Interpreting the Hawai‘i Seed Bank User’s Guide

The Hawai‘i Seed Bank Partnership (HSBP) created this User’s Guide for anyone interested to learn more about banking seeds of Hawai‘i’s native plants. However, we designed it with conservation professionals in mind, especially (1) people and organizations permitted to collect seeds in the field, who use the services of one of HSBP’s major seed banks, i.e. Lyon Arboretum, Army Natural Resources Program on O‘ahu, National Tropical Botanical Garden, Hawai‘i Island Seed Bank, Kaua‘i DOFAW Seed Bank, and Maui Nui Botanical Garden; and (2) people and organizations who store their own seeds at any other seed bank facility in Hawai‘i. If you fall into category 2 and are not already a HSBP member, we invite you to join us! Contact us to learn more.

Anyone interested in the 20+ years of collaborative scientific research behind the User’s Guide is encouraged to download (open access/free) the HSBP’s 2019 publication: *Seed freeze sensitivity and ex situ longevity of 295 species in the native Hawaiian flora*. (DOI:10.1002/ajb2.1351)

For seed bank users who are interested in the practical application of our research, instructions for the User’s Guide follow below. A few critical things to know about seed banking first:

Harrington’s Rule of Thumb: Longevity of seeds in storage approximately doubles for every 1% decrease in moisture content or every 6°C (10°F) decrease in temperature (to certain points). However,

- some seeds do not tolerate desiccation/drying,
- most seeds do not tolerate cold storage until **after** desiccation to appropriate levels,
- some seeds do not tolerate freezing temperatures, even with appropriate desiccation, and
- all seeds still age, even under optimal storage conditions, and will eventually lose viability.

Therefore, the most important aspects of the User’s Guide for conservation are the following, which are included in the simplified version of the User’s Guide:

- Recommended drying protocols (whether standard desiccation is tolerated or not)
- Recommended storage temperature (frozen at -18°C or refrigerated at 5°C)
- Re-Collection Intervals (RCI), or the period of time within which seeds are likely to drop below 70% of their highest recorded germination, when they should be withdrawn, used, and re-collected (or possibly regenerated through propagation) to replenish seed bank collections

For more information on recommended seed banking protocols that generally apply well to Hawai‘i, see the Center for Plant Conservation’s CPC Best Plant Conservation Practices to Support Species Survival in the Wild. The HSBP is also in the process of developing Hawai‘i-specific best practices and recommendations for partners.

Some important points and caveats are worth noting up front.

On seed storage behavior:

- Orthodox seeds should be frozen for long term storage (> 10 years), but they can also be desiccated and refrigerated, often for 10+ years, if short to medium term storage is your goal.
- Freeze sensitive seeds decline in frozen storage but can still be refrigerated, often for 10+ years.
- Research on seed freeze sensitivity is relatively new. Work is ongoing to better understand mechanisms and alternative storage methods. The National Laboratory for Genetic Resources Preservation reports that all tested Hawaiian species with desiccation tolerant (including orthodox or freeze sensitive) seeds survive cryopreservation in liquid nitrogen.
- We have included all Hawaiian species in this guide. For those that have not been tested, we either inferred storage behavior from congeneric/family behavior when reliable, or we stated that these categories are unknown.

On seed longevity in storage:

- Seed longevity can vary by species, population, individual plant, environmental conditions, seasonal conditions, harvest timing, and especially post-harvest handling of seeds. Thus, species with few tests may need further research. We also encourage users to follow the Hawai‘i Rare Plant Restoration Group’s Collecting & Handling Protocols to maximize collection quality.
- At some point within the RCI, there may be a steep/fast decline of seed viability in storage. If your seeds are especially valuable, we highly recommend at least withdrawing some seeds to test for viability early in the RCI period, if not before.
- **These RCI guidelines cannot replace direct testing of individual seed collections to monitor viability in storage over time.**

For further questions:

