### 4.1 PROJECT HIGHLIGHTS

During this reporting period, OANRP outplanted a total of 1,430 individuals of 17 MIP and OIP taxa. In the last year, OANRP made 784 observations at *in situ* sites and outplanting sites of IP taxa. In this chapter, a summary of this year's highlights are included, along with the explanation for understanding the Taxon Status, Threat Control, and Genetic Storage Summaries. Lastly, our five year stabilization plan for *Plantago princeps* var. *princeps* and *Cyanea superba* subsp. *superba* are presented. Some of this year's highlights include:

*Cyanea grimesiana* subsp. *obatae* (MIP & OIP): This is a continuation of the update from the controlled breeding study in 2014. This project was initiated to conduct supplemental pollination experiments to compare the fitness of progeny from self-pollinated, intra-population and inter-population hand crosses. This project was designed to address concerns for difficulty of *ex situ* propagation and poor survival and lack of recruitment at outplantings and wild sites. This study will continue into the next fiscal year, as we will outplant this coming winter, and report on the progress of those outplantings in future reports. We did not observe differences in fruit set, seed set, seed weight, or germination due to source populations). Fruit set, however, was higher than expected when the pollen used was collected on the same day. This suggests that our protocol for transporting and using pollen did not suffice for this species. It would be good to determine whether or not *C. grimesiana* subsp. *obatae* pollen can be dried and stored. Two outplantings of the progeny from this study will be planted this winter, one site at Palikea and another at Makaha. Locations and methods were approved by OANRP, NARS, and OPEPP staff.

*Eugenia koolauensis* (OIP): We have obtained material from the 150 maternal lines targeted for this species via salvaging seedlings and taking cuttings from trees in poor health at the *in situ* populations. Living collections now need to be cloned to meet genetic storage goals. A planting at Koko Crater Botanical Garden last winter was successful. Other than one initial rust treatment, plants appeared rust free for the last six months. Plants are growing, flowering, and fruiting. Due to this success, additional plants will be added this winter.

Cliff habitat and species (*Dubautia herbstobatae, Kadua parvula, Sanicula mariversa, Tetramolopium filiforme, Viola chamissoniana* subsp. *chamissoniana*): Declines in populations of cliff-dwelling IP taxa were observed this year. Of the 20 Population Units with the highest decline, cliff species comprise eight of the 20 PU, three of which experienced the highest declines this year. There has been a decline in all of the observed PU for these taxa except where outplantings occurred. There was such a decline in the *Kadua parvula* Ohikilolo PU that an overall decline still occurred despite the outplanting of 70 plants this year that yielded 20 matures. OANRP will strategize on ways to improve cliff habitats.

*Dubautia herbstobatae* (OIP): The first outplanting of this taxon was conducted this year. Over 50 plants were planted onto cliff habitat in Makaha via rappelling. The outplants are growing and little mortality has been observed. This is hopeful compared to the observations of *in situ* populations. Several days spent monitoring populations at Ohikilolo revealed a substantial decline in populations, along with decline in cliff habitat in general (see above). Surveys of Population Units are not complete and will be reported next year.

*Pritchardia kaalae* (MIP): Obtaining a bulk collection of fruits from this taxon to complete storage testing at the National Center for Genetic Resources Preservation (NCGRP) has been challenging. Restricted access to Makua Military Reservation last year allowed rat populations to increase in the absence of

OANRP control. Due to the long maturation time for fruit of *P. kaalae*, staff needed to wait for rat control to take effect and allow fruit to mature once again. In July 2016, the final shipment of seeds were mailed to NCGRP for one final test to confirm storage protocols for this taxon. Seeds will be subjected to various storage temperatures at various levels of processing (whole seed versus embryo removed) to determine the minimum amount of processing that will yield the longest re-collection interval.

*Cyanea superba* subsp. *superba* (MIP): A laboratory trial was conducted to examine seed germination reduction during *C. superba* subsp. *superba* fruit senescence (results are included in Appendix 4-1). Seed germination rates were relatively high among seeds sown from fresh fruit, however, viability from fruit that senesced for one to two weeks was less than half that of fresh material, followed by a total loss of viability after two weeks of fruit senescence. These results suggest a potential recruitment limitation in the event that fresh seeds are not dispersed by frugivores, as fruits tend to senesce on the plant for several days before falling to the ground.

