic Garden: Seed storage guidelines for California native plant species (Wall 2009)		✓		https://uploads-ssl.webflow.com/59613419dcbd9b40dceb57a6/5acd728da1daf8feb5427a0c_Seed%20Collecting%20and%20Storage%20Guidelines.pdf
Rancho Santa Ana Botanic Garden: Processing seeds of California native plants for conservation, storage, and restoration (Wall and MacDonald 2009)	✓	✓		http://www.hazmac.biz/Seed%20Processing/2009S.pdf
Royal Botanic Garden Kew: A field manual for seed collectors (RBG Kew 2001)	✓			http://brahmsonline.kew.org/Content/Projects/msbp/resources/Training/English_kppcont_035653_A-field-manual-for-seed-collectors.pdf
Royal Botanic Garden Kew: Assessing a population for seed collection (Way and Gold 2014a)	✓			http://brahmsonline.kew.org/Content/Projects/msbp/resources/Training/02-Assessing-population.pdf
Royal Botanic Garden Kew: Post-harvest handling of seed collections (Gold 2014)	✓	✓		http://brahmsonline.kew.org/Content/Projects/msbp/resources/Training/04-Post-harvest-handling.pdf

Table 4.1-1. Detailed Sources for Seed Collection, Banking, and Bulking.

Document	Seed Collection	Seed Banking	Seed Bulking	Document Link
Royal Botanic Garden Kew: Seed collecting techniques (Way and Gold 2014b)	✓			http://brahmsonline.kew.org/Content/Projects/msbp/resources/Training/03-Collecting-techniques.pdf https://sdmmp.com/upload/SDMMP_Repository/0/tj63vwd7xshfgrq205kz9pb4yn18mc.pdf
Santa Barbara Botanic Garden: Seed propagation of Native California plants (Emery 1988)			✓	https://calscape.org/seed_propagation.php
The Millennium Seed Bank Partnership: Seed Conservation Standards for 'MSB Partnership Collections' (Millennium Seed Bank Partnership 2015)	✓	✓		http://brahmsonline.kew.org/Content/Projects/msbp/resources/Training/MSBP-Seed-Conservation-Standards.pdf
U.S. Forest Service: Nursery manual for native plants; a guide for tribal nurseries (Dumroese et al. 2009)			✓	https://www.fs.fed.us/rm/pubs_series/wo/wo_ah730.pdf

Sampling Strategy

The objectives of seed collection are to maximize genetic diversity and provide high quality seed for conservation, research, or restoration purposes. A sampling strategy can help meet these objectives in a cost- and labor-efficient manner. In the context of this document, we focus primarily on seed collection for conservation and restoration.

The quality of a seed collection and thus, its value, depends on many elements, including location and size of the source population, target habitats, timing of seed collection, documentation, and interim storage. We recommend that land managers develop a sampling strategy to ensure that the final collection meets their needs, or that they work with professional seed collectors or a seed bank institution to develop a strategy.

All sampling strategies should address the elements discussed below.

- Collection Purpose
- Collection Type
- Seed Provenance
- Target Sample Size
- Materials and Methods
- Documentation
- Interim Storage
- Associated Costs

Strategies will vary in detail to accommodate differences in species biology, habitat(s), habit, occurrence size(s), and plant productivity. When developing a sampling strategy, it is advisable to include contingency funding or otherwise ensure that sampling may occur over multiple seasons, if needed (e.g., low population size, drought or other events).

Collection Purpose

A first step in developing a sampling strategy is to identify the purpose of the seed collection. For rare plant species, seed is most often collected for conservation (long-term storage), research or developing germination and/or propagation protocols (medium-term storage), and/or restoration (short-term storage) (Table 4.1-2).

The purpose of the collection will inform all other elements of the sampling strategy. For example, conservation or restoration collections should be relatively large to capture genetic diversity and account for seed loss during storage, bulking, or outplanting. Conversely, smaller collections may be appropriate for research or protocol development, and may be sourced from existing seed bank collections (if available) (Wall 2009, RBG Kew 2001).

Table 4.1-2. Purpose of Seed Collections.[1]

Purpose	Description	Duration
Conservation	Stores seed to preserve genetic diversity as a hedge against extinction or extirpation. May provide source material for conservation-related research or restoration if adequate seed is available.	Long-term
Research	Provides seed for research on seed biology or to develop germination/propagation protocols.	Medium-term
Restoration	Provides seed for bulking and/or outplanting to restore species.	Short-term

[1] Sources: CPC 2019, Wall 2009.

Collection Type

Collection type refers to whether the seed is collected in *bulk* or along *maternal lines*. A bulk collection packages and stores seed from all sampled plants as a single unit. A maternal line collection maintains seed from each parent plant separately. Bulk collections are less expensive to collect, process, and store; however, maternal line collections ensure that maximum genetic diversity will be reintroduced back into the field.

For MSP rare plants, experts recommend collecting along maternal lines for a conservation collection and either in bulk or along maternal lines for a restoration collection (Table 4.1-3). For restoration purposes, collecting along maternal lines is warranted where source occurrences are small (≤1,000 individuals) and/or genetic diversity is low or presumed low. In contrast, bulk collections are appropriate where occurrences are large (>1,000 individuals) and genetic diversity is high or presumed high.

Table 4.1-3. Seed Collection Type.

Collection Purpose	Collection Type	Source Population Size	Genetic Diversity
Conservation	Maternal	---	---
Restoration	Maternal	<1,000 plants	Low or unknown
Restoration	Maternal or Bulk	>1,000 plants	High or unknown

Seed Provenance

Seed provenance refers to the place of origin of the seed, or where the seed was collected. In developing a sampling strategy, identify (1) whether seed of a target species exists in a seed bank and (2) whether banked seed was collected at the target occurrence or within a suitable seed zone for the target species. If the provenance is suitable, assess the amount of seed available, age of the seed, and use restrictions on the collection to determine if the banked seed is adequate and available to meet specific objectives.

Based on this review, determine whether additional seed collection is warranted. Seed collection would be warranted under the following conditions:

- Collections of the target species do not exist in a seed bank.
- One or more collections exist, but they do not adequately represent genetic diversity across the species.
- A collection exists, but it is not duplicated in other seed banks.
- One or more collections exist, but they do not include adequate numbers of seed for long-term storage and testing.
- One or more collections exist, but they are more than 10 years old.
- Existing collections appear to be losing viability, based on seed testing.
- The provenance of the existing collection does not match the restoration site (i.e., the banked seed was not collected from within the same genetic cluster or seed zone as the target occurrence).
- A collection exists, but is from a translocated occurrence. While well-established translocations may serve as a seed source, these occurrences often have a small genetic pool, particularly if established from a limited number of founders.
- The banked seed is not available for restoration purposes.
- The banked seed is available for use, but there are not enough seeds available to meet seed bulking or restoration goals.

Note that availability of an existing seed collection is subject to many factors, including ownership or permit conditions, number of seeds, and research/recovery value of the requesting project. Contact seed bank managers directly to inquire about or request seed.

Target Sample Sizes

Establish minimum targets for the number of seeds to collect, based on the purpose of the seed collection. For a conservation collection, collect from a minimum of 30-50 unrelated plants per occurrence, with a goal of 2,500-3,000 seeds, to adequately represent the genetic diversity and provide enough material for storage and testing (e.g., CPC 2019, Wall 2009, and others).[3] If collecting seed from listed plant species, follow the requirements in the applicable permit.

[3] Guidelines vary on the minimum number of plants necessary for a conservation seed collection. Wall (2009) indicates that 30 plants would capture 95% of the genetic diversity in a population. Meyer (pers. comm.) suggested that sampling 40-50 plants would capture about 90% of the alleles in a population, based on studies at CBG. CPC guidelines (1991) recommend sampling 10-50 individuals per population. Within these ranges, minimum sampling size should be dependent on population size and history, life history, and intended use. Refer to Hoban and Schlarbaum 2014 and Hoban and Strand 2015 for additional information on sampling design for seed collection.

For a restoration collection, the same general guidelines apply with the following caveats: (1) the long-term success of the restoration effort depends, in part, on the number of seeds outplanted, so a larger collection (subject to seed availability) or subsequent seed bulking may be warranted to increase the number of seeds available and (2) the collecting location(s) may be more narrowly defined than the entire species range, depending on genetics and other factors (e.g., topography, habitat, climate).

Each occurrence that functions as a seed source will constitute a separate sample or collection, and will be stored in a seed bank (if appropriate) as a unique accession. In some cases, the collection may include seeds from a fragmented occurrence that was formerly connected.

Within an occurrence, collect no more than 5-10 percent (%) of the seed from an individual plant or from the reproductive population per season. Collect and count seed from several individuals first to determine what constitutes 5-10% of an individual for the target species. For both maternal line and bulk collections, collect seed randomly and evenly throughout the occurrence to maximize genetic diversity. For example, collect seed from:

- Plants that are widely spaced (e.g., center of occurrence, edge of occurrence)
- Plants in different habitats
- Plants that are large and small
- Plants that produce many versus few seeds
- Plants that fruit early and late in the season

Collect seed over several days, if needed. For some occurrences, it may be necessary to collect at different times during a growing season, or over multiple seasons to get enough seed. This is especially true for MSP rare plant occurrences that are small (<1,000 individuals), particularly if plant germination, survival, and seed production is affected by climate or other factors (e.g., fire, herbivory). Seed collecting guidelines incorporate safeguards against excessive seed collection; nonetheless, seed collectors should weigh the benefits of collecting seed against potential negative impacts for each occurrence.

Where an occurrence is extremely small (<50 plants) and at risk of extirpation, it may be advisable to collect all seed to preserve the remaining genetic diversity and subsequently, develop a restoration program to maximize diversity (e.g., composite provenancing, seed bulking, and outplanting; CPC 2019). In this case, seed collectors should secure land owner/manager and agency approval as needed for the collection effort before collecting seed.

Methods and Materials

Seed collecting methods are outlined below and discussed in more detail in species-specific chapters, as appropriate. Table 4.1-4 provides a list of materials used in the collection process.

Table 4.1-4. Recommended Equipment for Seed Collection.

Equipment	Description or Purpose
Pre-seed Collection	
Permits	Permission to collect voucher specimens and seed from regulatory agencies (if needed). Carry in the field.
Access letter	Permission to access the collecting location. Carry in the field.
Map or geographic coordinates of the target occurrence	Locate collecting site(s).
Flagging tape, pin flags, or other markers	Mark occurrence boundary or individual plants in flower to relocate easily when in fruit. In areas frequented by the public, use inconspicuous flagging or markers placed close to ground level to avoid drawing attention to rare plant occurrences.
Plant press, blotters, newspaper, notebook	Prepare and document voucher specimen(s).
Camera	Photograph occurrence/habitat and target species in flower.
Seed Collection	
Collecting strategy/instructions	Collect and store seed according to pre-determined strategy and standard collecting guidelines.
Map or geographic coordinates of the target occurrence	Locate collecting site(s).
GPS unit (preferably, sub-meter)	Accurately record location of occurrence and/or plants.
Camera	Photograph fruit and seed of target species.
Labels, markers, pencils	Record relevant information.
Seed collection forms	Record critical seed collection information.
Hand lens	Identify taxon or assess seed viability
Adhesive Tape	Secure small seeds when assessing viability
Safety razor or pocketknife	Cut seeds to assess viability
Container & water (optional)	Float test to assess seed for viability
Scissors, pruning shears, tweezers	Cut inflorescences or plant branches to collect seed, or collect seed from small plants.
Sieve	Recover fruits or seeds from sandy soil (use only for bulk collections).
Gloves	Protect hands while collecting seed.
Paper bags (dry fruit); various sizes	Collect dry seed. In general, use small paper bags or envelopes for maternal line collections and larger paper bags or envelopes for restoration collections.
Plastic bags or containers (fleshy fruit)	Collect fleshy fruit.
Breathable or mesh bags for bagging seed heads (optional)	Bag inflorescences of species with seed that dehisces continuously or explosively.
Post-seed Collection	
Storage container or cooler.	Keep collected seed from overheating.
Additional storage bags or containers.	Divide collected seed (if necessary) to promote air circulation and minimize seed degradation.
Pest strips (optional).	Place in collection bags temporarily if evidence of insects.

- Contact a seed bank or seed facility to obtain assistance/costs for seed collection (if desired), seed processing, seed testing, seed storage, and/or seed bulking, and enter into any agreements for these entities to receive and handle seed once it is collected. Coordinate with the selected entity on specific protocols to collect, document, or ship seeds. Table 4.1-5 lists facilities with experience collecting MSP rare plants.

Photo credit: Earth Discovery Institute.

- Obtain any necessary collecting permits from agencies or access agreements from land owners; and carry this documentation into the field. Note that it may take a year or more to obtain a permit.

- Monitor the source occurrence during the growing season to ensure that seeds are collected at the optimal time. For species that are difficult to identify or relocate when not in flower, flag or otherwise document flowering individuals to relocate them easily when in fruit (e.g., annual plants, *Dudleya* spp., *Brodiaea* spp.).

  As a general rule of thumb, Wall (2009) suggests that annuals and perennials will be ready for seed collection 2-5 weeks after peak flowering, while trees and shrubs may take 2 months or longer for seeds to mature. Time to maturity is also influenced by weather (warm weather speeds the process; cool weather delays it), elevation, or micro-habitats. In addition, some plants flower continuously over a period of time and thus, seed may ripen continuously over many weeks or months (Wall 2009).

  Characteristics of mature seed will vary by species, but may include changes in fruit color, changes in seed coat, fruits splitting or breaking open, seeds rattling, seeds that are hard and dry, or some seeds that have already dispersed (Way and Gold 2014a).

Table 4.1-5. Facilities with Experience Collecting MSP Rare Plants.[1]

Scientific Name[2]	Common Name	Seed Banks or Botanic Gardens[3]	Commercial Nurseries[4]
Acanthomintha ilicifolia	San Diego thornmint	SDZG	
Acmispon prostratus	Nuttall's acmispon	SDBG, SDZG	NW, S&S
Adolphia californica	California adolphia	SDZG	NW
Agave shawii var. *shawii*	Shaw's agave	SDBG	NW
Ambrosia pumila	San Diego ambrosia	SDZG	NW
Aphanisma blitoides	Aphanisma	SDZG	NW
Arctostaphylos glandulosa ssp. *crassifolia*	Del Mar manzanita	SDBG	
Arctostaphylos otayensis	Otay manzanita	SDZG	NW
Arctostaphylos rainbowensis	Rainbow manzanita		
Atriplex coulteri	Coulter's saltbush	SDZG	
Atriplex parishii	Parish brittlescale	SDZG	
Baccharis vanessae	Encinitas baccharis	SDBG, SDZG	NW
Bloomeria clevelandii	San Diego goldenstar	SDBG, SDZG	NW
Brodiaea filifolia	Thread-leaved brodiaea	SDZG	NW
Brodiaea orcuttii	Orcutt's brodiaea	SDZG	NW
Brodiaea santarosae	Santa Rosa brodiaea		
Calochortus dunnii	Dunn's mariposa lily	SDZG	
Ceanothus cyaneus	Lakeside ceanothus	SDZG	NW, S&S
Cenaothus verrucosus	Wart-stemmed ceanothus	SDBG, SDZG	NW, S&S
Centromadia parryi ssp. *australis*	Southern tarplant	CBG, SDZG	S&S
Chloropyron maritimum ssp. *maritimum*	Salt marsh bird's-beak	SDZG	S&S
Chorizanthe orcuttiana	Orcutt's spineflower	CBG	
Clinopodium chandleri	San Miguel savory	SDZG	
Comarostaphylis diversifolia ssp. *diversifolia*	Summer-holly	CBG, SDBG, SDZG	NW, S&S
Cylindropuntia californica var. *californica*	Snake cholla		NW
Deinandra conjugens	Otay tarplant	SDZG	NW, S&S
Dicranostegia orcuttiana	Orcutt's bird's-beak	SDZG	
Dudleya blochmaniae	Blochman's dudleya		NW
Dudleya brevifolia	Short-leaved dudleya	SDZG	
Dudleya variegata	Variegated dudleya	SDZG	NW
Dudleya viscida	Sticky dudleya	SDZG	

Table 4.1-5. Facilities with Experience Collecting MSP Rare Plants.[1]

Scientific Name[2]	Common Name	Seed Banks or Botanic Gardens[3]	Commercial Nurseries[4]
Ericameria palmeri ssp. *palmeri*	Palmer's goldenbush	SDZG	NW, S&S
Eryngium aristulatum var. *parishii*	San Diego button-celery	SDZG	NW
Erysimum ammophilum	Coast wallflower	SDZG	
Euphorbia misera	Cliff spurge	SDBG	NW
Ferocactus viridescens	San Diego barrel cactus	SDBG, SDZG	NW
Fremontodendron mexicanum	Mexican flannelbush	SDZG	NW
Hazardia orcuttii	Orcutt's hazardia	SDBG	NW
Hesperocyparis forbesii	Tecate cypress	SDBG, SDZG	NW
Iva hayesiana	San Diego marsh-elder		NW, S&S
Lepechinia cardiophylla	Heart-leaved pitcher sage		
Lepechinia ganderi	Gander's pitcher sage	SDZG	
Monardella hypoleuca ssp. *lanata*	Felt-leaved monardella	SDZG	
Monardella stoneana	Jennifer's monardella	SDZG	
Monardella viminea	Willowy monardella	SDZG	
Navarretia fossalis	Spreading navarretia	CBG, SDZG	NW
Nolina cismontana	Chaparral nolina		
Nolina interrata	Dehesa nolina	SDZG	
*Orcuttia californica**	California Orcutt grass	SDZG	NW
Packera ganderi	Gander's ragwort	SDZG	
Pinus torreyana ssp. *torreyana*	Torrey pine	SDBG, SDZG	NW
Pogogyne abramsii	San Diego mesa mint	SDZG	NW
*Pogogyne nudiuscula**	Otay mesa mint	SDZG	NW
Quercus dumosa	Nuttall's scrub oak	SDBG, SDZG	NW, S&S
Quercus engelmannii	Engelmann oak	SDZG	NW, S&S
Rosa minutifolia	Small-leaved rose	SDBG, SDZG	NW
Tetracoccus dioicus	Parry's tetracoccus	SDZG	NW

[1] MSP plant species as defined in the MSP Roadmap (SDMMP and TNC 2017). Note that there may be other institutions or organizations with experience collecting seed in the MSPA.
[2] Nomenclature generally follows Baldwin et al. 2012.
[3] Seed Banks or Botanic Gardens: **SDBG** = San Diego Botanic Garden, **SDZG** = San Diego Zoo Global, **CBG** = California Botanic Garden.
[4] Commercial Nurseries: **NW** = Native West Nursery (formerly Recon Native Plant Nursery), **S&S** = S&S Seeds, Inc.

- Collect voucher specimens of the target species, preferably while plants are in bloom. Collect vouchers only if none exist for the occurrence and if enough plants are available so that collecting does not adversely impact the occurrence.
- We include links (below) to two sources for collecting, documenting, storing, and labeling vouchers. The Smithsonian link includes instructions and photos for packaging and shipping specimens to an herbarium, which may be helpful for those unfamiliar with the process (note: plants collected in San Diego County should be deposited at the San Diego Natural History Museum).
    - San Diego County Plant Atlas project: How to collect and press proper plant specimens (http://sdplantatlas.org/pdfFiles/CollectPress.pdf).
    - Smithsonian National Museum of Natural History Guide to herbarium specimens for Bureau of Land Management (BLM) Seeds of Success (https://www.blm.gov/sites/blm.gov/files/programs_natural-resources_native-plant-communities_native-seed-development_collection_Herbarium%20Tutorial.pdf).

  Appendix B provides guidelines on collecting data for voucher specimens from the San Diego Natural History Museum. The 'Sample of Online Data Entry Form' is used by parabotanists in the Museum's Plant Atlas Program to collect specimens, while the 'Cheat Sheet' explains each data field in the data entry form.

  Submit voucher specimens and documentation to the San Diego Natural History Museum within 4 months of collection.

- Assemble materials for collecting seed, including appropriate bags or other storage containers (Table 4.1-4), and scissors, shears, or tweezers (depending on plant/seed size) if necessary to remove seed from the parent plant. In general, collect dry seeds in paper envelopes or bags and moist fruits in plastic bags or buckets.

- Test a small number of seeds in the field to ensure that the collection includes viable seeds. Quick methods for assessing viability are to (1) cut the seed to ensure it contains a well-formed embryo (hollow seeds are not viable), (2) squash the seeds on blotter paper and look for a ring of moisture that indicates a viable embryo, or (3) float a small number of seeds in water; viable (heavier) seeds will sink while nonviable (lighter) seeds will float. Secure very small seeds with tape before cutting or squashing.

- Collect seed using the method most appropriate for the target species (Table 4.1-6). Refer to species-specific chapters for more detailed information for each target species. For many species, ripe seed will detach easily from the inflorescence (Wall and MacDonald 2009). Figure 4.1-1 depicts volunteers collecting seed for native species in the MSPA.

In general, do not collect seeds from the ground unless the parental plant is known, seeds are recently dispersed and not damaged by pests or pathogens, or other methods are not available or practical (Way and Gold 2014).

Table 4.1-6. Common Methods for Collecting Seed.

Collecting Method	Description
Hand-pick Fruit	Collect fruit individually (e.g., *Quercus* spp.).
Collect Entire Plant	Collect fruit (and other plant material) by uprooting entire plant (e.g., annual species) (*note: use only for very small plants that are abundant and for which uprooting the plant would not damage soil*).
Clip Inflorescences or Branches	Selectively remove fruit attached to dry inflorescences or branches.
Shake Seed from Fruit	Open dried fruit and empty seeds into collecting container (e.g., *Brodiaea* spp.).
Strip Seed Head	Hand-strip or cut entire seed head (e.g., grasses).
Bag Fruits	Secure collecting bag around fruit that dehisces explosively or continuously, particularly if seed could be lost between monitoring or collecting visits.
Sieve Fruit	Sieve substrate (e.g., sand) to recover seeds or fruits. This method is most useful for larger seeds and applies only to bulk (not maternal line) collections.

Documentation

Detailed and accurate records are a critical component of any seed collection, and should be maintained during all stages of the seed collection, banking, and bulking process. At a minimum, seed collectors should record species name, collector name, collection date, collection location, and collection number (and maternal line plant numbers, if appropriate). In addition, use separate seed collection forms for maternal line and bulk collections. Refer to Appendix C for seed collection forms.

Table 4.1-7 lists information to record during seed collection to (1) provide insights into species biology and habitat requirements, (2) allow the seed bank to better assess seed condition and test results, and (3) match seed collections to appropriate sites for outplanting (ChBG 2019, CPC 2019, Wall 2009, RBG Kew 2001).

Figure 4.1-1. Seed Collection: Biologists and Volunteers
(Photo credits: Earth Discovery Institute (upper, lower left); Jessie Vinje, CBI (upper, lower right).

Table 4.1-7. Documentation for Seed Collection.[1]

Information	**Description[2]**
Species Name	Record the scientific name of the target species, including subspecies or variety (if applicable).
Occurrence Name	Record the MSP Occurrence Name (if assigned).
Occurrence Identification	Record MSP Occurrence Identification (ID) (if assigned). If the source occurrence does not have an MSP Occurrence ID, record the CNDDB EO number (if available).
Collection Date	Record seed collection date. If seed is collected at the same occurrence over several dates, include all dates on form and specific date on seed collection container (e.g., envelope, bag).
Collection Number	Assign each collection a unique number. If seeds are collected along maternal lines, number each collection container (e.g., envelope, bag) with both collection number and maternal line number. We recommend that the collection label include the Occurrence ID, collection number, and collecting date (see text for examples).
Collector(s)	Record name of each seed collector.
Collector's Affiliation	Record affiliated organization or institution (if any) of each seed collector.
Land Owner/Manager	Record name or organization of the land owner and land manager for the preserve or property where seed is collected.

Table 4.1-7. Documentation for Seed Collection.[1]

Information	**Description**[2]
Location	Record GPS coordinates of the collecting location. Include datum and coordinate system.
Habitat	Indicate habitat(s) at collecting location.
Associated Species	Indicate associated plant species at collecting location.
Site Conditions	Record site conditions at collecting location, including slope, aspect, elevation, and soil type.
Voucher Specimen(s)	Indicate whether voucher specimens were collected. If yes, provide voucher specimen numbers, and indicate where the specimens will be deposited.
Photographs	Take at least 3 photographs (landscape, plant, fruit/seed), and number photos consecutively. Include Occurrence ID and Collection Number in photo label. Record photograph numbers.
Population Size	Record population size (direct count or estimate) of source occurrence during year of collection.
Number of Plants Sampled	Indicate number of plants sampled for seed collection.
Fruiting Stage	Indicate fruiting stage of plants at time of sampling (early, ripe, late).
Seed Health	Note any signs of damage to fruits or seed from pests or disease.
Seed Viability	Estimate % seed viability based on visual assessment or squash test of 5-10 seeds.

[1] Information from SDMMP 2019, CBG 2019, CPC 2019, Wall 2009, RBG Kew 2001.
[2] Abbreviations: **CNDDB** = California Natural Diversity Database, **EO** = Element Occurrence, **GPS** = Global Positioning System, **MSP** = Management Strategic Plan.

Collect one or more voucher specimens of the target species at the collecting location, as discussed earlier. In addition, take at least three photographs at the collecting location to document (1) the occurrence and habitat, (2) the target species, and (3) fruit and/or seed. Figure 4.1-2 provides an example of a composite image of photos taken to document seed collecting location, plant, and fruit and seed. While this image includes cleaned fruits and seeds, collectors should document fruits and/or seeds in the field (prior to cleaning). Include photos as part of the documentation for the seed collection.

Many seed banks have a standard form for seed collection or instructions for data collection. Seed collectors should check with the institution where they plan to deposit seed to obtain forms or otherwise ensure that they adequately document the collection.

While seed bank institutions have their own numbering system, and will assign an accession number to incoming collections, we recommend that seed collectors/land managers label and track seed collections of MSP rare plants using a system similar to that used in the SDMMP's Inspect and Manage (IMG) rare plant monitoring program. All seed collected at an occurrence would have the same label, with variations for different collecting dates or maternal lines.

Figure 4.1-2. Seed Collection
Photo Documentation (composite photo credit: John MacDonald, RSA).

For a bulk seed collection, include the MSP occurrence ID, seed collection number, and collection date. For example, a collection of Otay tarplant seed from the Mother Miguel Grassland on July 14, 2019 would be labeled:

DECO13_3MMGR010_SC01_071419

In this case, **DECO13_3MMGR010** = MSP Occurrence ID, **SC01** indicates the seed collection code (SC) and number (01), and **071419** indicates the seed collection date (month/date/year). If there is no MSP occurrence ID for the target species, substitute Species_Preserve Name/Location Name_Collection Number_Date.

For a maternal line collection, use the same code but add the parental plant number (PL). In the collection above, envelopes or bags with seed collected from the 1st and 15th plants, respectively, would be labeled as follows:

DECO13_3MMGR010_SC01_**PL01**_071419

DECO13_3MMGR010_SC01_**PL15**_071419

Use similar labeling for voucher specimens (code = Voucher) and photographs (code = Photo) that accompany or support the seed collection. For example, voucher specimen 1 and photograph 1 for the collection above would be labeled as follows:

DECO13_3MMGR010_SC01_**Voucher01**_071419

DECO13_3MMGR010_SC01_**Photo01**_071419

When submitting a voucher specimen to an herbarium, label the plant with the established naming convention for that herbarium. Include the voucher label above as supplemental information, and maintain this information in the collector's records.

All MSP rare plant occurrence codes are available through the SDMMP web portal (https://sdmmp.com/spatial_search.php) or by contacting SDMMP staff.

Interim Seed Storage and Delivery

Once seed is collected, it should be sent to a seed bank or storage facility as soon as possible to retain maximum viability. In some cases, however, it will not be possible to deliver the seed immediately or the seed may need to be cleaned or dried before delivery. In this section, we summarize general guidelines for interim seed storage and delivery (e.g., ChBG 2019, CPC 2019, Gold 2014, Wall 2009, RBG Kew 2001). Seed collectors should coordinate with the selected seed bank or storage facility for any additional guidelines. For some guidelines presented below, more detailed information can be found elsewhere in this document (e.g., seed cleaning).

Interim Seed Storage

We provide the following guidelines for seed that is not delivered immediately to a storage facility:

- Store collected seed in a cool, dry location. Avoid exposure to high humidity, heat, or direct light.
- Do not store seed in vehicles.
- Do not freeze seeds.
- Remove moist seeds from plastic collecting containers and pack loosely in breathable collection bags. If necessary, clean and dry prior to shipping, or maintain seed in a refrigerator.
- If the collection is damp, dry seeds or fruits before packaging and sending to a storage facility to prevent mold. Spread fruits or seed on newspaper to dry (Gold 2014, RBG Kew 2001). Seeds can be dried outside (with protection from animals and wind), in a well-ventilated room, or with a fan to gently blow over the seeds (ChBG 2019). For fruits that

dehisce explosively, dry in sealed bags or otherwise cover so that seeds are not lost during drying.

- Periodically check the seeds for insect damage; where living insects are noted, add no-pest strips to collecting bags overnight. Do not spray insecticide directly onto seeds (CPC 2019).
- In general, seed cleaning for long-term storage or bulking is conducted most efficiently at a seed storage facility, where staff has access to specialized equipment and are experienced in cleaning different types of seeds to minimize damage during the cleaning process (RBG Kew 2001, ChBG 2019). However, seed collectors can minimize cleaning time at the storage facility by collecting carefully in the field or by some cleaning prior to sending the collection for storage (Wall 2009; Wall and MacDonald 2009). In some cases, land managers or volunteers can clean seed to an acceptable level for short-term storage and outplanting using relatively simple and cost-effective techniques (Figure 4.1-3).

Figure 4.1-3. Seed Cleaning: Volunteer Event
(Photo credit: Earth Discovery Institute)

Seed Delivery

Prepare seeds for delivery to a seed bank or seed storage facility as follows:

- Send seeds to a seed facility as soon as possible, or store the seeds in dry conditions free from light and moisture for several weeks before sending them for storage.
- Reduce the bulk of a shipment by removing extraneous plant material (e.g., branches, twigs, empty fruit) from the collection prior to shipping (Wall 2009; Wall and MacDonald 2009).
- Repackage seeds in appropriate (breathable) bags or containers (if necessary) so that each collection is loose with adequate air circulation.
- Label each collecting bag or container clearly. Make sure bags or containers are closed and/or sealed.
- Contact the seed bank or storage facility to inform them of the schedule for seed delivery and verify they will be able to accept and process the shipment quickly. Use special mail services such as 'signature confirmation' or 'registered mail' to ensure the seed is delivered to and received by the seed bank.
- Deliver seeds by hand or by mail. If sending by mail, place seed envelopes or bags inside a cardboard box for protection. Use an appropriately-sized box so that the seeds do not spill during shipment; include extra packing material, if needed. Use a 'breathable' shipment container (e.g., cardboard box rather than plastic container). Do not package seed until ready to ship (ChBG 2019). Use expedited shipping if possible to ensure that seeds arrive at the facility as soon as possible.
- Include collecting forms or other documentation (e.g., photographs) with the shipment. Include one copy of the documentation (e.g., collecting form, photographs) with the shipment and retain a copy as backup.
- Hand-deliver or ship voucher specimens to the San Diego Natural History Museum, along with all documentation (including seed collection number). Label voucher specimens and package them carefully to avoid damage during transport or shipping.

Photo credit: Colin Richard for Earth Discovery Institute.

4.2 SEED BANKING

We recognize that once seed is deposited at a seed bank or seed storage facility, it will be

processed by staff according to specific guidelines and with specific equipment that may not be available to land managers. It is not our intent, nor is it reasonable, to reproduce seed banking guidelines in detail in this document. However, we do discuss key elements of the process so that land managers or others involved in conserving seed of MSP rare plants have sufficient information to assess results, and to plan and budget for specific efforts.

Seed Cleaning

Seed must be cleaned prior to storage to remove undesirable or remaining plant material, including chaff,[4] seeds that are damaged, dead, or immature, or seeds of other species (CPC 2019). Cleaning protects the banked collection from pests or pathogens and reduces the amount of material placed in storage. Seed may be cleaned to different levels. For example, for a long-term conservation collection, seed may be cleaned moderately but retain some chaff. In this case, partial cleaning is cost-effective and enables the seed to be stored quickly. Alternatively, seed set aside for testing may be cleaned completely for easy access at a later date (CPC 2019).

Seed Biology Terms

Seed Dormancy: An adaptation to prevent a seed from germinating under unfavorable conditions. Dormant seeds are alive but not growing actively.

Seed Germination: The emergence and growth of a plant from a seed, as characterized by a seedling that develops from an embryo.

Seed Longevity: The length of time an ungerminated seed can remain viable.

Seed Viability: A measure of the potential of a seed to germinate and reproduce under suitable conditions.

Soil Seed Bank: All viable, ungerminated seeds present on or in the soil or associated litter which have the potential to form an adult plant.

There are many methods for cleaning seed, and different methods are used for different types of seed (Wall and MacDonald 2009). Refer to Table 4.2-1 for examples of cleaning methods for some common fruit types in southern California (Wall and MacDonald 2009). Wall and MacDonald (2009) provide illustrated examples of the cleaning process and equipment, along with specific cleaning methods for many California native plants, including some MSP rare plants or closely related species.

Seed Testing

Seeds are tested at various stages in the seed banking process for dormancy, germination, viability, and longevity. Seed bank institutions or commercial seed labs will conduct most seed tests. However, understanding the purpose of these tests and test results allows the land manager to make informed decisions about the (1) type of tests needed, (2) quality of seed collected, (3) length of time seed can be stored, (4) barriers to propagation, (5) target number of seeds needed for outplanting, and (6) realistic timeframes and/or metrics for reintroducing seed.

[4] Chaff refers to extraneous plant material (e.g., the seed coat or other structures surrounding the seed) that is separated from the seed during cleaning.

Seed Dormancy

Dormancy is a physical or chemical adaptation that prevents seeds from germinating under unfavorable conditions. Dormant seeds are alive but not growing actively. Dormancy prevents all seeds from germinating at one time and ensures that seeds germinate only when conditions are favorable for seedling survivorship.

Table 4.2-1. Seed Cleaning Methods.[1]

Fruit Type	Characteristic	Cleaning Method
Dry, Dehiscent Fruit (e.g., capsule, follicle, legume, silique or silicle)	Fruit splits, discharges dry seeds.	Allow seed to shed naturally in collecting bag or on sheeting; remove remaining seed from capsule manually or mechanically.
Dry, Indehiscent Fruit (e.g., caryposis, nut, utricle)	Fruit does not open to discharge seed, but decays or is eaten by animals.	Rub fruit to clean or to open and release seed.
Composites (Asteraceae)	Fruit an achene; often mixed with other plant material (bracts, pappus).	Collect mature achenes; remove pappus and break up chaff mechanically; separate seed from chaff with blowers or screens.
Fleshy fruit (e.g., drupes, berries)	Fruit is surrounded by pulp.	Soak dry fleshy fruit until soft; macerate moist fleshy fruit or softened dry fleshy fruit; spread onto a screen to dry. Once dry, separate seeds from pulp mechanically. If necessary, blow or sieve fruit to remove remaining pulp.
Cones	Seeds are held in cone scales which may open naturally or require heat to open.	Spread cones on mat or box in warm area to dry; shake or pry out seeds when cone scales open. Where heat is required to open, place cones in an oven for ≤ 20 minutes at 180°F, or immerse in boiling water for 30 seconds to one minute; remove and dry seeds.
Nutlets	Seeds loosely or tightly held within floral calyx.	Seeds may release in collecting bag. For seeds in calyces, break up mechanically; screen or blow material to separate seed from chaff.

[1] From Wall and MacDonald 2009.

Species that form persistent soil seed banks generally have some type of seed dormancy. Primary types of dormancy include physiological, physical, combinational (physiological + physical), morphological, and morphophysiological. In temperate zones, species that form persistent seed banks most often have seeds with physiological or physical dormancy mechanisms (Baskin and Baskin 1989); thus, we discuss these two types below.

Physiological Dormancy. For seeds with physiological dormancy, there is typically an afterripening period, with the seed passing through various states from dormant to non-

dormant and possibly, back to dormant if conditions become unfavorable once again (Baskin and Baskin 1989). Many obligate winter annuals exhibit this pattern of dormancy, although each species has a unique set of temperature, light, and/or precipitation cues for germination.

Physical Dormancy. For seeds with physical dormancy, the seed coat must become permeable to water before it can germinate. Physical seed coat dormancy is present in species with hard seed coats (e.g., legumes, some composites), and dormancy is relieved only when a portion of the seed coat softens.

Recognizing whether a species possesses seed dormancy is important for understanding results of germination and viability tests. Identifying dormancy cues is critical for relieving dormancy to (1) propagate seed efficiently and (2) establish realistic success criteria for outplanting/sowing. In a nursery setting, pre-treating dormant seed will maximize the number of plants that germinate. In the field, seeds with physical seed coat dormancy may experience a lag period of one or more years between sowing and significant seedling emergence.

Seed Germination

Seed germination occurs when the radicle, or seedling root, emerges from the seed coat. Germination is used as a simple test of seed viability, and can also determine whether the seed possesses any dormancy. The total number of seeds that germinate in a sample indicates the germination rate. Where all seeds in a sample germinate, we assume little or no dormancy.

In a nursery or lab setting, seeds that show some evidence of dormancy are often subjected to one or more pre-treatments to relieve dormancy and stimulate germination. Pre-treating seeds may relieve dormancy entirely or partially. Seeds that do not germinate after pre-treatment may be viable or nonviable. For seeds that are viable but do not germinate, dormancy has not been fully relieved. In other words, *high germination rates indicate high seed viability, whereas low germination rates require further testing to determine viability.*

Table 4.2-2 lists pre-treatment methods used to germinate seeds that are dormant. Note that seeds of some species may require a combination of pre-treatments for optimal germination. Seed banks and research institutions play a key role in identifying or developing germination protocols for native plants, including MSP rare plants.

Seed Viability

Viability is a measure of the potential for a seed to germinate and reproduce under suitable conditions. Viable seeds will germinate easily and under a range of conditions, unless they possess some type of dormancy. Seeds that are dormant but viable will germinate once conditions are appropriate. Non-viable seeds are dead or damaged and will never germinate.

Viability tests determine the percentage of sound (viable) seed in a collection; these tests range from non-destructive methods that estimate seed condition based on visual assessments to more precise but destructive tests (excision, staining) that examine the embryo.

Table 4.2-2. Pre-treatment Methods to Break Seed Dormancy.[1]

Treatment Type	Treatment Method(s)	Physiological Dormancy[2]	Physical Dormancy[2]
Acid/Other Chemicals	Soak seeds in bleach, vinegar, gibberellic acid	✓	✓
Charate/Smoke	Place burned plant residue over seeds, expose seeds to smoke	✓	
Fire	Place burning plant material over seeds		✓
Heat	Heat seeds in oven or autoclave	✓	
Light/Temperature	Expose seeds to various combinations of light/temperature	✓	
Scarification	Clip seed coat, excise seed/embryo, abrade seed coat mechanically (e.g., file, sandpaper) or with acid		✓
Stratification	Expose seeds to varying temperature-moisture conditions: warm-dry, warm-moist, cold-moist, warm-cold-warm	✓	
Water	Soak seeds in water (room temperature or boiling), pour boiling water over seeds, leach seeds under running water		✓

[1] Information from RSA 2018, Emery 1988.
[2] ✓ = treatment wholly or partially relieves dormancy.

Seeds are tested for viability at various stages in the seed collection and banking process, including in the field to estimate quality of seed prior to collecting and in a seed bank prior to storing. These tests are repeated on samples of stored seed at intervals to ensure that seeds remain viable and usable. We discuss some of the methods for assessing seed viability below.

Visual Assessments. Seed can be assessed visually in the field or at a seed facility. Visual assessments in the field are particularly useful for plant families known to have high levels of non-viable, empty, or damaged seeds (e.g., Asteraceae, Cyperaceae, Lamiaceae, Leguminosae, Poaceae; Way and Gold 2014a). Characteristics of viable seed include:

- Seed coat is hard and dark in color (tan, brown, or black).
- Seed is filled with healthy, firm, generally moist tissue.
- Seeds may rattle in fruit.
- Some seeds may have dispersed already.

Refer to Way and Gold (2014a) and Wall and MacDonald (2009) for additional details on visually assessing seed for viability. The CPC (2018) recommends assessing very small

collections (<100 seeds) by visual methods only (rather than by destructive methods) to preserve the limited amount of material available.

Squash Test. A 'squash' test also estimates seed viability. For this test, a ring of moisture will be present on blotter paper when healthy seeds are squashed. In contrast, seeds that are hollow will crack or pop and fail to leave a moisture residue when squashed (Wall and MacDonald 2009). The squash test is not as precise as other tests, but can be used quickly in the field.

Cut Test. A 'cut' test or seed dissection is another quick method for assessing seed viability in the field or the lab. In this test, seed is cut open using a pocket knife or safety (single-edge) razor blade and then examined with a hand lens or microscope. In the field, small seed can be held in place with double-sided tape. Viable seed is generally well-developed and fills the seed (Wall 2009).