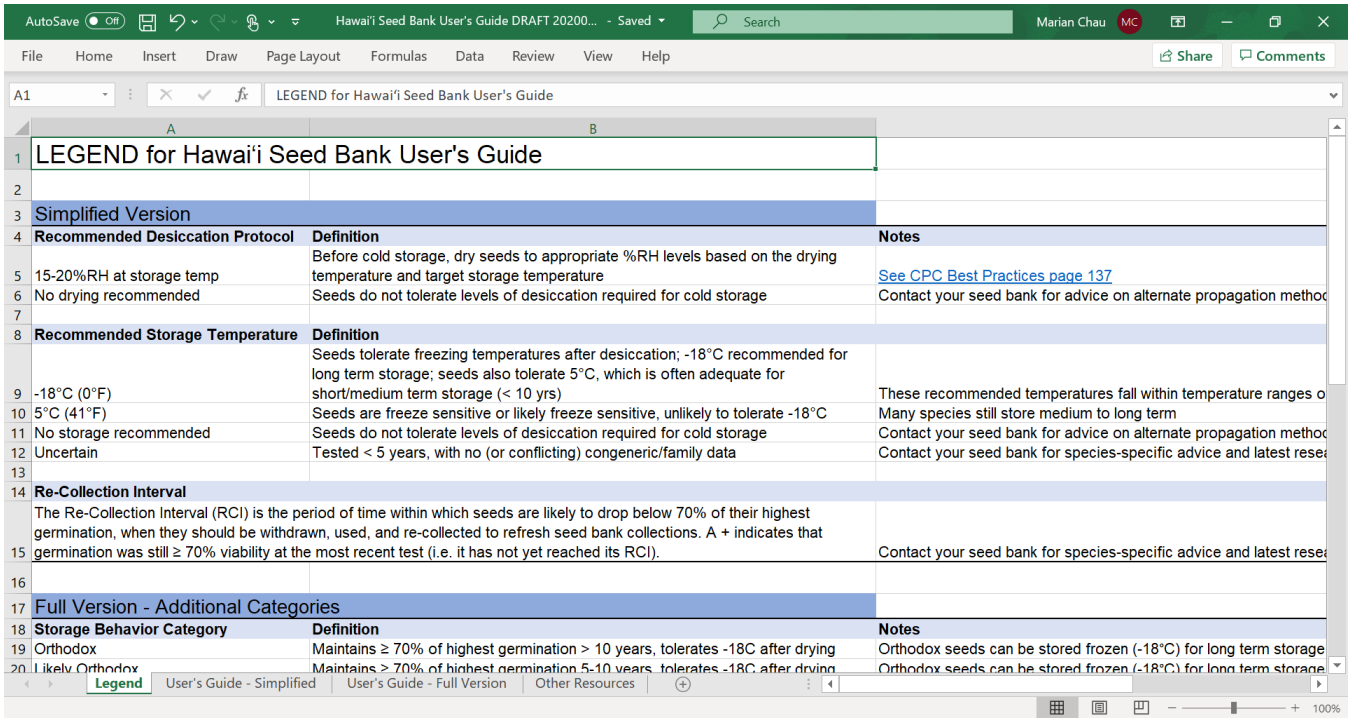
- Feel free to contact your local seed bank for collection-specific or species-specific advice and potential research updates.

Location	Seed Bank	Contact	Email
O‘ahu	Lyon Arboretum	Anna Sugiyama	asugi@hawaii.edu
O‘ahu	Army Natural Resources Program	Tim Chambers	tchambers.oanrp@gmail.com
Kaua‘i	National Tropical Botanical Garden	Dustin Wolkis	dwolkis@ntbg.org
Kaua‘i	Kaua‘i Division of Forestry & Wildlife	Denise Duenas	denise.h.duenas@hawaii.gov
Hawai‘i	Hawai‘i Island Seed Bank	Jill Wagner	jillwagner3@icloud.com
Maui	Maui Nui Botanical Garden	Tamara Sherrill	tamara@mnbg.org

Instructions for the HSBP User's Guide

Following is a summary of what is included in the HSBP User's Guide. In the future we plan to create a dynamic online database where users can search for seed storage information. In the meantime, the current User's Guide is an Excel spreadsheet that can be filtered, searched, and modified locally for your organization's own purposes.