### 4.2 TAXON STATUS SUMMARY

In the last year, there have been changes in the number of mature plants at 84/133 of the Manage for Stability Population Units managed by OANRP. Table 1 shows the Population Units where a change was observed in the last reporting period. The difference in the number of mature plants reported last year and this year is given (#Mat), with the percent change observed at each (%change). Most of the largest changes are due to fluctuations at outplanting sites when more plants are added, many plants in the same cohort mature at the same time, or are observed to have died at the same time. PU that are in **bold text** are wild *in situ* PUs that have not been augmented with outplants, so that the increase in the total number of plants is due to natural recruitment, the death of known plants OR better estimates from recent surveys. The largest increases occurred in PU that have been augmented with outplants, with a few exceptions. One exception was an increase in *in situ* populations of *Cyrtandra dentata* in the Koolau Mountains due to more thorough surveying in the past year. The next exception was in increase in the number of mature plants of *Euphorbia celastroides* var. *kaenana* at Kaena. This was due to a new population inside of the predator proof portion of the Natural Area Reserve. The last exception was for *Cyanea acuminata* at Kaala, which was also due to a more thorough survey, instead of an estimate, at one of the Population Reference Sites.

As mentioned in the Project highlights, many of the declines that were observed this year are due to thorough surveying of cliff dwelling species, particularly *Kadua parvula (in situ)*, *Plantago princeps* var. *princeps (in situ* and an outplanting), and *Tetramolopium filiforme* (an outplanting). Other substantial declines occurred at outplantings that appear to have failed (not on cliffs). These include *Schiedea obovata* at Makaha, *Phyllostegia mollis* at Ekahanui, and *Labordia cyrtandrae* at Koloa. Declines for these three species cannot be attributed to any one cause.

ige (ne	gative of positive) to the number of m	ature pr	%
IP	Species and MFS PU with DECREASES	$\Delta$ Mat	Change
MIP	TetFil - Puhawai	-18	-600%
MIP	PlaPriPri - Ekahanui	-41	-586%
MIP	KadPar - Halona	-62	-200%
MIP	SchObo - Makaha	-70	-92%
OIP	PhyMol - Ekahanui	-10	-91%
MIP	VioChaCha - Ohikilolo	-178	-86%
MIP	AleMacMac - Makua	-5	-83%
MIP	NerAng - Makua	-52	-76%
OIP	LabCyr - Koloa	-24	-73%
MIP	PlaPriPri - Halona	-4	-67%
MIP	SanMar - Kamaileunu	-2	-67%
MIP	SchObo - Keawapilau to West Makaleha	-22	-61%
MIP	VioChaCha - Halona	-7	-47%
MIP	KadDegDeg - Kahanahaiki to Pahole	-45	-44%
MIP	SchKaa - Pahole	-25	-43%
MIP	NotHum - Waianae Kai	-61	-39%
MIP	SchNut - Kapuna-Keawapilau Ridge	-19	-35%
MIP	TetFil - Ohikilolo	-492	-26%
MIP	CenAgrAgr - Kahanahaiki and Pahole	-64	-25%
MIP	EupCelKae - Puaakanoa	-30	-25%
MIP	AleMacMac - Makaha	-7	-24%
OIP	PhyHir - Puu Palikea	-27	-24%
MIP	SchNut - Kahanahaiki to Pahole	-20	-23%
MIP	SchObo - Kahanahaiki to Pahole	-51	-22%
MIP	CyaSupSup - Kahanahaiki	-10	-21%
MIP	NerAng - Waianae Kai Mauka	-2	-18%
OIP	HesSwe - Lower Opaeula	-3	-17%
OIP	PhyMol - Kaluaa	-14	-16%
OIP	CyaKoo - Kaipapau, Koloa & Kawainui	-13	-12%
MIP	CyrDen - Kahanahaiki	-13	-12%
MIP	FluNeo - Makaha	-1	-11%
MIP	HibBraMok - Manuwai	-15	-10%
MIP	DelWai - Kaluaa	-13	-10%
MIP	GouVit - Keaau	-32	-8%
MIP	SchKaa - South Ekahanui	-11	-7%
OIP	AbuSan - Kaawa to Puulu	-11	-6%
MIP	CenAgrAgr - Makaha and Waianae Kai	-10	-6%
MIP	EupCelKae - East of Alau	-10	-5%
OIP	CyaKoo - Poamoho	-1	-5%
MIP	NerAng - Manuwai	-1	-5%
OIP	CyaKoo - Opaeula to Helemano	-3	-3%
MIP	KadPar – Ohikilolo	-1	-4%
MIP		-4	-4%
	EupHer - Kapuna to Pahole		
MIP	CyaGriOba - Kaluaa	-4	-3%
MIP	NotHum - Manuwai	-3	-3%
MIP	MelTenf - Ohikilolo	-21	-2%
MIP	CenAgrAgr - Central Ekahanui	-3	-2%

