The SDZWA Native Plant Gene Bank:

How Ex-situ Collections Support In-situ Conservation

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San Diego Zoo Wildlife Alliance

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Conservation Science (8 Teams)

Plant Conservation Team

Native Plant Gene Bank

Center for Plant Conservation (CPC)

Headquartered at the San Diego Zoo Safari Park

Network of plant conservationists that strive to save native imperiled plants of the US and Canada

www.saveplants.org

Plant Conservation Team

Solutions to Plant Endangerment

- Ex-situ Conservation
  - Native Plant Gene Bank
  - Living Gene Banks
  - Exceptional Species
- Research
  - Experimental Reintroduction - TPSNR
  - Plant Genetics
  - Germination and Propagation Studies
- Restoration
  - Seed Bulking
  - Reintroductions


A HOUSE OF STRAW

Native Plant Gene Bank

What we do:

- Long-term conservation banking
  - Rare plant focus
- Germination testing
- Collections for plant material development
  - Propagation research
  - seed bulking/plant production
- Propagation and common garden experiments

Lupinus hirsutissimus


Hesperocyparis forbesii


Myosurus minmus ssp apus

Targeting

Overarching goals:

- All SD County Rare Plants
  - Species ranked 1B-4 by CNPS/State
  - California Plant Rescue (CaPR)
- Funding influences prioritization
- Access/permits
- Phenology/production
- Collaborator needs/projects

Ex-situ Conservation

Monardella viminea
Willowy monardella

	Unique Taxa Collected by NPGB	% of Total Species in SD County
CNPS Ranks		
1B	85	62%
2B	19	28%
3	2	20%
4	28	38%
Legal Status		
California Listed	23	74%
Federally Listed	15	75%
Endemism		
Strict	17	68%
Near	20	87%

Collecting Strategies

Follow Center for Plant Conservation Guidelines to maximize genetic diversity

- 50 individuals per population
- Collect randomly throughout the extent of a population along maternal lines
- Target quantity is 3000 seeds per collection
- No more than 5-10% of the total seed output is collected
- May collect over multiple years to capture diversity across space, time, appearance & ecology