The first sheet is the **Legend**, with detailed definitions of all headings and categories used in this guide:



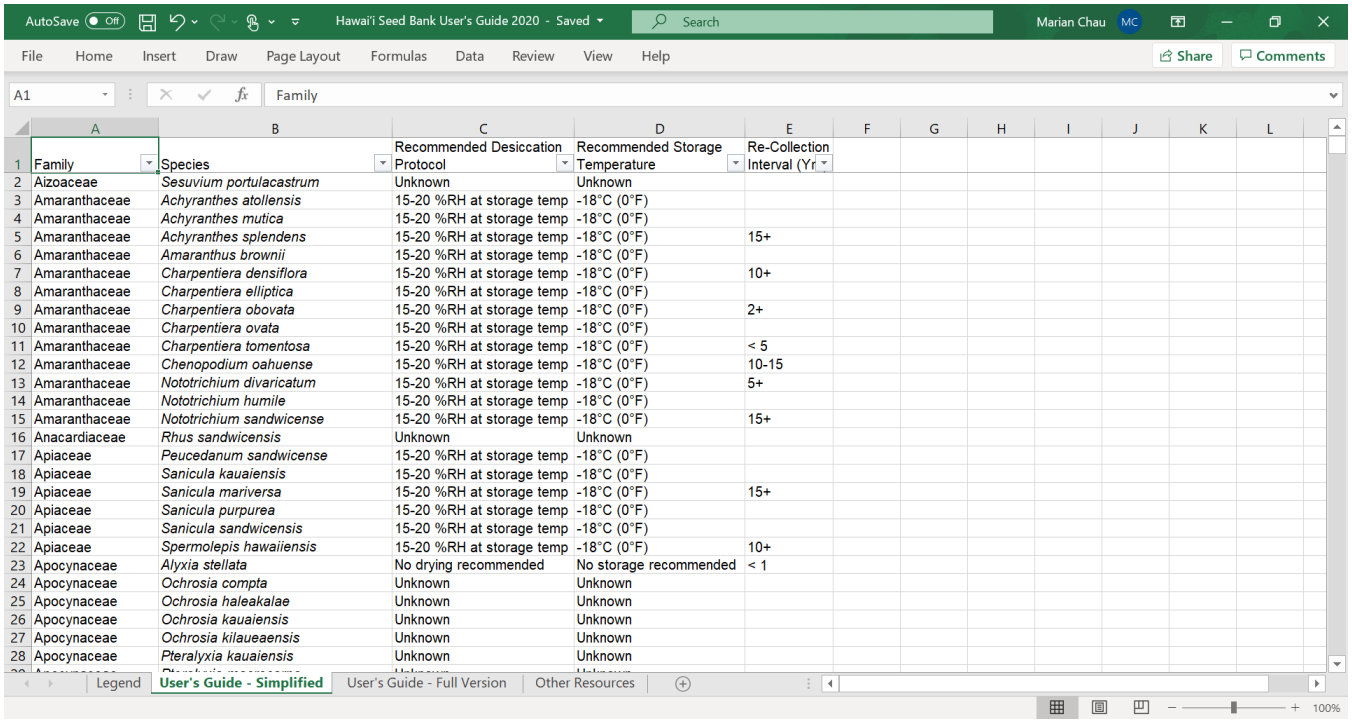
	A	B	C
1	LEGEND for Hawai'i Seed Bank User's Guide		
2			
3	Simplified Version		
4	**Recommended Desiccation Protocol**	**Definition**	**Notes**
5	15-20%RH at storage temp	Before cold storage, dry seeds to appropriate %RH levels based on the drying temperature and target storage temperature	See CPC Best Practices page 137
6	No drying recommended	Seeds do not tolerate levels of desiccation required for cold storage	Contact your seed bank for advice on alternate propagation metho
7			
8	**Recommended Storage Temperature**	**Definition**	
9	-18°C (0°F)	Seeds tolerate freezing temperatures after desiccation; -18°C recommended for long term storage; seeds also tolerate 5°C, which is often adequate for short/medium term storage (< 10 yrs)	These recommended temperatures fall within temperature ranges o
10	5°C (41°F)	Seeds are freeze sensitive or likely freeze sensitive, unlikely to tolerate -18°C	Many species still store medium to long term
11	No storage recommended	Seeds do not tolerate levels of desiccation required for cold storage	Contact your seed bank for advice on alternate propagation metho
12	Uncertain	Tested < 5 years, with no (or conflicting) congeneric/family data	Contact your seed bank for species-specific advice and latest resea
13			
14	**Re-Collection Interval**		
15	The Re-Collection Interval (RCI) is the period of time within which seeds are likely to drop below 70% of their highest germination, when they should be withdrawn, used, and re-collected to refresh seed bank collections. A + indicates that germination was still ≥ 70% viability at the most recent test (i.e. it has not yet reached its RCI).		Contact your seed bank for species-specific advice and latest resea
16			
17	Full Version - Additional Categories		
18	**Storage Behavior Category**	**Definition**	**Notes**
19	Orthodox	Maintains ≥ 70% of highest germination > 10 years, tolerates -18C after drying	Orthodox seeds can be stored frozen (-18°C) for long term storage
20	Likely Orthodox	Maintains ≥ 70% of highest germination 5-10 years, tolerates -18C after drying	Orthodox seeds can be stored frozen (-18°C) for long term storage