-600%OIPHibBraMok - Keaau-586%MIPKadPar - Ekahanui-200%MIPPlaPriPri - Ohikilolo-92%MIPSanMar - Ohikilolo-91%MIPHibBraMok - Haili to Kawaiu-86%MIPHesOah - Makaha-83%MIPDubHer - Makaha	1at 20 6 4 2 39	% Change 100% 100% 100%
-600%OIPHibBraMok - Keaau-586%MIPKadPar - Ekahanui-200%MIPPlaPriPri - Ohikilolo-92%MIPSanMar - Ohikilolo-91%MIPHibBraMok - Haili to Kawaiu-86%MIPHesOah - Makaha-83%MIPDubHer - Makaha	20 6 4 2 39	100% 100% 100%
-586%MIPKadPar - Ekahanui-200%MIPPlaPriPri - Ohikilolo-92%MIPSanMar - Ohikilolo-91%MIPHibBraMok - Haili to Kawaiu-86%MIPHesOah - Makaha-83%MIPDubHer - Makaha	6 4 2 39	100% 100%
-200%MIPPlaPriPri - Ohikilolo-92%MIPSanMar - Ohikilolo-91%MIPHibBraMok - Haili to Kawaiu-86%MIPHesOah - Makaha-83%MIPDubHer - Makaha	4 2 39	100%
-92%MIPSanMar - Ohikilolo-91%MIPHibBraMok - Haili to Kawaiu-86%MIPHesOah - Makaha-83%MIPDubHer - Makaha	2 39	
-91%MIPHibBraMok - Haili to Kawaiu-86%MIPHesOah - Makaha-83%MIPDubHer - Makaha	39	100%
-86%         MIP         HesOah - Makaha           -83%         MIP         DubHer - Makaha		10070
-83% MIP DubHer - Makaha	~	89%
	8	73%
-76% MIP NerAng - Makaha	51	65%
, , , , , , , , , , , , , , , , , , ,	90	63%
-73% MIP HesOah - Pualii	10	63%
-67% MIP CyrDen - Kawaiiki (Koolaus)	8	62%
-67% MIP CyaLong - Kapuna to West Makaleha	35	56%
-61% MIP HibBraMok - Makua	44	35%
-47% MIP PhyHir - Haleauau to Mohiakea	25	35%
-44% OIP NerAng - Kaluakauila	35	35%
-43% MIP CyrDen - Opaeula (Koolaus)	12	34%
-39% MIP EupCelKae - Kaena 3	301	34%
-35% MIP PriKaa – E. Ohikilolo East & W. Makaleha	2	33%
-26% MIP CyaAcu - Makaleha to Mohiakea	39	26%
-25% OIP SchNut - Makaha	23	25%
-25% MIP AbuSan - Ekahanui and Huliwai	11	24%
-24% OIP GarMan - Helemano and Poamoho	4	24%
-24% OIP EugKoo - Oio	1	20%
-23% OIP PhyHir - Koloa	17	18%
-22% OIP GarMan - Haleauau	8	12%
-21% OIP GarMan - Lower Peahinaia	1	11%
-18% OIP CyaGriOba - Palikea (South Palawai)	12	10%
-17% MIP MelTenf - Mt. Kaala NAR	10	8%
-16% MIP CyaLong - Makaha and Waianae Kai	9	8%
KadDegDeg - Central Makaleha and		
-12% MIP West Branch of East Makaleha	1	4%
-12% MIP KadDegDeg - Alaiheihe and Manuwai	3	4%
-11% MIP CyaLong - Pahole	2	3%
-10% MIP CyrDen - Pahole to West Makaleha	12	2%
	3	1%
-9% MIP LabCyr – E. Makaleha to North Mohiakea	5	

**Table 1:** MFS PUs sorted by Decreasing and Increasing numbers of Mature Plants. **Bold** PUs have only wild plants.  $\Delta$ Mat = the change (negative or positive) to the number of mature plants from 2014. %change = percent observed (negative or positive).