Maternal Lines

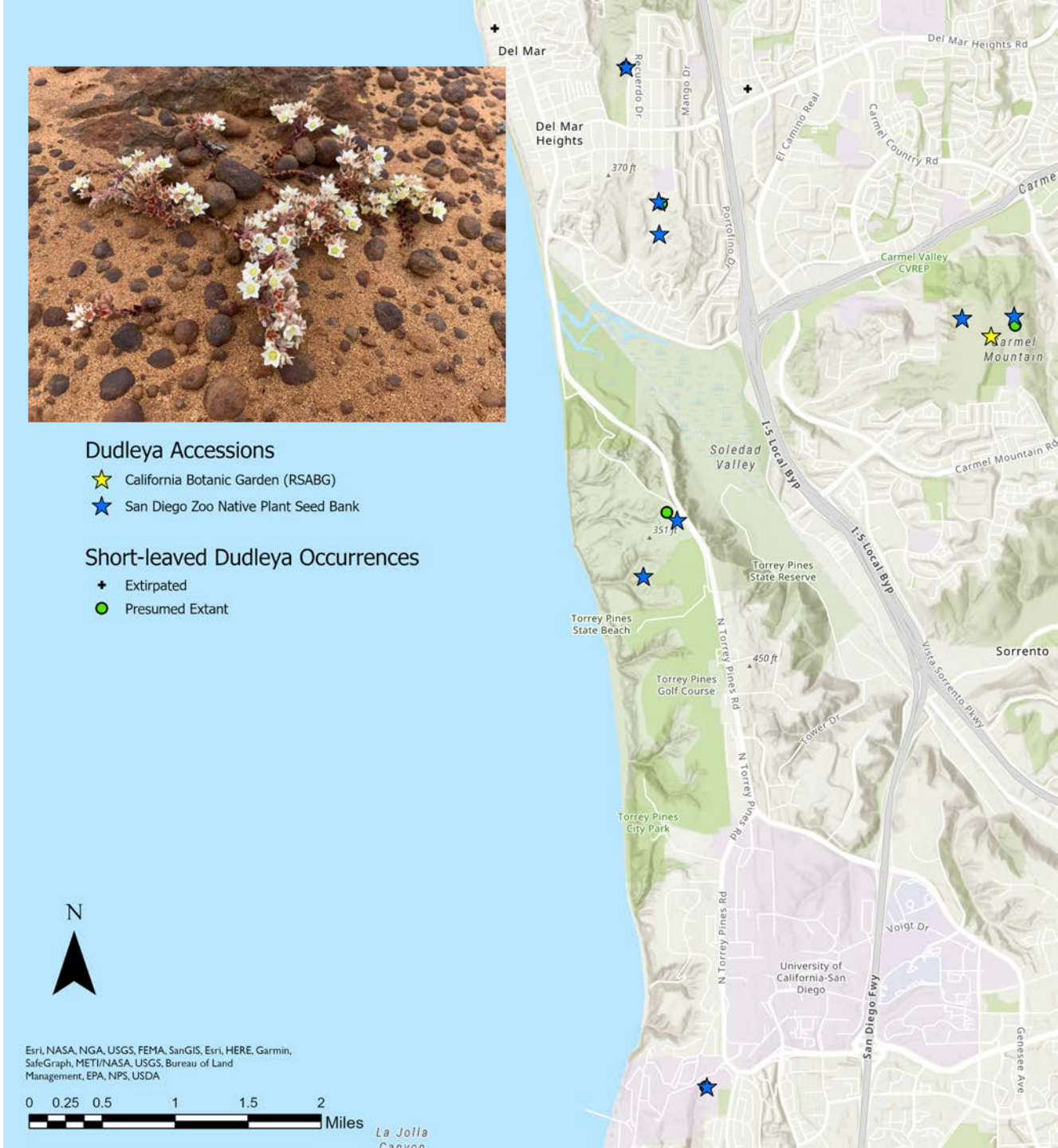
- Individuals kept separate
  - Cleaning
  - Counting
  - Storing
- Backup Lot
  - Duplication to federal facility : National Laboratory for Genetic Resource Preservation (NLGRP)
- Active Lot
  - Germination trials
  - Bulking
  - Restoration

Maternal Line	Acanthomintha ilicifolia		SDZG Acc# 0716		Dissection Test Results: 5 of 5 seeds filled with normal, viable emrbryos			
	Total		Active 30		Base 40		Backup 30	
Number	Quantity	Weight (g)	Quantity	Weight (g)	Quantity	Weight (g)	Quantity	Weight (g)
1	546	0.157	164		219		163	
2	535	0.164	161		214		160	
3	210	0.052	63		84		63	
4	252	0.062	76		100		75	
5	826	0.233	248		331		247	
6	277	0.072	84		110		83	
7	815	0.218	245		326		244	
8	349	0.099	105		140		104	
9	657	0.203	198		262		197	
10	109	0.023	33		44		32	
11	147	0.036	45		58		44	
12	292	0.076	88		117		87	
13	447	0.122	135		178		134	
14	431	0.117	130		172		129	
15	455	0.117	134		188		133	
16	209	0.054	63		84		62	
17	356	0.090	107		143		106	
18	444	0.121	134		177		133	
19	541	0.158	163		216		162	
20	192	0.057	58		77		57	
21	139	0.037	42		56		41	
22	284	0.064	86		113		85	
23	390	0.119	117		156		117	
24	654	0.175	197		261		196	
25	331	0.069	100		132		99	
26	197	0.069	60		78		59	
27	165	0.052	50		66		49	
28	259	0.067	78		104		77	
29	175	0.053	53		70		52	
30	314	0.088	95		125		94	
31	32	0.008						
32	~261	0.070						
33	~363	0.100						
34	~760	0.216						
35	89	0.021						
36	~466	0.130						
37	~305	0.083						
38	156	0.038						
39	63	0.016						
40	~575	0.162						
41	~199	0.052						
42	~145	0.036						