At the top are the 3 categories included in the **Simplified** version of the User's Guide. This version is for folks who just want to know what to do with their seeds, in a simple, clean format.

After that are several additional categories included in the **Full Version** of the User's Guide (which also includes the 3 categories from the Simplified version). Continue scrolling down in the **Legend** sheet for more definitions and notes for interpreting the User's Guide.

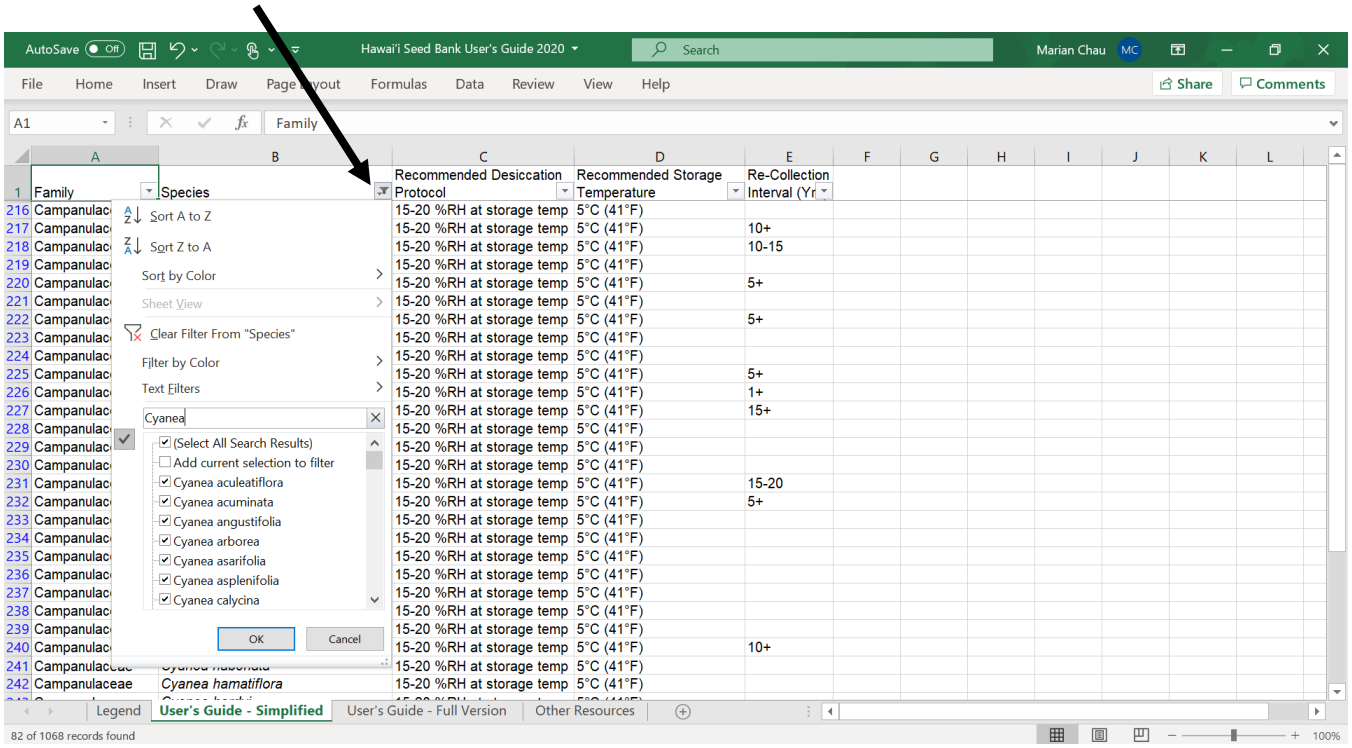
At the bottom of the screen, you can switch from the sheet titled **Legend** to the sheet titled **User's Guide – Simplified** to see a minimalist version of the guide.