The Taxon Status Summary for each IP taxon is included as Appendix 4-2. The example shown below (Table 2), displays the management designation, the original MIP or OIP population total, last year's reported total and the current status of the wild and outplanted plants for each PU. The PUs are grouped by those located inside the MIP or OIP AA (In) and PUs where all plants are outside of both AAs (Out). Definitions for each field are given below.

**Table 2:** Example of a Taxon Status Summary using Cenchrus agrimonioides var. agrimonioidesMakua Implementation Plan - Population Unit Status

TaxonName	: Cenchrus a	grim	onioi	ides v	ar. agrii	monioid	les		Та	rget # of	Matures	: 50		# MFS F	PU Met Go	bal: 3 of	3
Population Unit Name	Management Designation	Total Mature Original IP	Total Imm Original IP	Total Seedling Original IP	Total Mature 2014	Total Immature 2014	Total Seedling 2014	Total Mature Current	Total Immature Current	Total Seedling Current	Wild Mature Current	Wild Immature Current	Wild Seedling Current	Outplanted Mature Current	Outplanted Immature Current	Outplanted Seedling Current	PU LastObs Population Trend Date Notes
Kahanahaiki and Pahole	Manage for stability	210	66	0	327	138	128	319	61	79	80	42	70	239	19	9	2015-09-02 Thorough monito in the last year showed a decline
Kuaokala	Genetic Storage				1	3	0	1	3	0	1	3	0	0	0	0	2014-04-30 No monitoring in last year
	In Total:	210	66	0	328	141	128	320	64	79	81	45	70	239	19	9	
	: Out : Cenchrus a	•								rget # of					PU Met Go		
Action Area: FaxonName Population Unit Name		Total Mature Original	Total Imm Original	Total Seedling	ar. agrii Total Mature 2014	Total Immature 2014	Total Seedling 2014	Total Mature Current	Ta Total Immature Current	Total Seedling Current	Matures Wild Mature Current	: 50 Wild Immature Current	Wild Seedling Current	# MFS F Outplanted Mature Current	Outplanted Immature Current	oal: 3 of Outplanted Seedling Current	3 PU LastObs Population Trend Date Notes
FaxonName Population Unit Name	: Cenchrus a	Total Mature	Total Imm	Total Seedling Original	Total Mature	Total Immature	Total Seedling	Mature	Total	Total Seedling	Wild Mature	Wild	Seedling	Outplanted Mature	Outplanted Immature	Outplanted Seedling	PU LastObs Population Trend
FaxonName Population Unit Name Central Ekahanui Makaha and	Cenchrus a Management Designation	Total Mature Original IP	Total Imm Original IP	Total Seedling Original IP	Total Mature 2014	Total Immature 2014	Total Seedling 2014	Mature Current	Total Immature Current	Total Seedling Current	Wild Mature Current	Wild Immature Current	Seedling Current	Outplanted Mature Current	Outplanted Immature Current	Outplanted Seedling Current	PU LastObs Population Trend Date Notes
FaxonName Population Unit Name Central Ekahanui Wakaha and Walanae Kai	Cenchrus a Management Designation Manage for stability	Total Mature Original IP 20	Total Imm Original IP 0	Total Seedling Original IP 0	Total Mature 2014 168	Total Immature 2014 89	Total Seedling 2014 0	Mature Current 168	Total Immature Current 89	Total Seedling Current	Wild Mature Current 47	Wild Immature Current 72	Seedling Current 0	Outplanted Mature Current 121	Outplanted Immature Current 17	Outplanted Seedling Current	PU LastObs Population Trenc Notes 2014-09-02 Monitoring shown no change 2015-04-13 More plants were added to the
	Cenchrus a Management Designation Manage for stability Manage for stability	Total Mature Original IP 20 9	Total Imm Original IP 0 3	Total Seedling Original IP 0	Total           Mature           2014           168           10	Total Immature 2014 89 7	Total Seedling 2014 0 5	Mature Current 168 171	Total Immature Current 89 128	Total Seedling Current 0 5	Wild Mature Current 47 5	Wild Immature Current 72 7	Seedling Current 0 5	Outplanted Mature Current 121 166	Outplanted Immature Current 17 121	Outplanted Seedling Current 0	PU LastObs Population Trend Notes 2014-09-02 Monitoring show no change 2015-04-13 More plants were added to the outplanting site 2014-09-03 Monitoring show

**Population Unit Name:** Groupings of Population Reference Sites. Only PUs designated to be 'Manage for Stability' (MFS), 'Manage Reintroduction for Stability/Storage,' or 'Genetic Storage' (GS) are shown in the table. Other PUs with 'No Management' designations are not managed and their status will not be tracked or reported.