Ex-situ Conservation

Dudleya brevifolia
Short-leaved Dudleya

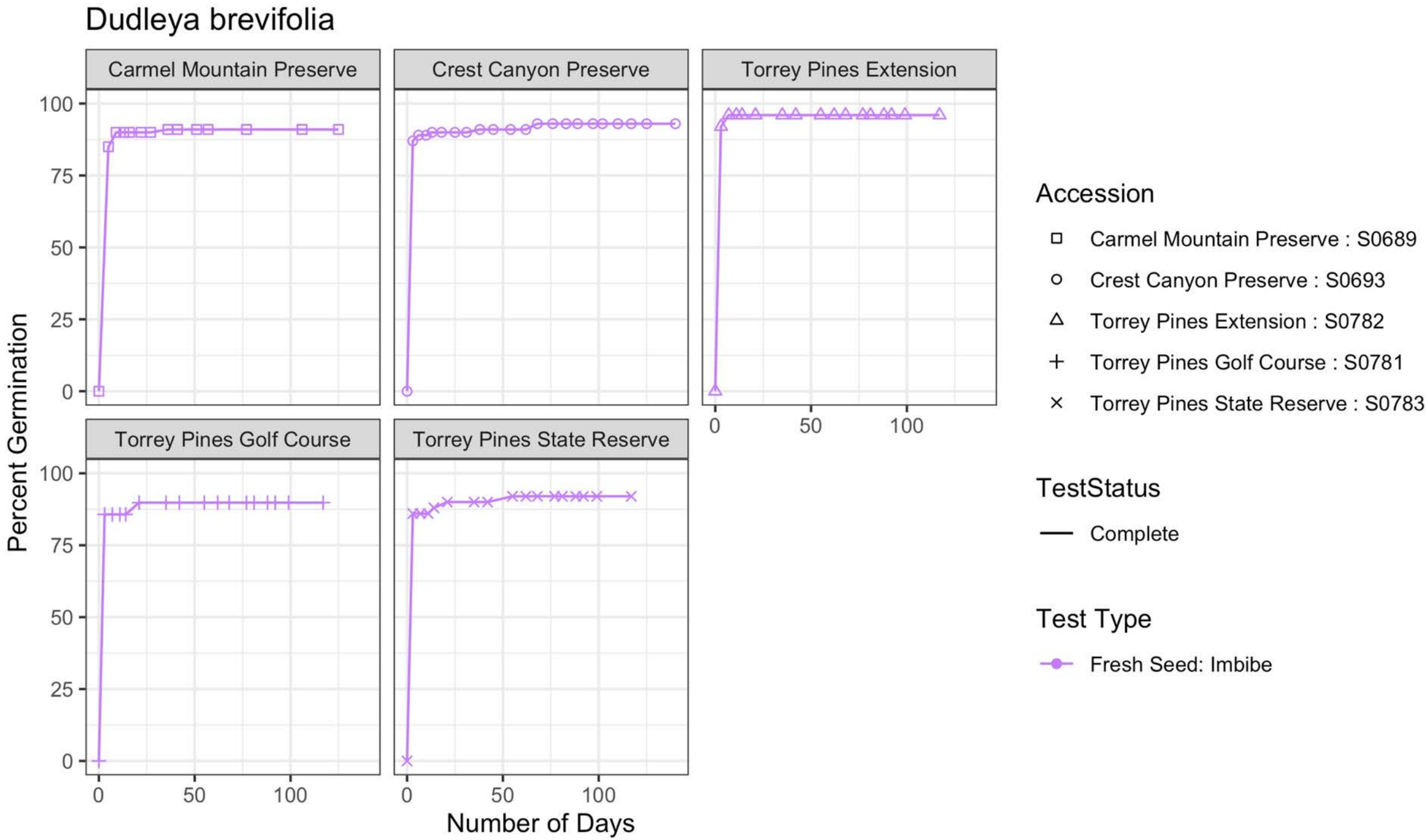
- Native Plant Gene Bank has 8 collections from 2016-2019
- California Botanic Garden has 1 collection from 1986



Dudleya brevifolia Collections Locality Name	Date Collected	Maternal Lines Collected	Total Seed Count
Carmel Mountain Preserve	7/01/16	158	7305
Crest Canyon Preserve	7/12/16	99	5184
Skeleton Canyon	7/21/17	11	94
Torrey Pines Extension	7/23/19	56	3981
Torrey Pines Golf Course	7/23/19	54	2838
Torrey Pines State Reserve	7/23/19	221	20100
Crest Canyon Preserve Sub	7/25/19	10	165