Floatation Test. A floatation test also assesses viability in the field quickly, and is most appropriate for species with large, smooth seeds. In this test, seeds are placed in a container filled with water. Viable seed will typically sink because of the presence of the healthy seed (embryo, endosperm), while non-viable seeds will float because they contain air pockets in place of developing seed (Wall 2009).

Seed Weight. Ripe, sound seed is heavier than immature or damaged seed, and can be separated by hand, blowers, or other methods. In a seed bank, seeds are separated by weight, and a small number of heavier seeds are then dissected and examined under a microscope to obtain a more precise measure of viability (CPC 2019, Wall and MacDonald 2009).

Germination. As discussed earlier, viability can be estimated, at least in some cases, through germination tests. Germination tests provide a fairly accurate measure of viability for non-dormant seeds. However, seeds that fail to germinate must be examined further to determine if they are viable or not, as illustrated in the following example using a sample of 100 seeds:

- If 30 of 100 seeds germinate in a simple germination test, the germination rate is 30%.
- If we then examine the 70 seeds that did not germinate by dissecting, staining, or other methods and determine that none are viable, then the germination rate is 30% and viability is 30%.

- If we examine the 70 ungerminated seeds and all are viable, then the germination rate is 30% and viability is 100%.

Tetrazolium Chloride (TZ). A specific test for seed viability is the tetrazolium chloride, or TZ test, in which a sample of seeds are cut to expose the embryo and then soaked in a solution of tetrazolium chloride (*2,3,5-triphenyl tetrazolium chloride*). Viable seeds stain a red color, whereas seeds with dead tissue stain poorly or not at all (Figure 4.2-1). The TZ test provides a good measure of viability and produces results faster than germination tests. The TZ test is often used after germination tests to determine the percentage of non-germinating seeds that are alive and dormant versus dead. However, the test requires specialized training and materials, and may not be practical for large or multiple collections.

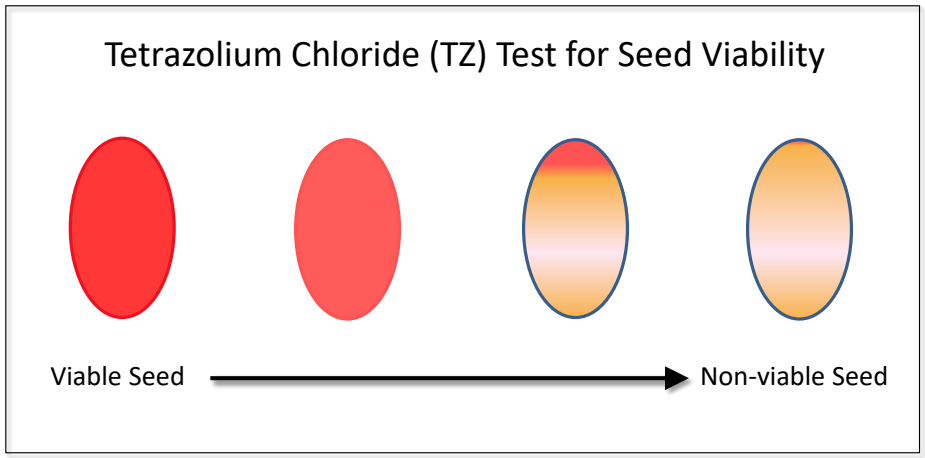

Figure 4.2-1. Tetrazolium (TZ) Test for Seed Viability: Staining Gradient.

Seed Longevity

Seed longevity is influenced by storage conditions and length of time stored; therefore, seeds in storage are tested at intervals to ensure they retain a certain level of viability. Seeds are tested initially for viability and then monitored at intervals (often, every 10 years). When viability drops to a certain level (e.g., 85% of initial viability), the seed collection may be replaced or regenerated with newly collected seed or bulked seed (MSBP 2015).

Longevity tests provide information on how long seed might reasonably persist under controlled conditions. The USDA (2019) estimates that undamaged, properly dried seeds will survive about 100 years in conventional storage (-18°C) and about 1,000 years in cryopreservation (liquid nitrogen). Seed longevity has not been determined definitively for most MSP rare plants.

F.W. Went started a long-term seed longevity study at the California Institute of Technology in 1947. Seeds from the study were subsequently stored at CBG before being moved to the NLGRP for follow-up testing (Birker pers. comm.). This longevity study includes seed from over 100 California native plant taxa that will be tested at 20-year intervals through 2307 (Went and Munz

1949). Although the study does not include any MSP plants, it does include several congeners and may shed light on potential long-term storage capabilities for these species. Results of the 50-year test are variable, with some species exhibiting higher germination rates than they did initially or at the 20-year mark, some exhibiting lower or no germination compared to previous testing, and others remaining relatively stable over the 50-year period (Wall 2009).

Seed Storage

Seed must be stored properly to maintain viability. We recommend storing seed at a recognized seed bank or seed storage facility under controlled conditions appropriate for the target species. Store seed as either a short-term (≤5 years) restoration collection or long-term (>5 years) conservation collection. In this section, we describe types of seeds, appropriate storage conditions, and packaging within the facility for long-term storage and testing.

Seed Type

Seed type influences how seed should be stored (storage conditions) and how long the seed is likely to last in storage (storage duration). We recognize three types of seeds with respect to storage capabilities: *orthodox, recalcitrant*, and *intermediate*.

- Orthodox seeds are common to arid regions and characteristic of annual or perennial species that form persistent seed banks, possess some seed dormancy, and have a relatively low seed moisture content when mature (CPC 2019). Orthodox seeds can be dried and safely stored in a conventional, long-term seed bank to retain viability. Most of the MSP rare plants possess orthodox (or presumed) orthodox seeds (RBG Kew 2019).
- Recalcitrant seeds are common in species that occur in wetland or riparian habitats, generally do not form a persistent seed bank, and exhibit no dormancy (CPC 2019). Recalcitrant seeds do not survive drying or freezing well and thus, do not lend themselves to storage in a conventional, long-term seed bank. One method for storing recalcitrant seeds is to remove and store the embryo in liquid nitrogen (CPC 2019). At least two MSP rare plants, Nuttall's scrub oak (*Quercus dumosa*) and Engelmann oak (*Quercus engelmannii*), have (or likely have) recalcitrant seeds.
- Intermediate seeds have a limited tolerance to drying. To retain viability, they are best stored in liquid nitrogen (CPC 2019). We do not currently know of any MSP rare plants that fall into this category.

Storage Conditions

Storage conditions will vary for each seed type, but all depend on drying seeds to slow the aging process and retain viability. Seed types differ in the amount of desiccation they can undergo and still remain viable.

Orthodox seeds are typically dried to a very low moisture content, frozen, and stored in sealed envelopes or containers at a low temperature (-18°C). The optimal moisture content (expressed as relative humidity or RH) of seeds for storage will depend on both drying and storage temperatures (CPC 2019, Wall 2009). Longevity in storage is typically higher for seeds collected when fully mature versus seeds collected when immature or old (Wall 2009). Refer to source documents (Table 2-1; e.g., CPC 2019, Wall 2009) for expanded discussions of recommended RH and temperatures for drying seeds. Orthodox seeds are the primary seed type in a conventional, long-term seed storage facility.

Recalcitrant seeds do not lend themselves to long-term storage at low temperatures. However, they can often be maintained at a storage facility for up to two years if kept moist and stored at a temperature just lower than the minimum temperature needed for germination (Wall 2009). Alternative methods to preserve material from recalcitrant seeds include tissue culture and cryopreservation. For example, embryonic tissue can be dried partially and cooled rapidly prior to storage (e.g., Ballesteros et al. 2019, CPC 2019, Walters et al. 2016). The SDZG is initiating a cryopreservation program to conserve recalcitrant and intermediate seeds; however, initial work is limited to Nuttall's scrub oak (*Quercus dumosa*).

Intermediate seeds can be stored for about 5 years in a conventional seed bank. They can withstand more drying that recalcitrant seeds, but not as much drying as orthodox seeds.

Packaging

After processing (cleaning, drying, testing), the seed collection will be packaged for storage by placing seed into packages as follows:

- *Storage package*. Most of the seed in a collection will be placed into a long-term storage package (or multiple packages if collected along maternal lines; Figure 4.2-2).
- *Curation package*. A smaller amount of seed from the same collection will be placed into a curation package. This seed will be tested over time for germination, viability, and longevity.

Figure 4.2-2. Maternal Line Seed Packages (Photo credit: Stacy Anderson, SDZG).

Seed collections may be split and stored at different facilities to protect against total loss if one facility is damaged or otherwise compromised. In this case, seed that is sent to a backup storage facility as a duplicate seed collection will generally include the same number of seeds as the primary collection, and will include its own curation package. Duplicate collections are most appropriate for conservation collections in long-term storage, but may be prepared for restoration collections if adequate seed and funding are available. The CPC (2018) provides the following guidelines for dividing seeds into primary, duplicate, and curation collections.

- For seed collections with >50 seeds, place 40% of the seed into a storage package and 10% into a curation package for storage at the primary facility, and 40% of the seed into a storage package and 10% of the seed into a curation package for storage at the backup facility.
- For maternal line collections where one or more samples has between 20-50 seeds, divide the seed along a 60:40 ratio for storage and curation packages, respectively, as follows:
    - For a collection of 50 seeds, place 15 seeds (30%) into a storage package and 10 seeds (20%) into a curation package for storage at the primary facility, and the same amount of seed into storage and curation packages for storage at the backup facility.
- For maternal line collections where one or more samples contain fewer than 20 seeds, do not divide for duplicate storage.

Note that the primary seed storage facility will typically arrange for a duplicate collection to be sent to a backup facility, often based on pre-existing agreements, and will provide documentation with that collection.

Seed Institutions

Table 4.2-3 lists seed banks or botanic gardens that currently maintain seed collections of one or more MSP rare plants in their long-term storage facilities or otherwise have seed for these species. This table is not necessarily comprehensive and seed at these facilities may or may not be available for use. Nonetheless, these facilities have the capability to process and store seeds for California native plants, including MSP rare plants. Refer to species-specific chapters for detailed information on stored seed for the target species.

Seed Banking Costs

Table 4.2-4 presents costs from different entities for various seed-related activities, from seed collection through storage. We present cost information in this section because costs for one activity often overlap or are incorporated into costs for another activity. Costs in Table 4.2-4 are estimates only for planning purposes and will vary based upon collecting location, amount of seed collected, type of seed, type of collection, and fruit/seed structure, among other factors. We include these estimates to provide land managers and others with a general idea of the costs involved in seed–related activities. We encourage land managers to contact one or more of these entities directly to discuss services and costs in greater detail.

Table 4.2-3. Seed Banks or Botanic Gardens.

Facility	Contact Information
Institute for Conservation Research, San Diego Zoo Global (SDZG) 15600 San Pasqual Valley Rd, Escondido, CA 92027 (760) 747-8702	Stacy Anderson Research Coordinator Native Plant Seed Bank sanderson@sandiegozoo.org (760) 796-5668
California Botanic Garden (CBG) 1500 N College Avenue, Claremont CA 91711 (909) 625-8767	Cheryl Birker Seed Conservation Program Manager cbirker@rsabg.org (909) 625-8767 ext. 259
Santa Barbara Botanic Garden (SBBG) 1212 Mission Canyon Road Santa Barbara, CA 93105-2126 (805) 682-4726	Heather Schneider, Ph.D. Rare Plant Biologist hschneider@sbbg.org (805) 682-4726 ext. 148
San Diego Botanic Garden (SDBG) 230 Quail Gardens Drive Encinitas, CA 92024-2702 (760) 436-3036	Tony Gurnoe Director of Horticulture tgurnoe@sdbgarden.org (760) 436-3036 x211

Table 4.2-4. Cost Estimates for Seed-related Activities.[1,2]

Entity	Seed Collection	Seed Cleaning	Seed Testing	Seed Bulking	Seed Storage
Seed Bank Institutions					
San Diego Zoo Global (SDZG)	$2,500 for one species at one occurrence. SDZG recommends making a full conservation collection (5 occurrences across the range; total cost = $12,500).	Included in collection cost.	Included in collection cost.	Varies by taxon. Focus is on rare species and research.	Usually included in the collection cost, but depends on the situation.
California Botanic Garden (CBG)	Varies by taxon, collection location, and amount of seed.	$60.00/hour (varies by species and number of seeds). Viability assessment included.	Germination testing = $300/test.	Varies by taxon.	Permanent Seed Collection = $3,000/accession[3]. Price includes germination testing. Temporary Research and Recovery Seed Collection (5 years) = $750/accession[2].
Santa Barbara Botanic Garden (SBBG)	Varies by taxon, collection location, and amount of seed.	Cost included in collection, unless species is very difficult to clean.	Germination testing = $300/collection. Includes testing at 1 and 10 years.	---	Cost varies by amount and length of time.
San Diego Botanic Garden (SDBG)	Varies by taxon, collection location, and amount of seed.	Included in collection cost.	Germination testing = $300/collection. Includes testing at 1 and 5 years.	Varies by taxon.	Varies by species, quantity, and storage time.
Commercial Seed Facilities					
AB Seed Laboratory, LLC	---	---	Purity + germination = $45/species; $5 to count seeds.	---	---
MD Seed Analysis, Inc.	---	---	Purity = $35/species. Germination = $35/species. Complete test = $90/species.	---	---

Table 4.2-4. Cost Estimates for Seed-related Activities.[1,2]

Entity	Seed Collection	Seed Cleaning	Seed Testing	Seed Bulking	Seed Storage
Ransom Seed Laboratory	---	---	Purity = $37-$300/species. Germination = $46-$70/species.	---	---
Hedgerow Farms, Inc.	Varies by taxon, collection location, and amount of seed.	Hand cleaning - $75.00/hour. Machine cleaning - $150.00/hour. Costs can vary by species though.	Purity + germination = $150/species (usually performed as part of the bulk seeding process)	$12,000/1 acre; 8,000/0.5-acre; 6,000/0.25-acre. Cost is usually higher for rare and uncommon species or planting plugs.	Cost is usually included in the collection process. Long-term storage is $50/month if not included in the original fee.
Native West Nursery (formerly Recon Native Plants)	Varies by taxon, collection location, and amount of seed.	Varies by species and quantity.	No testing unless requested.	Varies by species and quantity.	Varies by species, quantity, and storage time.
S&S Seeds, Inc.	Varies by taxon, collection location, and amount of seed.	Included in the seed collection cost, unless seed is not collected by them.	No testing for site-specific collections unless requested. They test their seed inventory regularly. Will outsource testing if requested by the client.	$12,000/1 acre 8,000/ ½-acre 6,000/¼-acre. Cost is usually higher for rare and uncommon species or planting plugs.	Cost is usually included in the collection process. Long-term storage is $50/month if not included in the original fee.
Stover Seed Company	Varies by taxon, collection location, and amount of seed.	Included in the seed collection cost.	No testing for site-specific collections unless requested. They test their seed inventory regularly. Will outsource testing if requested by the client.	---	Cost is usually included in the collection process. Long-term storage is species and duration-specific.

[1] Costs presented in this table are estimates only that are provided for planning purposes. They represent base costs and will vary by species, labor, and testing.

[2] Sources of information: S. Anderson (San Diego Zoo Global), M. Asghari (AB Seed Laboratory, LLC), C. Birker (California Botanic Garden), D. Grubisic (MD Seed Analysis, Inc.), T. Gurnoe (San Diego Botanic Garden), S. Knutson (Stover Seed Company), J. Miller (S&S Seeds, Inc.), M. Ranieri (Hedgerow Farms, Inc.), Ransom Seed Laboratory, P. Reynolds (Hedgerow Farms, Inc.), H. Schneider (Santa Barbara Botanic Garden), and R. West (Native West Nursery).

[3] Discuss cost with CBG since they will depend on the entity submitting the seed, the storage objective, and the size of the collection.

4.3 SEED BULKING

The purpose of seed bulking is to increase the number of seeds available by growing plants in a nursery setting and harvesting seeds for long-term storage and/or outplanting into the field. For species with a limited distribution and/or small or declining occurrences, *ex situ* seed bulking provides the means to produce enough material to restore a species or occurrence. Seed bulking is most often used to increase the amount of seed available for species with annual or biennial life cycles.

Seed bulking is an important tool for restoring rare plant species. However, it is important to structure the process to maintain (or increase) genetic diversity of the target species and ensure that the bulked seed does not differ significantly from wild seed as a result of bias in seed selection or nursery practice.

While this section deals primarily with *ex situ* seed bulking, we acknowledge that *in situ* (onsite) seed bulking may be appropriate for some occurrences. Onsite seed bulking requires long-term and sustained weed management and supplemental watering of germinated plants to maximize seed production in the field (see F-RPMP). Onsite seed bulking is a viable option where funds are not available for nursery bulking, and is appropriate for sites that are easily accessible and/or have access to a reliable water source, since plants will need to be watered multiple times during the growing season. If bulking seed onsite, consider annual weather patterns to avoid artificially selecting for plants adapted to specific conditions (e.g., drought). In addition, where the target occurrence is very small, consider adding seed from a genetically appropriate occurrence in proximity to maintain or increase diversity.

Seed Selection

Seed selection begins with seed provenance. As discussed in the F-RPMP and earlier sections of this document, practitioners should select seed for bulking that is sourced from an appropriate genetic cluster or seed zone for the target species. In some cases, it will be necessary or desirable to use a composite seed collection to obtain enough seed for bulking or to increase the genetic diversity of the seed sample used for bulking. Practitioners should not use seed from a seed bank or commercial facility if the provenance is unknown.

A primary concern during seed bulking is directional selection. Directional selection may be intentional or unintentional, but the outcome is to favor certain genotypes over others. As a result, some genes potentially important for species adaptation and survival in the wild may be lost from the bulked collection. Selection occurs under natural conditions. However, selection in the nursery may compound (not replace) natural selection and may select in a different direction than would occur naturally (USDA 2006).

Selection may occur at several points in the growing process. For example, selection may be introduced during seed germination if seeds are not pre-treated to relieve dormancy or the pre-treatment relieves dormancy only partially. In this case, dormant but viable seeds would not be represented in the bulked collection. Likewise, sowing only larger or heavier seeds might reduce the potential for lighter (but viable) seeds in the bulked collection that could be advantageous under certain conditions.

Selection may occur after germination, as well. We expect some phenotypic variation in nursery-grown plants if seeds were collected uniformly and randomly. Favoring some variants over others (e.g., tall plants over short plants, early-germinating plants over late-germinating plants) may influence the genotype of the bulked collection and again, potentially result in loss of genes important for survival.

Although we cannot avoid directional selection completely, we can minimize it by including the range of seeds in the germination process and as many phenotypes as possible in the nursery-grown plants.

Diversity of Bulked Samples

The size and genetic diversity of a seed source will influence the genetic diversity of the bulked seed sample. We can maximize genetic diversity in a bulked sample by (1) collecting seed from a large occurrence (2) collecting an adequate amount of seed (3) collecting seed randomly and uniformly (and possibly, along maternal lines), and (4) using an adequate number of seeds or seeds from an adequate number of plants for bulking. Where any of these conditions are not met, or an occurrence is otherwise known to have low genetic diversity based on genetic testing, the bulked seed may be genetically depauperate. Therefore, we need to structure the source seed appropriately to maximize genetic diversity in the bulked sample.

Small Seed Lots

For bulking purposes, a small seed lot is defined as fewer than 100 seeds or seeds from fewer than 50 plants (CPC 2019). Where the starting seed lot is small, there is a greater chance for bulked seed to have lower genetic diversity than a wild population. Measures to counteract this possibility include:

- Use seed for bulking that has been collected along maternal lines, and sow the same number of seed per maternal line to ensure that each line is adequately represented in the bulked sample.
- Consider collecting additional seed from the source occurrence in subsequent years to increase the amount of seed available for bulking. Alternatively, collect additional seed from a large occurrence within the same genetic cluster or seed zone as the target

occurrence, unless there is evidence of local adaptation at the target occurrence (possibly due to long-term isolation) or different ploidy levels between occurrences.

Low Genetic Diversity

Where there is evidence of low genetic diversity in a small occurrence that is declining even with management, use genetic structure as a guide to develop a bulking program to increase diversity. Refer to the discussions on genetic structure in Milano and Vandergast (2018) and the F-RPMP, and on seed zones in this document for additional guidelines.

- For species with low genetic differentiation among occurrences, use a genetically compatible seed source to increase the amount of seed available for bulking if adequate seed is not available at the target occurrence. Genetically compatible seed sources include large occurrences within the same genetic cluster or seed zone. If there is evidence of inbreeding, consider recovering additional seed from the soil seed bank at the target occurrence to use for bulking (Ottewell et al. 2016, Milano and Vandergast 2018).
- For species with high genetic differentiation, use seed from multiple, genetically compatible seed sources (composite provenancing) to increase the amount of seed available for bulking (if needed). Genetically compatible seed sources include medium or large occurrences within the same genetic cluster or seed zone. Manage potential risks from outbreeding depression by sourcing seed as locally as possible and from plants growing in similar habitats as the target occurrence.
- For occurrences with mixed ploidy levels, use only seed sourced from the restoration site (target occurrence) unless common garden studies indicate no local adaptation in the target occurrence (DeWoody et al. 2018).

Limits on Seed Bulking

Seed bulking is a cost-effective method for increasing the amount of seed available for conservation or restoration. As a result, it can be tempting to bulk seed for several generations to meet specific needs. As discussed above, however, selective pressures in the nursery can influence the genotype of bulked seed, often within one or two generations (e.g., Guerrant 1996, Mistretta and Burkhart 1990). At the same time, we need to balance the needs of rescuing a species or occurrence on the verge of extinction or extirpation against the potential to introduce seed that differs from wild occurrences.

For most MSP rare plants, we do not have data on the loss of diversity (if any) in bulked samples, and we may not know the effects until several years after outplanting. For this reason, we recommend the following approach to seed bulking:

- Bulk seed of MSP rare plants in a nursery setting for one generation only (F1 generation).[5] If more seed is needed, start with wild seed rather than seed from the F1 generation.
- If it is necessary to bulk seed for more than one generation due to lack of availability of wild seed, engage a plant geneticist to test and compare seed from later generations (e.g., F2 generation) to a genetic baseline (if one exists) prior to outplanting to ensure the bulked seed does not differ significantly from wild seed. Alternatively, outplant only a small sample of the F2 seed or sow into a small, discrete area and monitor for at least 5 years to track survival and reproduction. The 5-year monitoring period should include years of both low rainfall and average to above average rainfall to account for seed genotypes that are adapted to dry and wet conditions, respectively. If results are favorable, proceed with additional seeding. Favorable results include germination, flowering, *and* production of viable seed.

Documentation

Maintain detailed records on bulked seed collections to ensure they will be stored or used appropriately. Track seed after outplanting to provide information on both restoration success and failure to inform future efforts (Knapp and Rice 1994). Seed used in bulking will have source documentation (described above). Record additional, relevant information as the seed goes through the bulking process, including (but not necessarily limited to):

- Seed source (MSP occurrence ID or other site identifier)
- Seed collection date
- Amount of seed bulked
- Propagation method (e.g., pre-treatment, growth medium/containers, maternal lines)
- Propagation conditions
- Harvesting date
- Post-harvest storage location/duration/conditions
- Generation (F1, F2) of the bulked sample
- Receptor site location (if known)

Maintain a complete record of the documentation at the bulking facility and include a copy with seed sent for storage and/or for outplanting.

[5] Seed generations are denoted as F1, F2, etc. The F refers to the filial (offspring) of a parental plant and the number (1,2) refers to the generation. For example, the F1 generation is the first set of seed produced from a parental plant. The F1 seed, if grown to maturity, then produces a second, or F2, generation of seed.

Interim Seed Storage

Seed that is bulked but not outplanted immediately should be stored under controlled conditions to retain viability. Seed bank institutions or commercial seed facilities have storage protocols in place to store seed properly. These include storage in a freezer, refrigerator, or warehouse under conditions that maintain low relative humidity, low seed moisture content, and cool temperatures.

There may be cases in which a restoration practitioner or land manager will need to store seed collected and/or bulked for restoration purposes on an interim basis (e.g., a year or less). For those entities that do not have dedicated storage facilities or protocols in place, we provide the following recommendations (see Gold 2014, Dumroese et al. 2009, Houseal 2007, USDA-NRCS no date, and others).

- Dry seeds to reduce the moisture content before packaging them for storage. Refer to Section 4.1 (Seed Collecting, Interim Storage) for guidelines on drying damp seeds. Some seeds will have reached maturity but still have relatively high moisture content. For these seeds, air-drying in shallow trays or on tarps for several weeks should reduce the moisture content sufficiently (e.g., Gold 2014, Dumroese et al. 2009, Wall 2009).
- Store dried seeds in an airtight, moisture-resistant container and label the container. Suitable storage containers include (but are not limited to) sealed jars with rubber gaskets on the lids, envelopes placed into a sealed, plastic tub with an airtight lid, sealed, thick plastic bags, or heat-sealed foil-lined plastic pouches (e.g., the type used for seed storage). For seeds that are stored in a warehouse or open room, make sure the storage container is rodent-proof.
- For small seed lots, use a desiccant (e.g., silica gel packet) inside the storage container to help absorb moisture, particularly if the container is opened frequently. Replace or regenerate desiccants occasionally. Color indicator silica gels change color (e.g., from blue to pink) when saturated, thus providing a visual indicator that the desiccant needs to be replaced or regenerated.
- Store seeds under cool, dry conditions in a freezer, cooler, or at room temperature under low humidity. An appropriate storage unit can be created using a cold storage container with a commercial de-humidifier inside.
    - If using a freezer or refrigerator for storage, consider a backup power supply in case of power outages.
    - If storing seed in a refrigerator, use a self-defrosting refrigerator that maintains relative humidity between 10-40%, and that is not used for other purposes.
    - For seeds stored at room temperature, ensure that the storage area has low relative humidity. Store seeds at room temperature for a short time only because they will

deteriorate faster than seeds stored at lower temperatures. For example, seed stored at 60°F will remain viable twice as long as seed stored at 70°F (Houseal 2007).

Germination and Propagation Best Management Practices

Best Management Practices (BMPs) for seed bulking focus on growing conditions and nursery practices, because both may favor the survival of some seedlings over others and result in a bulked seed collection that differs genetically from the original collection. As described previously, this may reduce survivorship of outplanted seed (Knapp and Rice 1994, Campbell and Sorensen 1984).

Where the growing facility is situated in a different climatic zone than the seed collection site, bulked seed may not necessarily be adapted to the outplanting location. Indeed, genetic shifts have occurred in a single generation in plants from the same seed source grown in different locations (Knapp and Rice 1994).

Nursery growing practices often focus on maximum seed production, with plants subjected to different water, temperature, and nutrient regimes, as well as different soil medium than under natural conditions (USDA 2006, Knapp and Rice 1994). In addition, harvesting all seed at once (rather than over a period of time) can exclude early- and late-maturing seed from the bulked collection (Knapp and Rice 1994). These practices can select for genotypes that are not necessarily adapted to conditions at the outplanting location (USDA 2006, Knapp and Rice 1994). Other practices that may influence the quality of bulked seed and ultimately, outplanting success, include (1) pathogens or weed seeds introduced at the bulking facility and (2) cross-pollination during the growing process (Guerrant 1996).

We include the following guidelines to minimize directional selection and/or loss of genetic diversity and maximize seed quality in seed bulked at a nursery facility:

- Bulk seed at a facility as close to the source occurrence as possible or that is in an area with similar climatic conditions. If the growing facility is in proximity to a wild population of the target species, and there is the potential for cross-pollination, propagate only seed from the same seed zone as the wild population at that facility.
- Select seeds for sowing that represent the range of phenotypic variation in the seed collection, including (but not limited to) heavy and light seeds, large and small seeds, and variously colored seeds.
- Use germination protocols that maximize germination, including pre-treating seeds to relieve dormancy. Where protocols do not exist, conduct experimental germination trials prior to large-scale bulking or work with research or seed bank institutions to develop protocols.

- Sow seed into growing containers or flats at densities that avoid or minimize competition that excessively favors one phenotype over another.
- Grow seed under conditions as close to natural growing conditions as possible with respect to water, temperature, nutrients, and soil type. In some cases, it may be possible to import soil from the donor or receptor site to grow plants in the nursery.
- Follow standard nursery BMPs to minimize pathogens that can affect plant or seed health and survivorship.
- Remove weeds from planting containers to reduce the potential for non-desirable seeds in harvested material.
- If culling plants to improve growth and survivorship, make sure that retained plants represent the phenotypic variation in the sample rather than just the largest or most robust individuals.
- For outcrossing species, maintain a sufficient distance in the nursery to prevent cross-pollination between bulked collections from different seed zones or between bulked collections and other species that may hybridize with the target species.
- Harvest seed continuously throughout growing process.

Seed Bulking Facilities or Nurseries

Table 4.3-1 lists facilities with experience germinating, growing, or bulking seed for one or more MSP rare plants. This table is not necessarily comprehensive, nor it is it meant to endorse any entity. We include this information only as a guide for land managers interested in bulking seed to restore an occurrence of an MSP rare plant on their preserve. We encourage land managers to contact these or other facilities directly for additional information.

Potential Funding Sources

Table 4.3-2 lists potential funding sources that may be available to assist with seed collection, banking, and bulking activities identified as regional priorities in the F-RPMP. This list is not comprehensive, but focuses on sources that have funded activities in the region in the past and that are currently available. Refer to the SDMMP website for regularly updated information on grant opportunities:

https://sdmmp.com/events.php?type=Grants

Table 4.3-1. Facilities with Experience Growing MSP Rare Plants.[1]

Scientific Name[2]	Common Name	Seed Banks or Botanic Gardens[3]	Commercial Nurseries[4]
Acanthomintha ilicifolia	San Diego thornmint	CBG[5],SDZG	
Acmispon prostratus	Nuttall's acmispon	CBG, SDBG, SDZG	NW
Adolphia californica	California adolphia	SDZG	NW
Agave shawii var. *shawii*	Shaw's agave	SDBG	NW
Ambrosia pumila	San Diego ambrosia	SDZG	NW
Aphanisma blitoides	Aphanisma	CBG, SDZG	NW
Arctostaphylos glandulosa ssp. *crassifolia*	Del Mar manzanita	SDBG	NW
Arctostaphylos otayensis	Otay manzanita	SDZG	NW
Arctostaphylos rainbowensis	Rainbow manzanita		
Atriplex coulteri	Coulter's saltbush	CBG	
Atriplex parishii	Parish brittlescale	CBG	
Baccharis vanessae	Encinitas baccharis	SDBG	NW
Bloomeria clevelandii	San Diego goldenstar	SDBG, SDZG	NW
Brodiaea filifolia	Thread-leaved brodiaea	CBG	NW
Brodiaea orcuttii	Orcutt's brodiaea	SDZG	NW
Brodiaea santarosae	Santa Rosa brodiaea		
Calochortus dunnii	Dunn's mariposa lily	SDZG	
Ceanothus cyaneus	Lakeside ceanothus	SDZG	NW
Cenaothus verrucosus	Wart-stemmed ceanothus	SDBG, SDZG	NW
Centromadia parryi ssp. *australis*	Southern tarplant	CBG	
Chloropyron maritimum ssp. *maritimum*	Salt marsh bird's-beak	SDZG	
Chorizanthe orcuttiana	Orcutt's spineflower	CBG	
Clinopodium chandleri	San Miguel savory	SDZG	
Comarostaphylis diversifolia ssp. *diversifolia*	Summer-holly	SDBG, SDZG	NW
Cylindropuntia californica var. *californica*	Snake cholla	SDZG	NW
Deinandra conjugens	Otay tarplant	SDZG	NW
Dicranostegia orcuttiana	Orcutt's bird's-beak	SDZG	
Dudleya blochmaniae	Blochman's dudleya		NW
Dudleya brevifolia	Short-leaved dudleya	SDZG	
Dudleya variegata	Variegated dudleya	SDZG	NW
Dudleya viscida	Sticky dudleya	SDZG	NW
Ericameria palmeri ssp. *palmeri*	Palmer's goldenbush	SDZG[6]	NW

Table 4.3-1. Facilities with Experience Growing MSP Rare Plants.[1]

Scientific Name[2]	Common Name	Seed Banks or Botanic Gardens[3]	Commercial Nurseries[4]
Eryngium aristulatum var. *parishii*	San Diego button-celery	SDZG	NW
Erysimum ammophilum	Coast wallflower	SDZG	
Euphorbia misera	Cliff spurge	SDBG	NW
Ferocactus viridescens	San Diego barrel cactus	SDBG, SDZG	NW
Fremontodendron mexicanum	Mexican flannelbush	SDZG	NW
Hazardia orcuttii	Orcutt's hazardia	SDBG	NW
Hesperocyparis forbesii	Tecate cypress	CBG, SDBG, SDZG	NW
Iva hayesiana	San Diego marsh-elder	SDBG	NW
Lepechinia cardiophylla	Heart-leaved pitcher sage		
Lepechinia ganderi	Gander's pitcher sage	SDZG	
Monardella hypoleuca ssp. *lanata*	Felt-leaved monardella	SDZG	
Monardella stoneana	Jennifer's monardella	SDZG	
Monardella viminea	Willowy monardella	SDBG, SDZG	
Navarretia fossalis	Spreading navarretia	CBG, SDZG	NW
Nolina cismontana	Chaparral nolina		
Nolina interrata	Dehesa nolina	SDZG	NW
*Orcuttia californica**	California Orcutt grass	SDZG	NW
Packera ganderi	Gander's ragwort	SDZG	
Pinus torreyana ssp. *torreyana*	Torrey pine	SDBG, SDZG	NW
Pogogyne abramsii	San Diego mesa mint	SDZG	NW
*Pogogyne nudiuscula**	Otay mesa mint	SDZG	NW
Quercus dumosa	Nuttall's scrub oak	CBG, SDBG, SDZG	NW
Quercus engelmannii	Engelmann oak	CBG, SDZG	NW
Rosa minutifolia	Small-leaved rose	SDBG	NW
Tetracoccus dioicus	Parry's tetracoccus	SDZG	NW

[1] MSP plant species as defined in the MSP Roadmap (SDMMP and TNC 2017); information in table is obtained primarily from the California Plant Rescue conservation collections.
[2] Nomenclature generally follows Baldwin et al. 2012.
[3] Seed Banks or Botanic Gardens: **SDBG** = San Diego Botanic Garden, **SDZG** = San Diego Zoo Global, **CBG** = California Botanic Garden.
[4] Commercial Nurseries: **NW** = Native West Nursery (formerly Recon Native Plant Nursery).
[5] CBG conducted a series of germination tests and propagated San Diego thornmint in the 1990s (Mistretta and Burkhart 1990, https://www.rsabg.org/conservation/seed-conservation).
[6] SDZG conducted germination tests for this species, but these tests were not very successful.

Table 4.3-2. Potential Funding Sources.

Funding Source	Program	Focus	Eligible Organizations	Cycle
Preserve-specific Programs				
Land owner/manager	Annual budget allocation	Routine management, contingency funds.	Preserve-specific.	Annual
Land owner/manager	Endowments	Routine management, contingency funds.	Preserve-specific.	Annual
Regional Programs				
San Diego Association of Governments (SANDAG)	*Transnet* Environmental Mitigation Program – land management grants https://www.sandag.org/index.asp?projectid=447&fuseaction=projects.detail	Projects on conserved lands within MSPA; MSP species, habitats, threats.	Landowner/manager or representative.	Variable
San Diego Association of Governments (SANDAG)	*Transnet* Environmental Mitigation Program – land acquisition grants https://www.sandag.org/index.asp?projectid=447&fuseaction=projects.detail	Land acquisition that promotes regional habitat conservation plans.	Local jurisdictions, nonprofit organization, private land owners, consultants.	Variable
The San Diego Foundation	San Diego Foundation grants https://www.sdfoundation.org/grantseekers/	Variable; local or regional projects that benefit local residents (particularly, disadvantaged communities).	Any 501(c)(3) organization in or providing services to San Diego County.	Throughout the year
Federal and State Programs				
California Natural Resource Agency	Environmental enhancement mitigation grants http://resources.ca.gov/grants/environmental-enhancement-and-mitigation-eem/	Projects that mitigate the environmental impacts from public transportation facilities.	Local, state and federal governmental agencies and nonprofit organizations.	Annual
California State Coastal Conservancy	Coastal Conservancy grants https://scc.ca.gov/grants/	Biological diversity, water quality, habitat, and other natural resources in coastal watersheds.	Public agencies, federally-recognized tribes, nonprofit organizations.	Ongoing
California Department of Fish and Wildlife (CDFW)	Local assistance grants https://www.wildlife.ca.gov/Conservation/Planning/NCCP/Grants	High priority NCCP actions (identified in conjunction with the Wildlife Agencies).	Local jurisdictions or others implementing an approved NCCP.	Annual

Table 4.3-2. Potential Funding Sources.

Funding Source	Program	Focus	Eligible Organizations	Cycle
California Department of Fish and Wildlife (CDFW)	Prop 1: Watershed Restoration Grant Program (Water Bond 2014) https://www.wildlife.ca.gov/Conservation/Watersheds/Restoration-Grants	Reliable water supplies; resilient, sustainably managed water resources system; important species and habitat.	Public agencies, non-profit organizations, public utilities, Indian tribes,[1] and mutual water companies.	Annual
California Department of Fish and Wildlife (CDFW)	Prop 68: State of California Parks & Water Bond 2018 https://www.wildlife.ca.gov/Conservation/Watersheds/Prop-68	Climate change adaptation; economic development &protection, connectivity, recreation, drought tolerance, landscape resilience, water retention.	Public agencies, non-profit organizations, public utilities, Indian tribes,[1] and mutual water companies.	Annual
Natural Resource Conservation Service	Environmental Quality Incentives Program (EQIP) conservation innovation grants https://www.nrcs.usda.gov/wps/portal/nrcs/main/national/programs/financial/eqip/	Natural resource concerns, environmental benefits.	Agriculture and forestry producers.[2]	Annual
San Diego River Conservancy	Prop 1: Watershed Protection and Restoration Program (Water Bond 2014) http://sdrc.ca.gov/prop-1/	Shovel-ready, capital improvement projects in the San Diego River Watershed.	Public agencies, nonprofit organizations, Indian tribes.[1.]	Variable
San Diego River Conservancy	San Diego River Conservancy Proposition 68: The California Drought, Water, Parks, Climate, Coastal Protection, and Outdoor Access for all Act of 2018 http://sdrc.ca.gov/wp-content/uploads/2019/01/SDRC_prop-68-draft-guidelines-draft-012219-COMPLETE.pdf	Shovel-ready projects in the San Diego River Watershed; climate change adaptation; economic development & protection, connectivity, recreation, drought tolerance, landscape resilience, water retention.	Public agencies, nonprofit organizations, Indian tribes.[1.]	Variable
Southern California Wetlands Recovery Project (SCWRP)	Community wetland restoration grant program https://scwrp.org/community-wetland-restoration-grant-program/	Community-based restoration projects with an educational component; southern California coast.	Nonprofit organizations, universities, agencies.	Annual
U.S. Fish and Wildlife Service (USFWS)	Coastal Program https://www.fws.gov/coastal/	Native habitat restoration and acquisition (protection)	Local, state, and federal governmental agencies, Indian tribes,[1] nonprofit organizations, consultants, landowners/managers.	Annual

Table 4.3-2. Potential Funding Sources.

Funding Source	Program	Focus	Eligible Organizations	Cycle
U.S. Fish and Wildlife Service (USFWS)	Cost-sharing program (e.g., Partners for Fish and Wildlife grants) https://www.fws.gov/cno/conservation/Partners.html	Restore, protect habitat for native fish and wildlife species.	Private landowners, individuals, groups engaged in voluntary conservation efforts on private lands.	Annual
U.S. Fish and Wildlife Service (USFWS)	National Coastal Wetlands Conservation Grant https://www.fws.gov/coastal/CoastalGrants/	Native habitat restoration and acquisition (protection).	State agencies.	Annual
U.S. Fish and Wildlife Service (USFWS)	National Wildlife Refuge System Cooperative Recovery Initiative https://www.fws.gov/refuges/whm/cri/get-started/	Restore, recover federally or state-endangered species on National Wildlife Refuges and lands with a Refuge nexus.[3]	Internal grant program. Partners are encouraged to contact local or regional USFWS contacts.	Annual
U.S. Fish and Wildlife Service (USFWS)	Recovery Challenge http://www.federalgrants.com/FY-2018-Recovery-Challenge-72571.html	Enhance, increase partnerships to implement highest priority recovery actions identified in recovery plans (particular for breeding, rearing, and reintroduction programs).	State and local jurisdictions, public or private universities, Indian tribes, 1, nonprofit organizations, for-profit organizations and small businesses.	Annual
USFWS (funder), CDFW (administrator)	State wildlife grant program https://wsfrprograms.fws.gov/Subpages/GrantPrograms/SWG/SWG.htm https://www.wildlife.ca.gov/Grants/State-Wildlife-Grants	Programs that benefit wildlife and their habitats as identified in State Wildlife Action Plans.	Nonprofit organizations, local government agencies, colleges and universities, and state departments.	Annual
USFWS (funder), CDFW (administrator)	Cooperative Endangered Species Conservation Fund/Section 6 grants https://www.fws.gov/endangered/grants/index.html https://www.wildlife.ca.gov/Conservation/Planning/NCCP/Grants	Endangered species conservation, recovery; habitat acquisition for listed species per approved, draft species recovery plans.	Public agencies, state departments, colleges and universities, tribal governments, nonprofit entities working with resource agencies.	Annual
Wildlife Conservation Board (WCB), CDFW	Monarch butterfly and pollinator rescue program https://wcb.ca.gov/Programs/Pollinators	Monarch butterflies and other pollinators.	Private landowners, nonprofit organizations, resource conservation districts, public agencies.	Annual

[1] Including federally recognized Indian tribes, and state Indian tribes listed on the Native American Heritage Commission's California Tribal Consultation List.
[2] Including nonprofit organizations, indigenous tribes, private land owners, or individuals or groups engaged in conservation efforts on private lands.
[3] Program funds on-the-ground projects with high likelihood of success.