**Management Designation:** For PUs with naturally occurring (*in situ*) plants remaining, the designation is either 'Manage for Stability' or 'Genetic Storage'. Some MFS PUs will be augmented with outplantings to reach stability goals. When reintroductions alone will be used to reach stability, the designation is 'Manage Reintroduction for Stability.' When a reintroduction will be used for producing propagules for genetic storage, the designation is 'Manage Reintroduction for Storage'.

**Total Original IP Mature, Immature, Seedling:** These first three columns of numbers display the original population numbers as noted in the first Implementation Plan reports of MIP (2005), and OIP (2008). When no numbers are displayed, the PU was not known at the time of the IPs

**Total Mature, Immature and Seedling 2014:** This displays the **SUM** of the number of *wild and outplanted* mature, immature plants and seedlings from the previous year's report. These numbers should be compared to those in the next three columns to see the change observed over the last year.

**Total Current Mature, Immature, Seedling:** The **SUM** of the *current* numbers of *wild and outplanted* individuals in each PU. This number will be used to determine if each PU has reached stability goals for mature plants. These last three columns can be compared with the previous three columns to see the change observed over the last reporting period.

**Wild Current Mature, Immature, Seedling:** These set of three columns display the most up to date population estimates of the wild (*in situ*) plants in each PU. These numbers are generated from OANRP monitoring data, data from the Oahu Plant Extinction Prevention Program (OPEP), Koolau Mountains Watershed Partnership and Oahu NARS staff. The estimates may have changed from last year if estimates were revised after new monitoring data was taken or if the PUs have been split or merged since the last reporting period. The most recent estimate is used for all PUs, but some have not been monitored in several years. Several PU have not been visited yet by OANRP and no plants are listed in the population estimates. As these sites are monitored, estimates will be updated.

**Outplanted Current Mature, Immature, Seedling:** The third set of three columns display the numbers of individuals OANRP and partner agencies have outplanted into each PU. This includes augmentations of *in situ* sites, reintroductions into nearby sites and introductions into new areas.

**PU LastObs Date:** Last Observation Date of the most recent Population Reference Site observed within a PU. Where thorough monitoring was done, the estimates were updated.

**Population Trend Notes:** Comments on the general population trend of each PU are given here. This may include notes on whether the PU was monitored in the last year, a brief discussion of the changes in population numbers from the previous estimates, and some explanation of whether the change is due to new plants being discovered in the same site, a new site being found, reintroductions or augmentations that increased the numbers or fluctuations in the numbers of wild plants. In some cases where the numbers have not changed, OANRP has monitored the PU and observed no change. When the PU has not been monitored, the same estimate from the previous year is repeated.

### 4.3 THREAT CONTROL SUMMARY

The Threat Control Summary for each IP taxon is included as Appendix 4-3. An example shown below (Table 3), includes the current status of fence construction and removal of pigs and goats from Management Units, invasive plant, rat and slug control and preventing wildfire.

Several changes in ungulate threat control were due to pigs found in fence units including Makaha I, Ekahanui II and III, and Kapuna Upper. Fences have been repaired and pigs have been removed but it is uncertain whether or not all pigs have been removed from any of the four units at this time. The threat control status for ungulates for these affected PU has been changed to 'Partial' until the last pigs are removed.

Weed control continues at most MU, and is a threat to all taxa in all PU. See Chapter 3 for more details. This year we reported the weed control status by overlaying weed control efforts with IP taxa population sites in GIS. To receive a 'Yes', the entirety of a 50m radial buffer around a PU needed to be weeded. There are only four population sites for four different taxa that meet this goal. All other weed control efforts are described as 'Partial' for this reporting year. Of the 133 MFS PU, 95 PU receive 'Partial' weed control status.