Germination Testing

- Germination percentage from imbibed seed on agar
- Seeds may not germinate due to dormancy
- Pre-treatments may be necessary
  - Sanding, nicking seeds (mechanical)
  - Hot water treatment (heat)
  - Smoke water treatment (chemical)
  - Cold stratification

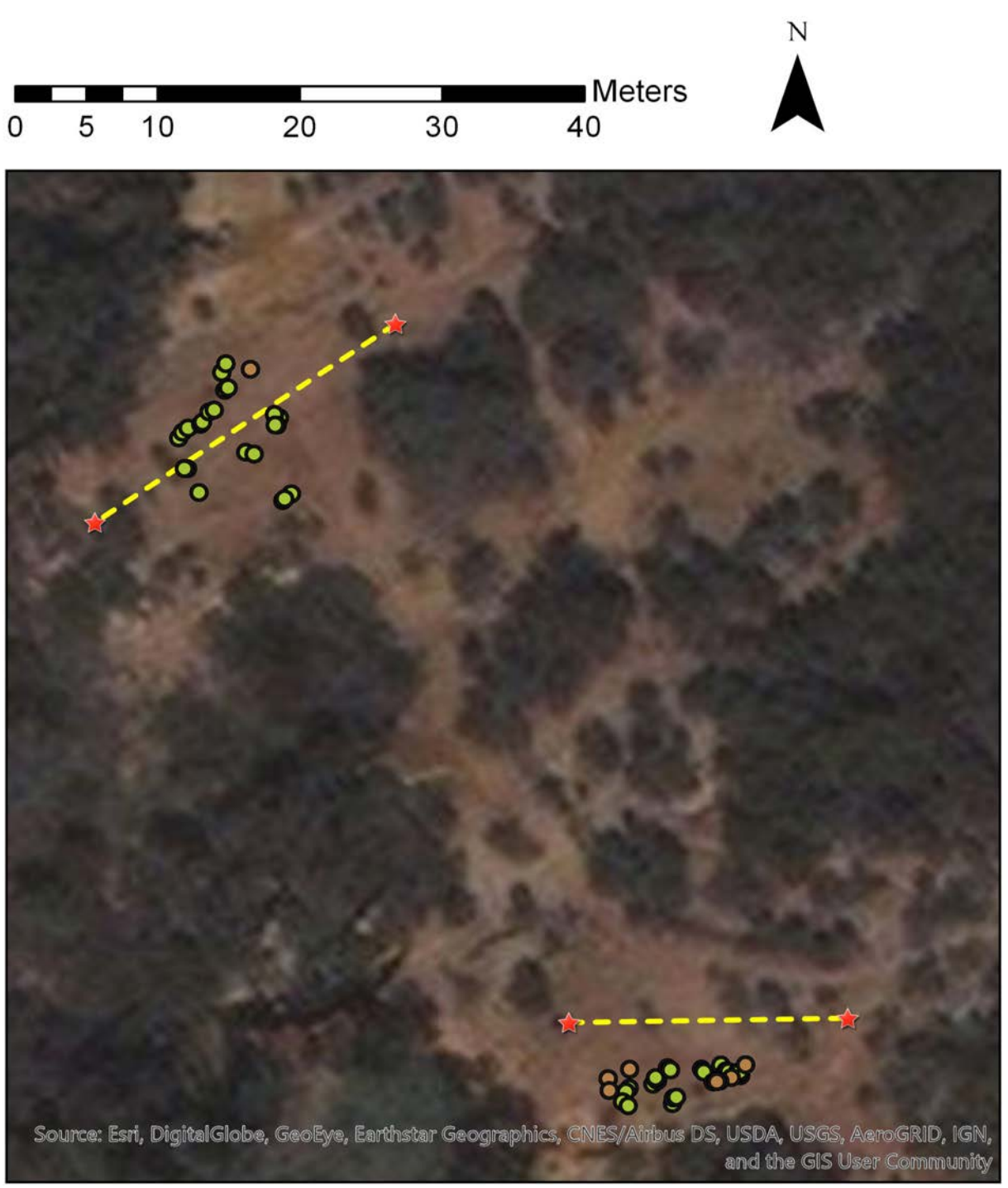

Dudleya brevifolia
Carmel Mountain Preserve
Crest Canyon Preserve
Torrey Pines Extension
Torrey Pines Golf Course
Torrey Pines State Reserve
Percent Germination
Number of Days
0
25
50
75
100
Accession
Carmel Mountain Preserve : S0689
Crest Canyon Preserve : S0693
Torrey Pines Extension : S0782
Torrey Pines Golf Course : S0781
Torrey Pines State Reserve : S0783
TestStatus
Complete
Test Type
Fresh Seed: Imbibe

Seed Bulking

Bulking short-leaved dudleya

- Excellent germination from trials and successfully transferred from agar to soil
- Ongoing collection of seed from mature plants growing ex-situ
- 2017 approached by David Hogan to bulk seed from the Torrey Pines Extension population

In-situ Out-planting



Monitoring

- Marked each with a nail and 23mm HDX pit tag
- Measured every two weeks during the growing season
- Long-term project
- Future plans to include seed experiments


Genetic Sampling

Dudleya brevifolia genetic analysis

Sample Collection

- 111 samples were collected from 5 *Dudleya brevifolia* populations.
- Unopened bud tissue was found to be optimal for extraction

DNA Extraction

Tissues were flash frozen in liquid nitrogen and ground to a fine powder