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5.0 SPECIES-SPECIFIC SEED COLLECTION, BANKING, AND BULKING

5.1 SAN DIEGO THORNMINT (*ACANTHOMINTHA ILICIFOLIA*)

Seed Collection

Seed Characteristics

San Diego thornmint flowers produce a maximum of four smooth, ovoid seeds (nutlets) that are held in a persistent, spine-tipped calyx (Miller and Jokerst 2012). Bauder and Sakrison (1997) observed 3-4 seeds per flower, with most calyces retaining seed.

Figure 5.1-1. San Diego Thornmint: Seed (Photo credit: John MacDonald, RSA)

Seed production is variable, but increases with plant size and flower production. Recent estimates range from 10-200 seeds per plant (DeWoody et al. 2018, Lippett et al. no date), although Bauder and Sakrison (1999) reported much higher seed production in experimentally-grown plants, including one individual that produced over 3,000 seeds.

Seeds mature in late spring and early summer. Mature seeds are light to medium brown with dark mottling (Figure 5.1-1). Lippett et al. (no date) found a higher percentage of filled seed and a higher germination rate for dark versus light seed, and suggested that light seed may not be fully mature. Seeds remain on the plants presumably until they desiccate completely or are released from the parental plant by weather (wind, rain), usually within one year. Although dry plants may be present from the previous year, they do not usually contain seeds.

Priority Occurrences

Table 5.1-1 prioritizes occurrences of San Diego thornmint on conserved lands in the MSPA for seed-related management actions. These priorities were developed in the F-RPMP based on monitoring data and specific studies, including genetic analyses. The decision to implement an action will depend on a number of factors, including status of the occurrence (stable or declining), habitat condition, threats management, and funding, among others.

In general, large occurrences with high genetic diversity are prioritized as potential seed sources, subject to land manager and/or agency approval. Small occurrences are high priority candidates for seed reintroduction, subject to response to management and site conditions, as discussed in the F-RPMP.

Currently, conservation seed collections exist in established seed banks for 14 thornmint occurrences (31%) on conserved lands in the MSPA (Table 5.1-1). Seed banks also hold collections for an additional six occurrences that are extirpated or not currently conserved.

We recommend conservation collections as a potential management action for all occurrences that are not currently represented in a seed bank *if adequate seed is available*. For occurrences where plants have not been detected recently, a conservation collection will only be possible if plants are detected in the future (Table 5.1-1). Large occurrences are the highest (or most immediate) priorities for seed conservation to maximize the amount of genetic diversity in long-term storage (e.g., ACIL_3PMA1013).

We recommend restoration collections as a potential management action for occurrences that are high or medium priorities for seed reintroduction as indicated in Table 5.1-1 and identified in the F-RPMP. We do not prioritize occurrences for seed reintroduction if they have not yet been monitored or if the location is questionable.[6]

We recommend reintroducing seed only if threats are controlled, habitat is likely to support this species in the future, and funding is available for short- and long-term management. Reintroducing seed is most appropriate for small occurrences that continue to decline, even with management. Seed may be introduced into extirpated occurrences if threats are managed and the habitat is likely to support thornmint in the future. For extirpated occurrences, source seed from a large occurrence in the same seed zone or from an appropriate seed source in storage (if available). Some threats (particularly, invasive plants) will likely require perpetual management; thus, ensure that adequate funding and/or labor are available to manage the occurrence after seed is reintroduced.

[6] For a few San Diego thornmint occurrences, the locality information appears to be incorrect based on monitoring data that shows both an absence of plants and a lack of suitable habitat to support the species. These problematic occurrences are included in Table 4.3-2, but highlighted with footnotes.

Table 5.1-1. San Diego Thornmint: Management Priorities for Seed-related Activities.

Occurrence ID[1]	Occurrence Name	Occurrence Size[2]	Genetic Structure[3]	Seed Reintroduction Priority[4]	Existing Seed Collection[5]	Seed-related Activities[6]			
						Seed Source	Conservation Collection	Restoration Collection	Seed Bulking
ACIL_2EDHI001	El Dorado Hills	Small	H+L+L	---			✓[7]		
ACIL_2EDHI002	El Dorado Hills	Small	H+L+L	---			✓[7]		
ACIL_3BOME003	Bonita Meadows	Medium	H+L+L	Medium			✓	✓	✓
ACIL_3CERE004	Crestridge Ecological Reserve	Small	H+L+L	High			✓	✓	✓
ACIL_3DREA005	Dennery Ranch East	Small	H+L+L	High	SDZG			✓	✓
ACIL_3HCWA006	Hollenbeck Wildlife Area	Medium	H+L+L	Medium	SDZG			✓	✓
ACIL_3LONC007	Long Canyon (PMA 4-2b)	Small	H+L+L	High			✓	✓	✓
ACIL_3MGMT008	McGinty Mountain	Small	H+L+L	High	SDZG			✓	✓
ACIL_3MGMT009	McGinty Mountain (southwest slope)	Small	H+L+L	High			✓	✓	✓
ACIL_3MGMT010	McGinty Mountain (summit, ridgeline)	Small	H+L+L	High	SDZG			✓	✓
ACIL_3OTLA011	Lower Otay Reservoir	Small	H+L+L	High			✓[7,8]	✓[8]	✓[8]
ACIL_3OTLA012	Lower Otay Reservoir	Small	H+L+L	High			✓	✓	✓
ACIL_3PMA1013	PMA1 (Rice Canyon)	Large	H+L+L	Low		✓			
ACIL_3PMA3014	PMA3 (Poggi Canyon)	Small	H+L+L	---			✓[7,8]		
ACIL_3RJER015	Rancho Jamul Ecological Reserve	Small	H+L+L	High			✓[7]	✓	✓
ACIL_3SOCR016	South Crest (Suncrest)	Medium	H+L+L	High	SDZG			✓	✓
ACIL_3WHRI017	Bonita, Wheeler Ridge (Long Canyon PMA 4-1cW)	Small	H+L+L	High			✓	✓	✓
ACIL_3WRFI018	Wright's Field (north & south)	Medium	H+L+L-M	Medium			✓	✓	✓
ACIL_4CSVI019	Canada San Vicente-Daney Canyon	Small	H+L+L	---			✓[7,8]		

Table 5.1-1. San Diego Thornmint: Management Priorities for Seed-related Activities.

Occurrence ID[1]	Occurrence Name	Occurrence Size[2]	Genetic Structure[3]	Seed Reintroduction Priority[4]	Existing Seed Collection[5]	Seed-related Activities[6]			
						Seed Source	Conservation Collection	Restoration Collection	Seed Bulking
ACIL_4CSVI020	Canada San Vicente--Monte Vista (Long's Gulch)	Small	H+L+L	High			✓[7]	✓	✓
ACIL_4MTRP021	Mission Trails Regional Park (MTRP)	Small	H+L+L	High	SDZG			✓	✓
ACIL_4MTRP022	MTRP (southwest Tierra Santa parcel, northwest of Mission Gorge)	Small	H+L+L	---			✓[7]		
ACIL_4POGR023	Poway Grade	Small	H+L+L	---			✓[7,8]		
ACIL_4POMT048	Poser Mountain	Small	H+L+L-M	---			✓[7]		
ACIL_4POMT049	Poser Mountain 35	Small	H+L+L-M	High			✓	✓	✓
ACIL_4POMT050	Poser Mountain	Small	H+L+L-M	---			✓[7]		
ACIL_4SASP024	Saber Springs (east)	Small	H+L+L	---			✓[7]		
ACIL_4SASP025	Sabre Springs (east, subpopulation 1)	Small	H+L+L	High	CBG, SDZG			✓	✓
ACIL_4SIPR026	Simon Preserve	Medium	H+L+L	Medium	SDZG			✓	✓
ACIL_4SYCA027	Sycamore Canyon	Large	H+L+L	---	SDZG	✓			
ACIL_4VIMT0028	Viejas Mountain (northwest slope)	Small	H+L+L-M	High			✓[7]	✓	✓
ACIL_4VIMT0029	Viejas Mountain (southwest slope)	Medium	H+L+L	Medium	CBG			✓	✓
ACIL_4VIMT0030	Viejas Mountain (west-southwest flank)	Small	H+L+L-M	High			✓	✓	✓
ACIL_5RAGR031	Ramona Grasslands/Hobbes Property	Small	H+L+L	---			✓[7]		
ACIL_6BLMO032	Black Mountain	Small	H+L+L	High			✓	✓	✓
ACIL_6CAHI033	Calavera Hills	Small	H+L+L	---			✓[7]		

Table 5.1-1. San Diego Thornmint: Management Priorities for Seed-related Activities.

Occurrence ID[1]	Occurrence Name	Occurrence Size[2]	Genetic Structure[3]	Seed Reintroduction Priority[4]	Existing Seed Collection[5]	Seed-related Activities[6]			
						Seed Source	Conservation Collection	Restoration Collection	Seed Bulking
ACIL_6CARA034	Carlsbad Racetrack (south)	Small	H+L+L	High			✓	✓	✓
ACIL_6CARL035	Southeast Carlsbad (east)	Small	H+L+L	---			✓[7]		
ACIL_6CARL036	Southeast Carlsbad (west)	Small	H+L+L	---			✓[7]		
ACIL_6EMPO037	Emerald Pointe	Small	H+L+L-M	High			✓	✓	✓
ACIL_6LCGR038	La Costa Greens	Small	H+L+L	High			✓	✓	✓
ACIL_6LPCA039	Los Peñasquitos Canyon	Small	H+L+L	High	CBG, SDZG			✓	✓
ACIL_6LUCA040	Lux Canyon (west)	Small	H+L+L-M	---			✓[7]		
ACIL_6LUCA042	Lux Canyon (east), Manchester Avenue Mitigation Bank	Small	H+L+L-M	---			✓[7]		
ACIL_6MAMI041	Lux Canyon (west of Manchester Avenue Mitigation Bank)	Medium	H+L+L-M	Medium	CBG			✓	✓
ACIL_6PARO043	Palomar Airport Road	Large	H+L+L-M	Low	SDZG	✓			
ACIL_6RACA044	El Fuerte Street (Rancho Carrillo)	Small	H+L+L	High			✓	✓	✓
ACIL_6RSFE045	Rancho Santa Fe	Small	H+L+L	---			✓[7]		
ACIL_6THCO046	Thornmint Court	Small	H+L+L	---	CBG				

[1] Occurrence ID: Occurrence identification (ID) code per the San Diego Management and Monitoring Program's (SDMMP) Master Occurrence Matrix (MOM) database.

[2] Occurrence Size: Recent size category as defined in the Framework-Rare Plant Management Plan (F-RPMP). **Small** = <1,000 individuals, **Medium** = 1,000-<10,000 individuals, **Large** = >10,000 individuals.

[3] Genetic Structure (per Milano and Vandergast 2018): Genetic differentiation + genetic diversity + inbreeding. **L** = low, **H** = High, **L-M** = Low or Mixed. Refer to F-RPMP for a discussion of the implications for seed movement.

[4] Seed Reintroduction Priority (per F-RPMP): **High** = high priority for seed reintroduction, **Medium** = medium priority for seed reintroduction, **Low** = low priority for seed reintroduction, **---** = not a priority for seed reintroduction at this time due to stable population (large occurrence) or lack of monitoring data.

[5] Existing Seed Collection: Indicates location of collection. **CBG** = California Botanic Garden, **SDZG** = San Diego Zoo Global.

[6] Seed-related Activities: ✓ indicates occurrence is suitable as a seed source (pending authorization from responsible entities) or management action is warranted. Where there is an existing seed collection, a restoration collection may not be needed, but land managers should contact the seed bank directly to make that determination. Likewise, seed bulking may not be necessary depending on the amount of seed available in an existing collection or collected specifically for restoration purposes.

[7] Occurrence has not been monitored or plants have not been observed recently. In this case, a conservation collection is warranted only if the occurrence is extant and plants are present in the future.

[8] Occurrence location is questionable (possibly mapped incorrectly) based on monitoring data that indicate an absence of both plants and suitable habitat for thornmint. Resolve issue prior to implementing seed-related management actions.

We recommend seed bulking as a potential management action for all occurrences requiring a restoration collection. However, the need for bulking will depend on the amount of seed available in storage and/or collected specifically for restoration, and the sowing strategy. For example, bulking will not be needed if enough seed is collected to sow directly into the target occurrence.

Seed Zones

We established five seed zones for San Diego thornmint to guide seed collecting, banking, and bulking efforts: North, South, East, Central-east, and Central-west. Seed zones correspond to the five genetic clusters and population groups identified for this species in the F-RPMP. Figure 5.1-2 shows the locations of the five seed zones; Table 5.1-2 lists the seed zone for each occurrence.

Seed zones place geographic limits on the transfer of seed between occurrences. *We do not recommend transfer of seed beyond a seed zone at this time*. Thus, if a small occurrence requires seed from another occurrence, the source (donor) occurrence should be located within the same seed zone as the target (recipient) occurrence. Within a seed zone, we recommend that seed transfer proceed between occurrences that are close to one another, particularly in larger seed zones. We designate subgroups (Table 5.1-2) to identify occurrences in proximity. Where it is not possible or practical to move seed within a subgroup, consider subgroups nearby for sourcing seed. The same limits on seed movement apply when using seed from a seed bank.

Collecting Plan

Refer to Section 4.1 of this document for guidelines on developing a sampling strategy for seed collection. Identify the purpose and type of collection needed for a specific project or restoration effort, as well as timeline and costs. In the section below, we highlight key steps in the process, including those specific to San Diego thornmint.

Sampling Strategy

Refer to Table 5.1-1 for prioritized seed-related activities for the target occurrence to identify whether seed is needed for a conservation collection, a restoration collection, or both.

For restoration, determine if seed is available in a seed bank to reintroduce directly into a target occurrence or for bulking. Proceed with the collecting plan if stored seed is not available, quantities are not sufficient, or the provenance is not appropriate for the target occurrence (e.g., different seed zone).

Assess whether the target occurrence will likely provide an adequate quantity of seed for restoration or if seed will need to be sourced from other occurrences in the seed zone. Refer to seed zones and seed zone subgroups (Figure 5.1-2, Table 5.1-2) for limits on seed movement and seed sources. Contact the appropriate land managers of potential seed source occurrences to obtain permission for seed collection.

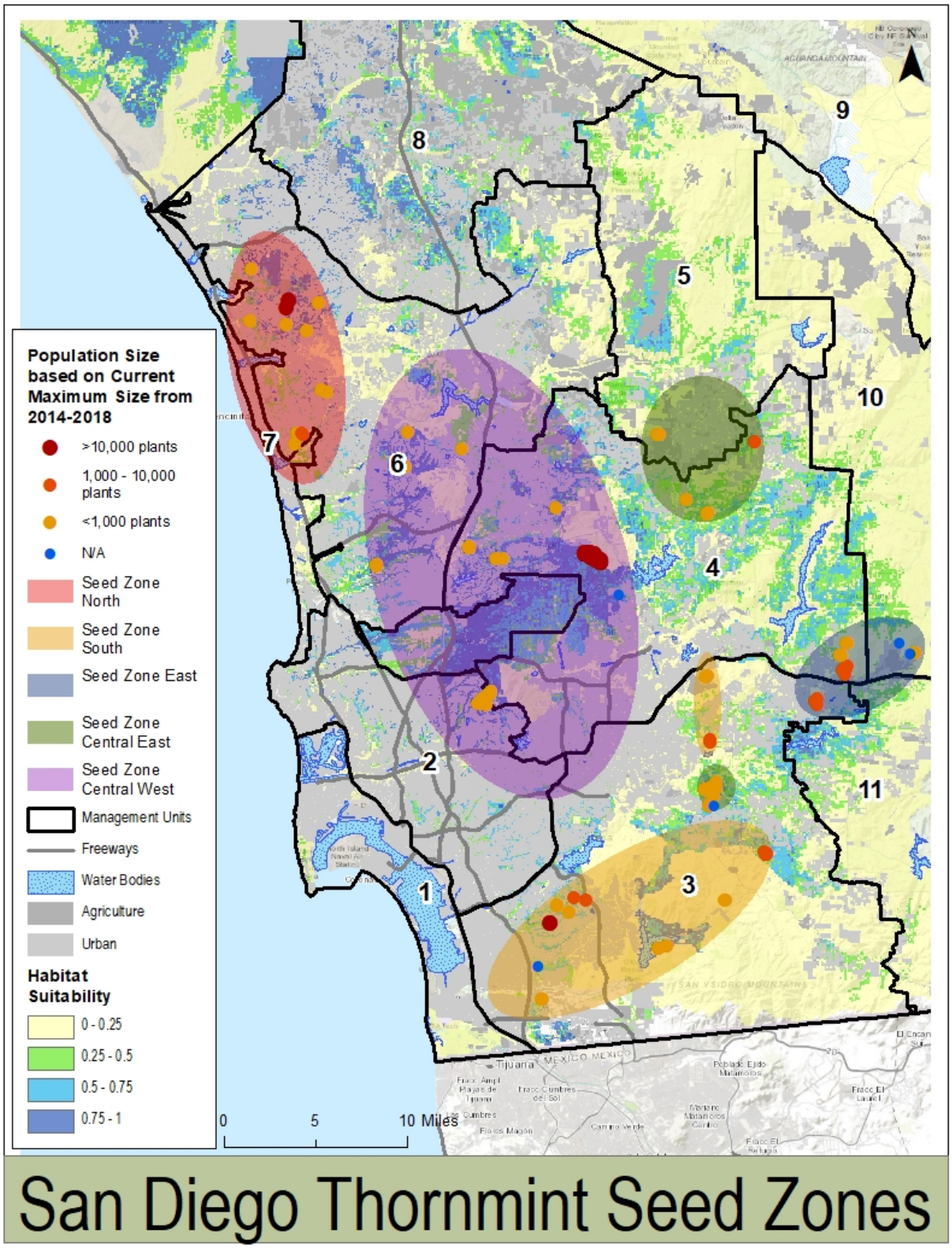

Figure 5.1-2. San Diego Thornmint: Seed Zones.

Table 5.1-2. San Diego Thornmint: Seed Zones.

Occurrence ID[1]	Occurrence Name	Seed Zone[2]	Seed Zone Subgroup[3]	Occurrence Size[4]
ACIL_6PARO043	Palomar Airport Road	North	1	Large
ACIL_6CAHI033	Calavera Hills	North	1	Small
ACIL_6CARA034	Carlsbad Racetrack (south)	North	1	Small[5]
ACIL_6EMPO037	Emerald Pointe	North	1	Small
ACIL_6LCGR038	La Costa Greens	North	1	Small[5]
ACIL_6RACA044	El Fuerte Street (Rancho Carrillo)	North	1	Small
ACIL_6CARL035	Southeast Carlsbad (east)	North	2	Small
ACIL_6CARL036	Southeast Carlsbad (west)	North	2	Small
ACIL_6MAMI041	Lux Canyon (west of Manchester Avenue Mitigation Bank)	North	3	Medium[5]
ACIL_6LUCA040	Lux Canyon (west)	North	3	Small
ACIL_6LUCA042	Lux Canyon (east), Manchester Avenue Mitigation Bank	North	3	Small
ACIL_3PMA1013	PMA1 (Rice Canyon)	South	1	Large
ACIL_3BOME003	Bonita Meadows	South	1	Medium
ACIL_3DREA005	Dennery Ranch East	South	1	Small
ACIL_3LONC007	Long Canyon (PMA 4-2b)	South	1	Small
ACIL_3OTLA012	Lower Otay Reservoir	South	1	Small
ACIL_3PMA3014	PMA3 (Poggi Canyon)	South	1	Small
ACIL_3WHRI017	Bonita, Wheeler Ridge (Long Canyon PMA 4-1cW)	South	1	Small
ACIL_3HCWA006	Hollenbeck Wildlife Area	South	2	Medium[5]
ACIL_3OTLA011	Lower Otay Reservoir	South	2	Small
ACIL_3RJER015	Rancho Jamul Ecological Reserve	South	2	Small
ACIL_3SOCR016	South Crest (Suncrest)	South	3	Medium
ACIL_3CERE004	Crestridge Ecological Reserve	South	3	Small
ACIL_3WRFI018	Wright's Field (north & south)	East	1	Medium
ACIL_4POMT048	Poser Mountain	East	1	Small[5]
ACIL_4POMT049	Poser Mountain 35	East	1	Small
ACIL_4POMT050	Poser Mountain	East	1	Small[5]
ACIL_4VIMT0028	Viejas Mountain (northwest slope)	East	1	Small
ACIL_4VIMT0029	Viejas Mountain (southwest slope)	East	1	Medium[5]
ACIL_4VIMT0030	Viejas Mountain (west-southwest flank)	East	1	Small

Table 5.1-2. San Diego Thornmint: Seed Zones.

Occurrence ID[1]	Occurrence Name	Seed Zone[2]	Seed Zone Subgroup[3]	Occurrence Size[4]
ACIL_3MGMT008	McGinty Mountain	Central-east	1	Small[5]
ACIL_3MGMT009	McGinty Mountain (southwest slope)	Central-east	1	Small[5]
ACIL_3MGMT010	McGinty Mountain (summit, ridgeline)	Central-east	1	Small[5]
ACIL_4SIPR026	Simon Preserve	Central-east	2	Medium
ACIL_4CSVI019	Canada San Vicente-Daney Canyon	Central-east	2	Small
ACIL_4CSVI020	Canada San Vicente--Monte Vista (Long's Gulch)	Central-east	2	Small
ACIL_5RAGR031	Ramona Grasslands/Hobbes Property	Central-east	2	Small
ACIL_2EDHI001	El Dorado Hills	Central-west	1	Small
ACIL_2EDHI002	El Dorado Hills	Central-west	1	Small
ACIL_4MTRP021	Mission Trails Regional Park (MTRP)	Central-west	1	Small
ACIL_4MTRP022	MTRP (southwest Tierra Santa parcel, northwest of Mission Gorge)	Central-west	1	Small
ACIL_4SYCA027	Sycamore Canyon	Central-west	2	Large
ACIL_4POGR023	Poway Grade	Central-west	2	Small
ACIL_4SASP024	Saber Springs (east)	Central-west	2	Small
ACIL_4SASP025	Sabre Springs (east, subpopulation 1)	Central-west	2	Small[5]
ACIL_6LPCA039	Los Peñasquitos Canyon	Central-west	2	Small[5]
ACIL_6BLMO032	Black Mountain	Central-west	3	Small[5]
ACIL_6RSFE045	Rancho Santa Fe	Central-west	3	Small
ACIL_6THCO046	Thornmint Court	Central-west	3	Small[5]

[1] Occurrence ID: Occurrence identification (ID) code per the San Diego Management and Monitoring Program's (SDMMP) Master Occurrence Matrix (MOM) database.

[2] Seed Zone: seed zones are defined in text and correspond to genetic clusters and population groups in the F-RPMP. Limit seed movement to within the same seed zone. Shading indicates seed zone group and subgroup.

[3] Seed Zone Subgroups are defined in text and correspond to population subgroups in the F-RPMP. Where feasible, limit seed movement to the same subgroup within a population group. Shading indicates seed zone subgroup and subgroup.

[4] Occurrence size: refers to population size category based on multiple years of monitoring data. Recent size category as defined in the Framework-Rare Plant Management Plan (F-RPMP). **Small** = <1,000 individuals, **Medium** = 1,000-<10,000 individuals, **Large** = >10,000 individuals.

[5] Indicates occurrences where monitoring indicates a decline in population size category over time. Refer to F-RPMP for additional details.

Based on the purpose and type of collection, and amount of seed expected to be available, determine whether the seed is likely to be collected along maternal lines or in bulk (Table 4.1-3), and set minimum targets for the number of plants to sample and amount of seed to collect. These targets are intended to provide a guide for planning, and may be modified based on the actual seed crop.

Permitting and Agreements

Ensure that all regulatory permits, memorandums, and access agreements are in place before collecting seed from an occurrence.

If unfamiliar with the collecting location, contact the land owner to obtain a map and coordinates of the occurrence, directions to the occurrence if necessary, gate codes, lock combinations, or keys, and any pertinent information about the occurrence (e.g., informing adjacent land owners, closures due to unsafe conditions).

Pre-collection Monitoring

Practitioners can collect San Diego thornmint seed as early as April and as late as August, depending on location, elevation, aspect, and weather and temperature conditions during the previous fall, winter, and spring. Visit the target occurrence multiple times to determine the best time to collect seed. Time the first visit when plants are in full flower and easier to locate. Talk to land owners and managers and refer to monitoring data and literature to determine the range of phenological variation for an occurrence to plan the first site visit.

Use pin flags to demarcate the boundary of the occurrence if plants are located in dense grassland, because they are very difficult to locate when dry. If necessary, place pin flags adjacent to a subset of plants to relocate them easily during subsequent visits. Avoid walking within the occurrence to the degree feasible to minimize damage to plants and soils. Return to the occurrence within two to four weeks to check phenology. Weather conditions will determine the rate of desiccation; plants will dry quicker in warm and dry conditions and slower if conditions remain cool and moist. Continue monthly site visits until the majority of plants in an occurrence are fully desiccated and seeds resemble those in Figure 5.1-1.

Voucher Specimens

Collect voucher specimens of San Diego thornmint if none exist for the source occurrence and if enough plants are available so that collecting a few specimens does not adversely affect the occurrence. Check the San Diego Natural History Museum or Consortium of California Herbaria to determine if a voucher specimen already exists:

San Diego Natural History Museum: http://sdplantatlas.org/publicsearch.aspx

Consortium of California Herbaria: http://ucjeps.berkeley.edu/consortium/

Note that recently collected specimens may not yet be accessed into an herbarium's collection. In addition, older specimens may not include sufficient locality information to definitively determine whether they were collected at the occurrence. If it is not appropriate to collect a voucher specimen, photograph the occurrence (landscape aspect) and an individual plant while in flower, and then again when the plant is in fruit.

Obtain all necessary permits and memorandums before collecting voucher specimens of endangered, threatened, and rare species. Collect specimens while plants are still in flower and before collecting seed. Refer to Section 4.1 of this document (Seed Collecting, Materials and Methods) and instructions and forms in Appendix B to guide your collection. Submit the voucher specimen(s) to the San Diego Natural History Museum as soon as possible.

Methods and Materials

Refer to Section 4.1 of this document for a detailed discussion of methods and materials and Table 4.1-4 for a comprehensive list of recommended equipment for collecting seed. At a minimum, bring the following to each San Diego thornmint collection location:

- Regulatory permits and access agreements
- Directions, map(s)
- Lock combination(s), key(s)
- Collecting strategy/instructions
- GPS unit (preferably, sub-meter)
- Camera
- Labels, markers, pens or pencils
- Seed collection forms
- Hand lens
- Adhesive tape
- Safety razor or pocketknife
- Container and water (optional)
- Scissors, snips
- Gloves
- Small paper envelopes or bags

Before collecting seed, map the seed collection area(s), record data on the seed collection form(s) and count or estimate the number of plants within an occurrence. Collect no more than 5-10% of the seed from an individual plant or from the reproductive population per season. Use the following method to determine the number of seeds to collect by plant and for an occurrence:

Step 1: Select one verticillaster (groups of flowers on the inflorescence) from 3-5 plants of different sizes spaced randomly across the occurrence (Spiegelberg 2019).

Step 2: Cut the verticillasters with scissors instead of removing the entire plant, place each verticillaster in a separate seed collection bag, and crush/rub to force seeds from dry calyces.

Step 3: Count the number of seeds in each verticillaster and multiply by the number of verticillasters on the plant to obtain an estimate of the *number of seeds per sampled plant*.

Step 4: Divide the total number of seeds per sampled plant (Step 3) by the number of plants sampled (Step 1) to calculate the *average number of seeds per plant* (Spiegelberg 2019).

Step 6: Multiply the average number of seeds per plant (Step 4) by the occurrence count or estimate to calculate the *estimated number of seeds per occurrence*. In most cases, the amount of seed available for collecting will be higher in years with optimal growing conditions (e.g., average or above-average rainfall) and lower during drought years.

Step 7: Calculate the maximum number of seeds to collect per plant or occurrence to stay within the 5-10% collecting guidelines.

Collect seeds when San Diego thornmint plants are fully dry. At this stage, plants will be tan to brown with no green parts showing, and seeds will be hard and tan with black patches (Figure 5.1-1). Seeds are not ready to collect if they are green, soft, or shriveled. Seeds may be ready to collect as early as one month after peak flowering.

Collect seed during early, mid, and late season to account for differences in maturity times and to capture the phenological variability among the occurrence. Collect seed randomly and evenly throughout the occurrence, taking care to collect from plants of various sizes and with variable inflorescences, as well as plants growing in unique or varying habitat conditions. Wear gloves when collecting seed from San Diego thornmint since the spines on the calyx lobes can easily penetrate skin.

Consider harvesting more seed than recommended by the CPC for small occurrences that are at serious risk of extirpation to capture the highest amount of genetic variation at the occurrence (McMillan 2019). In this case, coordinate with the regulatory agencies prior to the collecting effort.

Ideally, collect seed from 30-50 plants across the occurrence. If the number of plants present in a given year is below this number, then sample over multiple years to produce a genetically robust seed collection for restoration purposes. Where the occurrence is small and not expected to produce an adequate number of plants (and seed) in any given year, consider collecting seed from one or more occurrences within the same seed zone. Depending on project objectives and sampling strategy, collect seed along maternal lines or as a bulk collection.

Interim Seed Storage and Delivery

Refer to the general section of this document (Seed Collecting) for guidelines on interim seed storage and delivery. We provide additional guidelines for practitioners that choose to keep the seed for future restoration purposes rather than sending it immediately to a storage facility:

- Do not clean and sort seed unless the collection is very small since cleaning seed is time-consuming and often difficult, especially with large amounts of seed.
- Loosely pack the seed collection in envelopes or bags and store them in dark, dry, and moderate to cool conditions for several months before distributing seed onsite.
- If planning to hold seed for longer than one year, store the seed at a recognized seed bank (i.e., CBG, SDZG) to ensure proper storage conditions to retain viability.

Seed Banking

Existing Conservation and Restoration Collections

Refer to Table 5.1-3 for existing seed collections of San Diego thornmint. This table includes only thornmint occurrences with seed in storage, including some occurrences that are extirpated or on private lands. For planning purposes, we indicate the seed zone for each collection. We also indicate the number of seeds present in the seed bank and their potential availability for restoration. Note that only a portion of the seeds in a collection will be available for these purposes. Existing permits or contracts may also limit the use of some collections. We obtained information on each collection (seed bank, accession number and year, and seed quantity, provenance, and availability) from seed bank managers. Refer to Table 4.2-3 16 for contact information for these seed banks; we encourage land managers to contact facilities directly to get more information on seed availability or to request seed.

Best Management Practices

Seed Cleaning

One method for cleaning and sorting thornmint seed is as follows (per Wall and MacDonald 2009 for a related species of thornmint):

Table 5.1-3. San Diego Thornmint: Existing Seed Collections.

Occurrence ID[1]	Seed Zone[2]	Seed Bank[3]	Accession Number[4]	Collection Year	Seed Quantity[5]	Provenance Type[6]	Seed Available[7]
ACIL_3DREA005	South	SDZG	S0710	2017	~790	W	Yes
ACIL_3DREA005	South	SDZG	S0710-F12018	2018	~12,950	W	Yes
ACIL_3HCWA006	South	SDZG	S0713	2017	~1,550	W	Yes
ACIL_3MGMT008	Central-east	SDZG	S0773	2019	~2,445	W	Yes
ACIL_3MGMT010	Central-east	SDZG	S0746	2018	~210	W	Yes
ACIL_3MGMT010	Central-east	SDZG	S0780	2019	~150	W	Yes
ACIL_3SOCR016[7]	South	SDZG	S0792	2019	Unknown	W	Yes
ACIL_4MTRP021	Central-west	SDZG	S0716	2017	~2,496	W	Yes
ACIL_4MTRP021	Central-west	SDZG	S0716-F12018	2018	~1,834	---	No
ACIL_4MTRP021	Central-west	SDZG	S0788	2019	~500	W	Yes
ACIL_4SASP025	Central-west	CBG	16667-42	1991	5,847	W	No
ACIL_4SASP025	Central-west	CBG	19510-2274	1991	15,457	Z	Yes
ACIL_4SASP025	Central-west	CBG	17932-102	1992	209	W	No
ACIL_4SIPR026	Central-east	SDZG	S0768	2019	~500	W	Yes
ACIL_4SYCA027	Central-west	SDZG	S0742	2018	~525	W	Yes
ACIL_4SYCA027	Central-west	SDZG	S0777	2019	~1,200	W	Yes
ACIL_4VIMT0029	East	CBG	17823-216	1992	357	W	No
ACIL_6LPCA039	Central-west	CBG	17934-407	1992	634	W	No
ACIL_6LPCA039	Central-west	CBG	18532-563	1993	2,422	W	No
ACIL_6LPCA039	Central-west	SDZG	S0718	2017	~235	W	Yes
ACIL_6MAMI041	North	CBG	17931-101	1992	3,039	W	Limited
ACIL_6THCO046	Central-west	CBG	17935-280	1992	755	W	No
ACIL_6THCO046	Central-west	CBG	18531-562	1993	497	W	No
ACIL_6PARO043	North	SDZG	S0748	2018	~185	W	Yes

Table 5.1-3. San Diego Thornmint: Existing Seed Collections.

Occurrence ID[1]	Seed Zone[2]	Seed Bank[3]	Accession Number[4]	Collection Year	Seed Quantity[5]	Provenance Type[6]	Seed Available[7]
Black Mountain Road[8]	Central-west	CBG	15855-1242	1989	31,776	W	Limited
Black Mountain Road[8]	Central-west	CBG	18919-1243	1996	1,623,338	Z	Yes
Black Mountain Road[8]	Central-west	CBG	18920-1244	1996	31,133	Z	Yes
Palos Vista[8]	Between North and Central-west[9]	CBG	16184188	1990	4,606	W	No
San Diego Wild Animal Park[8]	Central-east	CBG	17936-355	1993	2,863	W	No
Southeast of Poway	Central-west	CBG	17933-269	1992	10,610	W	Limited
Southeast Viejas Mountain, lower population	East	CBG	17821-189	1992	585	W	No
Vicinity of Poser Mountain	East	CBG	17822-276	1992	211	W	No

[1] Occurrence identification (ID) per the SDMMP's MOM database. If no occurrence ID exists for a location, we provide the name of the location as indicated on collection in seed bank. The Black Mountain Road location is extirpated.

[2] Seed Zone: Seed zone corresponds to the five genetic clusters and population groups identified in the F-RPMP: South. Occurrences in italics were not addressed specifically in the F-RPMP and are placed in seed zones based on location.

[3] Seed bank: **CBG** = California Botanic Garden, **SDZG** = San Diego Zoo Global.

[4] Accession number = accession number of seed collection assigned by seed bank.

[5] Seed quantity indicates the number of seeds currently in the collection, according to seed bank records.

[6] Provenance type indicates the source location of the seed collection: **W** = seed collected directly from the wild (origin known), **Z** = seed collected from a cultivated plant of known wild origin, **---** = information not available.

[7] Seed available: **Yes** = seed is available for restoration use. **No** = seed is not available for restoration use, **Limited** = amount of seed available for restoration use is ≤400 seeds.

[8] Occurrence is extirpated or presumed extirpated.

[9] The Palos Vista location does not fall within a designated seed zone. This occurrence was located in Escondido, between the North seed zone and northern part of the Central-west seed zone.

- Rub dried verticillasters over a small screen to release seed from the dried calyx.
- Sort the remaining material through a series of increasingly fine filters to remove larger plant material.
- Use a blower at low speed to separate chaff from seed, and at a higher speed to separate hollow seed from filled seed.

Wall and MacDonald (2009) indicate that cleaning seed of thornmint species is easy, since most seed dehisces in the collection bag and the remaining seed separates from the calyx readily.

Seed Testing

Seed testing methods will vary by facility, but all seed banks conduct baseline germination tests on new accessions and then conduct follow up testing to check seed for viability.

At CBG, baseline tests for seed germination use a typical seed sample of 50 seeds, although sample size may vary depending on the size of the seed collection and methods to break dormancy (if needed). CBG provides abundant time for seeds to germinate and at the end of a germination test they conduct a cut test on a sample of 5 ungerminated seeds (Birker pers. comm.). CBG also conducts follow-up testing on seeds in storage at intervals (e.g., 1 year, 5 years, 10 years, 20 years, and every subsequent 10 years thereafter, as seed quantities allow). In December 2019, CBG conducted a 30-year follow-up germination trial for San Diego thornmint and 100% of the seeds germinated within one week with no pre-treatment (Birker pers. comm.).

At SDZG, a baseline germination test is conducted on all new accessions. The test is conducted primarily on fresh seed. Follow up testing is conducted on frozen seed after 1, 5, and 10 years in storage. Seed is sterilized with bleach and placed in reverse osmosis water at room temperature for 24 hours to fully imbibe (absorb) water. Seed is then plated onto agar trays (10 seeds per tray) and observed for signs of germination. The seed is counted as germinated when the radical emerges. SDZG does not use a cut test to determine viability. Rather, they provide abundant time for seeds to germinate (Anderson pers. comm.).

Seed Storage

All seed banks contacted store San Diego thornmint seed according to CPC guidelines. For example, SDZG dries seeds down to 23-35% moisture content. Seed is then stored in sealed, foil-lined envelopes in the freezer at a low temperature (-18°C to -23°C).

Each collection is separated into three packages that represent the primary, curation, and duplicate collections, as described in Section 4.1. The primary collection is the active seed collection and seeds in this package are available for research, testing, restoration, and other uses. The curation package is the base collection for long-term storage. This package is not

opened and is not available for other uses. It constitutes the long-term conservation collection. The duplicate collection is sent to another seed bank facility as backup storage to protect against catastrophic loss at the primary storage facility. Both SDZG and CBG send their duplicate seed collections to the NLGRP.

Seed Bulking

Seed Selection

Refer to Section 4.3 for guidelines on seed selection to maximize the quality of the bulked sample. Key considerations include:

- Use seed for bulking from the target occurrence or another occurrence in the same seed zone as the target occurrence. For very small occurrences (fewer than 100 plants), use seed from a large occurrence (if present) or from one or more smaller occurrences (composite provenancing) within the same seed zone.
- Use seed that was collected randomly and uniformly.
- Use an adequate amount of seed for bulking (ideally, ≥1,000 seeds or seed from 30-50 plants).
- For very small seed samples, use seed collected along maternal lines.

Best Management Practices

Germination and Propagation Methods

San Diego thornmint seed germinates readily without pre-treatment in the presence of adequate moisture, although some seeds take longer to germinate than others (Lippet et al. no date, Mistretta and Burkhart 1990). Practitioners have germinated seed in both native clay soil collected from near the occurrence and in horticultural potting mix (Lippet et al. [no date], Rice 2017, Vinje pers. obs.). After sowing, water the seeded flats or container pots several times a week to stimulate germination and continue watering, through flowering, until plants begin to senesce.

In a common garden study, Lippet et al. (no date) recorded a 40-60% germination rate for seed sown in a substrate of potting mix and washed sand (3:1), watered in initially with 500 milliliters (ml) of water, misted twice a week during germination, and subsequently irrigated using 1000 ml per pot per irrigation event.

San Diego thornmint is an outcrossing species. Mistretta and Burkhart (1990) observed that increased plant density in a nursery setting encouraged pollinators and possibly, increased seed production.

Limits on Generations

At this time, we recommend bulking thornmint seed for only one generation (F1) in a nursery setting. If additional seed is needed for restoration, start with wild seed rather than the F1 generation. Mistretta and Burkhart (1990) observed a sharp decline in germination between thornmint seed bulked for one generation in a nursery (45% germination rate) and wild-collected seed (95% germination rate). We recommend further studies to determine if these results were an anomaly or to be expected when bulking seed of this species.

Documentation

We encourage growing facilities and others bulking seed of San Diego thornmint for reintroduction purposes to document bulked samples as indicated in Section 4.3 and provide this information to land managers when seed is delivered. Likewise, we encourage land managers to provide information on seed collection and seed reintroductions to the SDMMP (contact information at https://sdmmp.com/about.php#contact) and include relevant information on IMG monitoring forms (i.e., translocated occurrence). Refer to Table 5.1-4 for key information for bulked seed samples.