Rat control continued around many PU in the last year. Although rats are considered a potential threat to most IP taxa, they are mainly controlled around sites where significant damage has been observed. There are situations where occasional damage to a few plants is observed. In those cases, if the damage is not observed again, control is not immediately installed and the site is monitored more closely. Substantial damage has been seen this year at multiple PU of *Delissea waianaeensis*, as well as the outplanting of *Labordia cyrtandrae* at Kaala. New rat control grids were established at these sites. Rats are considered a

threat to 20 of the 39 taxa in the MIP and OIP and are controlled at 83 population sites in 26 of the 63 MFS PU with those taxa. Last year we only conducted rat control at 15 MFS PU. A number of MFS PUs do receive year round or seasonal protection from rats where they are located within large rat control grids at Palikea, Kahanahaiki, Makaha, Ekahanui and Ohikilolo Ridge.

Slugs are a threat to seedlings and small immature plants of many native plants. They are noted as a threat to 25 of the 39 MIP and OIP taxa. Slugs are currently controlled at 21 of the 83 MFS PUs with those taxa, which is an increase from 10 MFS PU that received control last year. Decisions on where to initiate control are based on staff availability and can only occur at sites without native snails, thus meeting label restrictions. Future outplantings of IP taxa that may be dependent on slug control will be planned for areas that do not have those restrictions.

An example shown below (Table 3), summarizes the threat status at each Population Unit for every IP taxa. "Yes," "No," or "Partial" is used to indicate the level of threat management. Partial management has additional percentage based upon the number of mature plants being protected.

Table 3: Example of a Threat Control Summary using Cenchrus agrimonioides var. agrimonioides

### Threat Control Summary

#### Action Area: In

#### TaxonName: Cenchrus agrimonioides var. agrimonioides

PopulationUnitName	ManagementDesignation	# Mature Plants	Ungulates Managed	Weeds Managed	Rats Managed	Slugs Managed	Fire Managed
Kahanahaiki and Pahole	Manage for stability	319	Yes	Partial 2%	Partial 37%	No	No
Kuaokala	Genetic Storage	1	No	No	No	No	No

#### **Action Area: Out**

#### TaxonName: Cenchrus agrimonioides var. agrimonioides

PopulationUnitName	ManagementDesignation	# Mature Plants	Ungulates Managed	Weeds Managed	Rats Managed	Slugs Managed	Fire Managed
Central Ekahanui	Manage for stability	168	Yes	No	Yes	No	Νο
Makaha and Waianae Kai	Manage for stability	171	Partial 97%	Partial 96%	No	No	No
South Huliwai	Genetic Storage	15	No	No	No	No	No

= Threat to Taxon within Population Unit

No Shading = Absence of threat to Taxon within Population Unit

Ungulate Managed = Culmination of Cattle, Goats, and Pig threats

Yes=All PopRefSites within Population Unit have threat controlled

No=All PopRefSites within Population Unit have no threat control Partial%=Percent of mature plants in Population Unit that have threat controlled Partial 100%= All PopRefSites within Population Unit have threat partially controlled

Partial 0%= Threat partially controlled, but no mature plants

**Population Unit Name:** Groupings of Population Reference Sites. Only PUs designated to be 'Manage for Stability' (MFS), 'Manage Reintroduction for Stability/Storage,' or 'Genetic Storage' (GS) are shown in the table.

**Management Designation:** Designations for PUs with ongoing management are listed. Population Units that are MFS are the first priority for complete threat control. PUs that are managed in order to secure genetic storage collections receive the management needed for collection (ungulate and rodent control), but may be a lower priority for other threat control.

# Mature Plants: Number of Mature Plants within the Population Unit.

**Threat Columns:** The most common threats are listed in the next columns. To indicate if the threat is noted at each PU, a shaded box is used. If the threat is not present at that PU, it is not shaded. Threat control is defined as:

Yes = All sites within the PU have the threat controlled

No = All sites within the PU have no threat control

Partial %= Percent of mature plants in Population Unit that have threat controlled

Partial 100%= All PopRefSites within Population Unit have threat partially controlled

Partial (with no %) = All PopRefSites within Population Unit have threat partially controlled and only immature plants have been observed.

Partial 0% = Threat partially controlled, but no mature plants are currently present in the PU.