DNA extractions were performed using a modified CTAB protocol

Sequencing

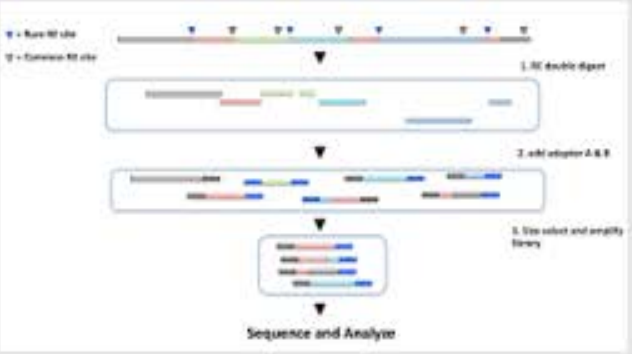


Image courtesy of: http://www.ucgenomi.com/whitepapers/radSequencing.pdf

Samples were sequenced at Texas A&M using double digest restriction-site associated DNA sequencing (ddRADseq).

Analysis

Currently working on genetic analysis of the data using the STACKS pipeline and R package 'adegenet'

Target list 2022-24 :

Acmispon prostratus	Nuttall's acmispon
Ambrosia pumila	San Diego ambrosia
Aphanisma blitoides	Aphanisma
Astragalus deanei	Deane's milkvetch
Atriplex coulteri	Coulter's saltbush
Atriplex parishii	Parish brittlescale
Baccharis vanessae	Encinitas baccharis
Brodiaea filifolia	Thread-leaved brodieae
Brodiaea santarosae	Santa Rosa brodiaea
Ceanothus foliosus	Viejas lilac
Chloropyron maritimum ssp maritimum	Salt marsh bird's beak
Clinopodium chandleri	San Miguel savory
Cryptantha wigginsii	Wiggin's cryptantha
Deinandra conjugens	Otay tarplant
Dudleya blochmaniae	Blochman's dudleya
Dudleya variegata	Variegated dudleya
Dudleya viscida	Sticky dudleya
Hazardia orcuttii	Orcutt's hazardia
Monardella viminea	Willowy monardella
Monardella stoneana	Jennifer's monardella
Nolina interrata	Dehesa nolina
Pogogyne nudiuscula	Otay mesa mint
Sphenopholis interrupta ssp californica	Prairie false oatgrass
Tetracoccus dioicus	Parry's tetracoccus

Sphenopholis interrupta ssp californica

The Chaparral Lands Conservancy

CALIFORNIA PLANT RESCUE

SANDAG

CENTER FOR PLANT CONSERVATION at SAN DIEGO ZOO GLOBAL