Table 5.1-4. Documentation for Bulked Seed Samples.

Key Information	Primary Responsibility
Seed Source	Seed Collector and/or Land Manager
Seed Collection Date	Seed Collector
Amount of Seed Bulked	Growing Facility
Propagation Method	Growing Facility
Propagation Conditions	Growing Facility
Harvesting Date(s)	Growing Facility
Post-harvest Storage Location/Duration/Conditions	Growing Facility
Generation of Bulked Sample	Growing Facility
Receptor Site Location	Growing Facility and/or Land Manager

5.2 NUTTALL'S ACMISPON (*ACMISPON PROSTRATUS*)

Seed Collection

Seed Characteristics

Nuttall's acmispon produces many slender legumes (fruits) that are round in cross-section and taper to a short, straight or curved beak. The fruits are reddish-brown in color and are usually constricted between each seed. Each fruit contains two hard, smooth, kidney-shaped seeds that are mottled tan and black (Figure 5.2-1). When the plant dries, fruits fall to the ground and eventually release the seeds. Neither fruits nor seeds have any apparent modifications for wind, water, or animal dispersal.

Photo credit: ©2018 Ron Vanderhoff.

Nuttall's acmispon seeds mature in spring through early fall. Seed production increases with plant size and flower production, with larger plants producing hundreds of seeds. Fruits can remain on dry plants throughout the summer and into early fall.

Figure 5.2-1. Nuttall's Acmispon: Seed (Photo credit: John MacDonald, RSA)

Priority Occurrences

Table 5.2-1 prioritizes occurrences of Nuttall's acmispon on conserved lands in the MSPA for seed-related management actions. These priorities were developed in the F-RPMP based on monitoring data and specific studies. The decision to implement an action will depend on a number of factors, including status of the occurrence (stable or declining), habitat condition, threats management, and funding, among others.

In general, large occurrences are prioritized as potential seed sources, subject to land manager and/or agency approval. Small occurrences are high priority candidates for seed reintroduction, subject to response to management and site conditions, as discussed in the F-RPMP.

Table 5.2-1. Nuttall's Acmispon: Management Priorities for Seed-Related Activities.

Occurrence ID[1]	Occurrence Name	Occurrence Size[2]	Seed Reintroduction Priority[3]	Existing Seed Collection[4]	Seed-related Activities[5]			
					Seed Source	Conservation Collection	Restoration Collection	Seed Bulking
ACPR_1BFSP014	Border Field SP	Small	M			✓	✓	✓
ACPR_1DSTR010	D Street Fill	Small	M			✓	✓	✓
ACPR_1DUTR005	Dune Triangle	Small	H			✓	✓	✓
ACPR_1FIIS007	Mission Bay (Fiesta Island)	Small	M			✓	✓	✓
ACPR_1FIIS029	Fiesta Island	Small	H			✓	✓	✓
ACPR_1HOPO002	Mission Bay (Hospitality Point)	Medium	L	SDZG				
ACPR_1MAPO004	Mission Bay (Mariner's Point)	Large	---		✓	✓		
ACPR_1NMLA001	Mission Bay (No Man's Land)	Small	M			✓	✓	✓
ACPR_1NOBE015	North Ocean Beach (Dog Beach)	Small	M	SDZG			✓	✓
ACPR_1RRSO003	Mission Bay (Rip Rap)	Small	M			✓	✓	✓
ACPR_1SBSA013	South Bay Salt Works Nuttall's Acmispon	Medium	L			✓		
ACPR_1SOSH006	Mission Bay (east of South Shores)	Medium	L			✓		
ACPR_1SSSB012	Silver Strand SB	Large	---		✓	✓		
ACPR_1SSSB027	Silver Strand SB	Small	M			✓	✓	✓
ACPR_1SSSB028	Silver Strand SB	Small	M			✓	✓	✓
ACPR_7AGHE024	Agua Hedionda	Small	H			✓	✓	✓
ACPR_7BALA020	Batiquitos Lagoon	Small	M			✓	✓	✓

Table 5.2-1. Nuttall's Acmispon: Management Priorities for Seed-Related Activities.

Occurrence ID[1]	Occurrence Name	Occurrence Size[2]	Seed Reintroduction Priority[3]	Existing Seed Collection[4]	Seed-related Activities[5]			
					Seed Source	Conservation Collection	Restoration Collection	Seed Bulking
ACPR_7CSPA018	San Elijo Lagoon	Large	---	CBG	✓			
ACPR_7SCSB025	South Carlsbad SB	Small	H			✓	✓	✓
ACPR_7SLRR017	San Luis Rey River	Small	M			✓	✓	✓
ACPR_7TPSR019	Torrey Pines SR (south)	Small	M			✓	✓	✓
ACPR_7TPSR023	Torrey Pines SR (north)	Small	M			✓	✓	✓

[1] Occurrence ID: Occurrence identification (ID) code per the San Diego Management and Monitoring Program's (SDMMP) Master Occurrence Matrix (MOM) database.
[2] Occurrence Size: Recent size category as defined in the Framework-Rare Plant Management Plan (F-RPMP). **Small** = <1,000 individuals, **Medium** = 1,000-<10,000 individuals, **Large** = >10,000 individuals.
[3] Seed Reintroduction Priority (per F-RPMP): **High** = high priority for seed reintroduction, **Medium** = medium priority for seed reintroduction, **Low** = low priority for seed reintroduction, **---** = not a priority for seed reintroduction at this time due to stable population (large occurrence) or lack of monitoring data.
[4] Existing Seed Collection: Indicates location of collection. **CBG** = California Botanic Garden, **SDZG** = San Diego Zoo Global.
[5] Seed-related Activities: ✓ indicates occurrence is suitable as a seed source (pending authorization from responsible entities) or management action is warranted. Where there is an existing seed collection, a restoration collection may not be needed, but land managers should contact the seed bank directly to make that determination. Likewise, seed bulking may not be necessary depending on the amount of seed available in an existing collection or collected specifically for restoration purposes.

Currently, conservation seed collections exist in established seed banks for three acmispon occurrences (14%) on conserved lands in the MSPA (Table 5.2-1). Seed banks also hold collections for an additional three occurrences that are extirpated or not currently conserved.

We recommend conservation collections as a potential management action for all occurrences that are not currently represented in a seed bank *if adequate seed is available*. For occurrences where plants have not been detected recently, a conservation collection will be possible only if plants are detected in the future (Table 5.2-1). Large occurrences are the highest (or most immediate) priorities for seed conservation to maximize the amount of genetic diversity in long-term storage (e.g., ACPR_1SSSB012).

We recommend restoration collections as a potential management action for occurrences that are high or medium priorities for seed reintroduction as indicated in Table 5.2-1 and identified in the F-RPMP. We do not prioritize occurrences for seed reintroduction if they have not yet been monitored.

We recommend reintroducing seed only if threats are controlled, habitat is likely to support this species in the future, and funding is available for short- and long-term management. Reintroducing seed is most appropriate for small occurrences that continue to decline, even with management. Seed may be introduced into extirpated occurrences if threats are managed and the habitat is likely to support acmispon in the future. For extirpated occurrences, source seed from a large occurrence in the same seed zone or from an appropriate seed source in storage (if available). Some threats (particularly, invasive plants) will likely require perpetual management; thus, ensure that adequate funding and/or labor are available to manage the occurrence after seed is reintroduced.

We recommend seed bulking as a potential management action for all occurrences potentially requiring a restoration collection. The need for seed bulking will depend on the amount of seed available in storage and/or collected specifically for restoration, as well as the sowing strategy. For example, bulking will not be needed if adequate seed is collected to sow directly into the target occurrence.

Seed Zones

We established three provisional seed zones for Nuttall's acmispon to guide seed collecting, banking, and bulking efforts: North, Central, South. Seed zones correspond to the three potential genetic clusters and three regional population groups identified for this species in the F-RPMP. Figure 5.2-2 shows the locations of the three seed zones; Table 5.2-2 lists the seed zone for each occurrence.

Seed zones place geographic limits on the transfer of seed between occurrences. *We do not recommend transfer of seed beyond a seed zone at this time*. Thus, if a small occurrence requires

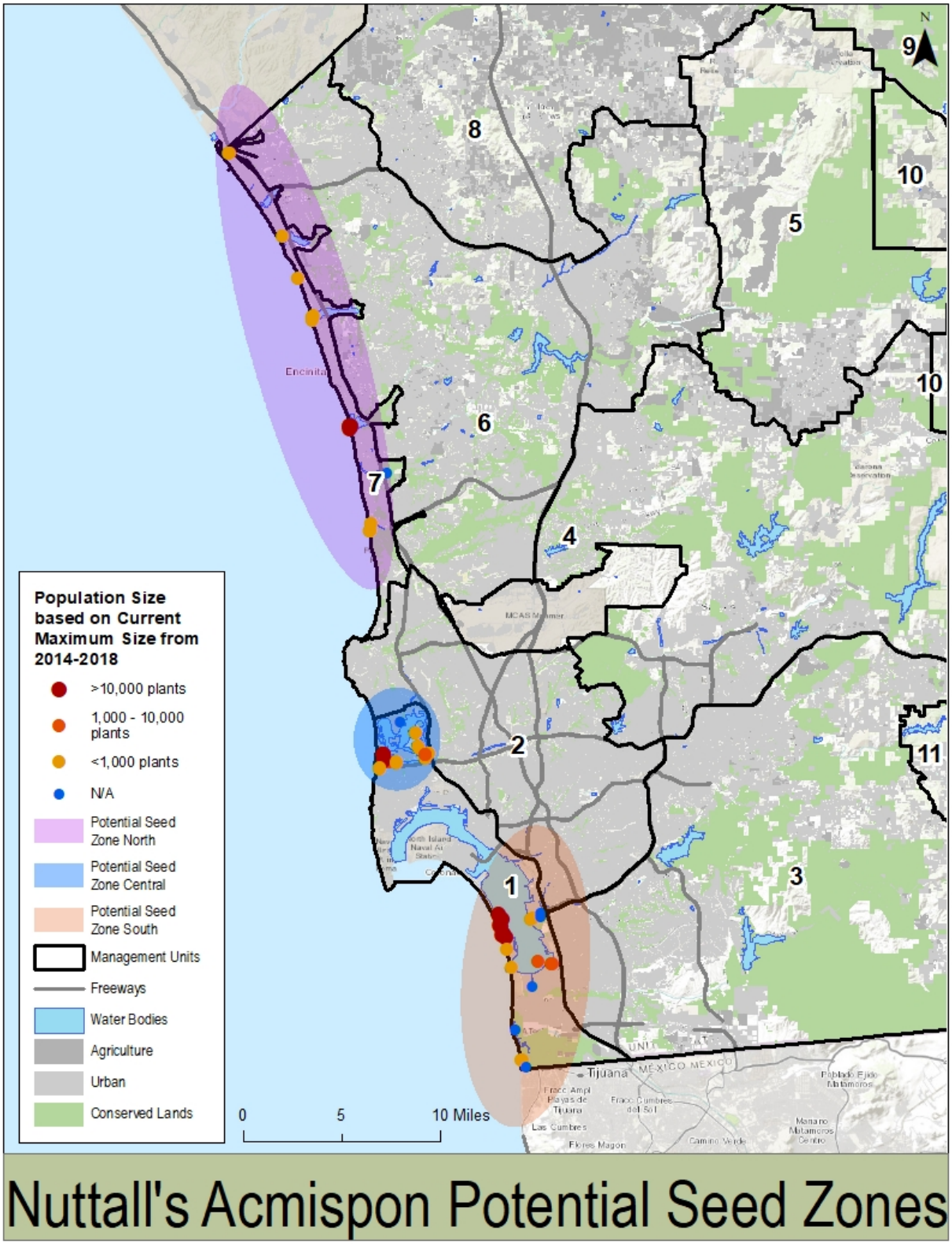

Figure 5.2-2. Nuttall's Acmispon: Seed Zones.

Table 5.2-2. Nuttall's Acmispon: Seed Zones.

Occurrence ID[1]	Occurrence Name	Seed Zone[2]	Occurrence Size[3]
ACPR_7AGHE024	Agua Hedionda	North	Small
ACPR_7BALA020	Batiquitos Lagoon	North	Small
ACPR_7CSPA018	San Elijo Lagoon	North	Large
ACPR_7SCSB025	South Carlsbad SB	North	Small
ACPR_7SLRR017	San Luis Rey River	North	Small
ACPR_7TPSR019	Torrey Pines SR (south)	North	Small
ACPR_7TPSR023	Torrey Pines SR (north)	North	Small
ACPR_1DUTR005	Dune Triangle	Central	Small
ACPR_1FIIS007	Mission Bay (Fiesta Island)	Central	Small
ACPR_1FIIS029	Fiesta Island	Central	Small
ACPR_1HOPO002	Mission Bay (Hospitality Point)	Central	Medium
ACPR_1MAPO004	Mission Bay (Mariner's Point)	Central	Large
ACPR_1NMLA001	Mission Bay (No Man's Land)	Central	Small
ACPR_1NOBE015	North Ocean Beach (Dog Beach)	Central	Small
ACPR_1RRSO003	Mission Bay (Rip Rap)	Central	Small
ACPR_1SOSH006	Mission Bay (east of South Shores)	Central	Medium
ACPR_1BFSP014	Border Field SP	South	Small
ACPR_1DSTR010	D Street Fill	South	Small
ACPR_1SBSA013	South Bay Salt Works	South	Medium
ACPR_1SSSB012	Silver Strand SB	South	Large
ACPR_1SSSB027	Silver Strand SB	South	Small
ACPR_1SSSB028	Silver Strand SB	South	Small

[1] Occurrence ID: Occurrence identification (ID) code per the San Diego Management and Monitoring Program's (SDMMP) Master Occurrence Matrix (MOM) database.

[2] Seed Zone: seed zones are defined in text and correspond to population groups in the F-RPMP. Limit seed movement to within the same seed zone. Shading indicates seed zone group.

[3] Occurrence size: refers to population size category based on multiple years of monitoring data. Recent size category as defined in the Framework-Rare Plant Management Plan (F-RPMP). **Small** = <1,000 individuals, **Medium** = 1,000-<10,000 individuals, **Large** = >10,000 individuals.

seed from another occurrence, the source (donor) occurrence should be located within the same seed zone as the target (recipient) occurrence. Within a seed zone, we recommend that seed transfer proceed between occurrences that are relatively close to one another, particularly in larger seed zones. The same limits on seed movement apply when using seed from a seed bank.

Collecting Plan

Refer to Section 4.1 of this document for guidelines on developing a sampling strategy for seed collection. Identify the purpose and type of collection needed for a specific project or restoration effort, as well as timeline and costs. In the section below, we highlight key steps in the process, including those specific to Nuttall's acmispon.

Sampling Strategy

Refer to Table 5.2-1 for prioritized seed-related activities for the target occurrence to identify whether seed is needed for a conservation collection, a restoration collection, or both.

For restoration, determine if seed is available in a seed bank to reintroduce directly into a target occurrence or for bulking. Proceed with the collecting plan if stored seed is not available, quantities are not sufficient, or the provenance is not appropriate for the target occurrence (e.g., different seed zone).

Assess whether the target occurrence will likely provide an adequate quantity of seed for restoration or if seed will need to be sourced from other occurrences in the seed zone. Refer to seed zones (Figure 5.2-2, Table 5.2-2) for limits on seed movement and seed sources. Contact the appropriate land managers of potential seed source occurrences to obtain permission for seed collection.

Based on the purpose and type of collection, and amount of seed expected to be available, determine whether the seed is likely to be collected along maternal lines or in bulk (Table 4.1-3), and set minimum targets for the number of plants to sample and amount of seed to collect. These targets are intended to provide a guide for planning, and may be modified based on the actual seed crop.

Permitting and Agreements

Ensure that all regulatory permits, memorandums, and access agreements are in place before collecting seed from an occurrence.

If unfamiliar with the collecting location, contact the land owner to obtain a map and coordinates of the occurrence, directions to the occurrence if necessary, gate codes, lock combinations, or keys, and any pertinent information about the occurrence (e.g., informing adjacent land owners, closures due to unsafe conditions).

Pre-collection Monitoring

Practitioners can collect Nuttall's acmispon seed as early as April and into early fall, depending on location, habitat, and weather and temperature conditions during the previous fall, winter and spring. Visit the target occurrence multiple times to determine the best time to collect seed. Time the first visit when plants are in full flower and easier to locate. Talk to land owners and managers and refer to monitoring data and literature to determine the range of phenological variation for an occurrence to plan the first site visit.

Use pin flags to demarcate the boundary of the occurrence if plants are located in coastal scrub habitat, because relocating plants after they dry can be difficult. If necessary, place pin flags adjacent to a subset of plants to relocate them easily during subsequent visits. Return to the occurrence within two to four weeks to check phenology. Weather conditions will determine the rate of desiccation; plants will dry quicker in warm and dry conditions and slower if conditions remain cool and moist. Continue monthly site visits until the majority of plants in an occurrence are fully desiccated and seeds resemble those in Figure 5.2-1.

Voucher Specimens

Collect voucher specimens of Nuttall's acmispon if none exist for the source occurrence and if enough plants are available so that collecting a few specimens does not adversely affect the occurrence. Check the San Diego Natural History Museum or Consortium of California Herbaria to determine if a voucher specimen already exists:

San Diego Natural History Museum: http://sdplantatlas.org/publicsearch.aspx

Consortium of California Herbaria: http://ucjeps.berkeley.edu/consortium/

Note that recently collected specimens may not yet be accessed into an herbarium's collection. In addition, older specimens may not include sufficient locality information to definitively determine whether they were collected at the occurrence. If it is not appropriate to collect a voucher specimen, photograph the occurrence (landscape aspect) and an individual plant while in flower, and then again when the plant is in fruit.

Obtain all necessary permits and memorandums before collecting voucher specimens of endangered, threatened, and rare species. Collect specimens while plants are still in flower and before collecting seed. Refer to Section 4.1 of this document (Seed Collecting, Materials and Methods) and instructions and forms in Appendix B to guide your collection. Submit the voucher specimen(s) to the San Diego Natural History Museum as soon as possible.

Methods and Materials

Refer to Section 4.1 of this document for a detailed discussion of methods and materials and Table 4.1-4 for a comprehensive list of recommended equipment for collecting seed. At a minimum, bring the following to each Nuttall's acmispon collection location:

- Regulatory permits and access agreements
- Directions, map(s)
- Lock combination(s), key(s)
- Collecting strategy/instructions
- GPS unit (preferably, sub-meter)
- Camera
- Labels, markers, pens or pencils
- Seed collection forms
- Hand lens
- Adhesive tape
- Safety razor or pocketknife
- Container and water (optional)
- Scissors, snips, sieve (optional)
- Small paper envelopes or bags

Before collecting seed, map the seed collection area(s), record data on the seed collection form(s) and count or estimate the number of plants within an occurrence. Collect no more than 5-10% of the seed from an individual plant or from the reproductive population per season. Use the following method to determine the number of seeds to collect by plant and for an occurrence:

Step 1: Select 5 plants of different sizes spaced randomly across the occurrence.

Step 2: Count the number of fruits on the 5 sample plants.

Step 3: Divide the total number of fruits counted (Step 2) by the number of plants sampled (Step 1) to calculate the *average number of fruits per plant*.

Step 4: Multiply the average number of fruits per plant (Step 3) by 2 (2 seeds/fruit) to calculate the *average number of seeds per plant*.

Step 5: Multiply the average number of seeds per plant (Step 4) by the occurrence count or estimate to calculate the *estimated number of seeds per occurrence*. In most cases, the amount of seed available for collecting will be higher in years with

optimal growing conditions (e.g., average or above-average rainfall) and lower during drought years.

Step 6: Use the numbers above to calculate the maximum number of fruits or seeds to collect to stay within the 5-10% collecting guidelines.

Note that where seed production is very heavy, seed collectors may choose to estimate fruit and seed quantities using subsamples.

Collect fruits when Nuttall's acmispon plants are fully dry. Remove dry fruits from the stems by hand and place them in a seed collecting envelope or bag. Refer to Figure 5.2-1 for ripe seeds. Fruits may be ready to collect as early as one month after peak flowering.

Collect seed during early, mid, and late season to account for differences in maturity times and to capture the phenological variability among the occurrence. Collect seed randomly and evenly throughout the occurrence, taking care to collect from plants of various sizes and with variable inflorescences, as well as plants growing in unique or varying habitat conditions.

The San Diego Audubon Society collects Nuttall's acmispon seed by gathering the soil and duff directly beneath senesced plants (Redfern and Flaherty 2018). They place the collected material in bags and save it for restoration purposes. They have had successful recruitment of Nuttall's acmispon using this method. This method does not allow for maternal line collections or an estimate of the percentage of seed collected per plant or occurrence, and is most appropriate where there are limitations on collecting seed at the optimal time.

Depending on the site, collect seed outside of the nesting season for the California least tern (*Sternula antillarum*) and avoid any other conflicts between collecting seed and sensitive resources. In some cases, it may be necessary to collect and bag pods before they are fully ripe.

Consider harvesting more seed than recommended by the CPC for small occurrences that are at serious risk of extirpation to capture the highest amount of genetic variation at the occurrence (McMillan 2019).

Ideally, collect seed from 30-50 plants across the occurrence. If the number of plants present in a given year is below this number, then sample over multiple years to produce a genetically robust seed collection for restoration purposes. Where the occurrence is small and not expected to produce an adequate number of plants (and seed) in any given year, consider collecting seed from one or more occurrences within the same seed zone. Depending on project objectives and sampling strategy, collect seed along maternal lines or as a bulk collection.

Interim Seed Storage and Delivery

Refer to the general section of this document (Seed Collecting) for guidelines on interim seed storage and delivery. We provide additional guidelines for practitioners that choose to keep the seed for future restoration purposes rather than sending it immediately to a storage facility:

- Do not clean and sort seed unless the collection is very small since cleaning seed is time-consuming and often difficult, especially with large amounts of seed.
- Loosely pack the seed collection in envelopes or bags and store them in dark, dry, and moderate to cool conditions for several months before distributing seed onsite.
- If planning to hold seed for longer than one year, store the seed at a recognized seed bank (i.e., CBG, SDZG) to ensure proper storage conditions to retain viability.

Seed Banking

Existing Conservation and Restoration Collections

Refer to Table 5.2-3 for existing seed collections of Nuttall's acmispon. This table includes only acmispon occurrences with seed in storage, including some occurrences that are on lands not included in the MSPA (e.g., military lands). For planning purposes, we indicate the seed zone for each collection. We also indicate the number of seeds present in the seed bank and their potential availability for restoration. Note that only a portion of the seeds in a collection will be available for these purposes. Existing permits or contracts may also limit the use of some collections. We obtained information on each collection (seed bank, accession number and year, and seed quantity, provenance, and availability) from seed bank managers. Refer to Table 4.2-3 for contact information for these seed banks; we encourage land managers to contact facilities directly to get more information on seed availability or to request seed.

Best Management Practices

Seed Cleaning

Wall and MacDonald (2009) suggest the following methods to clean and sort seeds of *Acmispon* spp.:

- Rub floral material over a medium screen to separate fruits from other plant material.
- Rub fruits over a sieve or rubber mat with a padded wooden block to open fruits and release seed.
- Shake released seed through a series of sieves to remove extraneous plant material.

Table 5.2-3. Nuttall's Acmispon: Exiting Seed Collections.

Occurrence ID[1]	Seed Zone[2]	Seed Bank[3]	Accession Number[4]	Collection Year	Seed Quantity[5]	Provenance Type[6]	Seed Available[7]
ACPR_1HOPO002	Central	SDZG	S0671	2016	~740	W	Yes
ACPR_1NOBE015	Central	SDZG	S0670	2016	~1,100	W	Yes
ACPR_7CSPA018	North	CBG	24216-5446	2015	4,877	W	Limited
North Island Naval Air Base)	South[8]	CBG	19341-2239	1996	132	W	No
EO#17 (North Island)	South[8]	SDZG	S0802	2019	Unknown	W	Limited
EO#3 (Silver Strand)	South	SDZG	S0803	2019	Unknown	W	Limited

[1] Occurrence identification (ID) per the SDMMP's MOM database. If no occurrence ID exists for a location, we provide the name of the location or the CNDDB element occurrence (EO) number, as indicated on collection in seed bank.

[2] Seed Zone: Seed zone corresponds to the three population groups identified in the F-RPMP: North, Central, and South. Occurrences in are placed in seed zones based on location.

[3] Seed bank: **CBG** = California Botanic Garden, **SDZG** = San Diego Zoo Global.

[4] Accession number = accession number of seed collection assigned by seed bank.

[5] Seed quantity indicates the number of seeds currently in the collection, according to seed bank records. Unknown indicates seed has not yet been processed and counted.

[6] Provenance type indicates the source location of the seed collection: **W** = seed collected directly from the wild (origin known),

[7] Seed available: **Yes** = seed is available for restoration use. **No** = seed is not available for restoration use, **Limited** = amount of seed available for restoration use is ≤400 seeds.

[8] The North Island Naval Air Station lies between the South and Central seed zones and was not included in a population group in the F-RPMP or seed zone in this document because of the absence of monitoring data. Based on location, it would likely be placed into the South seed zone.

- Use a blower at low speed to separate chaff from seed, and at a higher speed to separate hollow seed from filled seed.

Wall and MacDonald (2009) indicate that cleaning seed of *Acmispon* spp. ranges from moderate to high in difficulty, and caution that seeds can be broken during the cleaning process.

Seed Testing

Seed testing methods will vary by facility, but all seed banks conduct baseline germination tests on new accessions and then conduct follow up testing to check seed for viability.

At CBG, baseline tests for seed germination use a typical seed sample of 50 seeds, although sample size may vary depending on the size of the seed collection, and variable methods to break dormancy (if needed). They conduct follow-up testing on seeds in storage at intervals (e.g., 1 year, 5 years, 10 years, 20 years, and every subsequent 10 years thereafter, as seed quantities allow).

CBG conducted germination test results for two seed samples of Nuttall's acmispon from ACPR_7CSPA018. They conducted the first test on fresh seed and the second on seed that had been frozen for about a year. Both samples were pre-treated by placing seed in boiling water and then soaking the seed for 24 hours in the cooling water. Seeds were grown in a germination chamber at temperatures of 20°C during the day and 12°C at night. Germination was 70% for the fresh seed and 88% for the frozen seed.

At SDZG, a baseline germination test is conducted on all new accessions. The test is conducted primarily on fresh seed that has been scarified. Follow up testing is conducted on frozen seed after 1, 5, and 10 years in storage. Seed is sterilized with bleach and placed in reverse osmosis water at room temperature for 24 hours to fully imbibe (absorb) water. Seed is then plated onto agar trays (10 seeds per tray) and observed for signs of germination. The seed is counted as germinated when the radical emerges. SDZG does not use a cut test to determine viability. Rather, they provide abundant time for seeds to germinate (Anderson pers. comm.).

Seed Storage

All seed banks contacted store Nuttall's acmispon seed according to CPC guidelines. For example, SDZG dries seeds down to 23-35% moisture content. Seed is then stored in sealed, foil-lined envelopes in the freezer at a low temperature (-18°C to -23°C).

Each collection is separated into three packages that represent the primary, curation, and duplicate collections, as described in Section 4.1. The primary collection is the active seed collection and seeds in this package are available for research, testing, restoration, and other uses. The curation package is the base collection for long-term storage. This package is not opened and is not available for other uses. It constitutes the long-term conservation collection.

The duplicate collection is sent to another seed bank facility as backup storage to protect against catastrophic loss at the primary storage facility. Both SDZG and CBG send their duplicate seed collections to the NLGRP.

Seed Bulking

Seed Selection

Refer to Section 4.3 for guidelines on seed selection to maximize the quality of the bulked sample. Key considerations include:

- Use seed for bulking from the target occurrence or another occurrence in the same seed zone as the target occurrence. For very small occurrences (fewer than 100 plants), use seed from a large occurrence (if present) or from one or more smaller occurrences (composite provenancing) within the same seed zone.
- Use seed that was collected randomly and uniformly.
- Use an adequate amount of seed for bulking (ideally, ≥1,000 seeds or seed from 30-50 plants).
- For very small seed samples, use seed collected along maternal lines.

Best Management Practices

Germination and Propagation Methods

Nuttall's acmispon seed requires pre-treatment to germinate readily. CBG found that softening the seed coat with a boiling water soak resulted in germination rates of >70% in fresh and frozen seed. After sowing, water the seeded flats or container pots several times a week to stimulate germination and continue watering, through flowering, until plants begin to senesce.

Limits on Generations

At this time, we recommend bulking Nuttall's acmispon seed for only one generation (F1) in a nursery setting. If additional seed is needed for restoration, start with wild seed rather than the F1 generation.

Documentation

We encourage growing facilities and others bulking seed of Nuttall's acmispon for reintroduction purposes to document bulked samples as indicated in Section 4.3 and provide this information to land managers when seed is delivered. Likewise, we encourage land managers to provide information on seed collection and seed reintroductions to the SDMMP (contact information at https://sdmmp.com/about.php#contact) and include relevant information on IMG monitoring

forms (i.e., translocated occurrence). Refer to Table 5.2-4 for key information for bulked seed samples.

Table 5.2-4. Documentation for Bulked Seed Samples.

Key Information	Primary Responsibility
Seed Source	Seed Collector and/or Land Manager
Seed Collection Date	Seed Collector
Amount of Seed Bulked	Growing Facility
Propagation Method	Growing Facility
Propagation Conditions	Growing Facility
Harvesting Date(s)	Growing Facility
Post-harvest Storage Location/Duration/Conditions	Growing Facility
Generation of Bulked Sample	Growing Facility
Receptor Site Location	Growing Facility and/or Land Manager

5.3 SALT MARSH BIRD'S-BEAK (*CHLOROPYRON MARITIMUM* SSP. *MARITIMUM*)

Seed Collection

Seed Characteristics

Salt marsh bird's-beak plants produce loose to dense spike-like inflorescences that contain many flowers. Each flower produces 15-20 seeds that are approximately 2 millimeters (mm) in size. Seeds are brown to tan, kidney-shaped, and deeply netted (Figure 5.3-1).

Seeds are buoyant and may disperse primarily by floating in water, although animals may disperse seed, as well (USFWS 1985). Seed production is highly variable and affected by many factors, including insect pollinators, seed predators, and fungal diseases (USFWS 1985). Seed predators known to drastically and adversely affect seed production include larvae of the leaf roller moth (*Platynota stultana*), geranium plume moth (*Amblyptilia pica*), salt marsh plume moth (*Liphographus fenestrella),* and salt marsh leaf roller moth (*Saphenista* sp.) (USFWS 2013, Anderson 2019a).

Figure 5.3-1. Salt Marsh Bird's-beak: Seed
(Photo credit: John MacDonald, RSA)

Depending on climatic conditions, salt marsh bird's-beak plants can bloom from late May through mid-December; thus, seed production may begin in late spring and continue through winter as plants senesce (Figure 5.3-2). Seed production increases with plant size and flower production as long as the necessary insect pollinators are present.

Figure 5.3-2. Salt Marsh Bird's-beak: Dried Plants (arrow points to plant) (Photo credit: Heather Schneider, SBBG)

Priority Occurrences

Table 5.3-1 prioritizes occurrences of salt marsh bird's-beak on conserved lands in the MSPA for seed-related management actions. These priorities were developed in the F-RPMP based on monitoring data and specific studies, including genetic analyses. The decision to implement an action will depend on a number of factors, including status of the occurrence (stable or declining), habitat condition, threats management, and funding, among others.

In general, large occurrences are prioritized as potential seed sources, subject to land manager and/or agency approval. Small occurrences are high priority candidates for seed reintroduction, subject to response to management and site conditions, as discussed in the F-RPMP.

Currently, conservation seed collections exist in established seed banks for three salt marsh bird's-beak occurrences (30%) on conserved lands in the MSPA (Table 5.3-1). Seed banks also hold two additional seed collections without occurrence numbers.

We recommend conservation collections as a potential management action for all occurrences that are not currently represented in a seed bank *if adequate seed is available*. For occurrences where plants have not been detected recently, a conservation collection will be possible only if plants are detected in the future (Table 5.3-1). Large occurrences are the highest (or most immediate) priorities for seed conservation to maximize the amount of genetic diversity in long-term storage (e.g., COMAM3_1TIES002, COMAM3_1TISO011).

We recommend restoration collections as a potential management action for occurrences that are high or medium priorities for seed reintroduction as indicated in Table 5.3-1 and identified in the F-RPMP. We do not prioritize occurrences for seed reintroduction if they have not yet been monitored.

Table 5.3-1. Salt Marsh Bird's-beak: Management Priorities for Seed-related Activities.

Occurrence ID[1]	Occurrence Name	Occurrence Size[2]	Genetic Structure[3]	Seed Reintroduction Priority[4]	Existing Seed Collection[5]	Seed-related Activities[6]			
						Seed Source	Conservation Collection	Restoration Collection	Seed Bulking
COMAM3_1DOBE007	Dog Beach	Large	L+L+L	---	SDZG	✓			
COMAM3_1IMBE008	Camp Surf	Medium	L+L+L				✓		
COMAM3_1SDBA004	San Diego Bay, Naval Radar Receiving Facility, Naval Base Coronado	Small	L+H+L	High			✓	✓	✓
COMAM3_1SWMA005	Sweetwater Marsh (west of I-5 and north of Sweetwater River)	Large	L+L+L	---	CBG, SDZG[7]	✓			
COMAM3_1TIES001	Tijuana Estuary Area (at Boundary Monument #258)	Small	L+Hr+L				✓		
COMAM3_1TIES002	Tijuana Estuary Area (between mouth of Tijuana River & Coronado Avenue, Imperial Beach)	Large	L+Hr+L			✓	✓		
COMAM3_1TIES003	Tijuana Estuary Area (near mouth of Tijuana River and north part of Border Field State Park)	Small	L+Hr+L*	High			✓	✓	✓
COMAM3_1TIES009	Tijuana Slough	Small	L+Hr+L*	High	CBG, SDZG			✓	✓
COMAM3_1TISO010	Tijuana Slough NWA #2	Medium	L+Hr+L*				✓		
COMAM3_1TISO011	Tijuana Slough NWA #3	Large	L+Hr+L*	---		✓	✓		

[1] Occurrence ID: Occurrence identification (ID) code per the San Diego Management and Monitoring Program's (SDMMP) Master Occurrence Matrix (MOM) database.
[2] Occurrence Size: Recent size category as defined in the Framework-Rare Plant Management Plan (F-RPMP). **Small** = <1,000 individuals, **Medium** = 1,000-<10,000 individuals, **Large** = >10,000 individuals.
[3] Genetic Structure (per Milano and Vandergast 2018): Genetic differentiation + genetic diversity + inbreeding. **L** = low, **H** = High, **Hr** = Higher, * = some high relatedness. Refer to F-RPMP for a discussion of the implications for seed movement.
[4] Seed Reintroduction Priority (per F-RPMP): **High** = high priority for seed reintroduction. If no priority level is indicated, then management action is not a priority at this time, but may be undertaken at the discretion of the land manager and subject to adequate seed availability.
[5] Existing Seed Collection: Indicates location of collection. **CBG** = California Botanic Garden, SDZG = San Diego Zoo Global.

[6] Seed-related Activities: ✓ indicates occurrence is suitable (e.g., seed source) pending authorization from responsible entities, or action is warranted. In some cases, additional collecting for restoration may not be necessary if enough seed is available in an existing seed collection. Likewise, seed bulking may or may not be necessary depending on amount of seed available in existing collection or collected specifically for restoration purposes.

[7] Both seed banks (CBG, SDZG) list their collections from this location as COMAM3_1SWMA005. However, occurrence -005 was subsequently combined with occurrence -006. We now consider -005 and -006 to be the same occurrence, which is referenced as -006 in the IMG monitoring program.

We recommend reintroducing seed only if threats are controlled, habitat is likely to support this species in the future, and funding is available for short- and long-term management. Reintroducing seed is most appropriate for small occurrences that continue to decline, even with management. Seed may be introduced into extirpated occurrences if threats are managed and the habitat is likely to support salt marsh bird's-beak in the future. For extirpated occurrences, source seed from a large occurrence in the same seed zone or from an appropriate seed source in storage (if available). Some threats (particularly, invasive plants) will likely require perpetual management; thus, ensure that adequate funding and/or labor are available to manage the occurrence after seed is reintroduced.

We recommend seed bulking as a potential management action for all occurrences identified as potentially requiring a restoration collection. The need for seed bulking will depend on the amount of seed available in storage and/or collected specifically for restoration, as well as the sowing strategy. For example, bulking will not be needed if adequate seed is collected to sow directly into the target occurrence.

Seed Zones

We established one seed zone for salt marsh bird's-beak to guide seed collecting, banking, and bulking efforts. This seed zone corresponds to the genetic cluster and regional population group identified for this species in the F-RPMP. Figure 5.3-3 shows the locations of the seed zone; Table 5.3-2 lists all occurrences in the seed zone.

Seed zones place geographic limits on the transfer of seed between occurrences. *We do not recommend transfer of seed beyond the seed zone at this time* (e.g., between occurrences in San Diego County and counties to the north). Thus, if a small occurrence requires seed from another occurrence, the source (donor) occurrence should be located within the same seed zone as the target (recipient) occurrence. Within a seed zone, we recommend that seed transfer proceed between occurrences that are relatively close to one another. We designate subgroups (Table 5.3-2) to identify occurrences in proximity. The same limits on seed movement apply when using seed from a seed bank for restoration.

Collecting Plan

Refer to Section 4.1 of this document for guidelines on developing a sampling strategy for seed collection. Identify the purpose and type of collection needed for a specific project or restoration effort, as well as timeline and costs. In the section below, we highlight key steps in the process, including those specific to salt marsh bird's-beak.

Sampling Strategy

Refer to Table 5.3-1 for prioritized seed-related activities for the target occurrence to identify whether seed is needed for a conservation collection, a restoration collection, or both.

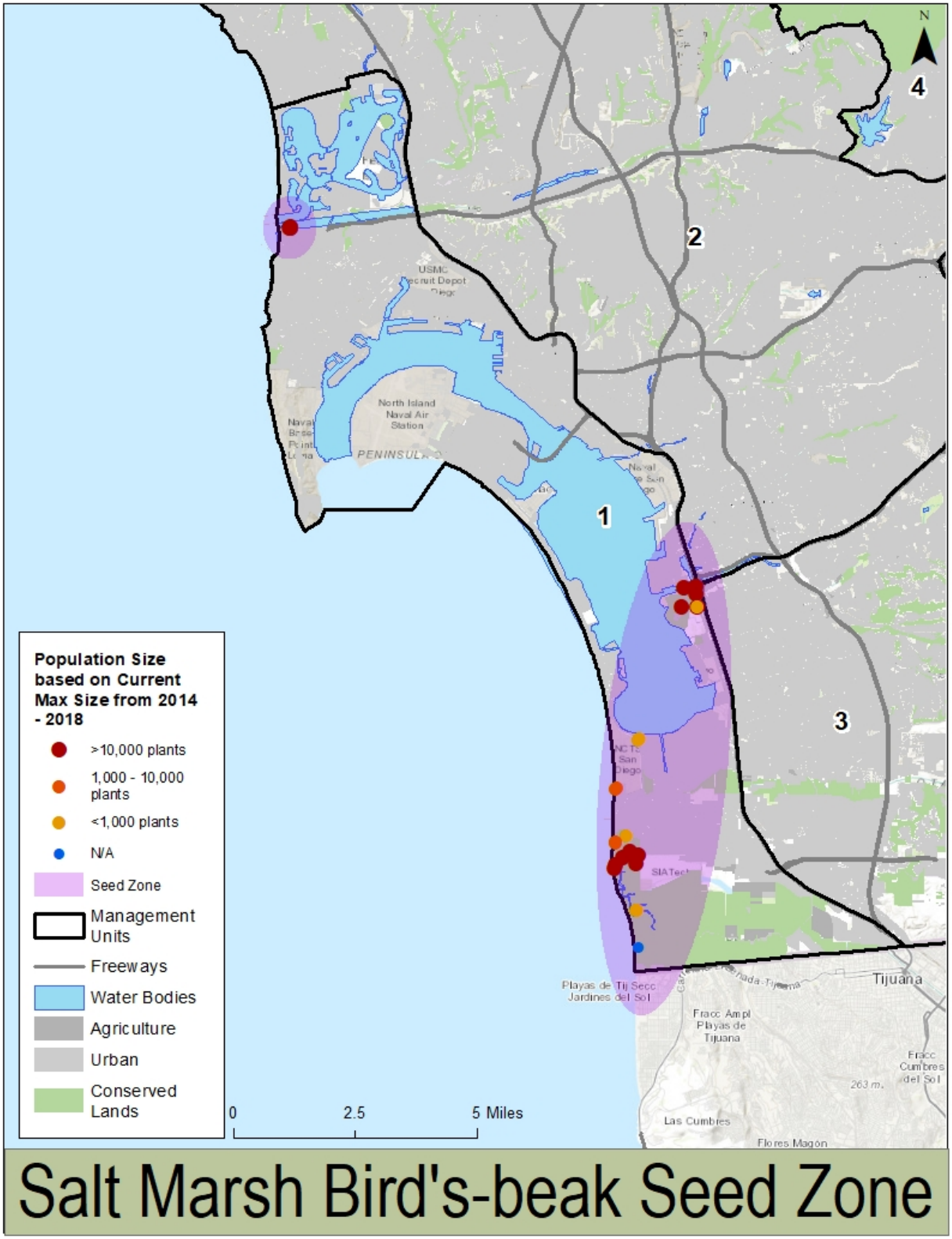

Figure 5.3-3. Salt Marsh Bird's-beak: Seed Zones.