The second sheet is the **User's Guide – Simplified** version:

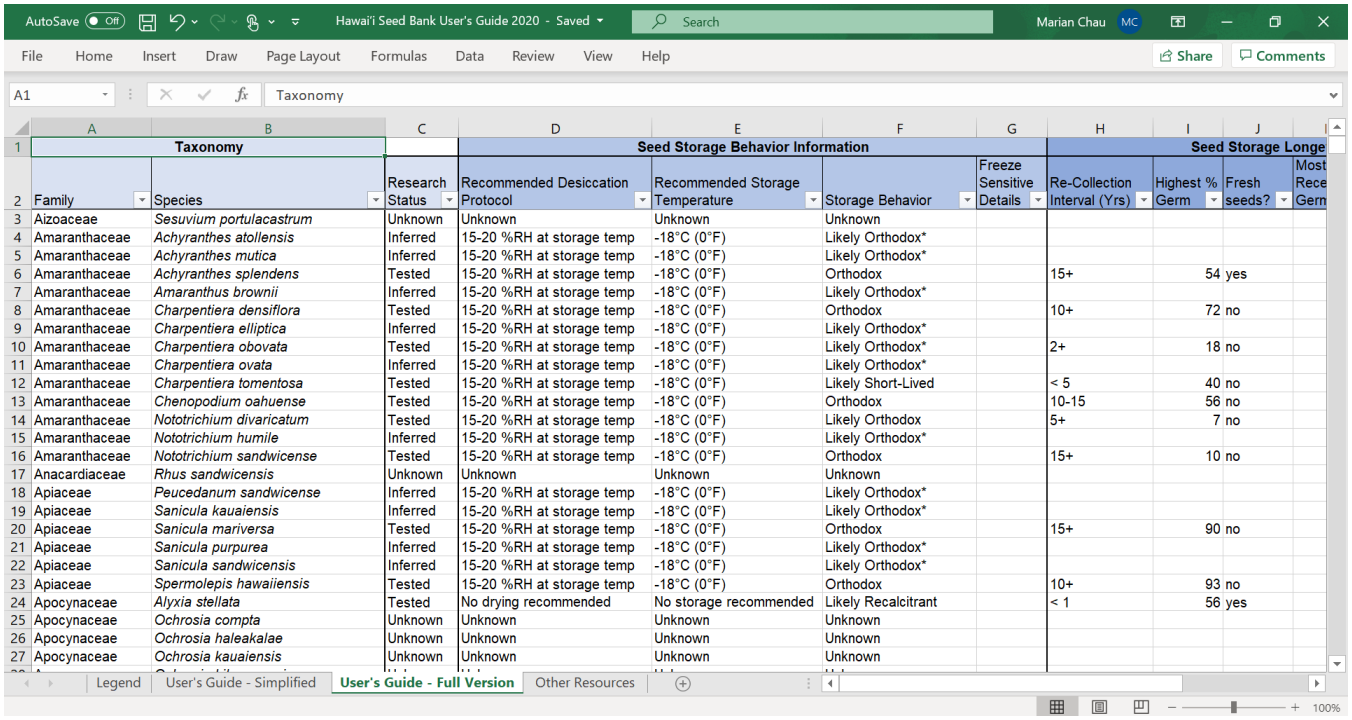


Family	Species	Recommended Desiccation Protocol	Recommended Storage Temperature	Re-Collection Interval (Yr
Aizoaceae	*Sesuvium portulacastrum*	Unknown	Unknown	
Amaranthaceae	*Achyranthes atollensis*	15-20 %RH at storage temp	-18°C (0°F)	
Amaranthaceae	*Achyranthes mutica*	15-20 %RH at storage temp	-18°C (0°F)	
Amaranthaceae	*Achyranthes splendens*	15-20 %RH at storage temp	-18°C (0°F)	15+
Amaranthaceae	*Amaranthus brownii*	15-20 %RH at storage temp	-18°C (0°F)	
Amaranthaceae	*Charpentiera densiflora*	15-20 %RH at storage temp	-18°C (0°F)	10+
Amaranthaceae	*Charpentiera elliptica*	15-20 %RH at storage temp	-18°C (0°F)	
Amaranthaceae	*Charpentiera obovata*	15-20 %RH at storage temp	-18°C (0°F)	2+
Amaranthaceae	*Charpentiera ovata*	15-20 %RH at storage temp	-18°C (0°F)	
Amaranthaceae	*Charpentiera tomentosa*	15-20 %RH at storage temp	-18°C (0°F)	< 5
Amaranthaceae	*Chenopodium oahuense*	15-20 %RH at storage temp	-18°C (0°F)	10-15
Amaranthaceae	*Nototrichium divaricatum*	15-20 %RH at storage temp	-18°C (0°F)	5+
