Science for a changing world

Rare Plant Propagation Research, Phase II (2021-USGS-2075A)

Seed Ecology of Three-Corner Milkvetch (2023-USGS-2385A)









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Photo: Todd Esque, USGS

Las Vegas Bearpoppy

Background

- Dormancy-breaking treatments have yielded low germination success (Meikle et al. 2006, Pereira et al. 2021, de Queiroz & Meyer 2023)
- Seeds have morphophysiological dormancy



Photo: Mikaela Gaskill & Bryce Usiak, USGS

<u>Approach</u>

- Identify treatments that influence embryo growth and germination in laboratory trials to inform propagation
 - Dry after-ripening, moist incubation, and gibberellic acid
 - Radicle and cotyledon emergence
- Adapt greenhouse emergence method to promote seedlings from soil seed bank (*start Fall 2024*)



Las Vegas Bearpoppy

Laboratory Trials



USGS

Las Vegas Bearpoppy Embryo Growth



Photos: Mikaela Gaskill & Bryce Usiak, USGS



Las Vegas Bearpoppy

Embryo Growth, Seed Viability, and Germination



- Embryos develop under prolonged exposure to dry, hot after-ripening
- · Seed viability steadily declines under warm, moist incubations
- Cotyledons emerge 55.2 ± 2.8 days after radicles emerge for 2 °C and 5/15 °C incubations (no cotyledons in warm, moist incubations)
- Continue to monitor radicle and cotyledon emergence and transfer into soil mixes









Photos: Sara Scoles-Sciulla & Lesley DeFalco, USGS

Blue Diamond Cholla

Background

- Seed production is variable, but plants can be propagated from joints (Scoles-Sciulla et al. 2023)
- Propagation from joint cuttings may preserve genetic diversity when seed collections are impractical

Approach

- Identify best practices for reintroducing joint-propagated plants into habitat
 - Season of planting (Spring vs. Fall)
 - Nurse plant association (Canopy cover vs. Open)
 - Supplemental watering (Frequent vs. Infrequent)
 - Herbivore protection (Cage vs Uncaged)



Blue Diamond Cholla Re-introduction into Habitat

- Season of collection/planting): Joint cuttings from Gold Butte population (Spring 2023/2024; Fall 2023)
- Supplemental watering: 3.8 L @ 2, 4, 6 months (Freq) and @ 4 mo only (Infreq)
- Nurse plant: Outplant 1-year old plants in spring (all caged)





Photos: Sara Scoles-Sciulla, USGS



Blue Diamond Cholla Re-introduction into Habitat

- Assessment of Spring outplants at 2 mo revealed 15% loss and 23% severed roots despite caging: replanted, and all cages intact
- Fall 2023 collection (growing in shadehouse for Fall outplanting)





Photos: Sara Scoles-Sciulla, USGS Rare Plant Propagation Research, Phase II



White-margined Penstemon



Background

- Clark County subpopulations are threatened by development and climate change (Miller 2021)
- Propagation from cuttings may preserve genetic diversity when seed collections are impractical
- Reintroduction may be necessary for conservation areas

Approach

- Identify best practices for producing robust stock from stem cuttings
- Identify best practices for reintroducing cuttingpropagated plants into habitat to maximize survival



Photos: Alex Stosich. USU

White-margined Penstemon Production of Robust Nursery Stock

- Pre-reproductive cuttings from Ivanpah, Jean Lake, and Hidden Valley
- Full stem cutting (versus basal or terminal cuttings from 2023)
- 18% rooting: balance between cutting size and rooting success
 - \circ $\,$ Hidden Valley cuttings and longer cuttings have greater rooting





White-margined Penstemon Re-introduction into Habitat

- Outplant in Feb 2024 using 2023 plant propagation
- Compare nursery treatments: lean vs. organic soil mix and shallow vs. deep pot
- 30 active / 86 dormant assigned to watering treatments "Low" = 1.9 L biweekly; "High" = 7.6 L biweekly (8 weeks total)



Photo: Lesley DeFalco, USGS



Photo: Alex Stosich, USU



White-margined Penstemon Re-introduction into Habitat

- 77% of active plants survived to May 2024 (23 of 30 plants)
- 26% of active plants flowered (6 of 23 plants)
- No watering treatment effect (rainfall: 31 mm in Feb 33 mm in Mar)
- No obvious nursery soil mix or pot dimension effects



Photo: Alex Stosich, USU







Photos: Lesley DeFalco, USGS, Alex Stosich, USU

Sticky Buckwheat

Background

- Plant occurrence and abundance are variable between populations and across years
- Seed bank study demonstrates reproductive plants can be raised from soil samples, even those from historic populations (2019-USGS-1990A)

<u>Approach</u>

- Create seed collections from minimum of 4 populations for conservation and research by:
 - o Collecting from seed-bearing plants in habitat,
 - Collecting seed bank in suitable microsites, and growing seed-bearing plants from soil in greenhouse



Sticky Buckwheat Seed Collection from Habitat

- Collect seed from adult plants (matrilines) from habitat during 2023 2026 (Center for Plant Conservation guidelines)
- Germination and viability testing on collections (AOSA 2010)

Year	Population	# Matrilines	Total # Seeds	% Viability	Est # Viable Seeds
2024	Middle Muddy River	45	10,964	93.3	10,229
2023	Upper Virgin Valley	42	3,204	98.7	3,162
2023	Toquop Wash	18	8,550	98.7	8,439
2023	Upper Muddy River	33	1,385	100.0	1,385



Positive tetrazolium test

Photos: Sara Scoles-Sciulla, USGS, Alex Stosich, USU

Sticky Buckwheat Seed Collection from Habitat

 Meeting seed collection targets can be difficult to estimate because of large variability in individual plant seed production