**Ungulates:** This threat is indicated if pigs, goats or cattle have been observed at any sites within the PU. This threat is controlled (Yes) if a fence has been completed and all ungulates removed from the site. Most PUs are threatened by pigs, but others are threatened by goats and cattle as well. The same type of fence is used to control for all three types of ungulates on Oahu. Partial indicates that the threat is controlled for some but not all plants in the PU or only one of the ungulate threats has been controlled. If some of the mature plants in a MFS PU are outside of the fence, the threat is partially controlled for the percentage of mature plants inside the fence. If all plants are fenced, but only goats have been eliminated, the threat has been partially controlled for 100% of the mature plants.

**Weeds:** This threat is indicated at all PUs for all IP taxa. This threat is controlled if weed control has been conducted in the vicinity of the sites for each PU. If only some of the sites have had weed control, 'Partial' is used to indicate what portion of the PU has had control.

**Rats:** This threat is indicated for any PUs where damage from rodents has been confirmed by OANRP staff. This includes fruit predation and damage to stems or any part of the plant. The threat is controlled if the PU is protected by snap traps and bait stations. For some taxa, rats are not known to be a threat, but the sites are within rat control areas for other taxa so the threat is considered controlled. In these cases, the box is not shaded but control is 'Yes' or 'Partial.' Partial indicates that the threat is fully controlled over part of the PU.

**Slugs:** This threat is indicated for IP taxa as confirmed by OANRP staff. Currently, slug control is conducted under an Experimental Use Permit from Hawaii State Department of Agriculture, which permits the use of Sluggo<sup>®</sup>. Partial indicates that the threat is fully controlled over part of the PU.

**Fire:** This threat is indicated for PUs that occur on Army lands within the high fire threat area of the Makua AA, and some PUs within the Schofield West Range AA and Kahuku Training Area that have been threatened by fire within the last ten years. Similarly, PUs that are not on Army land were included if there is a history of fires in that area. This includes the PUs below the Honouliuli Contour Trail, the gulches above Waialua where the 2007 fire burned including Puulu, Kihakapu, Palikea, Kaimuhole, Alaiheihe, Manuwai, Kaomoku iki, Kaomoku nui and Kaawa and PUs in the Puu Palikea area that were threatened by the Nanakuli fire. Threat control conducted by OANRP includes removing fuel from the area with pesticides, marking the site with Seibert Stakes for water drops, and installing fuel-breaks in fallow agricultural areas along roads. 'Partial' means that the threat has been partially controlled to the

whole PU, not that some plants are fully protected. Firebreaks and other control measures only partially block the threat of fire which could make it into the PU from other unprotected directions.

### 4.4 GENETIC STORAGE SUMMARY

The Genetic Storage Summary for each IP taxon is included as Appendix 4-4. Every year, OANRP collects propagules from IP taxa for *ex situ* genetic storage. The amount of propagules to meet these goals were pre-determined in the MIP and OIP. In general, each wild plant (up to 50 plants from each PU) needs either 50 viable seeds (as estimated at the time of collection) or 3 explants/plants in tissue culture or nursery. This year we reported only the collections that have not expired, *i.e.* have not been stored for longer than the species re-collection interval.

This year there were 66 PU that reached their storage goal, representing 898 plants. This is a slight decline from last year, and attributed to the removal of expired collections from the seed bank inventory. There are an additional 1,351 plants that meet their storage goal in 217 other PU (where the PU genetic storage effort is not 100% complete. Sixty-nine new plants met their genetic storage goal this year.

In the example below (Table 4), estimates of seeds remaining in genetic storage account for the expected viability of the stored collections. The viability rates of a sample of most collections are measured prior to storage. These rates are used to estimate the number of viable seeds in the rest of the stored collection. If the product of (the total number of seeds stored) and (the initial percentage of viable seeds) is >50, that founder is considered secured in genetic storage. If each collection of a species is not tested, the initial viability is determined from the mean viability of (preference in descending order): 1. Other founders in that collection; 2. That founder from other collections; 3. All founders in that population reference site; 4. All founders of that species.