Table 5.3-2. Salt Marsh Bird's-beak: Seed Zones.

Occurrence ID[1]	Occurrence Name	Seed Zone[2]	Seed Zone Subgroup[3]	Occurrence Size[4]
COMAM3_1DOBE007	Dog Beach	South	1	Large
COMAM3_1IMBE008	Camp Surf	South	2	Medium
COMAM3_1SDBA004	San Diego Bay, Naval Radar Receiving Facility, Naval Base Coronado	South	2	Small
COMAM3_1SWMA005	Sweetwater Marsh (west of I-5 and north of Sweetwater River)	South	2	Large
COMAM3_1TIES001	Tijuana Estuary Area (at Boundary Monument #258)	South	2	Small
COMAM3_1TIES002	Tijuana Estuary Area (between mouth of Tijuana River & Coronado Avenue, Imperial Beach)	South	2	Large
COMAM3_1TIES003	Tijuana Estuary Area (near mouth of Tijuana River and north part of Border Field State Park)	South	2	Small[5]
COMAM3_1TIES009	Tijuana Slough	South	2	Small
COMAM3_1TISO010	Tijuana Slough NWA #2	South	2	Medium
COMAM3_1TISO011	Tijuana Slough NWA #3	South	2	Large

[1] Occurrence ID: Occurrence identification (ID) code per the San Diego Management and Monitoring Program's (SDMMP) Master Occurrence Matrix (MOM) database.
[2] Seed Zone: seed zones are defined in text and correspond to genetic clusters and population groups in the F-RPMP. Limit seed movement to within the same seed zone.
[3] Seed Zone Subgroups are defined in text and correspond to population subgroups in the F-RPMP. Where feasible, limit seed movement to the same subgroup within a population group.
[4] Occurrence size: refers to size category based on multiple years of monitoring data. Recent size category as defined in the Framework-Rare Plant Management Plan (F-RPMP). **Small** = <1,000 individuals, **Medium** = 1,000-<10,000 individuals, **Large** = >10,000 individuals.
[5] Indicates occurrences where monitoring indicates a decline in population size category over time. Refer to F-RPMP for additional details.

For restoration, determine if seed is available in a seed bank to reintroduce directly into a target occurrence or for bulking. Proceed with the collecting plan if stored seed is not available, quantities are not sufficient, or the provenance is not appropriate for the target occurrence (e.g., different seed zone).

Assess whether the target occurrence will likely provide an adequate quantity of seed for restoration or if seed will need to be sourced from other occurrences in the seed zone. Refer to seed zones (Figure 5.3-3, Table 5.3-2) for limits on seed movement and seed sources. Contact the appropriate land managers of potential seed source occurrences to obtain permission for seed collection.

Based on the purpose and type of collection, and amount of seed expected to be available, determine whether the seed is likely to be collected along maternal lines or in bulk (Table 4.1-3), and set minimum targets for the number of plants to sample and amount of seed to collect. These targets are intended to provide a guide for planning, and may be modified based on the actual seed crop.

Permitting and Agreements

Ensure that all regulatory permits, memorandums, and access agreements are in place before collecting seed from an occurrence.

If unfamiliar with the collecting location, contact the land owner to obtain a map and coordinates of the occurrence, directions to the occurrence if necessary, gate codes, lock combinations, or keys, and any pertinent information about the occurrence (e.g., informing adjacent land owners, closures due to unsafe conditions).

Pre-collection Monitoring

Practitioners can collect salt marsh bird's-beak seed as early as July and into winter, depending on location, habitat, and weather and temperature conditions during the previous fall, winter, and spring. Visit the target occurrence multiple times to determine the best time to collect seed. Time the first visit when salt marsh bird's-beak is in full flower to make it easier to locate. Talk to land owners and managers and refer to monitoring data and literature to determine the range of phenological variation for an occurrence to plan the first site visit.

Use pin flags to demarcate the boundary of the occurrence and place pin flags adjacent to salt marsh bird's-beak individuals to relocate them easily during subsequent visits. Return to the occurrence within two to four weeks to check phenology. Weather conditions will determine the rate of desiccation; plants will dry quicker in warm and dry conditions and slower if conditions remain cool and moist. Continue monthly site visits until the majority of plants in an occurrence are fully desiccated and seeds resemble those in Figure 5.3-2.

Voucher Specimens

Collect voucher specimens of salt marsh bird's-beak if none exist for the source occurrence and if enough plants are available so that collecting a few specimens does not adversely affect the occurrence. Check the San Diego Natural History Museum or Consortium of California Herbaria to determine if a voucher specimen already exists:

San Diego Natural History Museum: http://sdplantatlas.org/publicsearch.aspx

Consortium of California Herbaria: http://ucjeps.berkeley.edu/consortium/

Note that recently collected specimens may not yet be accessed into an herbarium's collection. In addition, older specimens may not include sufficient locality information to definitively determine whether they were collected at the occurrence. If it is not appropriate to collect a voucher specimen, photograph the occurrence (landscape aspect) and an individual plant while in flower, and then again when the plant is in fruit.

Obtain all necessary permits and memorandums before collecting voucher specimens of endangered, threatened, and rare species. Collect specimens while plants are still in flower and before collecting seed. Refer to Section 4.1 of this document (Seed Collecting, Materials and Methods) and instructions and forms in Appendix B to guide your collection. Submit the voucher specimen(s) to the San Diego Natural History Museum as soon as possible.

Methods and Materials

Refer to Section 4.1 of this document for a detailed discussion of methods and materials and Table 4.1-4 for a comprehensive list of recommended equipment for collecting seed. At a minimum, bring the following to each salt marsh bird's-beak collection location:

- Regulatory permits and access agreements
- Directions, map(s)
- Lock combination(s), key(s)
- Collecting strategy/instructions
- GPS unit (preferably, sub-meter)
- Camera
- Labels, markers, pens or pencils
- Seed collection forms
- Hand lens
- Adhesive tape
- Safety razor or pocketknife

- Container and water (optional)
- Scissors, snips
- Small paper envelopes or bags

Before collecting seed, map the seed collection area(s), record data on the seed collection form(s) and count or estimate the number of plants within an occurrence. Collect no more than 5-10% of the seed from an individual plant or from the reproductive population per season. Use the following method to determine the number of seeds to collect by plant and for an occurrence:

Step 1: Select 5 plants of different sizes spaced randomly across the occurrence.

Step 2: Select 2-3 inflorescences per plant and count the number of seeds per inflorescence.

Step 3: Divide the total number of seeds counted (Step 2) by the number of sampled inflorescences (Step 2) to calculate the *average number of seeds per inflorescence*.

Step 4: Count or estimate the number of inflorescences on each sampled plant. Divide the total number of inflorescences by the number of plants sampled (Step 1) to calculate the *average number of inflorescences per plant*.

Step 5: Multiply the average number of seeds per inflorescence (Step 3) by the average number of inflorescences per plant (Step 4) to obtain an *average number of seeds per plant*.

Step 6: Multiply the average number of seeds per plant (Step 5) by the occurrence count or estimate to calculate the estimated *number of seeds per occurrence*. In most cases, the amount of seed available for collecting will be higher in years with optimal growing conditions (e.g., average or above-average rainfall) and lower during drought years.

Step 7: Calculate the maximum number of fruits or seeds to collect per plant and for an occurrence to stay within the 5-10% collecting guidelines.

Collect seeds when salt marsh bird's-beak plants are fully dry. At this stage, plants will be tan to brown with no green parts showing (Figure 5.3-2), and seeds will be hard, kidney-shaped, netted, and tan to light brown (Figure 5.3-4). Fruits may be ready to collect as early as one month after peak flowering. Remove dry fruits from the stems by hand and place them in a seed collecting envelope or bag.

Figure 5.3-4. Salt Marsh Bird's-beak: Seed Detail
(Photo credit: Heather Schneider, SBBG)

Collect seed during early, mid, and late season to account for differences in maturity times and to capture the phenological variability among the occurrence. Collect seed randomly and evenly throughout the occurrence, taking care to collect from plants of various sizes and with variable inflorescences, as well as plants growing in unique or varying habitat conditions.

Consider harvesting more seed than recommended by the CPC for small occurrences that are at serious risk of extirpation to capture the highest amount of genetic variation at the occurrence (McMillan 2019). In this case, coordinate with the regulatory agencies prior to the collecting effort.

Ideally, collect seed from 30-50 plants across the occurrence. If the number of plants present in a given year is below this number, then sample over multiple years to produce a genetically robust seed collection for restoration purposes. Where the occurrence is small and not expected to produce an adequate number of plants (and seed) in any given year, consider collecting seed from one or more occurrences within the same seed zone. Depending on project objectives and sampling strategy, collect seed along maternal lines or as a bulk collection.

Interim Seed Storage and Delivery

Refer to the general section of this document (Seed Collecting) for guidelines on interim seed storage and delivery. We provide additional guidelines for practitioners that choose to keep the seed for future restoration purposes rather than sending it immediately to a storage facility:

- Do not clean and sort seed unless the collection is very small since cleaning seed is time-consuming and often difficult, especially with large amounts of seed.
- Loosely pack the seed collection in envelopes or bags and store them in dark, dry, and moderate to cool conditions for several months before distributing seed onsite.
- If planning to hold seed for longer than one year, store the seed at a recognized seed bank (i.e., CBG, SDZG) to ensure proper storage conditions to retain viability.

Seed Banking

Existing Conservation and Restoration Collections

Refer to Table 5.3-3 for existing seed collections of salt marsh bird's-beak. This table includes only bird's-beak occurrences with seed in storage. For planning purposes, we indicate the seed zone for each collection. We also indicate the number of seeds present in the seed bank and their potential availability for restoration. Note that only a portion of the seeds in a collection will be available for these purposes. Existing permits or contracts may also limit the use of some collections. We obtained information on each collection (seed bank, accession number and year, and seed quantity, provenance, and availability) from seed bank managers. Refer to Table 4.2-3 for contact information for these seed banks; we encourage land managers to contact facilities directly to get more information on seed availability or to request seed.

Best Management Practices

Seed Cleaning

Wall and MacDonald (2009) suggest the following methods to clean and sort seeds of salt marsh bird's-beak:

- Rub dried floral material over a medium screen to open capsules and release seeds.
- Sort material through sieves several times to remove large chaff.
- Use a blower at low speed to separate chaff and sterile seed from viable seed.

Wall and MacDonald (2009) indicate that cleaning seed of this species is moderately difficult and may require some hand-cleaning.

Seed Testing

Seed testing methods will vary by facility, but all seed banks conduct baseline germination tests on new accessions and then conduct follow up testing to check seed for viability.

At CBG, baseline tests for seed germination use a typical seed sample of 50 seeds, although sample size may vary depending on the size of the seed collection, and variable methods to break dormancy (if needed). They conduct follow-up testing on seeds in storage at intervals (e.g., 1 year, 5 years, 10 years, 20 years, and every subsequent 10 years thereafter, as seed quantities allow).

Table 5.3-3. Salt Marsh Bird's-beak: Exiting Seed Collections.

Occurrence ID[1]	Seed Zone[2]	Seed Bank[3]	Accession Number[4]	Collection Year	Seed Quantity[5]	Provenance Type[6]	Seed Available[7]
COMAM3_1DOBE007	South	SDZG	S0735	2017	~335	W	Yes
COMAM3_1SWMA006[8]	South	CBG	21772-3317	2005	41,209	W	Limited
COMAM3_1SWMA006[8]	South	SDZG	S0740	2017	~1,595	W	Yes
COMAM3_1TIES009	South	CBG	21771-3318	2005	4,929	W	Limited
COMAM3_1TIES009	South	SDZG	S0739	2017	~1,615	W	Yes
Sweetwater Marsh	South	SDZG	S0083	2005	~12,000	W	Yes
Tijuana Estuary	South	SDZG	S0082	2005	~15,000	W	Yes

[1] Occurrence identification (ID) per the SDMMP's MOM database. If no occurrence ID exists for a location, we provide the name of the location as indicated on collection in seed bank.
[2] Seed Zone: Seed zone corresponds to the one genetic cluster and population group identified in the F-RPMP: South.
[3] Seed bank: **CBG** = California Botanic Garden, **SDZG** = San Diego Zoo Global.
[4] Accession number = accession number of seed collection assigned by seed bank.
[5] Seed quantity indicates the number of seeds currently in the collection, according to seed bank records. Unknown indicates seed has not yet been processed and counted.
[6] Provenance type indicates the source location of the seed collection: **W** = seed collected directly from the wild (origin known).
[7] Seed available: **Yes** = seed is available for restoration use. **No** = seed is not available for restoration use, **Limited** = amount of seed available for restoration use is ≤400 seeds.
[8] For this occurrence, the number -006 was assigned in the early stages of IMG rare plant monitoring. With subsequent monitoring, SDMMP combined -005 and -006 based on distance. As a result, we addressed only -005 in the F-RPMP and earlier in this document because these two locations were considered to be a single occurrence. For restoration purposes, seed from -005 and -006 are considered the same seed source.

CBG has conducted a number of seed germination tests on salt marsh bird's-beak (https://www.rsabg.org/conservation/seed-conservation). Germination is relatively high in this species. CBG observed germination rates >70% on fresh seed, including 71% germination with no pre-treatment and 81% germination after soaking seed in water. Germination rates were somewhat lower on frozen seed (40-72%), although they appeared to increase over time. For example, 3-year old seed had a germination rate of 40% while 20-year old seed had a germination rate of 72% (Wall 2009).

At SDZG, a baseline germination test is conducted on all new accessions. The test is conducted primarily on fresh seed. Follow up testing is conducted on frozen seed after 1, 5, and 10 years in storage. Seed is sterilized with bleach and placed in reverse osmosis water at room temperature for 24 hours to fully imbibe (absorb) water. Seed is then plated onto agar trays (10 seeds per tray) and observed for signs of germination. The seed is counted as germinated when the radical emerges. SDZG does not use a cut test to determine viability. Rather, they provide abundant time for seeds to germinate (Anderson pers. comm.).

Seed Storage

All seed banks contacted store salt marsh bird's-beak seed according to CPC guidelines. For example, SDZG dries seeds down to 23-35% moisture content. Seed is then stored in sealed, foil-lined envelopes in the freezer at a low temperature (-18°C to -23°C).

Each collection is separated into three packages that represent the primary, curation, and duplicate collections, as described in Section 4.1. The primary collection is the active seed collection and seeds in this package are available for research, testing, restoration, and other uses. The curation package is the base collection for long-term storage. This package is not opened and is not available for other uses. It constitutes the long-term conservation collection. The duplicate collection is sent to another seed bank facility as backup storage to protect against catastrophic loss at the primary storage facility. Both SDZG and CBG send their duplicate seed collections to the NLGRP.

Seed Bulking

Seed Selection

Refer to Section 4.3 for guidelines on seed selection to maximize the quality of the bulked sample. Key considerations include:

- Use seed for bulking from the target occurrence or another occurrence in the same seed zone as the target occurrence. For very small occurrences (fewer than 100 plants), use seed from a large occurrence (if present) or from one or more smaller occurrences (composite provenancing) within the same seed zone.
- Use seed that was collected randomly and uniformly.

- Use an adequate amount of seed for bulking (ideally, ≥1,000 seeds or seed from 30-50 plants).
- For very small seed samples, use seed collected along maternal lines.

Best Management Practices

Germination and Propagation Methods

Salt marsh bird's-beak seed germinates without pre-treatment although the seed does appear to possess some dormancy that is relieved with after-ripening, scarification, or vernalization (artificially cooling seeds to mimic cold temperatures and induce germination), or by germinating in water with lower levels of salinity (Zedler et al. 1992, USFWS 1985). Although seed germinates readily without pre-treatment, soaking seed in water for 24-72 hours may improve the germination rate (Zedler 2001 *in* Zahn 2015).

Limits on Generations

At this time, we recommend bulking salt marsh bird's-beak seed for only one generation (F1) in a nursery setting. If additional seed is needed for restoration, start with wild seed rather than the F1 generation.

Documentation

We encourage growing facilities and others bulking seed of salt marsh bird's-beak for reintroduction purposes to document bulked samples as indicated in Section 4.3 and provide this information to land managers when seed is delivered. Likewise, we encourage land managers to provide information on seed collection and seed reintroductions to the SDMMP (contact information at https://sdmmp.com/about.php#contact) and include relevant information on IMG monitoring forms (i.e., translocated occurrence). Refer to Table 5.3-4 for key information for bulked seed samples.

Table 5.3-4. Documentation for Bulked Seed Samples.

Key Information	Primary Responsibility
Seed Source	Seed Collector and/or Land Manager
Seed Collection Date	Seed Collector
Amount of Seed Bulked	Growing Facility
Propagation Method	Growing Facility
Propagation Conditions	Growing Facility
Harvesting Date(s)	Growing Facility
Post-harvest Storage Location/Duration/Conditions	Growing Facility
Generation of Bulked Sample	Growing Facility
Receptor Site Location	Growing Facility and/or Land Manager

5.4 OTAY TARPLANT (*DEINANDRA CONJUGENS*)

Seed Collection

Seed Characteristics

Otay tarplant inflorescences include both ray and disk flowers. Each inflorescence possesses 7-10 ray flowers and 13-21 disk flowers. Most fruits are produced by ray flowers. Baldwin et al. (2012) indicate that disk flowers are staminate (male) and do not typically produce seed. Where disk flowers do form fruits, however, they germinate more readily than fruit of ray flowers, possibly due to differences in the thickness of the seed coat (USFWS 2009, Bauder et al. 2002).

Otay tarplant fruits (achenes) are dry and indehiscent. Each achene contains one seed. The rough-textured, triangular to obovate seeds are brown, tan, and black (Figure 5.4-1). The short, papery pappus (modified calyx) remains attached to each seed and its role is unknown, but it may assist in animal- or possibly, wind-dispersal.

Figure 5.4-1. Otay Tarplant: Seed. (left: disk seeds with pappus, right: ray seeds) (Photo credit: John MacDonald, RSA

Otay tarplant seed forms in late spring and matures through summer as the plant senesces. Seed production increases with plant size and flower production, with larger plants producing hundreds of seeds. Fruits can remain on dry plants throughout the fall.

Priority Occurrences

Table 5.4-1 prioritizes occurrences of Otay tarplant on conserved lands in the MSPA for seed-related management actions. These priorities were developed in the F-RPMP based on monitoring data and specific studies, including genetic analyses. The decision to implement an action will depend on a number of factors, including status of the occurrence (stable or declining), habitat condition, threats management, and funding, among others.

In general, large occurrences are prioritized as potential seed sources, subject to land manager and/or agency approval. Small occurrences are high priority candidates for seed reintroduction, subject to response to management and site conditions, as discussed in the F-RPMP.

Currently, conservation seed collections exist in established seed banks for six Otay tarplant occurrences (22%) on conserved lands in the MSPA (Table 5.4-1). Seed banks also hold collections for an additional occurrence that is extirpated or not currently conserved.

We recommend conservation collections as a potential management action for all occurrences that are not currently represented in a seed bank *if adequate seed is available*. For occurrences where plants have not been detected recently, a conservation collection will be possible only if plants are detected in the future (Table 5.4-1). Large occurrences are the highest (or most immediate) priorities for seed conservation to maximize the amount of genetic diversity in long-term storage (e.g., DECO13_3JABO028, DECO13_3MMGR010).

We recommend restoration collections as a potential management action for occurrences that are high or medium priorities for seed reintroduction as indicated in Table 5.4-1 and identified in the F-RPMP. We do not prioritize occurrences for seed reintroduction if they have not yet been monitored.

We recommend reintroducing seed only if threats are controlled, habitat is likely to support this species in the future, and funding is available for short- and long-term management. Reintroducing seed is most appropriate for small occurrences that continue to decline, even with management. Seed may be introduced into extirpated occurrences if threats are managed and the habitat is likely to support Otay tarplant in the future. For extirpated occurrences, source seed from a large occurrence in the same seed zone or from an appropriate seed source in storage (if available). Some threats (particularly, invasive plants) will likely require perpetual management; thus, ensure that adequate funding and/or labor are available to manage the occurrence after seed is reintroduced.

We recommend seed bulking as a potential management action for all occurrences identified as potentially requiring a restoration collection. The need for seed bulking will depend on the amount of seed available in storage and/or collected specifically for restoration, as well as the sowing strategy. For example, bulking will not be needed if adequate seed is collected to sow directly into the target occurrence.

Table 5.4-1. Otay Tarplant: Management Priorities for Seed-related Activities.

Occurrence ID[1]	Occurrence Size[2]	Genetic Structure[3]	Seed Reintroduction Priority[4]	Existing Seed Collection[5]	Seed-related Activities[6]			
					Seed Source	Conservation Collection	Restoration Collection	Seed Bulking
DECO13_2PAVA001	Small	L+H+L	High			✓	✓	✓
DECO13_2PAVA030	Small	L+H+L	High			✓	✓	✓
DECO13_3BOME009	Large	L+H+L		CBG	✓		✓	✓
DECO13_3DENC022	Small	L+H+L				✓		
DECO13_3DERA020	Small	L+H+L	High			✓	✓	✓
DECO13_3DREA021	Large	L+H+L		SDZG	✓			
DECO13_3JABO028	Large	L+H+L			✓			
DECO13_3JAHI006	Medium	L+H+L	Low	SDZG				
DECO13_3JOCA019	Medium	L+H+L	Low			✓		
DECO13_3LOST027	Medium	L+H+L	Low			✓		
DECO13_3MMGR010	Large	L+H+L	Low		✓	✓		
DECO13_3OMEA026	Small	L+H+L	Medium			✓	✓	✓
DECO13_3ORVA017	Small	L+H+L				✓		
DECO13_3ORVA018	Large	L+H+L			✓			
DECO13_3PMA1002	Large	L+H+L			✓			
DECO13_3PMA2003	Medium	L+H+L	Medium			✓	✓	✓
DECO13_3PMA4005	Large	L+H+L	Medium		✓			
DECO13_3PRVA013	Small	L+H+L	Medium			✓	✓	✓
DECO13_3PRVA014	Small	L+H+L				✓		
DECO13_3RHRA012	Medium	L+H+L	Low			✓		
DECO13_3RJER015	Large	L+H+L		CBG, SDZG	✓			

Table 5.4-1. Otay Tarplant: Management Priorities for Seed-related Activities.

Occurrence ID[1]	Occurrence Size[2]	Genetic Structure[3]	Seed Reintroduction Priority[4]	Existing Seed Collection[5]	Seed-related Activities[6]			
					Seed Source	Conservation Collection	Restoration Collection	Seed Bulking
DEC013_3SCPA016	Small	L+H+L				✓		
DECO13_3SMHA024	Small	L+H+L	Medium			✓	✓	✓
DECO13_3SMHA025	Small	L+H+L	Medium			✓	✓	✓
DECO13_3SVPC007	Large	L+H+L		CBG	✓			
DECO13_3TRIM008	Large	L+H+L		CBG	✓			
DECO13_3WMCA023	Small	L+H+L				✓		

[1] Occurrence ID: Occurrence identification (ID) code per the San Diego Management and Monitoring Program's (SDMMP) Master Occurrence Matrix (MOM) database.
[2] Occurrence Size: Recent size category as defined in the Framework-Rare Plant Management Plan (F-RPMP). **Small** = <1,000 individuals, **Medium** = 1,000-<10,000 individuals, **Large** = >10,000 individuals.
[3] Genetic Structure (per Milano and Vandergast 2018): Genetic differentiation + genetic diversity + inbreeding. **L** = low, **H** = High. Refer to F-RPMP for a discussion of the implications for seed movement.
[4] Seed Reintroduction Priority (per F-RPMP): **High** = high priority for seed reintroduction, **Medium** = medium priority for seed reintroduction, **Low** = low priority for seed reintroduction. If no priority level is indicated, then management action is not a priority at this time, but may be undertaken at the discretion of the land manager and subject to adequate seed availability.
[5] Existing Seed Collection: Indicates location of collection. **CBG** = California Botanic Garden.
[6] Seed-related Activities: ✓ indicates occurrence is suitable (e.g., seed source) pending authorization from responsible entities, or action is warranted. In some cases, additional collecting for restoration may not be necessary if enough seed is available in an existing seed collection. Likewise, seed bulking may or may not be necessary depending on amount of seed available in existing collection or collected specifically for restoration purposes.

Seed Zones

We established one seed zone for Otay tarplant to guide seed collecting, banking, and bulking efforts. This seed zone corresponds to the genetic cluster and regional population group identified for this species in the F-RPMP. Figure 5.4-2 shows the locations of the seed zone, while Table 5.4-2 lists all occurrences in the seed zone.

Seed zones place geographic limits on the transfer of seed between occurrences. *We do not recommend transfer of seed beyond the seed zone at this time* (e.g., between occurrences in San Diego County and Baja California). Thus, if a small occurrence requires seed from another occurrence, the source (donor) occurrence should be located within the same seed zone as the target (recipient) occurrence. Within a seed zone, we recommend that seed transfer proceed between occurrences that are relatively close to one another. We designate subgroups (Table 5.4-2) to identify occurrences in proximity. The same limits on seed movement apply when using seed from a seed bank for restoration.

Collecting Plan

Refer to Section 4.1 of this document for guidelines on developing a sampling strategy for seed collection. Identify the purpose and type of collection needed for a specific project or restoration effort, as well as timeline and costs. In the section below, we highlight key steps in the process, including those specific to Otay tarplant.

Sampling Strategy

Refer to Table 5.4-1 for prioritized seed-related activities for the target occurrence to identify whether seed is needed for a conservation collection, a restoration collection, or both.

For restoration, determine if seed is available in a seed bank to reintroduce directly into a target occurrence or for bulking. Proceed with the collecting plan if stored seed is not available, quantities are not sufficient, or the provenance is not appropriate for the target occurrence (e.g., different seed zone).

Assess whether the target occurrence will likely provide an adequate quantity of seed for restoration or if seed will need to be sourced from other occurrences in the seed zone. Refer to seed zones (Figure 5.4-2, Table 5.4-2) for limits on seed movement and seed sources. Contact the appropriate land managers of potential seed source occurrences to obtain permission for seed collection.

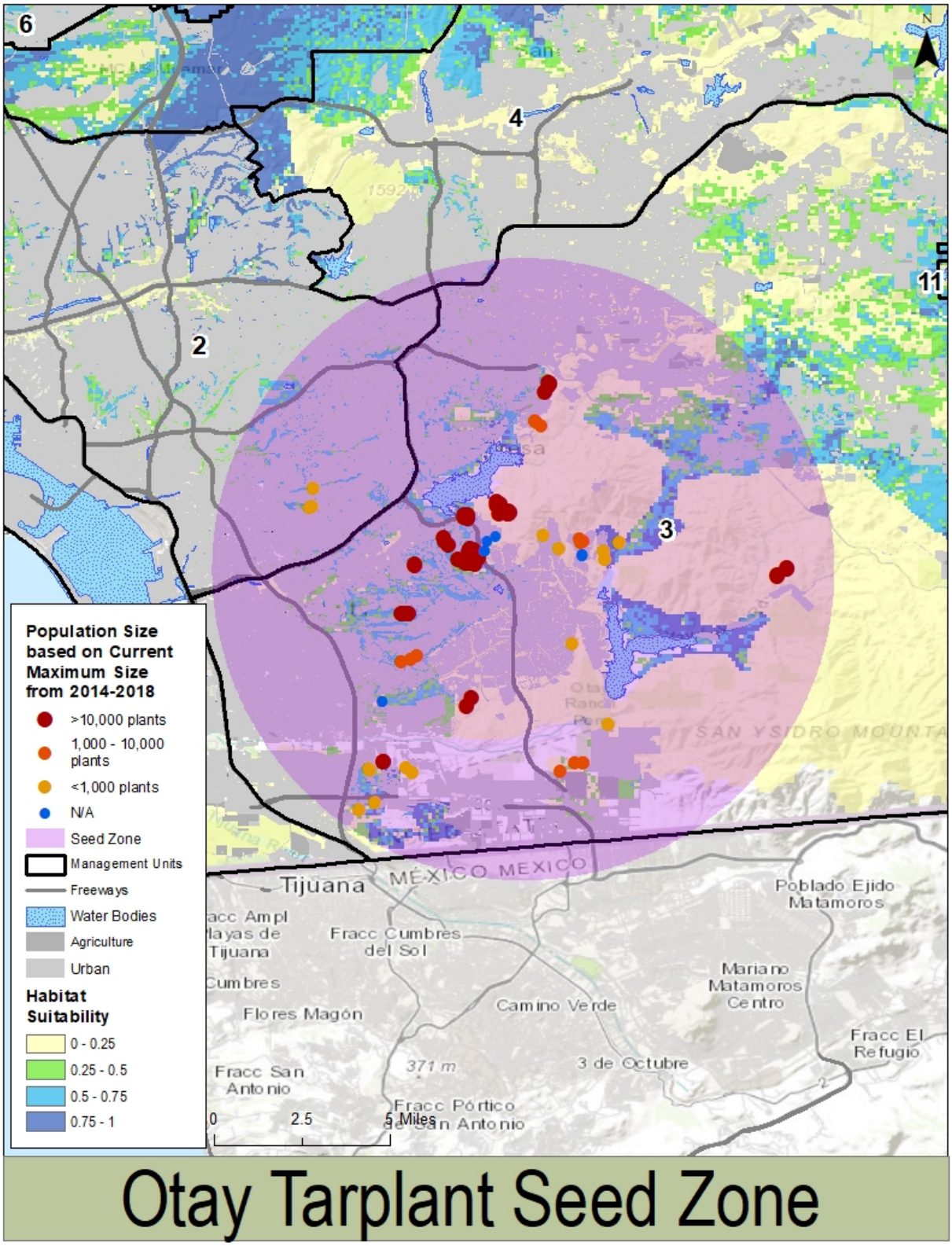

Figure 5.4-2. Otay Tarplant: Seed Zone.

Table 5.4-2. Otay Tarplant: Seed Zones.

Occurrence ID[1]	Occurrence Name	Seed Zone[2]	Seed Zone Subgroup[3]	Occurrence Size[4]
DECO13_3JABO028	Jamacha Boulevard	South	1	Large
DECO13_3JAHI006	Jamacha Hills	South	1	Medium
DECO13_3PAVA001	Paradise Valley	South	2	Small[5]
DECO13_3PAVA030	Paradise Gardens	South	2	Small
DECO13_3MMGR010	Mother Miguel Grassland	South	3	Large
DECO13_3PRVA013	Proctor Valley	South	3	Small[5]
DECO13_3PRVA014	Proctor Valley (Bella Lago)	South	3	Small[5]
DECO13_3RHRA012	Rolling Hills Ranch	South	3	Medium
DECO13_3SMHA024	San Miguel HMA West - DECO13	South	3	Small
DECO13_3SMHA025	San Miguel HMA West - DECO13	South	3	Small
DECO13_3SVPC007	Shinohara Vernal Pool Complex (southeast Sweetwater Reservoir)	South	3	Large
DECO13_3BOME009	Bonita Meadows	South	4	Small[5]
DECO13_3PMA1002	PMA1 (Rice Canyon & Other Canyons)	South	4	Large
DECO13_3PMA2003	PMA2	South	4	Medium
DECO13_3PMA4005	PMA4	South	4	Large
DECO13_3TRIM008	Trimark/Gobbler's Knob/Horseshoe Bend	South	4	Large
DECO13_3DENC022	Dennery Canyon South	South	5	Small[6]
DECO13_3DERA020	Dennery Ranch	South	5	Small
DECO13_3DREA021	Dennery Ranch East	South	5	Large
DECO13_3OMEA026	Furby North	South	5	Small
DECO13_3WMCA023	West of Moody Canyon	South	5	Small[5]
DECO13_3JOCA019	Johnson Canyon	South	6	Medium[5]
DECO13_3LOST027	Lonestar	South	6	Medium[5]
DECO13_3ORVA017	Otay Valley (east end)	South	6	Small
DECO13_3ORVA018	North side of Otay River Valley near Wolf Canyon	South	6	Large
DECO13_3SCPA016	Salt Creek Parcel	South	6	Small[6]
DECO13_3RJER015	Rancho Jamul ER Subpopulation #1	South	7	Large

[1] Occurrence ID: Occurrence identification (ID) code per the San Diego Management and Monitoring Program's (SDMMP) Master Occurrence Matrix (MOM) database.

[2] Seed Zone: seed zones are defined in text and correspond to genetic clusters and population groups in the F-RPMP. Limit seed movement to within the same seed zone.

[3] Seed Zone Subgroups are defined in text and correspond to population subgroups in the F-RPMP. Where feasible, limit seed movement to the same subgroup within a seed zone.

[4] Occurrence size: refers to size category based on multiple years of monitoring data. Recent size category as defined in the Framework-Rare Plant Management Plan (F-RPMP). **Small** = <1,000 individuals, **Medium** = 1,000-<10,000 individuals, **Large** = >10,000 individuals.
[5] Indicates occurrences where monitoring indicates a decline in population size category over time. Refer to F-RPMP for additional details.
[6] Occurrence size presumed small at this time; no population size data available.

Based on the purpose and type of collection, and amount of seed expected to be available, determine whether the seed is likely to be collected along maternal lines or in bulk (Table 4.1-3), and set minimum targets for the number of plants to sample and amount of seed to collect. These targets are intended to provide a guide for planning, and may be modified based on the actual seed crop.

Permitting and Agreements

Ensure that all regulatory permits, memorandums, and access agreements are in place before collecting seed from an occurrence.

If unfamiliar with the collecting location, contact the land owner to obtain a map and coordinates of the occurrence, directions to the occurrence if necessary, gate codes, lock combinations, or keys, and any pertinent information about the occurrence (e.g., informing adjacent land owners, closures due to unsafe conditions).

Pre-collection Monitoring

Practitioners can collect Otay tarplant seed as early as June and into early fall, depending on location, habitat, and weather and temperature conditions during the previous fall, winter, and spring. Visit the target occurrence multiple times to determine the best time to collect seed. Talk to land owners and managers and refer to monitoring data and literature to determine the range of phenological variation for an occurrence to plan the first site visit.

Otay tarplant and fascicled tarplant (*Deinandra fasciculata*) co-occur at several San Diego County occurrences. Otay tarplant is also easily confused with paniculate tarplant (*Deinandra paniculata*), a species that typically occurs further north, but occurs sporadically in northern Baja California and southern San Diego County, where it is found in proximity to Otay tarplant occurrences.[7] Differentiating between these three species is possible when they are in flower, but is very difficult after senescence. Therefore, time the first visit when Otay tarplant is in full flower to make it easier to locate and identify this species correctly. Use pin flags to demarcate the boundary of the occurrence, particularly in areas where Otay tarplant and fascicled tarplant co-occur. Place pin flags adjacent to Otay tarplant individuals to relocate them easily during subsequent visits.

[7] The Consortium of California Herbaria list four specimens of paniculate tarplant from southern San Diego County, and all were collected prior to 1960. At least two of these locations have been developed.

Return to the occurrence within two to four weeks to check phenology. Weather conditions will determine the rate of desiccation; plants will dry quicker in warm and dry conditions and slower if conditions remain cool and moist. Continue monthly site visits until the majority of plants in an occurrence are fully desiccated and seeds resemble those in Figure 5.4-1.

Voucher Specimens

Collect voucher specimens of Otay tarplant if none exist for the source occurrence and if enough plants are available so that collecting a few specimens does not adversely affect the occurrence. Check the San Diego Natural History Museum or Consortium of California Herbaria to determine if a voucher specimen already exists:

San Diego Natural History Museum: http://sdplantatlas.org/publicsearch.aspx

Consortium of California Herbaria: http://ucjeps.berkeley.edu/consortium/

Note that recently collected specimens may not yet be accessed into an herbarium's collection. In addition, older specimens may not include sufficient locality information to definitively determine whether they were collected at the occurrence. If it is not appropriate to collect a voucher specimen, photograph the occurrence (landscape aspect) and an individual plant while in flower, and then again when the plant is in fruit.

Obtain all necessary permits and memorandums before collecting voucher specimens of endangered, threatened, and rare species. Collect specimens while plants are still in flower and before collecting seed. Refer to Section 4.1 of this document (Seed Collecting, Materials and Methods) and instructions and forms in Appendix B to guide your collection. Submit the voucher specimen(s) to the San Diego Natural History Museum as soon as possible.

Methods and Materials

Refer to Section 4.1 of this document for a detailed discussion of methods and materials and Table 4.1-4 for a comprehensive list of recommended equipment for collecting seed. At a minimum, bring the following to each Otay tarplant collection location:

- Regulatory permits and access agreements
- Directions, map(s)
- Lock combination(s), key(s)
- Collecting strategy/instructions

- GPS unit (preferably, sub-meter)
- Camera
- Labels, markers, pens or pencils
- Seed collection forms
- Hand lens
- Adhesive tape
- Safety razor or pocketknife
- Container and water (optional)
- Scissors, snips
- Paint sanding respirator mask (optional; for allergic reactions)
- Safety goggles (optional; for allergic reactions)
- Small paper envelopes or bags

Before collecting seed, map the seed collection area(s), record data on the seed collection form(s) and count or estimate the number of plants within an occurrence. Collect no more than 5-10% of the seed from an individual plant or from the reproductive population per season. Use the following method to determine the number of seeds to collect by plant and for an occurrence:

Step 1: Select 5-10 plants of different sizes spaced randomly across the occurrence.

Step 2: Count the number of seeds in 3-5 flower heads per plant.

Step 3: Divide the total number of seeds counted (Step 2) by the total number of flower heads sampled (Step 2) to calculate an *average number of seeds per flower head*.

Step 4: Count or estimate the number of flower heads on each sampled plant. Divide the total number of flower heads by the number of plants sampled to estimate the *average number of flower heads per plant*.

Step 5: Multiply the average number of seeds per flower head (Step 3) by the average number of flower heads per plant (Step 4) to generate an *average number of seeds per plant*.

Step 6: Multiply the average number of seeds per plant (Step 5) by the occurrence count or estimate to calculate the *estimated number of seeds per occurrence*. In most cases, the amount of seed available for collecting will be higher in years with optimal growing conditions (e.g., average or above-average rainfall) and lower during drought years.

Step 7: Calculate the maximum number of fruits or seeds to collect per plant or occurrence to stay within the 5-10% collecting guidelines.

Collect seeds when plants are fully dry (Figure 5.4-3). At this stage, plants will be tan to brown with no green parts showing, and seeds will be hard and tannish-brown to black (Figure 5.4-1).

Figure 5.4-3. Otay Tarplant: Dried Inflorescence. (Photo credit: Stacy Anderson, SDZG)

Fruits may be ready to collect as early as one month after peak flowering. Remove dry fruits from the stems by hand and place them in a seed collecting envelope or bag. Note that some collectors have reported allergic reactions while collecting Otay tarplant seeds; thus, we recommend wearing a paint sanding respirator mask and goggles if an allergic reaction is suspected.

Collect seed during early, mid, and late season to account for differences in maturity times and to capture the phenological variability among the occurrence. Collect seed randomly and evenly throughout the occurrence, taking care to collect from plants of various sizes and with variable inflorescences, as well as plants growing in unique or varying habitat conditions.

Consider harvesting more seed than recommended by the CPC for small occurrences that are at serious risk of extirpation to capture the highest amount of genetic variation at the occurrence (McMillan 2019). In this case, coordinate with the regulatory agencies prior to the collecting effort.

Ideally, collect seed from 30-50 plants across the occurrence. If the number of plants present in a given year is below this number, then sample over multiple years to produce a genetically robust seed collection for restoration purposes. Where the occurrence is small and not expected to produce an adequate number of plants (and seed) in any given year, consider collecting seed from one or more occurrences within the same seed zone. Depending on project objectives and sampling strategy, collect seed along maternal lines or as a bulk collection.

Interim Seed Storage and Delivery

Refer to the general section of this document (Seed Collecting) for guidelines on interim seed storage and delivery. We provide additional guidelines for practitioners that choose to keep the seed for future restoration purposes rather than sending it immediately to a storage facility:

- Do not clean and sort seed unless the collection is very small since cleaning seed is time-consuming and often difficult, especially with large amounts of seed.
- Loosely pack the seed collection in envelopes or bags and store them in dark, dry, and moderate to cool conditions for several months before distributing seed onsite.
- If planning to hold seed for longer than one year, store the seed at a recognized seed bank (i.e., CBG, SDZG) to ensure proper storage conditions to retain viability.