Amaranthaceae	*Nototrichium humile*	15-20 %RH at storage temp	-18°C (0°F)	
Amaranthaceae	*Nototrichium sandwicense*	15-20 %RH at storage temp	-18°C (0°F)	15+
Anacardiaceae	*Rhus sandwicensis*	Unknown	Unknown	
Apiaceae	*Peucedanum sandwicense*	15-20 %RH at storage temp	-18°C (0°F)	
Apiaceae	*Sanicula kauaiensis*	15-20 %RH at storage temp	-18°C (0°F)	
Apiaceae	*Sanicula mariversa*	15-20 %RH at storage temp	-18°C (0°F)	15+
Apiaceae	*Sanicula purpurea*	15-20 %RH at storage temp	-18°C (0°F)	
Apiaceae	*Sanicula sandwicensis*	15-20 %RH at storage temp	-18°C (0°F)	
Apiaceae	*Spermolepis hawaiiensis*	15-20 %RH at storage temp	-18°C (0°F)	10+
Apocynaceae	*Alyxia stellata*	No drying recommended	No storage recommended	< 1
Apocynaceae	*Ochrosia compta*	Unknown	Unknown	
Apocynaceae	*Ochrosia haleakalae*	Unknown	Unknown	
Apocynaceae	*Ochrosia kauaiensis*	Unknown	Unknown	
Apocynaceae	*Ochrosia kilaueaensis*	Unknown	Unknown	
Apocynaceae	*Pteralyxia kauaiensis*	Unknown	Unknown	

Each column can be filtered using the triangle icons next to header fields in row 1. In the example below, we have filtered for all *Cyanea* species.



The third sheet is the **User's Guide – Full Version**:



	A	B	C	D	E	F	G	H	I	J
1	Taxonomy			Seed Storage Behavior Information				Seed Storage Longe		
2	Family	Species	Research Status	Recommended Desiccation Protocol	Recommended Storage Temperature	Storage Behavior	Freeze Sensitive Details	Re-Collection Interval (Yrs)	Highest % Germ	Fresh seeds?
3	Aizoaceae	*Sesuvium portulacastrum*	Unknown	Unknown	Unknown	Unknown				
4	Amaranthaceae	*Achyranthes atollensis*	Inferred	15-20 %RH at storage temp	-18°C (0°F)	Likely Orthodox*				
5	Amaranthaceae	*Achyranthes mutica*	Inferred	15-20 %RH at storage temp	-18°C (0°F)	Likely Orthodox*				
6	Amaranthaceae	*Achyranthes splendens*	Tested	15-20 %RH at storage temp	-18°C (0°F)	Orthodox		15+	54	yes
7	Amaranthaceae	*Amaranthus brownii*	Inferred	15-20 %RH at storage temp	-18°C (0°F)	Likely Orthodox*				
8	Amaranthaceae	*Charpentiera densiflora*	Tested	15-20 %RH at storage temp	-18°C (0°F)	Orthodox		10+	72	no
9	Amaranthaceae	*Charpentiera elliptica*	Inferred	15-20 %RH at storage temp	-18°C (0°F)	Likely Orthodox*				
10	Amaranthaceae	*Charpentiera obovata*	Tested	15-20 %RH at storage temp	-18°C (0°F)	Likely Orthodox*		2+	18	no
11	Amaranthaceae	*Charpentiera ovata*	Inferred	15-20 %RH at storage temp	-18°C (0°F)	Likely Orthodox*				
12	Amaranthaceae	*Charpentiera tomentosa*	Tested	15-20 %RH at storage temp	-18°C (0°F)	Likely Short-Lived		< 5	40	no
13	Amaranthaceae	*Chenopodium oahuense*	Tested	15-20 %RH at storage temp	-18°C (0°F)	Orthodox		10-15	56	no
14	Amaranthaceae	*Nototrichium divaricatum*	Tested	15-20 %RH at storage temp	-18°C (0°F)	Likely Orthodox		5+	7	no
15	Amaranthaceae	*Nototrichium humile*	Inferred	15-20 %RH at storage temp	-18°C (0°F)	Likely Orthodox*				
16	Amaranthaceae	*Nototrichium sandwicense*	Tested	15-20 %RH at storage temp	-18°C (0°F)	Orthodox		15+	10	no
17	Anacardiaceae	*Rhus sandwicensis*	Unknown	Unknown	Unknown	Unknown				
18	Apiaceae	*Peucedanum sandwicense*	Inferred	15-20 %RH at storage temp	-18°C (0°F)	Likely Orthodox*				
19	Apiaceae	*Sanicula kauaiensis*	Inferred	15-20 %RH at storage temp	-18°C (0°F)	Likely Orthodox*				
20	Apiaceae	*Sanicula mariversa*	Tested	15-20 %RH at storage temp	-18°C (0°F)	Orthodox		15+	90	no
21	Apiaceae	*Sanicula purpurea*	Inferred	15-20 %RH at storage temp	-18°C (0°F)	Likely Orthodox*				
22	Apiaceae	*Sanicula sandwicensis*	Inferred	15-20 %RH at storage temp	-18°C (0°F)	Likely Orthodox*				
23	Apiaceae	*Spermolepis hawaiiensis*	Tested	15-20 %RH at storage temp	-18°C (0°F)	Orthodox		10+	93	no
24	Apocynaceae	*Alyxia stellata*	Tested	No drying recommended	No storage recommended	Likely Recalcitrant		< 1	56	yes
25	Apocynaceae	*Ochrosia compta*	Unknown	Unknown	Unknown	Unknown				
26	Apocynaceae	*Ochrosia haleakalae*	Unknown	Unknown	Unknown	Unknown				
27	Apocynaceae	*Ochrosia kauaiensis*	Unknown	Unknown	Unknown	Unknown				

Columns A-H give the most critical information that most of our users need to know to plan for banking their seed collections.

- A and B list plant family and species names
- C provides the research status of this species (see above, and legend)
- D and E provide our recommended seed storage conditions (desiccation and temperature)
- F and G provide seed storage behavior categories inferred from our research
- H provides information on how long seeds may last in storage (RCIs)

Other columns beyond those give more detailed information about % germination, seed longevity, and species-specific notes that may affect longevity.

Not included in this version of the User's Guide is information about seed dormancy and germination requirements, treatments, or timing. The HSBP is currently working on synthesizing this research to provide it in the next version of the User's Guide.

The fourth and last sheet provides links to a few **Other Resources** we recommend for seed banking in Hawai‘i.