CDC guidelines: "3,000 seeds/population from 50 matrilines collected in multiple years"



Rare Plant Propagation Research, Phase II

Sticky Buckwheat Seed Collection from Seed Bank Grow-out

- Only 3 sticky buckwheat seedlings emerged from seed bank and failed to thrive and produce seed-bearing plants
- 7 empty seed coats recovered from soil samples
- Similarly low seed bank as Black Mountain and Upper Muddy River (2019-USGS-1990A)
- Attempt seed bank in Fall 2024 to diversify seed collection





Photos: Lesley DeFalco, USGS





Photos: Lesley DeFalco, USGS

Three-corner Milkvetch

Background

- Plant occurrence and abundance low and variable during 2020 – 2023 (2017-IRONWOOD-1755A)
- No seed bank detected at four populations examined in 2020 – 2021 (2019-USGS-1990A)

Approach

- Create seed collections from minimum of 4 populations for conservation and research by:
 - Irrigating habitat patches to promote seed-bearing plants
 - Collecting soil from suitable microsites, and growing plants from seed bank in greenhouse (*start Fall 2024*)



Three-corner Milkvetch Seed Collection from Habitat

- Establish supplemental watering plots during Fall/Winter 2023/24
- Monitor and remove Sahara mustard (BLM request)
- Select milkvetch plants and collect seeds in Spring 2024/2025 (n = 50)



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Three-corner Milkvetch Seed Collection from Habitat

- Collect seed from adult plants (= matrilines) from habitat during 2024 and 2025 (Center for Plant Conservation guidelines)
- Irrigated plots; amended permits to include good rainfall year ٠
- Germination and viability testing on collections (AOSA 2010) •
- Use seed for continued research (2023-USGS-2385A)

Population	# Matrilines	Total # Seeds	Viability	Est # Viable Seeds	Proportion Dark			
Mud Lake	34	745	97%	723	0.12			
Mormon Mesa	46	9,542	78%	7,443	0.41			
*Sandy Cove	50	1,357	96%	1,303	0.66			
* No irrigation plots								



Photos: Lesley DeFalco, USGS





Three-corner Milkvetch Seed Collection from Seed Bank Grow-out

- Collect soils from Mud Lake and Mormon Mesa (start Fall 2024)
- Seedling grow-out using emergence method



Photos: Lesley DeFalco, USGS



Rare Plant Propagation Research, Phase II

Rare Plant Propagation Research, Phase II 2024 Progress

Las Vegas bearpoppy: Nursery propagation from seed

• Embryo growth and germination completed, seedling transfer to soils on-going

Blue Diamond cholla: Joint propagation and reintroduction into habitat

 Collections of joints during spring and fall completed; spring outplanting completed and watering/monitoring on-going; fall joint collection growing in shadehouse

White-margined penstemon: Cutting propagation and reintroduction into habitat

- Second cohort of nursery growth in progress in greenhouse
- First cohort outplanted; second cohort planned for Spring 2025

Sticky buckwheat: Seed collections for conservation and research

- Seed collections from plants in habitat (2 populations completed and 2 partial populations; viability testing completed; further collections in 2025/2026)
- Propagate seed-bearing plants from soil seed bank (continue in 2024 2026)

Three-cornered milkvetch: Seed collections for conservation and research

 Habitat watering at 2 populations and seed collections from 3 populations (complete in Fall 2024/Spring 2025)



Propagate seed-bearing plants from soil seed bank, irrigated plots (start Fall 2024)

Seed Ecology of Three-corner Milkvetch 2023-USGS-2385A





Photos: Lesley DeFalco, USGS

Background

- Multiple factors may influence milkvetch but are not wellunderstood
- Seeds have physical dormancy (seed coat impermeable to water)

Approach

- Integrate with on-going research (2021-USGS-2075A) at Mud Lake and Mormon Mesa populations
- Explore roles of pollen limitation, herbivore pressure, competition with invasive species, and seed longevity in milkvetch persistence
- Identify how seed coat permeability changes with treatments to influence germination and viability



Pollen Limitation

- Pollen transfer treatments before flowering (n = 10 plants)
- Determine ability for milkvetch to self-pollinate and seed set from different pollen donor flowers



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Photos: Lesley DeFalco, USGS



Herbivory and Competition

- Herbivory (caged vs uncaged) and competition treatments (neighbors removed vs intact) (n = 40 young plants)
- Measure growth and flowering through spring and fruit/seed production





Photos: Lesley DeFalco, USGS



Confirmed and Potential Herbivores





Photos: Lesley DeFalco, USGS



Seed Longevity

- Nylon bags containing seeds for burial at Mud Lake, Mormon Mesa, and Sandy Cove
- Exhume bags at 3, 6, 9, 12, 15, 24, 36 and 48 months and test for permeability, germination, and viability







Photos: Lesley DeFalco, USGS

- Seed permeability influenced by maternal environment and therefore vulnerable to environmental stressors
- Potential for cycling between "sensitive" and "insensitive" dormancy states



Seed Ecology of Three-corner Milkvetch 2024 Progress

Pollen Limitation, Herbivory and Competition

 Pollen transfer, caging, and neighbor removal treatments deployed and resulting plant growth and fruit collection completed at Mud Lake and Mormon Mesa during spring

Seed Longevity

 Seed bags created for deployment at Mud Lake, Mormon Mesa, and Sandy Cove in summer



Photo: Lesley DeFalco, USGS

Seed Permeability, Germination, and Viability

• Laboratory trials for field treatments and buried bags are in progress



Questions?

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