Seed Banking

Existing Conservation and Restoration Collections

Refer to Table 5.4-3 for existing seed collections of Otay tarplant. This table includes only tarplant occurrences with seed in storage. For planning purposes, we indicate the seed zone for each collection. We also indicate the number of seeds present in the seed bank and their potential availability for restoration. Note that only a portion of the seeds in a collection will be available for these purposes. Existing permits or contracts may also limit the use of some collections. We obtained information on each collection (seed bank, accession number and year, and seed quantity, provenance, and availability) from seed bank managers. Refer to Table 4.2-3 for contact information for these seed banks; we encourage land managers to contact facilities directly to get more information on seed availability or to request seed.

Best Management Practices

Seed Cleaning

Wall and MacDonald (2009) suggest the following methods to clean and sort seeds of Otay tarplant:

- Rub and sort material through graduated sieves to separate achenes and chaff from other plant material.
- Gently rub material again over a sieve with a padded wooden block to separate ray achenes from involucres and to break up large chaff.
- Use a blower at low speed to separate sterile or parasitized seed from fertile seed.

Wall and MacDonald (2009) indicate that cleaning seed of this species is difficult, and found that most ray achenes but very few disk achenes were fertile.

Table 5.4-3. Otay Tarplant: Exiting Seed Collections.

Occurrence ID[1]	Seed Zone[2]	Seed Bank[3]	Accession Number[4]	Collection Year	Seed Quantity[5]	Provenance Type[6]	Seed Available[7]
DECO13_3BOME009	South	CBG	23634-5082	2013	1,498	W	No
DECO13_3DREA021	South	SDZG	S0804	2019	Unknown	W	Yes
DECO13_3JAHI006	South	SDZG	S0805	2019	Unknown	W	Yes
DECO13_3RJER015	South	CBG	23625-5079	2013	530	W	No
DECO13_3RJER015	South	CBG	23626-5080	2013	536	W	No
DECO13_3RJER015	South	CBG	23633-5109	2013	448	W	No
DECO13_3RJER015	South	CBG	24320-5495	2016	52,368	Z	Limited
DECO13_3RJER015	South	SDZG	S0806	2019	Unknown	W	Yes
DECO13_3SVPC007	South	CBG	23632-5085	2013	593	W	No
DECO13_3SVPC007	South	CBG	23632-5086	2013	924	W	No
DECO13_3SVPC007	South	CBG	24322-5496	2016	36,852	Z	Limited
DECO13_3TRIM008	South	CBG	24321-5494	2016	128,760	Z	Limited
State Route 54	South	CBG	20347-2885	1999	56,400	W	No

[1] Occurrence identification (ID) per the SDMMP's MOM database. If no occurrence ID exists for a location, we provide the name of the location as indicated on collection in seed bank.
[2] Seed Zone: Seed zone corresponds to the one genetic cluster and population group identified in the F-RPMP: South. Occurrences in italics are placed in seed zones based on location.
[3] Seed bank: **CBG** = California Botanic Garden, **SDZG** = San Diego Zoo Global.
[4] Accession number = accession number of seed collection assigned by seed bank.
[5] Seed quantity indicates the number of seeds currently in the collection, according to seed bank records. Unknown indicates seed has not yet been processed and counted.
[6] Provenance type indicates the source location of the seed collection: **W** = seed collected directly from the wild (origin known), **Z** = seed collected from a cultivated plant of known wild origin.
[7] Seed available: **Yes** = seed is available for restoration use. **No** = seed is not available for restoration use, **Limited** = seed is only available for use by Land IQ/South County land managers.

Seed Testing

Seed testing methods will vary by facility, but all seed banks conduct baseline germination tests on new accessions and then conduct follow up testing to check seed for viability.

At CBG, baseline tests for seed germination use a typical seed sample of 50 seeds, although sample size may vary depending on the size of the seed collection, and variable methods to break dormancy (if needed). They conduct follow-up testing on seeds in storage at intervals (e.g., 1 year, 5 years, 10 years, 20 years, and every subsequent 10 years thereafter, as seed quantities allow).

CBG has conducted a number of seed germination tests on Otay tarplant (https://www.rsabg.org/conservation/seed-conservation). Tarplant seed germinates poorly with no pre-treatment to soften the hard seed coat. However, CBG achieved germination rates between 38-68% with a hot water treatment (i.e., seeds were soaked in hot water for 24 hours).

At SDZG, a baseline germination test is conducted on all new accessions. The test is conducted primarily on fresh seed. Follow up testing is conducted on frozen seed after 1, 5, and 10 years in storage. Seed is sterilized with bleach and placed in reverse osmosis water at room temperature for 24 hours to fully imbibe (absorb) water. Seed is then plated onto agar trays (10 seeds per tray) and observed for signs of germination. The seed is counted as germinated when the radical emerges. SDZG does not use a cut test to determine viability. Rather, they provide abundant time for seeds to germinate (Anderson pers. comm.).

Seed Storage

All seed banks contacted store Otay tarplant seed according to CPC guidelines. For example, SDZG dries seeds down to 23-35% moisture content. Seed is then stored in sealed, foil-lined envelopes in the freezer at a low temperature (-18°C to -23°C).

Each collection is separated into three packages that represent the primary, curation, and duplicate collections, as described in Section 4.1. The primary collection is the active seed collection and seeds in this package are available for research, testing, restoration, and other uses. The curation package is the base collection for long-term storage. This package is not opened and is not available for other uses. It constitutes the long-term conservation collection. The duplicate collection is sent to another seed bank facility as backup storage to protect against catastrophic loss at the primary storage facility. Both SDZG and CBG send their duplicate seed collections to the NLGRP.

Seed Bulking

Seed Selection

Refer to Section 4.3 for guidelines on seed selection to maximize the quality of the bulked sample. Key considerations include:

- Use seed for bulking from the target occurrence or another occurrence in the same seed zone as the target occurrence. For very small occurrences (fewer than 100 plants), use seed from a large occurrence (if present) or from one or more smaller occurrences (composite provenancing) within the same seed zone.
- Use seed that was collected randomly and uniformly.
- Use an adequate amount of seed for bulking (ideally, ≥1,000 seeds or seed from 30-50 plants).
- For very small seed samples, use seed collected along maternal lines.

Best Management Practices

Germination and Propagation Methods

Otay tarplant seed generally requires pre-treatment (i.e., scarification, hot water soak) to germinate readily. In the absence of pre-treatment, germination rates may be somewhat higher with fresh versus frozen seed. Propagation efforts that do not pre-treat seed should take into account the low germination rate when determining the number of seeds to sow.

Photo credit: Recon Native Plant Nursery.

In a nursery setting, seeds from mature plants fell into containers and germinated immediately. This suggests that germination occurred before dormancy set in (Recon Native Plant Nursery 2014), and reinforces that germination rates may be higher with fresh seed.

Limits on Generations

At this time, we recommend bulking Otay tarplant seed for only one generation (F1) in a nursery setting. If additional seed is needed for restoration, start with wild seed rather than the F1 generation.

Documentation

We encourage growing facilities and others bulking seed of Otay tarplant for reintroduction purposes to document bulked samples as indicated in Section 4.3 and provide this information to land managers when seed is delivered. Likewise, we encourage land managers to provide information on seed collection and seed reintroductions to the SDMMP (contact information at https://sdmmp.com/about.php#contact) and include relevant information on IMG monitoring forms (i.e., translocated occurrence). Refer to Table 5.4-4 for key information for bulked seed samples.

Table 5.4-4. Documentation for Bulked Seed Samples.

Key Information	Primary Responsibility
Seed Source	Seed Collector and/or Land Manager
Seed Collection Date	Seed Collector
Amount of Seed Bulked	Growing Facility
Propagation Method	Growing Facility
Propagation Conditions	Growing Facility
Harvesting Date(s)	Growing Facility
Post-harvest Storage Location/Duration/Conditions	Growing Facility
Generation of Bulked Sample	Growing Facility
Receptor Site Location	Growing Facility and/or Land Manager

5.5 ORCUTT'S SPINEFLOWER (*CHORIZANTHE ORCUTTIANA*)

Seed Collection

Seed Characteristics

Orcutt's spineflower flowers produce one small, black, obovate seed that forms in single chambered superior ovaries that do not dehisce. The seeds are held within an achene located inside a hard, rigid involucre (Figures 5.5-1 and 5.5-2) that disperse by wind or animals after breaking from the plant (USFWS 2014). A portion of the seed produced in a given year does not germinate the following winter, but instead becomes part of the soil seed bank. Soil seed bank longevity is currently unknown; however, Bauder (2000) speculated that seeds may remain viable for decades in the soil seed bank. The United States Navy is currently conducting multi-site research studies to understand soil seed bank dynamics.

Figure 5.5-1. Orcutt's Spineflower Involucres (Photo credit: California Botanic Garden)

Orcutt's spineflower seed forms by April and matures through May and into early summer. Seed production increases with plant size and flower production with easterly aspects supporting larger plants in larger populations than westerly aspects (Kaur et al. 2020). While the specific pollinators and pollination mechanisms are unknown for Orcutt's spineflower, seed production for a related rare taxon, *Chorizanthe parryi* var. *fernandina*, was high at 50-60% seed set achieved through a combination of ants, flying insects, and self-pollination (Jones et al. 2009). *In situ*, the species exhibits higher germination and plant fitness under cool, moist soil conditions, but prefers drier environments during after-ripening (Kaur et al. 2020).

Figure 5.5-2. Orcutt's Spineflower Involucres
(Photo credit: Jyotsna Sharma)

Priority Occurrences

Table 5.5-1 prioritizes occurrences of Orcutt's spineflower on conserved lands in the MSPA for seed-related management actions. These priorities were developed in the F-RPMP based on monitoring data and specific studies. The decision to implement an action will depend on a number of factors, including status of the occurrence (stable or declining), habitat condition, threats, previous management actions (if any), and funding, among others.

In general, large occurrences with high genetic diversity are prioritized as potential seed sources, subject to land manager and/or agency approval. Small occurrences are high priority candidates for seed reintroduction, subject to response to management and site conditions, as discussed in the F-RPMP.

Currently, conservation seed collections exist at CBG for three Orcutt's spineflower occurrences (50%) on conserved lands in the MSPA (Table 5.5-1). Seed collections for additional occurrences outside of the MSPA (i.e., military lands) also exist; however, these collections are not discussed at length in this plan.

We recommend conservation collections as a potential management action for all occurrences that are not currently represented in a seed bank *if adequate seed is available*. For occurrences where plants have not been detected recently, a conservation collection will only be possible if plants are detected in the future (Table 5.5-1). Large occurrences (>10,000 individuals) are the highest (or most immediate) priorities for seed conservation to maximize the amount of genetic diversity in long-term storage although no large occurrences of Orcutt's spineflower are currently known.

We recommend restoration collections as a potential management action for occurrences that are high or medium priorities for seed reintroduction as indicated in Table 5.5-1 and identified in the F-RPMP. We do not prioritize occurrences for seed reintroduction if they have not yet been monitored or if the location is questionable (e.g., CHOR_6OAPA003).

We recommend reintroducing seed only if threats are controlled, habitat is likely to support this species in the future, and funding is available for short- and long-term management. Reintroducing seed is most appropriate for small occurrences that continue to decline, even with management. Seed may be introduced into extirpated occurrences if threats are managed and the habitat is likely to support Orcutt's spineflower in the future. For extirpated occurrences, source seed from a large occurrence in the same seed zone or from an appropriate seed source in storage (if available). If seed is not available to source from a large occurrence in the same seed zone or in storage, bulk seed using collected seed from the same seed zone. Some threats (particularly, invasive plants) will likely require perpetual management; thus, ensure that adequate funding and/or labor are available to manage the occurrence after seed is reintroduced.

Table 5.5-1. Orcutt's Spineflower: Management Priorities for Seed-related Activities on Conserved Lands within the MSPA.

Occurrence ID[1]	Occurrence Name	Occurrence Size[2]	Seed Reintroduction Priority[3]	Existing Seed Collection[4]	Seed-related Activities[5]			
					Seed Source	Conservation Collection	Restoration Collection	Seed Bulking
CHOR_6GOCA001	Gonzales Canyon	Medium	L	CBG				
CHOR_6OAPA003[6]	Oakcrest Park	Small[6]	---			✓[7]		
CHOR_6SOHI002	Sorrento Hills	Small	H			✓	✓	✓
CHOR_7GUTR004	Gully Trail	Medium	L	CBG				
CHOR_7CRCA005	Crest Canyon Preserve	Medium	L	CBG				
CHOR_7TPSR007	Torrey Pines State Reserve South	Small	H			✓[8]	✓	✓

[1] Occurrence ID: Occurrence identification (ID) code per the San Diego Management and Monitoring Program's (SDMMP) Master Occurrence Matrix (MOM) database.

[2] Occurrence Size: Recent size category as defined in the Framework-Rare Plant Management Plan (F-RPMP). **Small** = <1,000 individuals, **Medium** = 1,000-<10,000 individuals, **Large** = >10,000 individuals.

[3] Seed Reintroduction Priority (per F-RPMP): High = high priority for seed reintroduction, Medium = medium priority for seed reintroduction, Low = low priority for seed reintroduction, --- = not a priority for seed reintroduction at this time due to stable population (large occurrence) or lack of monitoring data.

[4] Existing Seed Collection: Indicates location of collection. **CBG** = California Botanic Garden

[5] Seed-related Activities: ✓ indicates occurrence is suitable as a seed source (pending authorization from responsible entities) or management action is warranted. Where there is an existing seed collection, a restoration collection may not be needed, but land managers should contact the seed bank directly to make that determination. Likewise, seed bulking may not be necessary depending on the amount of seed available in an existing collection or collected specifically for restoration purposes.

[6] Indicates occurrences with at least one IMG monitoring event or survey during the 6-year period from 2014-2019, but 0 plants detected.

[7] Occurrence has not been monitored or plants have not been observed recently. In this case, a conservation collection is warranted only if the occurrence is extant and plants are present in the future.

[8]. CHOR_7TPSR007 was established using seed from CHOR_7GUTR004.

Seed Zones

We established six provisional seed zones for Orcuttôs spineflower to guide seed collecting, banking, and bulking efforts within the MSPA: North, Central, East, West, South, Southeast. Seed zones correspond to the six geographic clusters that might represent genetic clusters (to be confirmed) and six regional population groups identified for this species in the F-RPMP. Figure 5.5-3 shows the locations of the six seed zones; Table 5.5-2 lists the seed zone for each occurrence.

Seed zones place geographic limits on the transfer of seed between occurrences. *We do not recommend transfer of seed beyond a seed zone at this time*. Thus, if a small occurrence requires seed from another occurrence, the source (donor) occurrence should be located within the same seed zone as the target (recipient) occurrence. Within a seed zone, we recommend that seed transfer proceed between occurrences that are relatively close to one another, particularly in larger seed zones. The same limits on seed movement apply when using seed from a seed bank.

For occurrences of unknown status (CHOR_6OAPA003) and where no seed source is available within the same seed zone, we recommend restoring habitat and controlling threats for a minimum of five years to determine if the soil seed bank is extant. If the soil seed bank is no longer extant, procure seed from medium-sized occurrences within the nearest seed zone or consider composite provenancing from within the nearest seed zone to develop a genetically appropriate seed source.

Collecting Plan

Refer to Section 4.1 of this document for guidelines on developing a sampling strategy for seed collection. Identify the purpose and type of collection needed for a specific project or restoration effort, as well as timeline and costs. In the section below, we highlight key steps in the process, including those specific to Orcuttôs spineflower.

Sampling Strategy

Refer to Table 5.5-1 for prioritized seed-related activities for the target occurrence to identify whether seed is needed for a conservation collection, a restoration collection, or both.

For restoration, determine if seed is available in a seed bank to reintroduce directly into a target occurrence or for bulking. Proceed with the collecting plan if stored seed is not available, quantities are not sufficient, or the provenance is not appropriate for the target occurrence (e.g., different seed zone).

Assess whether the target occurrence will likely provide an adequate quantity of seed for restoration or if seed will need to be sourced from other occurrences in the seed zone. Refer to seed zones (Figure 5.5-3, Table 5.5-2) for limits on seed movement and seed sources. Contact the appropriate land managers of potential seed source occurrences to obtain permission for seed collection.

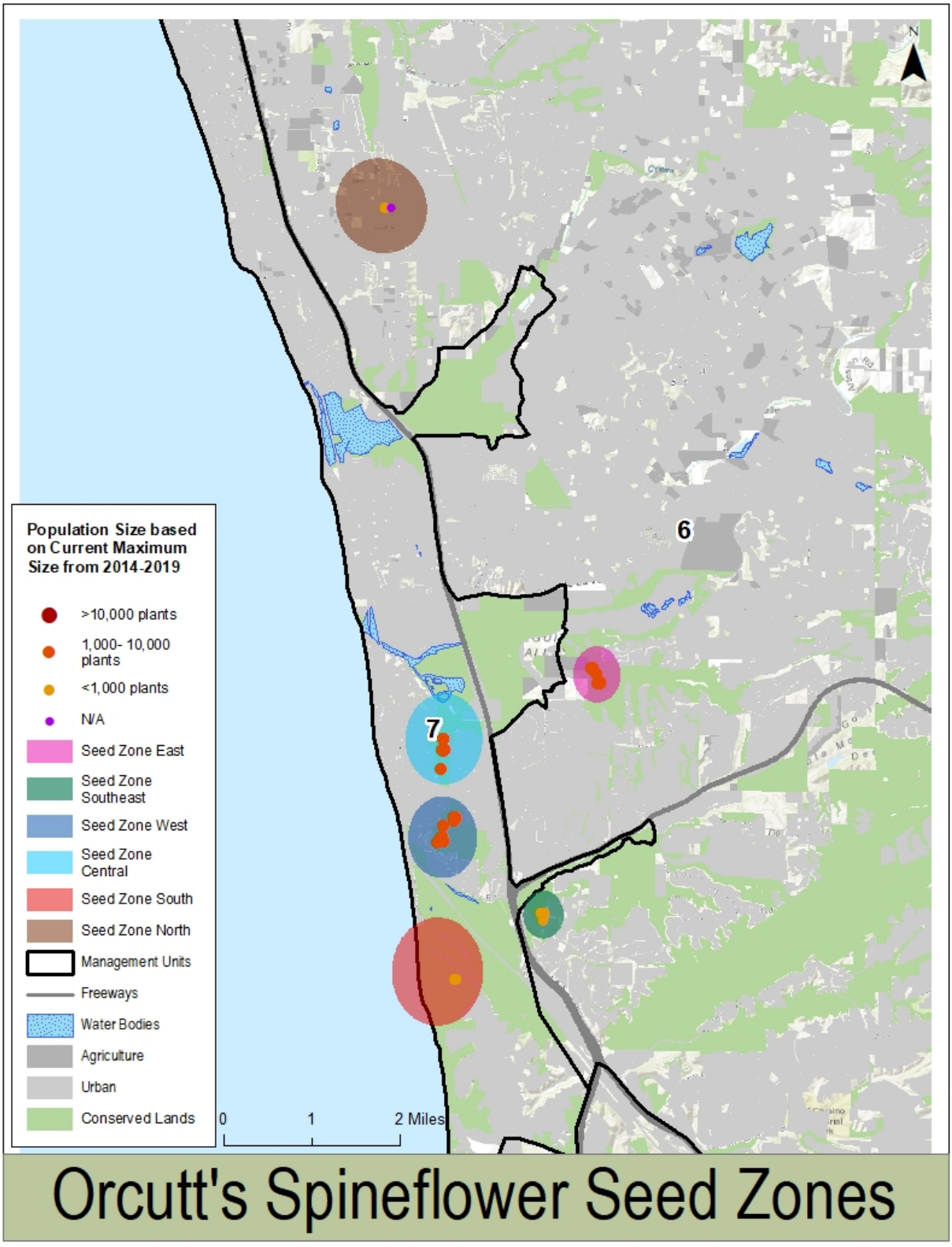

Figure 5.5-3. Orcuttôs Spineflower: Seed Zones.

Based on the purpose and type of collection, and amount of seed expected to be available, determine whether the seed is likely to be collected along maternal lines or in bulk (Chapter 4, Table 4.1-3), and set minimum targets for the number of plants to sample and amount of seed to collect. These targets are intended to provide a guide for planning and may be modified based on the actual seed crop.

Table 5.5-2. Orcuttôs Spineflower: Seed Zones.

Occurrence ID[1]	Occurrence Name	Seed Zone[2]	Occurrence Size[3]
CHOR_6OAPA0036	Oakcrest Park	North	Small
CHOR_7CRCA005	Crest Canyon Preserve	Central	Medium
CHOR_7TPSR007	Torrey Pines State Reserve South	South	Small
CHOR_6SOHI002	Sorrento Hills	Southeast	Small
CHOR_6GOCA001	Gonzales Canyon	East	Medium
CHOR_7GUTR004	Gully Trail	West	Medium

[1] Occurrence ID: Occurrence identification (ID) code per the San Diego Management and Monitoring Programôs (SDMMP) Master Occurrence Matrix (MOM) database.

[2] Seed Zone: seed zones are defined in text and correspond to population groups in the F-RPMP. Limit seed movement to within the same seed zone. Shading indicates seed zone group.

[3] Occurrence size: refers to population size category based on multiple years of monitoring data. Recent size category as defined in the Framework-Rare Plant Management Plan (F-RPMP). **Small** = <1,000 individuals, **Medium** = 1,000-<10,000 individuals, **Large** = >10,000 individuals.

Permitting and Agreements

Ensure that all regulatory permits, memorandums, and access agreements are in place before collecting seed from an occurrence.

If unfamiliar with the collecting location, contact the land owner to obtain a map and coordinates of the occurrence; directions to the occurrence if necessary; gate codes, lock combinations, or keys; and any pertinent information about the occurrence (e.g., informing adjacent land owners, closures due to unsafe conditions).

Pre-collection Monitoring

Practitioners can collect Orcuttôs spineflower seed beginning in late April and into early summer, depending on location, habitat, and weather and temperature conditions during the previous fall, winter, and spring. Visit the target occurrence multiple times to determine the best time to collect seed. Time the first visit when plants are fully grown and easier to identify and locate. Talk to landowners and managers and refer to monitoring data and literature to determine the range of phenological variation for an occurrence to plan the first site visit.

Use temporary markers that are least intrusive (e.g., in color, material, size) to demarcate the boundary of the occurrence because relocating plants after they dry can be difficult depending on the occurrence location and habitat. If necessary, place markers adjacent to a subset of plants to relocate them easily during subsequent visits. Return to the occurrence within two to four weeks to check phenology. Weather conditions will determine the rate of desiccation; plants will dry quicker in warm and dry conditions and slower if conditions remain cool and moist. Continue monthly site visits until the majority of plants in an occurrence are fully desiccated (Figure 5.5-4) and involucres resemble those in Figure 5.5-5.

Figure 5.5-4. Fully desiccated Orcuttôs Spineflower Plants
(Credit: Jyotsna Sharma).

Figure 5.5-5. Dry Orcuttôs Spineflower Involucres
(Credit: Jyotsna Sharma)

Voucher Specimens

Collect voucher specimens of Orcuttôs spineflower if none exist for the source occurrence and if enough plants are available so that collecting a few specimens does not adversely affect the occurrence. Consider collecting a few stems from individual plants instead of removing the entire plant. Check the San Diego Natural History Museum or Consortium of California Herbaria to determine if a voucher specimen already exists:

San Diego Natural History Museum: http://sdplantatlas.org/publicsearch.aspx

Consortium of California Herbaria: http://ucjeps.berkeley.edu/consortium/

Note that recently collected specimens may not yet be accessed into an herbariumôs collection. In addition, older specimens may not include sufficient locality information to definitively determine whether they were collected at the occurrence. If it is not appropriate to collect a voucher specimen, photograph the occurrence (landscape aspect) and an individual plant while in flower, and then again when the plant is in fruit. Ensure that key identifying characteristics are clearly visible in the photographs (e.g., hooked involucral bracts).

Obtain all necessary permits and memorandums before collecting voucher specimens of endangered, threatened, and rare species. Collect specimens while plants are still in flower and before collecting seed. Refer to Section 4.1 of this document (Seed Collecting, and Materials and Methods) and instructions and forms in Appendix B to guide your collection. Submit the voucher specimen(s) to the San Diego Natural History Museum as soon as possible.

Methods and Materials

Refer to Section 4.1 of this document for a detailed discussion of methods and materials and Table 4.1-4 for a comprehensive list of recommended equipment for collecting seed. At a minimum, bring the following to each Orcutt's spineflower collection location:

- Regulatory permits and access agreements
- Directions, map(s)
- Lock combination(s), key(s)
- Collecting strategy/instructions
- GPS unit (preferably, sub-meter)
- Camera
- Labels, markers, pens or pencils
- Seed collection forms
- Hand lens
- Adhesive tape
- Safety razor or pocketknife
- Container and water (optional)
- Scissors, snips, sieve (optional)
- Small paper envelopes or bags

Before collecting seed, map the seed collection area(s), record data on the seed collection form(s), and count or estimate the number of plants within an occurrence. Collect no more than 5-10% of the seed from an individual plant or from the reproductive population per season. Use the following method to determine the number of seeds to collect by plant and for an occurrence:

Step 1: Select five plants of different sizes spaced randomly across the occurrence.

Step 2: Count the number of involucres on the five sample plants.

Step 3: Divide the total number of involucres counted (Step 2) by the number of plants sampled (Step 1) to calculate the *average number of involucres per plant*.

Step 4: Multiply the average number of involucres per plant (Step 3) by 1 (one achene/involucre) to calculate the average number of seeds per plant (one seed/achene).

Step 5: Multiply the average number of seeds per plant (Step 4) by the occurrence count or estimate to calculate the *estimated number of seeds per occurrence*. In most cases, the amount of seed available for collecting will be higher in years with optimal growing conditions (e.g., average or above-average rainfall) and lower during drought years.

Step 6: Use the numbers above to calculate the maximum number of seeds to collect to stay within the 5-10% collecting guidelines.

Note that where seed production is very heavy, seed collectors may choose to estimate seed quantities using subsamples.

Collect involucres when Orcuttôs spineflower plants are fully dry (Figure 5.5-4). Remove dry involucres from the plants by hand and place them in a seed collecting envelope or bag. Refer to Figure 5.5-2 and Figure 5.5-5 for ripe involucres. Involucres may be ready to collect as early as one month after peak flowering.

Collect involucres during early, mid, and late season to account for differences in maturity times and to capture the phenological variability among the occurrence. Collect randomly and evenly throughout the occurrence, taking care to collect from plants of various sizes and with variable inflorescences, as well as plants growing in unique or varying habitat conditions.

Species experts recommend seed remain in the involucres because removing it can damage the small achenes (Sharma pers. comm.). Successful restoration of Orcuttôs spineflower occurrences is possible by scattering involucres and by sowing cleaned seed into a site (Sharma pers. comm., Hogan pers. comm.). Refer to the F-RPMP for detailed instructions on preparing a site before reintroducing Orcuttôs spineflower involucres (seed).

Consider harvesting more seed than recommended by the CPC for small occurrences that are at serious risk of extirpation to capture the highest amount of genetic variation at the occurrence (McMillan pers. comm.).

Ideally, collect involcures from 30-50 plants across the occurrence. If the number of plants present in a given year is below this number, then sample over multiple years to produce a genetically robust collection for restoration purposes. Where the occurrence is small and not expected to produce an adequate number of plants (and seed) in any given year, consider collecting involcures from one or more occurrences within the same seed zone. Depending on project objectives and sampling strategy, collect along maternal lines or as a bulk collection.

Interim Seed Storage and Delivery

Refer to the Section 4.1 of this document (Seed Collecting) for guidelines on interim seed storage and delivery. We provide additional guidelines for practitioners that choose to keep the seed for future restoration purposes rather than sending it immediately to a storage facility:

- Do not remove seed from the involucres because it can damage the seed and make handling/processing it more difficult.
- Loosely pack the collection in envelopes or bags and store them in dark, dry, and moderate to cool conditions for several months before distributing involucres onsite.
- If planning to hold involucres for longer than one year, store at a recognized seed bank (i.e., CBG, SDZG) to ensure proper storage conditions to retain viability.

Seed Banking

Existing Conservation and Restoration Collections

Refer to Table 5.5-3 for existing seed collections of Orcuttôs spineflower. This table includes only spineflower occurrences with seed in storage, including some occurrences that are on lands not included in the MSPA (e.g., military lands). For planning purposes, we indicate the seed zone for each collection. We also indicate the number of seeds present in the seed bank and their potential availability for restoration. Note that only a portion of the seeds in a collection will be available for these purposes. Existing permits or contracts may also limit the use of some collections. We obtained information on each collection (seed bank; accession number and year; and seed quantity, provenance, and availability) from seed bank managers and other sources. Refer to Table 4.2-3 for contact information for these seed banks and other sources; we encourage land managers to contact sources directly to get more information on seed availability or to request seed.

Best Management Practices

Seed Cleaning

We recommend leaving Orcuttôs spineflower seed in the involucre instead of removing it. Removing the seed is very difficult, time consuming, and often leads to damaged seed (Sale pers. comm., Sharma pers. comm.). Store collected involucres and other plant material (Figure 5.5-6) and contact a seed facility that has experience excising seeds from involucres for reintroduction efforts that require pure seed. CBG (Sale pers. comm.) uses the following methods to clean and sort Orcuttôs spineflower seeds when pure seed is required:

- Place involucres in a soil sieve and rub to break stems, branches, and involucres.
- Place resulting seed and chaff in a seed blower.

Table 5.5-3. Orcuttôs Spineflower: Existing Seed Collections.

Occurrence ID[1]	Seed Zone[2]	Seed Bank[3]	Accession Number[4]	Collection Year	Seed Quantity[5]	Provenance Type[6]	Seed Available[7]
CHOR_1NBPL008	N/A	US Navy	N/A	2019 & 2020	Ó60,000	Z[8]	Limited[9]
CHOR_1NBPL010	N/A	US Navy	N/A	2019 & 2020	Ó90,000	Z[8]	Limited[9]
CHOR_7GUTR004 (*Torrey Pines State Reserve*)	West	CBG	23619	2013	924	W	Limited
CHOR_7GUTR004 (*Torrey Pines State Reserve*)	West	CBG	24020	2014	11,979	Z[10]	Limited
CHOR_7GUTR004 (*Torrey Pines State Reserve*)	West	CBG	24020	2014	5,211	Z[10]	Limited
CHOR_7CRCA005 (*Crest Canyon*)	Central	CBG	24115	2015	1,140	W	Limited
CHOR_6GOCA001 (*Gonzalez Canyon*)	East	CBG	24116	2015	960	W	Limited
CHOR_6GOCA001 (*Gonzalez Canyon*)	East	CBG	24117	2015	1,500	W	Limited
CHOR_6GOCA001 (*Gonzalez Canyon*)	East	CBG	24670	2015	36,033	Z	Limited

[1] Occurrence identification (ID) per the SDMMPôs MOM database. If no occurrence ID exists for a location, we provide the name of the location as indicated on collection in seed bank.
[2] Seed zone: Seed zone corresponds to the potential genetic clusters and population groups identified in the F-RPMP: South, North, Central, East, Southeast, and West. N/A = Not applicable because occurrence occurs outside of MSPA.
[3] Seed bank: **CBG** = California Botanic Garden, **US Navy** = United States Navy (Naval Base Point Loma).
[4] Accession number = accession number of seed collection assigned by seed bank.
[5] Seed quantity indicates the number of seeds currently in the collection, according to seed bank records. Unknown indicates seed has not yet been processed and counted.
[6] Provenance type indicates the source location of the seed collection: **W** = seed collected directly from the wild (origin known), **Z** = seed collected from a cultivated plant of known wild origin.
[7] Seed available: **Limited** = amount of seed available for Land Managers (owner of seed).
[8] Seed was generated *in situ*.
[9] Availability of seed to researchers and land managers not employed by or affiliated with the United States Navy requires specific protocols and permissions.
[10] Bulked *in situ* using seed from accession 23619 and kept separate because of different germination rates.

- Remove by hand any remaining stem, leaf, and branch fragments.
- Continue to blow until only clean, filled seed remains.

Land managers can use the same steps above to sort dry involucres from other plant material when pure seed is not required for reintroduction efforts.

Figure 5.5-6. Dry Orcutt's Spineflower Involucres and Plant Material
(Photo credit: Jyotsna Sharma)

Seed Testing

Seed testing methods will vary by facility, but all seed banks conduct baseline germination tests on new accessions and then conduct follow-up testing to check seed for viability.

At CBG, baseline tests for seed germination use a typical seed sample of 50 seeds, although sample size may vary depending on the size of the seed collection and methods to break dormancy (if needed). CBG provides abundant time for seeds to germinate and, at the end of a germination test, they conduct a cut test (see Section 4.2) on a sample of 5 ungerminated seeds (Birker pers. comm.). CBG also conducts follow-up testing on seeds in storage at intervals (e.g., 1 year, 5 years, 10 years, 20 years, and every subsequent 10 years thereafter, as seed quantities allow).

CBG conducted germination tests on Orcutt's spineflower seed in spring 2014 and found that excised seeds germinated at a higher rate in agar trays in growth chambers than intact seeds. Germination rates were 90% for excised seed and 80% for intact seed, and seed viability remains high (84%) after eight years in cold storage (Sale 2020, USFWS 2014). CBG also conducted two rounds of soil propagation testing and determined that excised seed performed better than intact seed with a germination rate of 30% versus 5% and 8% for intact seed (Sale 2020).

At SDZG, a baseline germination test is conducted on all new accessions. The test is conducted primarily on fresh seed. Follow-up testing is conducted on frozen seed after 1, 5, and 10 years in storage. Seed is sterilized with bleach and placed in reverse osmosis water at room temperature for 24 hours to fully imbibe (absorb) water. Seed is then plated onto agar trays (10 seeds per tray) and observed for signs of germination. The seed is counted as germinated when the radical emerges. SDZG does not use a cut test to determine viability. Rather, they provide abundant time for seeds to germinate (Anderson pers. comm.).

Seed Storage

All seed banks contacted store Orcuttôs spineflower seed according to CPC guidelines. For example, SDZG dries seeds down to 23-35% moisture content. Seed is then stored in sealed, foil-lined envelopes in the freezer at a low temperature (-18ÁC to -23ÁC).

Each collection is separated into three packages that represent the primary, curation, and duplicate collections, as described in Section 4.1. The primary collection is the active seed collection and seeds in this package are available for research, testing, restoration, and other uses. The curation package is the base collection for long-term storage. This package is not opened and is not available for other uses. It constitutes the long-term conservation collection. The duplicate collection is sent to another seed bank facility as backup storage to protect against catastrophic loss at the primary storage facility. Both SDZG and CBG send their duplicate seed collections to the NLGRP.

Seed Bulking

Seed Selection

Refer to Section 4.3 for guidelines on seed selection to maximize the quality of the bulked sample. Key considerations include:

- Use seed for bulking from the target occurrence or another occurrence in the same seed zone as the target occurrence. For very small occurrences (fewer than 100 plants), use seed from a large occurrence (if present) or from one or more smaller occurrences (composite provenancing) within the same seed zone.
- Use seed that was collected randomly and uniformly.
- Use an adequate amount of seed for bulking (ideally, Ó1,000 seeds or seed from 30-50 plants).
- For very small seed samples, use seed collected along maternal lines.

Best Management Practices

Germination and Propagation Methods

CBG performed two Orcuttôs spineflower seed bulking events in fall 2016 using excised and intact seed placed in soil-filled 2-inch pots in growth chambers at 20 degrees Celsius (Figure 5.5-7). Intact seed produced germination rates ranging from 4-11% while excised seed produced germination rates ranging from 20-22%. CBG transplanted surviving plants to soil-filled 6-inch pots, moved them to a greenhouse and eventually moved them outdoors for pollination. Orcuttôs spineflower plants grew larger with increased pot size; however, transplanting from small to large pots resulted in high mortality. Surviving plants were very large and produced seed

amounts ranging from 2,000 to 132,000 for three Orcuttôs spineflower occurrences confirming that bulking works well for Orcuttôs spineflower. Bulking also produces significantly more seed than collecting from the wild (Sale 2020); however, viability differences between wild-collected and first generation seed is currently unknown. Sharma (2020) recommends bulking seed in a shade house or *in situ* where plants have access to pollinators.

Limits on Generations

At this time, we recommend bulking Orcuttôs spineflower seed for only one generation (F1) in a nursery setting. If additional seed is needed for restoration, start with wild seed rather than the F1 generation.

Figure 5.5-7. Propagated Orcuttôs Spineflower Seedlings
(Photo credit: California Botanic Garden)

Documentation

We encourage growing facilities and others bulking seed of Orcuttôs spineflower for reintroduction purposes to document bulked samples as indicated in Section 4.3 and provide this information to land managers when seed is delivered. Likewise, we encourage land managers to provide information on seed collection and seed reintroductions to the SDMMP (contact information at https://sdmmp.com/about.php#contact) and include relevant information on IMG monitoring forms (i.e., translocated occurrence). Refer to Table 5.5-4 for key information for bulked seed samples.

Table 5.5-4. Documentation for Bulked Seed Samples.

Key Information	Primary Responsibility
Seed Source	Seed Collector and/or Land Manager
Seed Collection Date	Seed Collector
Amount of Seed Bulked	Growing Facility
Propagation Method	Growing Facility
Propagation Conditions	Growing Facility
Harvesting Date(s)	Growing Facility
Post-harvest Storage Location/Duration/Conditions	Growing Facility
Generation of Bulked Sample	Growing Facility
Receptor Site Location	Growing Facility and/or Land Manager

5.6 SHORT-LEAVED DUDLEYA (*DUDLEYA BREVIFOLIA*)

Seed Collection

Seed Characteristics

Short-leaved dudleya flowers produce five follicles that contain many seeds. Seeds are very small, obovate, shiny and brown, with parallel grooves and ridges (Figure 5.6-1). The California Department of State Parks funded a project that included counting seeds. Results determined a range of 0-27 seeds per flower (Dodero 2020).

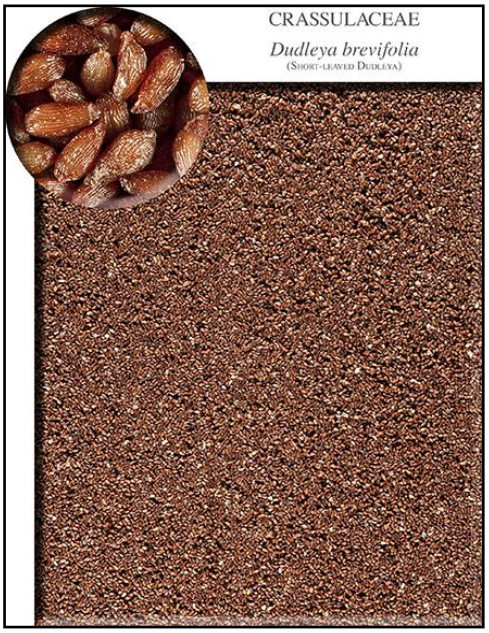

Figure 5.6-1. Short-leaved Dudleya Seed
(Photo credit: California Botanic Garden)

Short-leaved dudleya is likely insect pollinated with the center of the flower turning red post pollination (Figure 5.6-2). Seed dispersal mechanisms likely include wind and water aided by gravity. Dodero (1995) observed short-leaved dudleya seeds moving across the soil surface in windy weather and dried inflorescences moving along the ground dropping seeds in windy weather. Short-leaved dudleya seedlings are often concentrated on sandstone bluffs in depressions that pond with water during sheet flooding events indicating that water moves seed to these depressions (Dodero 1995).

Figure 5.6-2. Short-leaved Dudleya Flowers
(Photo credit: Jessie Vinje)

Soil seed bank longevity is currently unknown; however, short-leaved dudleya seed stored under controlled conditions have remained viable for many years (Dodero pers. comm.), suggesting that a soil seed bank may be present in natural populations.

Seeds mature in late spring through summer and seed production increases with plant size and flower production with larger plants producing many seeds. Under ideal conditions, seeds germinate in as little as three days (Dodero 2020).

Priority Occurrences

Table 5.6-1 prioritizes occurrences of short-leaved dudleya on conserved lands in the MSPA for seed-related management actions. These priorities were developed in the F-RPMP based on

Table 5.6-1. Short-leaved Dudleya: Management Priorities for Seed-Related Activities

Occurrence ID[1]	Occurrence Name	Occurrence Size[2]	Seed Reintroduction Priority[3]	Existing Seed Collection[4]	Seed-related Activities[5]			
					Seed Source	Conservation Collection	Restoration Collection	Seed Bulking
DUBLB2_6CMPR001	Carmel Mtn Preserve - Obs #1	Large	---	CBG, SDZG	✓			
DUBLB2_7CRCA003	Crest Canyon Preserve	Large	---	SDZG	✓			
DUBLB2_7SKCA002	Skeleton Canyon	Small	High	SDZG			✓	✓
DUBLB2_7TPEX004	Torrey Pines Extension	Small	High	SDZG			✓	✓
DUBLB2_7TPSR005	Torrey Pines State Natural Reserve	Large	---	SDZG	✓			

[1] Occurrence ID: Occurrence identification (ID) code per the San Diego Management and Monitoring Program's (SDMMP) Master Occurrence Matrix (MOM) database.
[2] Occurrence Size: Recent size category as defined in the Framework-Rare Plant Management Plan (F-RPMP). **Small** = <1,000 individuals, **Medium** = 1,000-<10,000 individuals, **Large** = >10,000 individuals.
[3] Seed Reintroduction Priority (per F-RPMP): **High** = high priority for seed reintroduction, **Medium** = medium priority for seed reintroduction, **Low** = low priority for seed reintroduction, --- = not a priority for seed reintroduction at this time due to stable population (large occurrence) or lack of monitoring data.
[4] Existing Seed Collection: Indicates location of collection. **CBG** = California Botanic Garden, **SDZG** = San Diego Zoo Global.
[5] Seed-related Activities: ✓ indicates occurrence is suitable as a seed source (pending authorization from responsible entities) or management action is warranted. Where there is an existing seed collection, a restoration collection may not be needed, but land managers should contact the seed bank directly to make that determination. Likewise, seed bulking may not be necessary depending on the amount of seed available in an existing collection or collected specifically for restoration purposes.

monitoring data and specific studies. The decision to implement an action will depend on a number of factors, including status of the occurrence (stable or declining), habitat condition, threats management, and funding, among others.

In general, large occurrences are prioritized as potential seed sources, subject to land manager and/or agency approval. Small occurrences are high priority candidates for seed reintroduction, subject to response to management and site conditions, as discussed in the F-RPMP.

Currently, conservation seed collections exist in established seed banks for all short-leaved dudleya occurrences on conserved lands in the MSPA (Table 5.6-1); therefore, we do not recommend collecting additional seed for conservation collections.

We recommend restoration collections as a potential management action for occurrences that are high priorities for seed reintroduction as indicated in Table 5.6-1 and identified in the F-RPMP.

We recommend reintroducing seed only if threats are controlled, habitat is likely to support this species in the future, and funding is available for short- and long-term management. Reintroducing seed is most appropriate for small occurrences that continue to decline, even with management. Seed may be introduced into extirpated occurrences if threats are managed and the habitat is likely to support dudleya in the future. For extirpated occurrences, source seed from a large occurrence in the same seed zone or from an appropriate seed source in storage (if available), or through seed bulking using seed collected from the same seed zone. Some threats (particularly, invasive plants) will likely require perpetual management; thus, ensure that adequate funding and/or labor are available to manage the occurrence after seed is reintroduced.

We recommend seed bulking as a potential management action for all occurrences potentially requiring a restoration collection. The need for seed bulking will depend on the amount of seed available in storage and/or collected specifically for restoration. For example, bulking will not be needed if adequate seed is collected from source occurrences.

Seed Zones

We established five provisional seed zones for short-leaved dudleya to guide seed collecting, banking, and bulking efforts: North, Central, East, West, and South. Seed zones correspond to the five *potential* genetic clusters identified for this species in the F-RPMP. Figure 5.6-3 shows the locations of the five seed zones; Table 5.6-2 lists the seed zone for each occurrence.

Seed zones place geographic limits on the transfer of seed between occurrences. *We do not recommend transfer of seed beyond a seed zone at this time*. Thus, if a small occurrence requires seed from another occurrence, the source (donor) occurrence should be located within the same seed zone as the target (recipient) occurrence. Within a seed zone, we recommend that seed

transfer proceed between occurrences that are relatively close to one another, particularly in larger seed zones. The same limits on seed movement apply when using seed from a seed bank.

Collecting Plan

Refer to Section 4.1 of this document for guidelines on developing a sampling strategy for seed collection. Identify the purpose and type of collection needed for a specific project or restoration effort, as well as timeline and costs. In the section below, we highlight key steps in the process, including those specific to short-leaved dudleya.

Sampling Strategy

Refer to Table 5.6-1 for prioritized seed-related activities for the target occurrence to identify whether seed is needed for a conservation collection, a restoration collection, or both.

For restoration, determine if seed is available in a seed bank to reintroduce directly into a target occurrence or for bulking. Proceed with the collecting plan if stored seed is not available, quantities are insufficient, or the provenance is not appropriate for the target occurrence (e.g., different seed zone).

Assess whether the target occurrence will likely provide an adequate quantity of seed for restoration or if seed will need to be sourced from other occurrences in the seed zone. Refer to seed zones (Figure 5.6-3, Table 5.6-2) for limits on seed movement and seed sources. Contact the appropriate land managers of potential seed source occurrences to obtain permission for seed collection.

Based on the purpose and type of collection, and amount of seed expected to be available, determine whether the seed is likely to be collected along maternal lines or in bulk (Table 4.1-3, Chapter 4), and set minimum targets for the number of plants to sample and amount of seed to collect. These targets are intended to provide a guide for planning and may be modified based on the actual seed crop.

Permitting and Agreements

Ensure that all regulatory permits, memorandums, and access agreements are in place before collecting seed from an occurrence.

If unfamiliar with the collecting location, contact the land owner to obtain a map and coordinates of the occurrence; directions to the occurrence if necessary; gate codes, lock combinations, or keys; and any pertinent information about the occurrence (e.g., informing adjacent land owners, closures due to unsafe conditions).

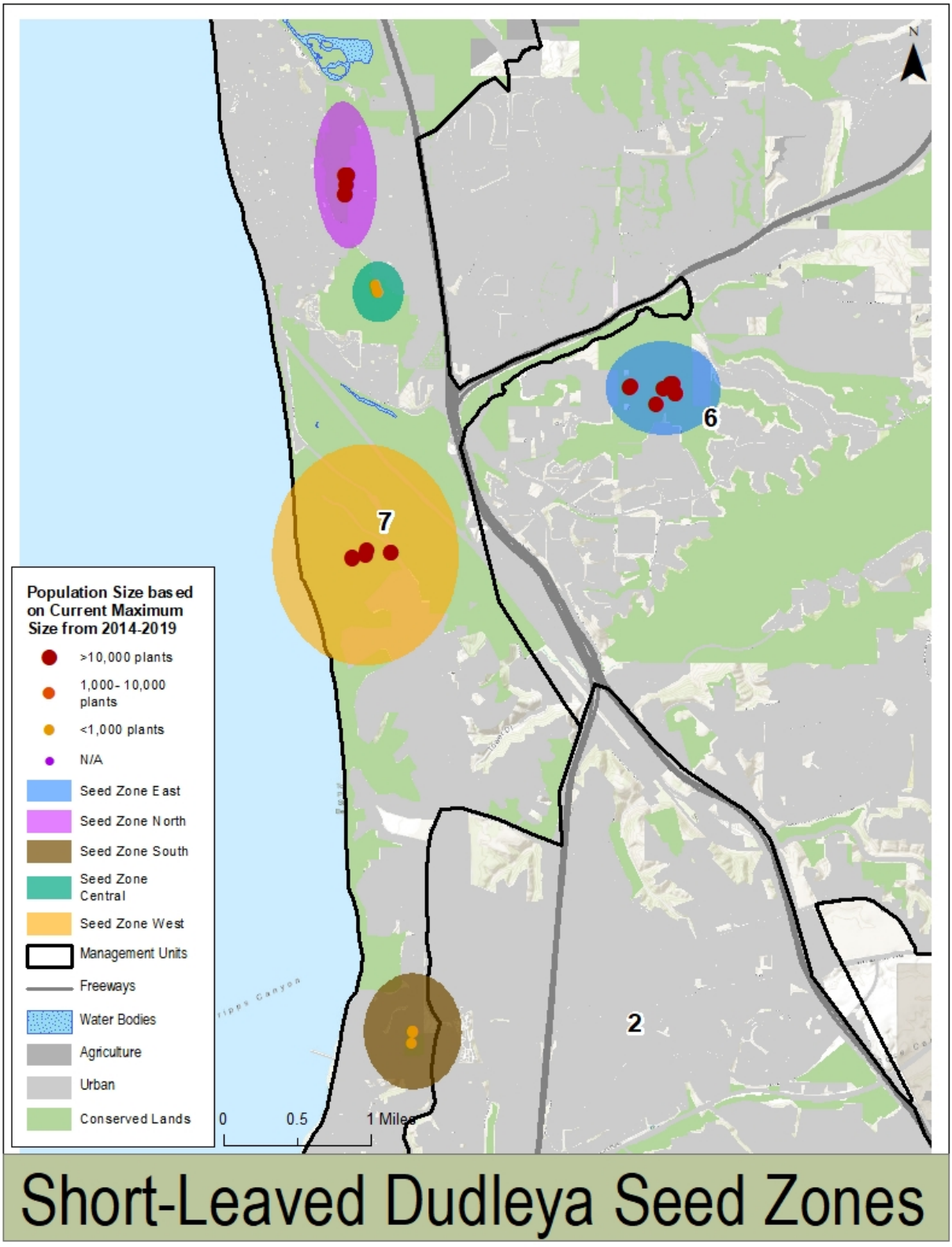

Figure 5.6-3. Short-leaved Dudleya: Seed Zones.

Table 5.6-2. Short-leaved Dudleya: Seed Zones

Occurrence ID[1]	Occurrence Name	Seed Zone[2]	Occurrence Size[3]
DUBLB2_7CRCA003	Crest Canyon Preserve	North	Large
DUBLB2_7TPEX004	Torrey Pines Extension	Central	Small
DUBLB2_7SKCA002	Skeleton Canyon	South	Small
DUBLB2_6CMPR001	Carmel Mtn Preserve - Obs #1	East	Large
DUBLB2_7TPSR005	Torrey Pines State Natural Reserve	West	Large

[1] Occurrence ID: Occurrence identification (ID) code per the San Diego Management and Monitoring Programôs (SDMMP) Master Occurrence Matrix (MOM) database.

[2] Seed Zone: seed zones are defined in text and correspond to population groups in the F-RPMP. Limit seed movement to within the same seed zone. Shading indicates seed zone group.

[3] Occurrence size: refers to population size category based on multiple years of monitoring data. Recent size category as defined in the Framework-Rare Plant Management Plan (F-RPMP). **Small** = <1,000 individuals, **Medium** = 1,000-<10,000 individuals, **Large** = >10,000 individuals.

Pre-collection Monitoring

Practitioners can collect short-leaved dudleya seed as early as late June and into early July, depending on location, habitat, and weather and temperature conditions during the previous fall, winter, and spring (Dodero pers comm.). Visit the target occurrence multiple times to determine the best time to collect seed. Talk to land owners and managers and refer to monitoring data and literature to determine the range of phenological variation for an occurrence to plan the first site visit.

Time the first visit when short-leaved dudleya is in full flower to make it easier to locate. Use pin flags to demarcate the boundary of the occurrence and place pin flags adjacent to short-leaved dudleya individuals to relocate them easily during subsequent visits.

Return to the occurrence within two to four weeks to check phenology. Weather conditions will determine the rate of desiccation; plants will dry quicker in warm and dry conditions and slower if conditions remain cool and moist. Continue monthly site visits until the majority of plants in an occurrence are fully desiccated and seeds resemble those in Figure 5.6-1.

Voucher Specimens

Collect voucher specimens of short-leaved dudleya if none exist for the source occurrence and if enough plants are available so that collecting a few specimens does not adversely affect the

occurrence. Check the San Diego Natural History Museum or Consortium of California Herbaria to determine if a voucher specimen already exists:

San Diego Natural History Museum: http://sdplantatlas.org/publicsearch.aspx

Consortium of California Herbaria: http://ucjeps.berkeley.edu/consortium/

Note that recently collected specimens may not yet be accessed into an herbariumôs collection. In addition, older specimens may not include sufficient locality information to definitively determine whether they were collected at the occurrence. If it is not appropriate to collect a voucher specimen, photograph the occurrence (landscape aspect) and an individual plant while in flower, and then again when the plant is in fruit.

Obtain all necessary permits and memorandums before collecting voucher specimens of endangered, threatened, and rare species. Collect specimens while plants are still in flower and before collecting seed. Refer to Section 4.1 of this document (Seed Collecting, and Materials and Methods) and instructions and forms in Appendix B to guide your collection. Submit the voucher specimen(s) to a recognized herbarium (i.e., San Diego Natural History Museum) as soon as possible.

Methods and Materials

We recommend that experts familiar with harvesting short-leaved dudleya seed perform seed collections since the seed is very delicate and easily damaged (Anderson pers. comm.). Follow the methods provided in this section to collect short-leaved dudleya seed.

Refer to Section 4.1 of this document for a detailed discussion of methods and materials and Table 4.1-4 for a comprehensive list of recommended equipment for collecting seed. At a minimum, bring the following to each short-leaved dudleya collection location:

- Regulatory permits and access agreements
- Directions, map(s)
- Lock combination(s), key(s)
- Collecting strategy/instructions
- GPS unit (preferably, sub-meter)
- Camera
- Labels, markers, pens or pencils
- Seed collection forms
- Hand lens
- Adhesive tape
- Safety razor or pocketknife

- Container and water (optional)
- Scissors, snips, sieve (optional)
- Small paper envelopes or bags

Before collecting seed, map the seed collection area(s), record data on the seed collection form(s), and count or estimate the number of plants within an occurrence. Collect no more than 5-10% of the seed from an individual plant or from the reproductive population per season. Use the following method to determine the number of seeds to collect by plant and for an occurrence:

Step 1: Select 3-5 plants of different sizes spaced randomly across the occurrence.

Step 2: Count or estimate the number of seeds on the 3-5 sampled plants. Add together the number of seeds counted or estimated from each sampled plant to obtain the *total number of seeds sampled*.

Step 3: Divide the total number of seeds sampled (Step 2) by the number of plants sampled (Step 1) to calculate the *average number of seeds per plant*.

Step 4: Multiply the average number of seeds per plant (Step 3) by the occurrence count or estimate to calculate the *estimated number of seeds per occurrence*. In most cases, the amount of seed available for collecting will be higher in years with optimal growing conditions (e.g., average or above-average rainfall) and lower during drought years.

Step 5: Calculate the maximum number of fruits or seeds to collect per plant or occurrence to stay within the 5-10% collecting guidelines.

Note that where seed production is very heavy, seed collectors may choose to estimate fruit and seed quantities using subsamples.

Collect seed when short-leaved dudleya plants are fully dry. Remove dry flower heads from the plants by hand and place them in a seed collecting envelope or bag. Refer to Figure 5.6-1 for ripe seeds. Seed may be ready to collect as early as one month after peak flowering.

Collect seed during early, mid, and late season to account for differences in maturity times and to capture the phenological variability among the occurrence. Collect randomly and evenly throughout the occurrence, taking care to collect from plants of various sizes and with variable inflorescences, as well as plants growing in unique or varying habitat conditions.

Consider harvesting more seed than recommended by the CPC for small occurrences that are at serious risk of extirpation to capture the highest amount of genetic variation at the occurrence (McMillan per. comm.).

Ideally, collect seed from 30-50 plants across the occurrence. If the number of plants present in a given year is below this number, then sample over multiple years to produce a genetically robust collection for restoration purposes. Where the occurrence is small and not expected to produce an adequate number of plants (and seed) in any given year, consider collecting seed from one or more occurrences within the same seed zone. Depending on project objectives and sampling strategy, collect along maternal lines or as a bulk collection.

Interim Seed Storage and Delivery

Refer to the Section 4.1 of this document (Seed Collecting) for guidelines on interim seed storage and delivery. We provide additional guidelines for practitioners that choose to keep the seed for future restoration purposes rather than sending it immediately to a storage facility:

- Do not clean seed unless the collection is very small because cleaning seed is time-consuming, difficult (especially with large amounts of seed), and can damage seed since the seed coat is very thin (Anderson pers. comm.).
- If choosing to clean seed, use a microscope and tweezers to remove seed from follicles to avoid damaging the seed (Anderson pers. comm.).
- Loosely pack the seed collection in envelopes or bags and store them in dark, dry, and moderate to cool conditions for several months before distributing seed onsite.
- If planning to hold seed for longer than one year, store the seed at a recognized seed bank (i.e., CBG, SDZG) to ensure proper storage conditions to retain viability.

Seed Banking

Existing Conservation and Restoration Collections

Refer to Table 5.6-3 for existing seed collections of short-leaved dudleya. This table includes only dudleya occurrences with seed in storage. For planning purposes, we indicate the seed zone for each collection. We also indicate the number of seeds present in the seed bank and their potential availability for restoration. Note that only a portion of the seeds in a collection will be available for these purposes. Existing permits or contracts may also limit the use of some collections. We obtained information on each collection (seed bank; accession number and year; and seed quantity, provenance, and availability) from seed bank managers. Refer to Table 4.2-3 (Section 4.2) for contact information for these seed banks; we encourage land managers to contact facilities directly to get more information on seed availability or to request seed.

Table 5.6-3. Short-leaved Dudleya: Existing Seed Collections.

Occurrence ID[1]	Seed Zone[2]	Seed Bank[3]	Accession Number[4]	Collection Year	Seed Quantity[5]	Provenance Type[6]	Seed Available[7]
DUBLB2_6CMPR001 (*Carmel Mountain Preserve*)	East	SDZG	S0689	2016	7,305	W	Limited
DUBLB2_6CMPR001 (*Carmel Mountain Preserve*)	East	CBG	15350	1986	101,025	W	Limited
DUBLB_7CRCA003 (*Crest Canyon Preserve*)	North	SDZG	S0693	2016	5,184	W	Limited
DUBLB_7CRCA003 (*Crest Canyon Preserve Sub*)	North	SDZG	S0786	2019	165	W	Limited
DUBLB2_SKCA002 (*Skeleton Canyon*)	South	SDZG	S0728	2017	94	W	Limited
DUBLB2_7TPEX004 (*Torrey Pines Extension*)	Central	SDZG	S0782	2019	3,981	W	Limited
DUBLB2_7TPSR005 (*Torrey Pines State Reserve*)	West	SDZG	S0783	2019	20,100	W	Limited

[1] Occurrence identification (ID) per the SDMMPôs MOM database.
[2] Seed Zone: Seed zone corresponds to the potential genetic clusters and population groups identified in the F-RPMP: North, Central, East, West, and South.
[3] Seed bank: **CBG** = California Botanic Garden, **SDZG** = San Diego Zoo Global.
[4] Accession number = accession number of seed collection assigned by seed bank.
[5] Seed quantity indicates the number of seeds currently in the collection, according to seed bank records.
[6] Provenance type indicates the source location of the seed collection: **W** = seed collected directly from the wild (origin known).
[7] Seed available: **Limited** = seed is only available for use by Land Manager (owner of seed).

Best Management Practices

Seed Cleaning

We recommend leaving short-leaved seed in the follicles instead of removing it. The seed is very small (<1mm) and delicate, and has a thin seed coat. Removing the seed from the follicle and cleaning it is difficult, time consuming, and may damage seed (Anderson pers. comm.). Store collected follicles and contact a seed facility that has experience cleaning short-leaved dudleya seeds for reintroduction efforts that require pure seed. SDZG cleans short-leaved dudleya seed under a microscope using tweezers (Anderson pers. comm.). CBG uses the following methods to clean and sort seeds of variegated dudleya (*Dudleya variegata*), a species similar to short-leaved dudleya in size and habit:

- Place follicles and floral material in a #45 soil sieve and rub.
- Place resulting seed and chaff in a seed blower (1.25 blower speed).
- Remove by hand any remaining chaff.
- Continue to blow until only clean, filled seed remains.

Seed Testing

Seed testing methods will vary by facility, but all seed banks conduct baseline germination tests on new accessions and then conduct follow-up testing to check seed for viability.

At CBG, baseline tests for seed germination use a typical seed sample of 50 seeds, although sample size may vary depending on the size of the seed collection and methods to break dormancy (if needed). CBG provides abundant time for seeds to germinate and at the end of a germination test they conduct a cut test (see Section 4.2) on a sample of 5 ungerminated seeds (Birker pers. comm.). CBG also conducts follow-up testing on seeds in storage at intervals (e.g., 1 year, 5 years, 10 years, 20 years, and every subsequent 10 years thereafter, as seed quantities allow).

At SDZG, a baseline germination test is conducted on all new accessions. The test is conducted primarily on fresh seed. Follow-up testing is conducted on frozen seed after 1, 5, and 10 years in storage. Seed is sterilized with bleach and placed in reverse osmosis water at room temperature for 24 hours to fully imbibe (absorb) water. Seed is then plated onto agar trays (10 seeds per tray) and observed for signs of germination. The seed is counted as germinated when the radical emerges. SDZG does not use a cut test to determine viability. Rather, they provide abundant time for seeds to germinate (Anderson pers. comm.).

SDZG produced high seed germination rates for short-leaved dudleya ranging from >85% - <100% using fresh seed (imbibe) (Anderson 2020).

Seed Storage

All seed banks contacted store short-leaved dudleya seed according to CPC guidelines. For example, SDZG dries seeds down to 23-35% moisture content. Seed is then stored in sealed, foil-lined envelopes in the freezer at a low temperature (-18ÁC to -23ÁC).

Each collection is separated into three packages that represent the primary, curation, and duplicate collections, as described in Section 4.1. The primary collection is the active seed collection and seeds in this package are available for research, testing, restoration, and other uses. The curation package is the base collection for long-term storage. This package is not opened and is not available for other uses. It constitutes the long-term conservation collection. The duplicate collection is sent to another seed bank facility as backup storage to protect against catastrophic loss at the primary storage facility. Both SDZG and CBG send their duplicate seed collections to the NLGRP.

Seed Bulking

Seed Selection

Refer to Section 4.3 for guidelines on seed selection to maximize the quality of the bulked sample. Key considerations include:

- Use seed for bulking from the target occurrence or another occurrence in the same seed zone as the target occurrence. For very small occurrences (fewer than 100 plants), use seed from a large occurrence (if present) or from one or more smaller occurrences (composite provenancing) within the same seed zone.
- Use seed that was collected randomly and uniformly.
- Use an adequate amount of seed for bulking (ideally, Ó1,000 seeds or seed from 30-50 plants).
- For very small seed samples, use seed collected along maternal lines.

Best Management Practices

Germination and Propagation Methods

Fresh short-leaved dudleya seeds will germinate in approximately three days with continuous moisture, 11 hours of sunlight, and temperatures ranging between 10-15 degrees Celsius. Under these conditions, >90% of seeds will germinate (Dodero 2020).

Germinate seeds toward the end of October when temperatures are cool. Do not attempt to germinate seeds in summer when temperatures are high. Although untested, Dodero (2020) suggests dormancy mechanisms may prevent seed from germinating during high temperatures.

Propagate short-leaved dudleya seed in soil-filled flats or pots in a greenhouse (Anderson 2020, Dodero 2020) (Figure 5.6-4. Thoroughly and regularly water seeded flats and pots. Greenhouse-produced plants can reach flowering size in 6 months, while *Hasseanthus* seedlings in the wild can reach flowering size in 3-5 years (Dodero 2020). During the first year of growth and under natural conditions, short-leaved dudleya plants focus on enlarging the tuberous caudex and typically produce only two cotyledons (Dodero 2020). The tuberous caudex grows 5-10 mm during the first season.

Figure 5.6-4. Propagated Short-leaved Dudleya Seedlings
(Photo credit: San Diego Zoo Global)

Although researchers and experts have not propagated short-leaved dudleya using leaf cuttings, the method is successful for species of *Hasseanthus,* especially those with narrow/thin petioles (Dodero 2020).

Limits on Generations

At this time, we recommend bulking short-leaved dudleya seed for only one generation (F1) in a nursery setting. If additional seed is needed for restoration, start with wild seed rather than the F1 generation.

Documentation

We encourage growing facilities and others bulking seed of short-leaved dudleya for reintroduction purposes to document bulked samples as indicated in Section 4.3 and provide this information to land managers when seed is delivered. Likewise, we encourage land managers to provide information on seed collection and seed reintroductions to the SDMMP (contact information at https://sdmmp.com/about.php#contact) and include relevant information on IMG monitoring forms (i.e., translocated occurrence). Refer to Table 5.6-4 for key information for bulked seed samples.

Table 5.6-4. Documentation for Bulked Seed Samples.

Key Information	Primary Responsibility
Seed Source	Seed Collector and/or Land Manager
Seed Collection Date	Seed Collector
Amount of Seed Bulked	Growing Facility
Propagation Method	Growing Facility
Propagation Conditions	Growing Facility
Harvesting Date(s)	Growing Facility
Post-harvest Storage Location/Duration/Conditions	Growing Facility
Generation of Bulked Sample	Growing Facility
Receptor Site Location	Growing Facility and/or Land Manager

5.7 WILLOWY MONARDELLA (*MONARDELLA VIMINEA*)

Seed Collection

Seed Characteristics

Willowy monardella flower heads can support up to 80 flowers with each flower producing up to four smooth, shiny, brown and speckled, ovoid seeds that are less than 1.9 mm long (Epling 1925, Elvin and Sanders 2009). One willowy monardella individual can produce hundreds to thousands of seeds (Figure 5.7-1) depending on plant size (Anderson pers. comm.).

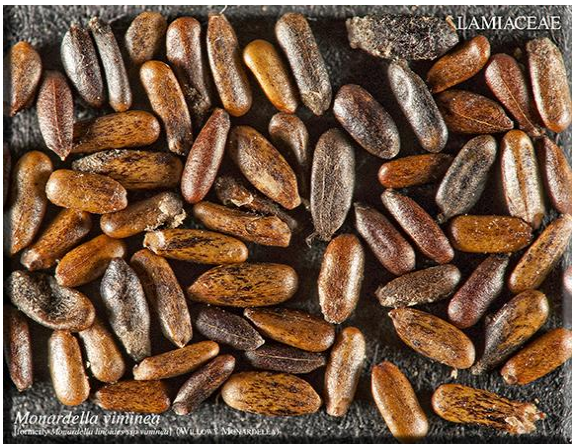

Figure 5.7-1. Willowy monardella seed (Credit: California Botanic Garden)

Germination and establishment of plants in the wild are very low despite high seed production and lab germination rates (Anderson pers. comm., Kelly pers. comm.). Reasons for low germination and establishment are unknown, but biologists have identified nonnative species, erosion, altered hydrology, herbivory, and drought as factors that may affect germination and establishment (Kelly 2020).

Insects likely pollinate willowy monardella and seeds are possibly transported by flowing water (Akiba et al. 2018, Elvin and Sanders 2003). Willowy monardella seed forms throughout the summer months and matures in mid-August.

Priority Occurrences

Table 5.7-1 prioritizes occurrences of willowy monardella on conserved lands in the MSPA for seed-related management actions. These priorities were developed in the F-RPMP based on monitoring data and specific studies, including genetic analyses. The decision to implement an action will depend on a number of factors, including status of the occurrence (stable or declining), habitat condition, threats management, and funding, among others.

In general, large occurrences with high genetic diversity are prioritized as potential seed sources, subject to land manager and/or agency approval. Small occurrences are high priority candidates for seed reintroduction, subject to response to management and site conditions, as discussed in the F-RPMP.

Currently, conservation seed collections exist in established seed banks for four willowy monardella occurrences (57%) on conserved lands in the MSPA (Table 5.7-1).

Table 5.7-1. Willowy Monardella: Management Priorities for Seed-Related Activities

Occurrence ID[1]	Occurrence Name	Occurrence Size[2]	Genetic Structure[3]	Seed Reintroduction Priority[4]	Existing Seed Collection[5]	Seed-related Activities[6]			
						Seed Source	Conservation Collection	Restoration Collection	Seed Bulking
MOLIV_4SYCA001[7]	Sycamore Cyn	Small	L+H+L	H	SDZG			✓	✓
MOLIV_4SYCA002[7]	Sycamore Cyn at Sycamore Cyn Rd	Small	L+H+L	H			✓	✓	✓
MOLIV_4WSCA003[8]	West Sycamore Cyn	Small	L+H+L	H		✓[9]	✓	✓	✓
MOLIV_4SYCA006[8]	Sycamore Cyn East	Medium	L+H+L	L	SDZG	✓[9]		✓	✓
MOLIV_4SPCA008[8]	Spring Cyn	Small	L+L+L	H			✓	✓	✓
MOLIV_6LOCA004	Lopez Cyn	Small	L+H+L	H	SDZG			✓	✓
MOLIV_6FLCA007	Flanders Cyn	Small	L+H+L	H	SDZG			✓	✓

[1] Occurrence ID: Occurrence identification (ID) code per the San Diego Management and Monitoring Program's (SDMMP) Master Occurrence Matrix (MOM) database.
[2] Occurrence Size: Recent size category as defined in the Framework-Rare Plant Management Plan (F-RPMP). **Small** = <100 plants**, Medium** = 100-500 plants**, Large** = >500 plants.
[3] Genetic Structure (per Milano and Vandergast 2018): Genetic differentiation + genetic diversity + inbreeding. **L** = low, **H** = High, **L** = Low. Refer to F-RPMP for a discussion of the implications for seed movement.
[4] Seed Reintroduction Priority (per F-RPMP): **High** = high priority for seed reintroduction, **Medium** = medium priority for seed reintroduction, **Low** = low priority for seed reintroduction, --- = not a priority for seed reintroduction at this time due to stable population (large occurrence) or lack of monitoring data.
[5] Existing Seed Collection: Indicates location of collection. **SDZG** = San Diego Zoo Global.
[6] Seed-related Activities: ✓ indicates occurrence is suitable as a seed source (pending authorization from responsible entities) or management action is warranted. Where there is an existing seed collection, a restoration collection may not be needed, but land managers should contact the seed bank directly to make that determination. Likewise, seed bulking may not be necessary depending on the amount of seed available in an existing collection or collected specifically for restoration purposes.
[7] Occurrences are upstream and separated from larger Marine Corps Air Station (MCAS) Miramar occurrences by >0.5-mile.
[8] Occurrences are part of larger populations located on MCAS Miramar or private land.
[9] Occurrences on conserved lands are small but are connected to large genetically diverse populations on MCAS Miramar and therefore are suitable seed collection sources.

We recommend conservation collections as a potential management action for all occurrences that are not currently represented in a seed bank *if adequate seed is available*. Large occurrences are the highest (or most immediate) priorities for seed conservation to maximize the amount of genetic diversity in long-term storage. Although there are no large willowy monardella occurrences on conserved lands, one medium and one small occurrence are part of larger populations on MCAS Miramar or private land and therefore are suitable seed collection sources (i.e., MOLIV_4WSCA003, MOLIV_4SYCA006).

We recommend restoration collections as a potential management action for occurrences that are high priorities for seed reintroduction as indicated in Table 5.7-1 and identified in the F-RPMP.

We recommend reintroducing seed only if threats are controlled, habitat is likely to support this species in the future, and funding is available for short- and long-term management. Reintroducing seed is most appropriate for small occurrences that continue to decline, even with management. Seed may be introduced into extirpated occurrences if threats are managed and the habitat is likely to support willowy monardella in the future. For extirpated occurrences, source seed from the nearest occurrence or from an appropriate seed source in storage (if available). Some threats (particularly, invasive plants) will require perpetual management; thus, ensure that adequate funding and/or labor are available to manage the occurrence after seed is reintroduced.

Seed Zones

Seed zones place geographic limits on the transfer of seed between occurrences of different or unknown genetic structure. Genetic studies of willowy monardella indicate that the species has generally low genetic differentiation (divergence), high genetic diversity within occurrences, and low levels of inbreeding (Milano and Vandergast 2018). Spring Canyon (MOLIV_4SPCA008) is the exception with low genetic diversity. Milano and Vandergast (2018) did not find distinct genetic clusters or evidence of isolation by distance, including Spring Canyon, and concluded that the species has a high rate of gene flow and low risk of outbreeding depression (Milano and Vandergast 2018). Thus, we established one seed zone for willowy monardella that encompasses all occurrences and corresponds to the one genetic cluster identified for this species by Milano and Vandergast (2018) (Figure 5.7-2).

Within the seed zone, we recommend that seed transfer proceed between occurrences that are relatively close to one another. The same limit on seed movement applies when using willowy monardella seed from a seed bank. Since Spring Canyon has low genetic diversity, we do not recommend transferring seed from this occurrence to other occurrences or to establish new occurrences.

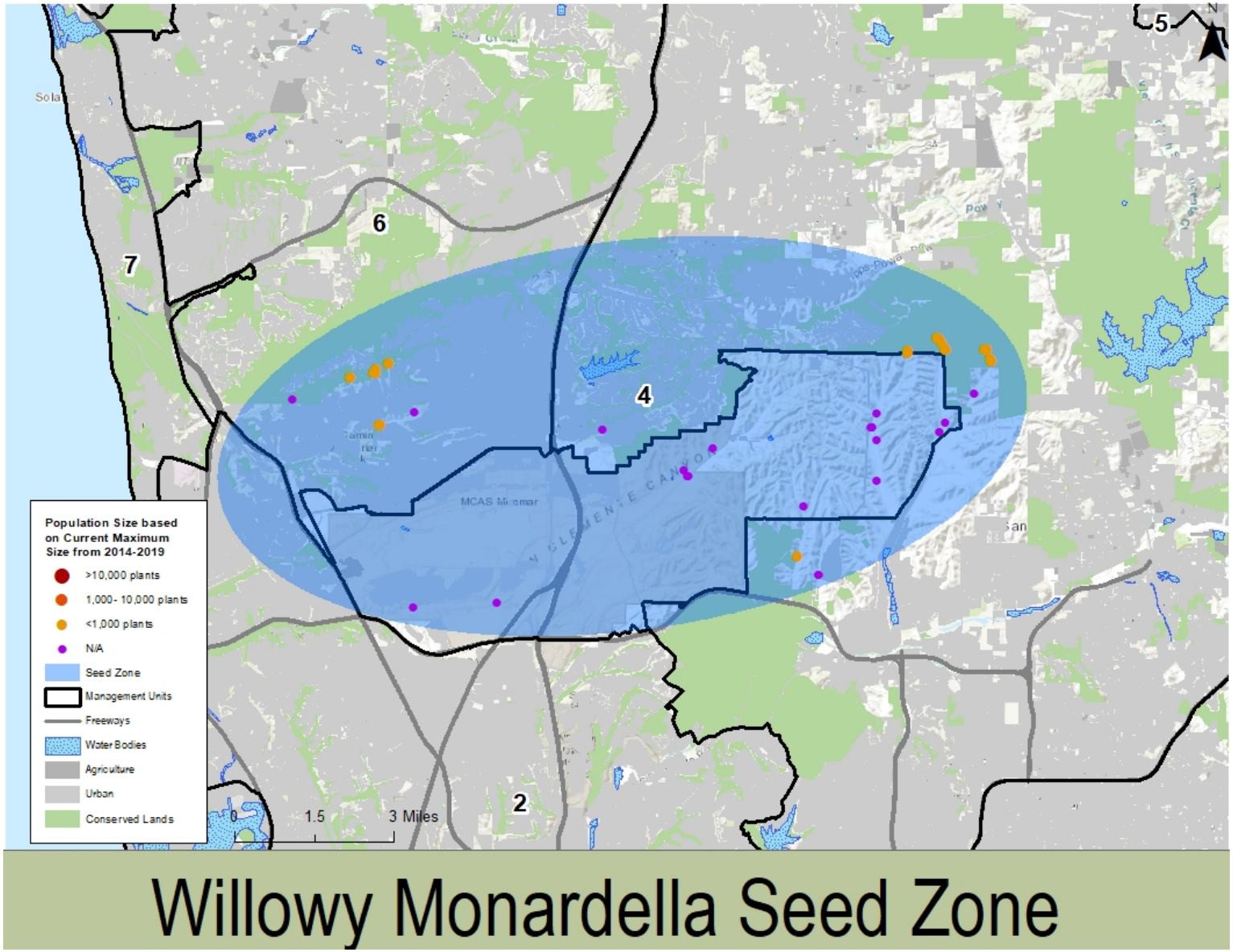

Figure 5.7-2. Willowy Monardella: Seed Zone.

Collecting Plan

Refer to Section 4.1 of this document for guidelines on developing a sampling strategy for seed collection. Identify the purpose and type of collection needed for a specific project or restoration effort, as well as timeline and costs. In the section below, we highlight key steps in the process, including those specific to willowy monardella.

Sampling Strategy

Refer to Table 5.7-1 for prioritized seed-related activities for the target occurrence to identify whether seed is needed for a conservation collection, a restoration collection, or both.

For restoration, determine if seed is available in a seed bank to reintroduce directly into a target occurrence or for bulking. Proceed with the collecting plan if stored seed is not available, quantities are insufficient, or the provenance is not appropriate for the target occurrence (e.g., different seed zone).

Assess whether the target occurrence will likely provide an adequate quantity of seed for restoration or if seed will need to be sourced from other occurrences in the seed zone. Refer to the seed zone (Figure 5.7-2) for limits on seed movement and seed sources. Contact the appropriate land managers of potential seed source occurrences to obtain permission for seed collection.

Based on the purpose and type of collection, and amount of seed expected to be available, determine whether the seed is likely to be collected along maternal lines or in bulk (Chapter 4, Table 4.1-3), and set minimum targets for the number of plants to sample and amount of seed to collect. These targets are intended to provide a guide for planning and may be modified based on the actual seed crop.

Permitting and Agreements

Ensure that all regulatory permits, memorandums, and access agreements are in place before collecting seed from an occurrence.

If unfamiliar with the collecting location, contact the land owner to obtain a map and coordinates of the occurrence; directions to the occurrence if necessary; gate codes, lock combinations, or keys; and any pertinent information about the occurrence (e.g., informing adjacent land owners, closures due to unsafe conditions).

Pre-collection Monitoring

Practitioners can collect willowy monardella seed beginning in August and into early fall, depending on location, habitat, and weather and temperature conditions during the previous fall, winter, and spring. Visit the target occurrence multiple times to determine the best time to collect

seed. Talk to land owners and managers and refer to monitoring data and literature to determine the range of phenological variation for an occurrence to plan the first site visit.

Place pin flags adjacent to willowy monardella plants within small occurrences because relocating plants after they dry can be difficult depending on the occurrence location and habitat. Return to the occurrence within four to six weeks to check phenology. Weather conditions will determine the rate of seed set; seed will dry quicker in warm and dry conditions and slower in cool and moist conditions. Continue monthly site visits until the majority of flower heads on willowy monardella plants are fully desiccated and seeds are shiny, brown, and specked, resembling those in Figure 5.7-1.

Voucher Specimens

Collect voucher specimens of willowy monardella if none exist for the source occurrence. Check the San Diego Natural History Museum or Consortium of California Herbaria to determine if a voucher specimen already exists:

San Diego Natural History Museum: http://sdplantatlas.org/publicsearch.aspx

Consortium of California Herbaria: http://ucjeps.berkeley.edu/consortium/

Note that recently collected specimens may not yet be accessed into an herbarium's collection. In addition, older specimens may not include sufficient locality information to definitively determine whether they were collected at the occurrence. If it is not appropriate to collect a voucher specimen, photograph the occurrence (landscape aspect) and an individual plant while in flower, and then again when the plant is in fruit.

Obtain all necessary permits and memorandums before collecting voucher specimens of endangered, threatened, and rare species. Collect specimens while plants are still in flower and before collecting seed. Refer to Section 4.1 of this document (Seed Collecting, and Materials and Methods) and instructions and forms in Appendix B to guide your collection. Submit the voucher specimen(s) to the San Diego Natural History Museum as soon as possible.

Methods and Materials

We recommend that experts familiar with harvesting willowy monardella seed perform seed collections since the seed is very delicate and easily damaged (Anderson pers. comm.). Follow the methods provided in this section to collect willowy monardella seed.

Refer to Section 4.1 of this document for a detailed discussion of methods and materials and Table 4.1-4 for a comprehensive list of recommended equipment for collecting seed. At a minimum, bring the following to each willowy monardella collection location:

- Regulatory permits and access agreements
- Directions, map(s)
- Lock combination(s), key(s)
- Collecting strategy/instructions
- GPS unit (preferably, sub-meter)
- Camera
- Labels, markers, pens or pencils
- Seed collection forms
- Hand lens
- Adhesive tape
- Safety razor or pocketknife
- Container and water (optional)
- Scissors, snips, sieve (optional)
- Small paper envelopes or bags

Before collecting seed, map the collection area(s), record data on the seed collection form(s), and count or estimate the number of plants within an occurrence. The number of seeds per willowy monardella plant will vary since each flower head contains multiple flowers and up to 80 seeds; therefore, we recommend collecting ripe flower heads versus counting and collecting seed in the field. Collect no more than 5-10% of flower heads from an individual plant per season. Figure 5.7-3 depicts desiccating willowy monardella flower heads.

Figure 5.7-3. Willowy monardella senescing flower heads
(Credit: Jessie Vinje)

Use the following method to determine the number of flower heads to collect by plant:

Step 1: Select 3-5 plants of different sizes spaced randomly across the occurrence.

Step 2: Count or estimate the number of flower heads per sampled plant.

Step 3: Calculate the maximum number of flower heads to collect per plant to stay within the 5-10% collecting guidelines. For example, if a sampled plant supports 100 flower heads, then collect 5-10 flower heads to remain within the 5-10% collecting guidelines.

In most cases, the number of flower heads (i.e., amount of seed) available for collecting will be higher in years with optimal growing conditions (e.g., average or above-average rainfall) and lower during drought years.

Collect fully dry flower heads during early, mid, and late season to account for differences in maturity times and to capture the phenological variability among the occurrence. Visually inspect a sub-sample of flower heads to ensure that seed is fully dry, shiny, light to dark brown and speckled (Figure 5.7-3).

Collect randomly and evenly throughout the occurrence, taking care to collect flower heads from plants of various sizes and with variable inflorescences, as well as plants growing in unique or varying habitat conditions. Remove dry flower heads from the stems by hand and place them in a seed collecting envelope or bag.

Consider harvesting more seed than recommended by the CPC for small occurrences that are at serious risk of extirpation to capture the highest amount of genetic variation at the occurrence (McMillan pers. comm.). In this case, coordinate with the regulatory agencies prior to the collecting effort.

Ideally, collect seed from 30-50 plants across the occurrence. If the number of plants present in an occurrence is below this number, then sample over multiple years to produce a genetically robust seed collection for restoration purposes. Where the occurrence is small and not expected to produce an adequate amount of seed in any given year, consider collecting seed from one or more occurrences within the population sub-group. Depending on project objectives and sampling strategy, collect seed along maternal lines or as a bulk collection.

Interim Seed Storage and Delivery

Refer to Section 4.1 of this document (Seed Collecting) for guidelines on interim seed storage and delivery. We provide additional guidelines for practitioners that choose to keep the seed for future restoration purposes rather than sending it immediately to a storage facility:

- Do not clean and sort seed unless the collection is very small because cleaning seed is time-consuming and often difficult, especially with large amounts of seed.
- Loosely pack the seed collection in envelopes or bags and store them in dark, dry, and moderate to cool conditions for several months before distributing seed onsite.
- If planning to hold seed for longer than one year, store the seed at a recognized seed bank (i.e., CBG, SDZG) to ensure proper storage conditions to retain viability.

Seed Banking

Existing Conservation and Restoration Collections

Refer to Table 5.7-2 for existing seed collections of willowy monardella. This table includes only monardella occurrences with seed in storage. For planning purposes, we indicate the seed zone for each collection. We also indicate the number of seeds present in the seed bank and their potential availability for restoration. Note that only a portion of the seeds in a collection will be available for these purposes. Existing permits or contracts may also limit the use of some collections. We obtained information on each collection (seed bank; accession number and year; and seed quantity, provenance, and availability) from seed bank managers. Refer to Table 4.2-3 (Section 4.2) for contact information for these seed banks; we encourage land managers to contact facilities directly to get more information on seed availability or to request seed.

Best Management Practices

Seed Cleaning

Wall and MacDonald (2009) suggest the following methods to clean and sort seeds of *Monardella* spp.:

- Rub floral material over a small to medium-size screen.
- Rub and shake floral material through a series of sieves to remove extraneous plant material and release seeds.
- Use a blower at low speed to separate chaff from seed, and successively higher speeds to separate hollow, sterile seeds from fertile seeds.

Wall and MacDonald (2009) indicate that cleaning *Monardella* spp. seed is generally easy because many thousands of seeds can be processed in a relatively short period of time.

Seed Testing

Seed testing methods will vary by facility, but all seed banks conduct baseline germination tests on new accessions and then conduct follow-up testing to check seed for viability.

Table 5.7-2. Willowy Monardella: Existing Seed Collections.

Occurrence ID[1]	Seed Zone[2]	Seed Bank[3]	Accession Number[4]	Collection Year	Seed Quantity[5]	Provenance Type[6]	Seed Available[7]
San Diego - Carrol Canyon	Central	CBG	21117	2003	65	W	Limited
MOLIV_4SYCA001 (*Sycamore Canyon*)	Central	SDZG	S0696	2016	19,545	W	Limited
MOLIV_6FLCA007 (*Flanders Canyon*)	Central	SDZG	S0732	2017	10,788	W	Limited
MOLIV_6LOCA004 (*Lopez Canyon*)	Central	SDZG	S0734	2017	5,109	W	Limited
MOLIV_4SYCA006 (*Sycamore Canyon*)	Central	SDZG	S0750	2018	26,381	W	Limited

[1] Occurrence identification (ID) per the SDMMP's MOM database. If no occurrence ID exists for a location, we provide the name of the location as indicated on collection in seed bank.
[2] Seed zone: Seed zone corresponds to the one genetic cluster and population group identified in the F-RPMP: Central.
[3] Seed bank: **CBG** = California Botanic Garden, **SDZG** = San Diego Zoo Global.
[4] Accession number = accession number of seed collection assigned by seed bank.
[5] Seed quantity indicates the number of seeds currently in the collection, according to seed bank records.
[6] Provenance type indicates the source location of the seed collection: **W** = seed collected directly from the wild (origin known).
[7] Seed available: **Limited** = amount of seed available for Land Managers (owner of seed).

At CBG, baseline tests for seed germination use a typical seed sample of 50 seeds, although sample size may vary depending on the size of the seed collection and methods to break dormancy (if needed). CBG provides abundant time for seeds to germinate and at the end of a germination test they conduct a cut test (see Section 4.2) on a sample of 5 ungerminated seeds (Birker pers. comm.). CBG also conducts follow-up testing on seeds in storage at intervals (e.g., 1 year, 5 years, 10 years, 20 years, and every subsequent 10 years thereafter, as seed quantities allow).

At SDZG, a baseline germination test is conducted on all new accessions. The test is conducted primarily on fresh seed. Follow-up testing is conducted on frozen seed after 1, 5, and 10 years in storage. Seed is sterilized with bleach and placed in reverse osmosis water at room temperature for 24 hours to fully imbibe (absorb) water. Seed is then plated onto agar trays (10 seeds per tray) and observed for signs of germination. The seed is counted as germinated when the radical emerges. SDZG does not use a cut test to determine viability. Rather, they provide abundant time for seeds to germinate (Anderson pers. comm.).

SDZG produced seed germination rates for willowy monardella ranging from approximately 60%-90% using fresh seed (imbibe). Germination of most seeds occurred by day 25 (Anderson 2020).

Seed Storage

All seed banks contacted store willowy monardella seed according to CPC guidelines. For example, SDZG dries seeds down to 23-35% moisture content. Seed is stored in sealed, foil-lined envelopes in the freezer at a low temperature (-18°C to -23°C).

Each collection is separated into three packages that represent the primary, curation, and duplicate collections, as described in Section 4.1. The primary collection is the active seed collection and seeds in this package are available for research, testing, restoration, and other uses. The curation package is the base collection for long-term storage. This package is not opened and is not available for other uses. It constitutes the long-term conservation collection. The duplicate collection is sent to another seed bank facility as backup storage to protect against catastrophic loss at the primary storage facility. Both SDZG and CBG send their duplicate seed collections to the NLGRP.

Seed Bulking

Currently, we do not recommend bulking willowy monardella since practitioners can collect adequate amounts of seed from the wild, although the species is easy to grow. Plants germinated on agar trays are easily transplanted into fast draining nursery grade potting soil (Anderson pers com. 2020). Seeds also germinate in containers filled with nursery grade potting soil and on wet coir inside of plastic containers placed in a refrigerator (Gordon pers. com. 2020).

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Appendix A
Meeting Participants

Rare Plant Management Group Steering Committee

Rare Plant Working Groups

Appendix A

Participants in Rare Plant Management Group, Steering Committee Meeting and Working Group Meetings

Rare Plant Management Group Steering Committee Meeting: June 12, 2019.

Steering Committee Participant	Organization
Sara Allen	City of San Diego
Mark Berninger	City of San Diego
Mary Crawford	U.S. Fish and Wildlife Service
Mark Dodero	RECON Environmental
Patricia Gordon-Reedy	Conservation Biology Institute
Jenna Hartsook	AECOM
Christa Horn	San Diego Zoo Global
Joyce Maschinski	San Diego Zoo Global
Sarah McCutcheon	San Diego Management and Monitoring Program
Scott McMillan	Dudek
Thomas Oberbauer	AECOM
Chelsea Ohanesian	AECOM
Kris Preston	San Diego Management and Monitoring Program
Kyle Rice	California Department of Fish and Wildlife
Fred M. Roberts	Botanist
Kim Smith	San Diego Association of Governments
Amy Vandergast	U.S. Geological Survey
Susan Wynn	U.S. Fish and Wildlife Service

San Diego Thornmint Working Group Meeting: June 25, 2019.

Working Group Participant	Organization
Sara Allen	City of San Diego
Stacy Anderson	San Diego Zoo Global
Mark Berninger	City of San Diego
Cindy Burrascano	California Native Plant Society-San Diego
Carol Crafts	Friends of Goodan Ranch & Sycamore Canyon
Mark Dodero	RECON Environmental
Justin Daniel	California Native Plant Society-San Diego
John Ekhoff	California Department of Fish and Wildlife
Sarah Godfrey	Center for Natural Lands Management
Patricia Gordon-Reedy	Conservation Biology Institute
Christa Horn	San Diego Zoo Global
Mike Kelly	Friends of Los Peñasquitos Canyon Preserve
Anna Leavitt	RECON Environmental
Chris Manzuk	Endangered Habitats Conservancy
John Martin	San Diego National Wildlife Refuge
Joyce Maschinski	San Diego Zoo Global
Sarah McCutcheon	San Diego Management and Monitoring Program
Scott McMillan	Dudek
Margie Mulligan	Mulligan Biological Consulting
Tracie Nelson	California Department of Fish and Wildlife
Thomas Oberbauer	AECOM
Chelsea Ohanesian	AECOM
Meredith Osborne	California Department of Fish and Wildlife
Eric Piehel	AECOM
Kathleen Pollett	San Diego Habitat Conservancy
Kristine Preston	San Diego Management and Monitoring Program
Kyle Rice	California Department of Fish and Wildlife
Jonathan Snapp-Cook	U.S. Fish and Wildlife Service
Markus Spiegelberg	Center for Natural Lands Management
Fred Sproul	AECOM
Amy Vandergast	U.S. Geological Survey
Jessie Vinje	Conservation Biology Institute
Phoenix Von Hendy	Friends of Goodan Ranch & Sycamore Canyon
Gina Washington	City of San Diego

Otay Tarplant Working Group Meeting: June 25, 2019.

Working Group Participant	Organization
Sara Allen	City of San Diego
Mark Berninger	City of San Diego
Mark Dodero	RECON Environmental
John Ekhoff	California Department of Fish and Wildlife
Patricia Gordon-Reedy	Conservation Biology Institute
Christa Horn	San Diego Zoo Global
Anna Leavitt	RECON Environmental
John Martin	San Diego National Wildlife Refuge
Sarah McCutcheon	San Diego Management and Monitoring Program
Margie Mulligan	Mulligan Biological Consulting
Tracie Nelson	California Department of Fish and Wildlife
Chelsea Ohanesian	AECOM
Meredith Osborne	California Department of Fish and Wildlife
Kristine Preston	San Diego Management and Monitoring Program
Trish Smith	The Nature Conservancy
Linnea Spears-Lebrun	ICF
Jessie Vinje	Conservation Biology Institute

Nuttall's Acmispon Working Group Meeting: June 27, 2019.

Working Group Participant	Organization
Sara Allen	City of San Diego
Stacy Anderson	San Diego Zoo Global
Alys Arenas	Nature Collective
Christine Beck	California Department of Fish and Wildlife
Mark Berninger	City of San Diego
Cindy Burrascano	California Native Plant Society-San Diego
Megan Flaherty	San Diego Audubon
Patricia Gordon-Reedy	Conservation Biology Institute
Christa Horn	San Diego Zoo Global
Frank Landis	California Native Plant Society-San Diego
Carolyn Lieberman	U.S. Fish and Wildlife Service
Joyce Maschinski	San Diego Zoo Global
Sarah McCutcheon	San Diego Management and Monitoring Program
Andrew Meyer	Audubon
Margie Mulligan	Mulligan Biological Consulting
Tracie Nelson	California Department of Fish and Wildlife
Thomas Oberbauer	AECOM
Kris Preston	San Diego Management and Monitoring Program
Debbie Schafer	San Diego Gas & Electric
Julie Simonsen	Nature Collective
Darren Smith	California State Parks
Jessie Vinje	Conservation Biology Institute

Salt Marsh Bird's-beak Working Group Meeting: June 27, 2019.

Working Group Participant	Organization
Sara Allen	City of San Diego
Stacy Anderson	San Diego Zoo Global
Alys Arenas	Nature Collective
Mark Berninger	City of San Diego
Cindy Burrascano	California Native Plant Society-San Diego
Araceli Dominguez	City of San Diego
Patricia Gordon-Reedy	Conservation Biology Institute
Mark Hannaford	Tidal Influence
Christa Horn	San Diego Zoo Global
Sarah Hutmacher	San Diego River Park Foundation
Carolyn Lieberman	U.S. Fish and Wildlife Service
Sarah McCutcheon	San Diego Management and Monitoring Program
Margie Mulligan	Mulligan Biological Consulting
Daniel North	Tidal Influence
Thomas Oberbauer	AECOM
Chelsea Ohanesian	AECOM
Bronti Patterson	California State Parks
Kris Preston	San Diego Management and Monitoring Program
Heather Schneider	Santa Barbara Botanic Garden
Julie Simonsen	Nature Collective
Amy Vandergast	U.S. Geological Survey (U.S. Geological Survey)
Jessie Vinje	Conservation Biology Institute
Carol Williams	California Department of Fish and Wildlife

Orcutt's Spineflower Working Group Virtual Meeting: August 18, 2020.

Working Group Participant	Organization
Sara Allen	City of San Diego
Stacy Anderson	San Diego Zoo Global
Mark Berninger	City of San Diego
Diana Brand Ramirez	AECOM
Jeremy Bugarchich	San Diego Botanic Garden
Edward Christensen	City of San Diego
Mary Crawford	U.S. Fish and Wildlife Service
Naomi Fraga	California Botanic Garden
Tony Gurnoe	San Diego Botanic Garden
Jenna Hartsook	AECOM
David Hogan	Chaparral Lands Conservancy
Christa Horn	San Diego Zoo Global
Paula Jacks	AECOM
Daniel Leavitt	U. S. Navy
Michelle Maley	U. S. Navy
Joyce Maschinski	San Diego Zoo Global
Sarah McCutcheon	San Diego Management and Monitoring Program
Betsy Miller	San Bernardino Valley Water Conservation District
Tom Oberbauer	AECOM
Chelsea Ohanesian	AECOM
Meredith Osborne	California Department of Fish and Wildlife
Kristine Preston	San Diego Management and Monitoring Program
Jyotsna Sharma	Texas Tech University
Kim Smith	SANDAG
Amy Vandergast	U.S. Geological Survey
Jessie Vinje	Conservation Biology Institute

Short-leaved Dudleya Working Group Virtual Meeting: August 18, 2020.

Working Group Participant	Organization
Sara Allen	City of San Diego
Stacy Anderson	San Diego Zoo Global
Diana Brand Ramirez	AECOM
Jeremy Bugarchich	San Diego Botanic Garden
Mary Crawford	U.S. Fish and Wildlife Service
Mark Dodero	RECON Environmental
Tony Gurnoe	San Diego Botanic Garden
Jenna Hartsook	AECOM
David Hogan	Chaparral Lands Conservancy
Christa Horn	San Diego Zoo Global
Paula Jacks	AECOM
Joyce Maschinski	San Diego Zoo Global
Betsy Miller	San Bernardino Valley Water Conservation District
Tom Oberbauer	AECOM
Chelsea Ohanesian	AECOM
Meredith Osborne	California Department of Fish and Wildlife
Kristine Preston	San Diego Management and Monitoring Program
Kim Smith	SANDAG
Cara Stafford	California State Parks
Jessie Vinje	Conservation Biology Institute
Gina Washington	City of San Diego

Willowy Monardella Working Group Virtual Meeting: August 19, 2020.

Working Group Participant	Organization
Sara Allen	City of San Diego
Stacy Anderson	San Diego Zoo Global
Diana Brand Ramirez	AECOM
Cindy Burrascano	California Native Plant Society-San Diego
Mary Crawford	U.S. Fish and Wildlife Service
Lee Gordon	California Native Plant Society
Tony Gurnoe	San Diego Botanic Garden
Jenna Hartsook	AECOM
Christa Horn	San Diego Zoo Global
Paula Jacks	AECOM
Katy Kughen	U.S. Fish and Wildlife Service
Joyce Maschinski	San Diego Zoo Global
Betsy Miller	San Bernardino Valley Water Conservation District
Tom Oberbauer	AECOM
Chelsea Ohanesian	AECOM
Meredith Osborne	California Department of Fish and Wildlife
Kristine Preston	San Diego Management and Monitoring Program
Bethany Principe	County of San Diego
Kim Smith	SANDAG
Amy Vandergast	U.S. Geological Survey
Jessie Vinje	Conservation Biology Institute

Appendix B

Collecting Voucher Specimens

San Diego Natural History Museum

San Diego Zoo Native Plant Seed Bank

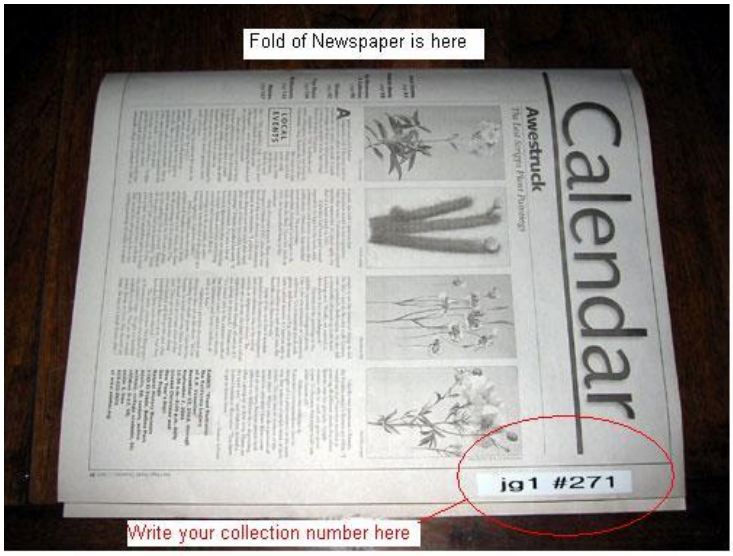
How to Collect and Press Proper Plant Specimens

As a parabotanist, your task is to contribute to our overall project goal, which is to curate one flowering/reproducing specimen of each plant species that occurs in each atlas grid square. Once you submit a pressed and dried specimen to the museum, we will mount it on a sheet of special paper with its label, and file it in the SD Herbarium. Herbarium specimens will last for hundreds of years if properly maintained and they are an effective way to scientifically document floristic diversity. Here is some general guidance on how to collect and press museum-quality plant specimens.

Field Collecting

- Plan ahead. Please review the maps and other information provided in your handout packet, and make sure you carry with you copies of your letter identifying you as a parabotanist, and any permits that are required for access and/or collecting. **Phone your contacts in advance** to make arrangements if you are working on public land.
- Before you go out, visit our website at www.sdplantatlas.org to check out which plant species have already been collected from your square. You can choose "Search the Database" to find this out. Print the list if you like, and carry it with you for reference.
- Once in the field, evaluate whether or not a species *should* be collected. Be able to recognize any possible "sensitive" species, i.e., those that are rare, threatened, or endangered that may be protected by law and may not be collected legally without special permits. *Do NOT collect sensitive species*!
- Do not endanger the local population if there are only a few individuals present. Use the "1 to 20" rule of thumb: for every one specimen you collect, there should be at least 20 more present in the surrounding population. (For herbs, the rule applies to individual plants; for shrubs and trees, it applies to shoots removed.)
- For herbs, dig up at least one whole plant to show roots that can help the botanist determine whether the plant is an annual, biennial, or perennial and identify the type of root (e.g., fibrous or tap) or underground stem (e.g., corm, bulb, rhizome, etc.).
- For shrubs, trees, or vines, clip one or more branches. The ideal plant specimen includes flowers (or other reproductive parts for ferns and non-vascular plants), fruit, leaves, and branches. Reproductive structures are often necessary to positively identify the plant, but it is not always possible to find flowers and fruit on the same plant at the same time. Do the best you can but do not mix together cuttings from different plants (i.e., don't take a branch from one plant and then take the fruits or flowers from another).
- Get enough of a sample to distribute over your 11x17 sheet in your plant press (e.g., a few branches of larger shrubs, or several small plants that can be distributed over the sheet).

- For cacti, slice and press the flowers, but place the stems and fruits into a paper bag. Label the bag with the same collection number as the flowers and submit them both to us. Similarly, large cones cannot be pressed so they may be placed into a paper bag with the same collection number as the rest of the specimen.
- Place each plant specimen inside a folded sheet of newspaper (like *The Reader*) and write its unique collection number on the upper outside edge of the newspaper, facing outwards (please see diagram below).



The "**collection number**" consists of your own "Login Initials", the # sign, and the specimen's unique collection number. In this example, the assigned login initials were "jg1", and the newspaper contains specimen number 271. Please write each collection number in the location and format shown here.

- Stack the newspapers and place in a field press with a few cardboard ventilators for support, or tie up the bundle with string, straps, or bungee cords.
- Record the field data for each specimen in your field notebook (including the collection number, and detailed information about the collecting location, surrounding vegetation, and characteristics of the plant itself).

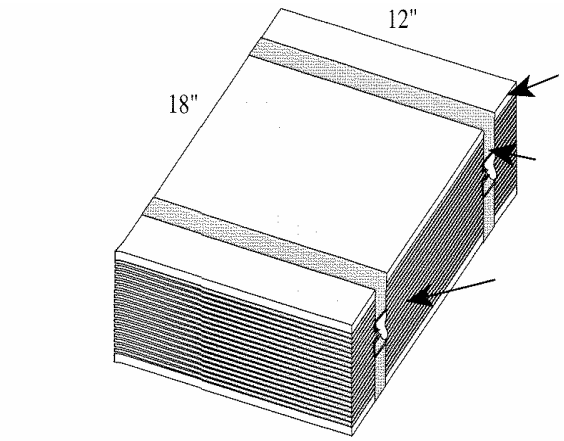
Plant Pressing

After you get your plants home from the field, they will have relaxed a bit and will be ready for you to transfer from your field setup to a plant press where they can be properly laid out, pressed and dried. Plants should be pressed within about 12 hours of their collection (otherwise the leaves may become moldy or the plant may shrivel up and not press nicely). If the newspaper is wet, you will need to change the paper, ensuring that you also transfer the collection number to the fresh paper.

- A basic plant press consists of two boards 12" by 18" (half-inch plywood or even thinner will do fine), plus two adjustable straps (or even ropes).
- Clean up the specimens (e.g., shake off excess soil from the roots and pick off dead leaves etc.) and if necessary trim or bend into a "V", "N" or "M" shape to neatly fit inside the press.
- Arrange the plants exactly as you want them to appear once they are mounted. Make sure leaves are spread out and not overlapping, that fruits and flowers are

showing, and turn over a few leaves so that the underside of several can be seen.

- Plants are pressed by placing each specimen inside one of the single sheets of folded newspapers, and separating each newspaper sheet with a cardboard ventilator so you have an alternating stack of newspaper and cardboard.
- Place the endboards on the outside of the stack, and tie straps around the outside as shown in diagram below. Tighten the straps down very hard.



Standard Plant Press

1/2" plywood endboards

Straps with buckles

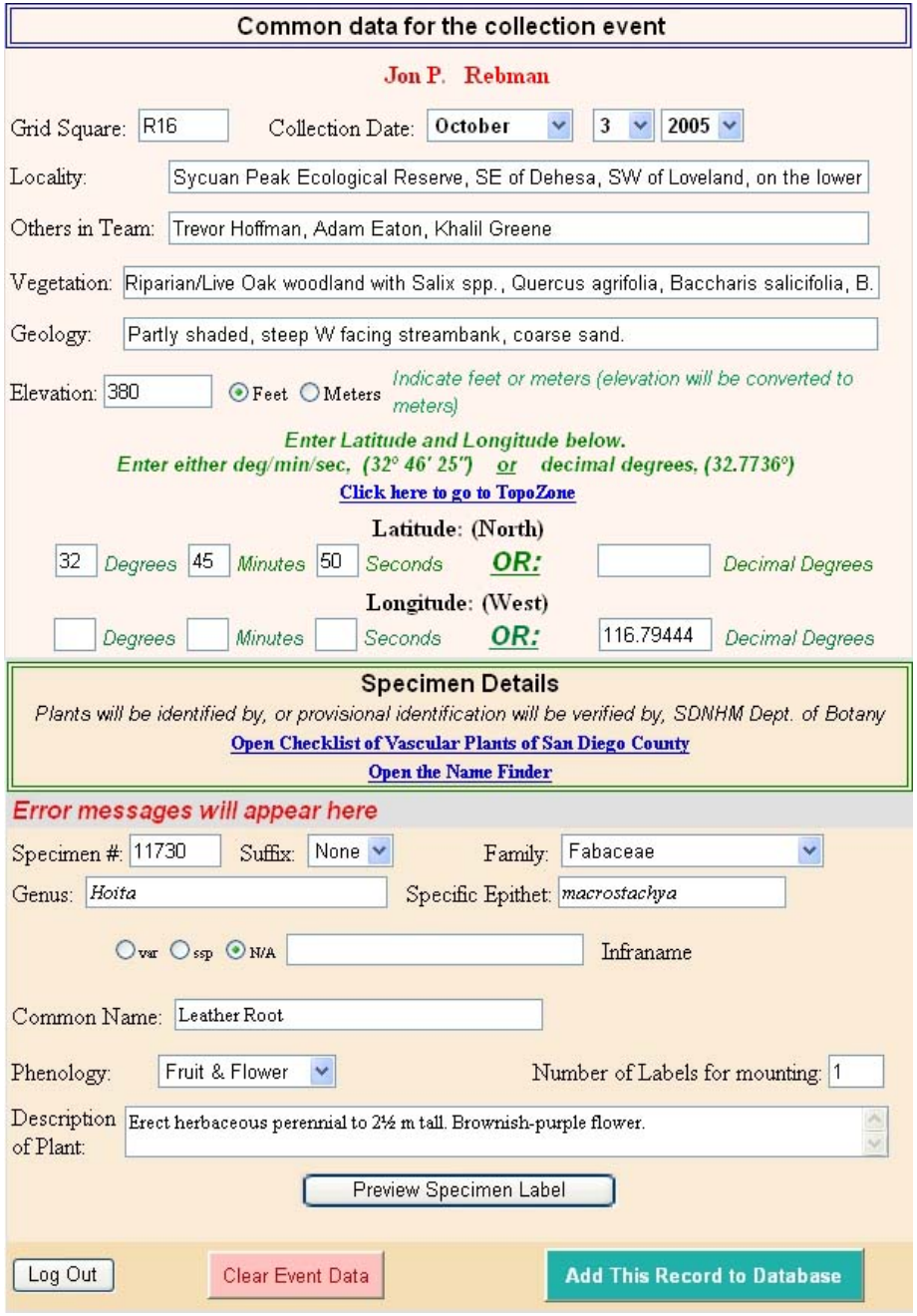
Layers of cardboard ventilators

From: Simpson, M.G. 1997 *Plant Collecting and Documentation Field Notebook*. SDSU Herbarium Press.

- Air dry by placing the plant press in a well ventilated location. It may take days to weeks for the plants to dry completely.
- Do NOT put the plants or plant press into a microwave or conventional oven. Heat is not necessary but good air circulation is.
- If required, change the paper every few days to prevent molding, especially for succulent plants. Plants are dry when they don't feel cool to the touch.
- For the health of those who must handle the dried plants and the specimens, please do not use chemicals of any kind on the plants (e.g., use no mothballs, insecticides etc.). During the verification, mounting, and accessioning process in the herbarium, all specimens are routinely frozen to kill any insect pests.
- Do not use any tape, glue or other adhesives.
- Sit down ASAP and enter the field data online at our website www.sdplantatlas.org. Timely data entry is important because it will permit other parabotanists sharing your square to see what has already been collected, thus helping to prevent duplication of effort.
- When it is time to submit your pressed and dried plants to the herbarium, please look on our website and print out the handout titled "*Guide to Specimen Submission*" and the "*Specimen Submission Form*".

For more details, refer to Simpson, Michael G. (1997) *Plant Collecting and Documentation Field Notebook*, SDSU Herbarium Press, for an excellent explanation of plant collection techniques.

Sample of Online Data Entry Form



Common data for the collection event

Jon P. Rebman

Grid Square: R16 Collection Date: October 3 2005

Locality: Sycuan Peak Ecological Reserve, SE of Dehesa, SW of Loveland, on the lower

Others in Team: Trevor Hoffman, Adam Eaton, Khalil Greene

Vegetation: Riparian/Live Oak woodland with Salix spp., Quercus agrifolia, Baccharis salicifolia, B.

Geology: Partly shaded, steep W facing streambank, coarse sand.

Elevation: 380 Feet Meters *Indicate feet or meters (elevation will be converted to meters)*

Enter Latitude and Longitude below.
Enter either deg/min/sec, (32° 46' 25") or decimal degrees, (32.7736°)
Click here to go to TopoZone

Latitude: (North)
32 Degrees 45 Minutes 50 Seconds OR: Decimal Degrees

Longitude: (West)
Degrees Minutes Seconds OR: 116.79444 Decimal Degrees

Specimen Details
Plants will be identified by, or provisional identification will be verified by, SDNHM Dept. of Botany
Open Checklist of Vascular Plants of San Diego County
Open the Name Finder

Error messages will appear here

Specimen #: 11730 Suffix: None Family: Fabaceae

Genus: *Hoita* Specific Epithet: *macrostachya*

var ssp N/A Infraname

Common Name: Leather Root

Phenology: Fruit & Flower Number of Labels for mounting: 1

Description of Plant: Erect herbaceous perennial to 2½ m tall. Brownish-purple flower.

Preview Specimen Label

Log Out Clear Event Data Add This Record to Database

CHEAT SHEET: What to Record in Your Field Notebook

Entry	Instructions
Date	Date you collected the specimen
Grid Square	Atlas grid square you are collecting in (e.g., A3, G14)
Locality	Provide directions clear enough to relocate the site on a topographic map by recording approx distance (miles) and compass direction from two or more geographical landmarks (such as the closest town/city, and a natural feature like a mountain, river, or lake). Roads/intersections may be included (e.g., "NW corner of A St and 1st Ave" or "5 mi W of I-5")
Others in Team	Name(s) of people who were with you
Vegetation	The dominant vegetation in the vicinity of the collected specimen. You may use general community or vegetation types (e.g., chaparral, coastal sage scrub etc.) and/or name specific plants if you like (from the tree, shrub, & herb layers).
Geology	Physical habitat/substrate (i.e., the abiotic or non-living factors, like "dry creek bed" or "granite outcrop" etc.). Record other details about the terrain in the vicinity, such as the slope and aspect ("steep S-facing slope") or shading. Record soil texture (e.g., "medium sandy loam") and parent material (e.g., "granitic", "alluvium", "gabbro" etc.) if you know it.
Elevation	Record elevation (from GPS and/or map contours).
Latitude Longitude	Record the coordinates of each collecting location (called the "collection event" on the online data entry form). Mark the spot on your map! Your lat/long can be in either of two formats: Degrees-Minutes-Seconds, for example 32° 46' 25" or Decimal Degrees, for example 32.77361° *Note: if using a GPS*, routinely verify the readings on a map or by using our Topozone tool on the website!
Specimen Number	The collecting number you have assigned to the specimen (e.g., if this is your specimen #352 then enter 352 in the box). NOTE: Remember to write the "**index number**" on the outside edge of the newspaper in which you are pressing the specimen (e.g., if your login initials are "jg1" the index number is jg1#352). We will not accept specimens that don't have the index numbers clearly written down on the newspaper along with each specimen that comes in!
Plant Family, Genus, Species	If you know the family, scientific, and/or common name of the plant, record it now. You may correct it or look it up later.
Description of Plant	Describe the plant (esp. info that may be lost when it is mounted, such as height, width, branching pattern etc.) Was it erect, climbing, or prostrate (sprawling)? Was it parasitic (if so, on what host)? Does it have flowers and/or fruits? Include colors of flowers or fruit that often fade when dried. Record the habit (herb, shrub, tree, bulb, vine, etc.) and whether it was an annual, perennial, or biennial. (Max. 255 characters allowed).

Note: Use field time to record information that is only apparent to you while there. Other items like plant family, elevation, or lat/long can always be looked up and filled in later. For more detailed instructions about what field data to record, visit the FAQ section on our website.

Herbarium Voucher & Genetic Sampling Form 2018:
San Diego Zoo Native Plant Seed Bank

Soil Sampling (enter data on Locality form)			
Occurrence Accession Code:		**Date Collected:**	/ /
Notes			

Genetic Sampling (enter data on Locality form)			
Occurrence Accession Code:		**Date Collected:**	/ /
Number of Plants Sampled:		**Notes**	
Observers Present:	☐ Stacy Anderson ☐ Joe Davitt ☐ Tobin Weatherson ☐ _____, _____, _____, _____		

Herbarium Voucher Collection			
Occurrence Accession Code:		**Date Collected:**	/ /
Herbarium Collection Code:			
Observers Present:	☐ Stacy Anderson ☐ Joe Davitt ☐ Tobin Weatherson ☐ _____, _____, _____, _____		
Phenology	☐ Not fertile ☐ Flowering ☐ Fruiting ☐ Fruit & Flower	**Plant Form**	☐ Forb ☐ Shrub ☐ Tree ☐ Grass ☐ Aquatic
Plant Height		**Height units**	
Specimen Description (Condition, Color, etc.)			

Seed Collection			
Occurrence Accession Code:		**Date Collected:**	/ /
Seed Collection Code:		**Date of Second Collection:**	/ /
Observers Present:	☐ Stacy Anderson ☐ Joe Davitt ☐ Tobin Weatherson ☐ _____, _____, _____, _____		
Maternal Lines			
Collection Notes			

Appendix C

Seed Collection Forms

Rancho Santa Ana Botanic Garden

Santa Barbara Botanic Garden

San Diego Zoo Native Plant Seed Bank

Rancho Santa Ana Botanic Garden
California's Native Garden

Accession # _____

Collection Date:		**Received Date:**	
Species Name:			
Collector Name:		**Collection #:**	
Associate Collectors:			
Voucher? (Yes or No)**:**		**Voucher Location:**	
Country:		**State:**	**County:**
Jepson Geographic Subregion:		**Elevation:**	**Unit** (ft or m)**:**
Locality:			
CNDDB EO#:		**Land Owner:**	
Latitude: N		**Longitude: W**	**Datum**: NAD83 NAD27 WGS84
Map Quad:		**T** **R** **SEC** **¼ SEC**	

Sampled population size:	**Collection type** (circle/underline one)**:**
Number of individuals sampled:	Seed Division Other:
Local abundance (common, scattered, rare)**:**	Spore Cutting _____
	Plant Bulb/Corm
Associated species:	

Floristic Province (circle/underline one)**:**	**Horticultural Source** (if cultivated)**:**	**Collection Method** (circle/underline one)**:**
Californian Sonoran		Collected Along Maternal Lines
Mojave		Bulk Collected

Habitat (circle/underline one)**:**	**Slope:**	**Exposure:**	**Moisture:**
Alpine Chaparral	Flat	Full sun	Dry
Sub Alpine Scrub	Gentle	Semi shade	Moist
Forest Riparian	Steep	Shade	Wet
Woodland Type:	Cliff		Seasonally Moist
Grassland	Aspect:		

Geology (circle/underline one)**:**			**Soil:**		
Gabbro	Shale	Other:	Sand	Clay	Other:
Granite	Volcanic	_____	Gravel	Humus	_____
Limestone	Serpentine		Rock	Alluvium	
Sandstone			Loam		

Collector notes and observations:	

Santa Barbara Botanic the GARDEN

Survey & Seed Collection Form

FOR OFFICE USE ONLY: **Accession # _____ USDA Plants Code: _____**

Date of survey (mm/dd/yyyy): _____ **Date of seed collection:** _____

Field collector name (s): _____

Scientific name: _____

Rare? Y/N Status if rare _____ **Voucher collected? Y / N #**_____

Location information

Site name: _____ **Population (circle):** wild restored planted

County: _____ **Land Owner:** _____

Elevation (m):_____ **Aspect:** _____ **Slope (circle):** flat gentle moderate steep cliff

GPS Coordinates

Datum: _____ **Latitude:** _____ **Longitude:**_____

Habitat description

Plant community & habitat notes:_____

_____ **Soil:** _____

Associated species: _____

_____

Pollinators obs.? (if Y, describe): _____ **#of individuals/stems** _____

Percent cover of target species: <5% 5-10% 10-25% 25-50% 50-75% 75-100%

Phenology: % vegetative _____% flowering _____ % fruiting _____ % other (list)_____

Site information

Overall site viability/quality (site + population): Excellent Good Fair Poor

Immediate and surrounding land use: _____

Visible disturbances: _____

Threats to population: _____

Other notes & location information:

Seed information

Seeds collected? Y / N **Collection timing:** early perfect late

Type of collection (circle): Individual maternal lines Bulk (seeds from multiple individuals together)

 # plants sampled: _____ **Evidence of seed predation?** _____

Tissue collected? Y / N Notes: _____

FOR LAB USE ONLY

Cut test results: _____ out of 5 filled

Total seed count:

Average individual seed weight (g):

Notes/observations during cleaning:

Fumigation date:

Accession data entry date:

Frozen date:

the Santa Barbara Botanic GARDEN

Survey & Seed Collection Form

FOR OFFICE USE ONLY: **Accession # ___________________________ USDA Plants Code: _____________**

See back of page for maternal line information

Maternal line #	# of seeds	Maternal line #	# of seeds	Maternal line #	# of seeds

Total number of seeds: **Use extra pages, if needed*

Herbarium Voucher & Genetic Sampling Form 2018:
San Diego Zoo Native Plant Seed Bank

Soil Sampling (enter data on Locality form)			
Occurrence Accession Code:		**Date Collected:**	/ /
Notes			

Genetic Sampling (enter data on Locality form)			
Occurrence Accession Code:		**Date Collected:**	/ /
Number of Plants Sampled:		**Notes**	
Observers Present:	☐ Stacy Anderson ☐ Joe Davitt ☐ Tobin Weatherson ☐ _____, _____, _____, _____		

Herbarium Voucher Collection			
Occurrence Accession Code:		**Date Collected:**	/ /
Herbarium Collection Code:			
Observers Present:	☐ Stacy Anderson ☐ Joe Davitt ☐ Tobin Weatherson ☐ _____, _____, _____, _____		
Phenology	☐ Not fertile ☐ Flowering ☐ Fruiting ☐ Fruit & Flower	**Plant Form**	☐ Forb ☐ Shrub ☐ Tree ☐ Grass ☐ Aquatic
Plant Height		**Height units**	
Specimen Description (Condition, Color, etc.)			

Seed Collection			
Occurrence Accession Code:		**Date Collected:**	/ /
Seed Collection Code:		**Date of Second Collection:**	/ /
Observers Present:	☐ Stacy Anderson ☐ Joe Davitt ☐ Tobin Weatherson ☐ _____, _____, _____, _____		
Maternal Lines			
Collection Notes			

Occurrence Establishment Form 2018: San Diego Native Plant Seed Bank

Locality Information			
Occurrence Accession Code		**Date Established:**	/ /
Focal Species:		**Locality Short Name:**	
Locality Description:			
Landowner/Manager		**County**	
Observers Present:	☐ Stacy Anderson ☐ Joe Davitt ☐ Tobin Weatherson ☐ _____, _____, _____, _____		

Associated Occurrence Codes			
MOM Code(s)		**CNDDB EO(s)**	

Site Characteristics	
Immediate Land use	☐ Nature Preserve ☐ Farmland/Ranchland ☐ Urban Development
Surrounding Land use	☐ Nature Preserve ☐ Farmland/Ranchland ☐ Urban Development
Land use Notes	
Ecological Community:	Southern Foredunes, / Riparian Woodland / Meadow and Seep / Desert Scrub Coastal Sage Scrub / Riparian Forest / Marsh / Desert Chaparral Chaparral / Pinyon Juniper Woodlands / Coniferous Forest / Dry Wash Woodland Grassland / Other Woodlands / Desert Dunes / Water Riparian Scrub / Oak Forest / Playas/Badlands/ Mudhill Forbs / Urban, Disturbed Habitat _____
Slope	☐ Flat ☐ Gentle Slope ☐ Medium Slope ☐ Steep Slope ☐ Cliff Face
Aspect	☐ Flat ☐ North ☐ South ☐ East ☐ West
Sun Exposure	☐ Full Sun ☐ Semi Shade ☐ Shade — **Elevation (m)**

Geographic Coordinates (Revise at time of Voucher Collection)

1. Use Geode to take point 2. Longitude should be negative 3. Record in Decimal Degrees (-33.222241)

Latitude		**Longitude**	
Horizontal Accuracy (m)		**GPS Type**	☐ Geode ☐ Phone ☐ _____

Data Collection Checklist

Seed Accession:	☐ yes ☐ no	Genetic Sample	☐ yes ☐ no	Population Polygon:	☐ yes ☐ no
Herbarium Voucher:	☐ yes ☐ no	Soil Voucher:	☐ yes ☐ no		