						Partial Stora	age Status			Storage	Goals		Storage Goals Met	
Population Unit Name	Management Designation	# of Po Current Mature	Current	Dead and Repres.	# Plants >= 10 in SeedLab	# Plants >= 10 Est Viable in SeedLab	# Plants >=1 Microprop	# Plants >=1 Army Nursery	# Plants >= 50 in SeedLab	# Plants >= 50 Est. Viable in SeedLab	# Plants >=3 in Microprop	# Plants >=3 Army Nursery	# Plants that Met Goal	% Completed Genetic Storage Requirement
Action Area: In														
Cenchrus agrimonioid	es var. agrimonioides													
Kahanahaiki and Pahole	Manage for stability	80	42	40	74	56	0	2	34	10	0	1	11	22%
Kuaokala	Genetic Storage	1	3	0	0	0	0	1	0	0	0	1	1	100%
Action Area: Out														
Cenchrus agrimonioid	es var. agrimonioides													
Central Ekahanui	Manage for stability	47	72	18	36	19	0	40	12	1	0	38	38	76%
Makaha and Waianae Kai	Manage for stability	5	7	6	3	2	0	9	0	0	0	9	9	82%
South Huliwai	Genetic Storage	15	13	13	18	10	0	20	6	3	0	17	19	68%
		Total Current Mature	Total Current Imm.	Total Dead and Repres.	Total # Plants w/ >=10 Seeds in SeedLab	Total # Plants w/ >=10 Est Vaible Seeds In SeedLab	Total # Plants w/ >=1 Microprop	Total # Plants w/ >=1 Army Nursery	Total # Plants w/ >=50 Seeds in SeedLab	Total # Plants w/ >=50 Est Viable Seeds in SeedLab	Total # Plants w/ >=3 in Microprop	Total # Plants w/ >=3 Army Nursery	Total # Plants that Met Goal	
		148	137	77	131	87	0	72	52	14	0	66	78	

 Table 4: Example of a Genetic Storage Summary using Cenchrus agrimonioides var. agrimonioides

 Genetic Storage Summary

**Number (#) of Potential Founders:** These first columns list the current number of live *in situ* immature and mature plants in each PU. These plants have been collected from already, or may be collected from in the future. The number of dead plants from which collections were made in the past is also included to show the total number of plants that could potentially be represented in genetic storage for each PU since collections began. Immature plants are included as founders for all taxa, but they can only serve as founders for some. For example, for *Hibiscus brackenridgei* subsp. *mokuleianus*, cuttings can be taken

from immature plants for propagation. In comparison, for *Sanicula mariversa*, cuttings cannot be taken and seed is the only propagule used in collecting for genetic storage. Therefore, including immature plants in the number of potential founders for *S. mariversa* gives an over-estimate. The 'Manage reintroduction for stability/storage' PUs have no potential founders. The genetic storage status of the founder stock used for these reintroductions is listed under the source PU.

**Partial Storage Status:** To meet the IP genetic storage goal for each PU for taxa with seed storage as the preferred genetic storage method, at least 50 seeds must be stored from 50 plants. This year, the number of seeds needed for each plant (50) accounts for the original viability (Estimate Viability) of seed collections. In order to show intermediate progress, this column displays the number individual plants that have collections of >10 seeds in storage. For taxa where vegetative collections will be used to meet storage goals, a minimum of three clones per plant in either the Lyon Micropropagation Lab, the Army nurseries or the State's Pahole Mid-elevation Nursery is required to meet stability goals. Plants with one or more representatives in either the Lyon Micropropagation Lab or a nursery are considered to partially meet storage goals. The number of plants that have met this goal at each location is displayed.

**# Plants that Met Goal:** This column displays the total number of plants in each PU that have met the IP genetic storage goals. As discussed above, a plant is considered to meet the storage goal if it has 50 seeds in storage or three clones in micropropagation or three in a nursery. For some PUs, the number of founders has increased in the last year; therefore, it is feasible that NRS could be farther from reaching collection goals than last year. Also, as seeds age in storage, plants are outplanted, or explants contaminated, this number will drop. In other PUs where collections have been happening for many years, the number of founders represented in genetic storage may exceed the number of plants currently extant in each PU. In some cases, plants that are being grown for reintroductions are also being counted for genetic storage. These plants will eventually leave the greenhouse and the genetic storage goals will be met by retaining clones of all available founders or by securing seeds in storage. This column does not show the total number of seeds in storage; in some cases thousands of seeds have been removed from one plant. For the first time this year, collections that have expired in the seed bank, have been removed from the inventory and are not reflected here as represented. These collections have been flagged for *in situ* seed dispersal as collections have aged past adequate genetic representation of founder lines without high levels of artificial selection.

**% Completed Genetic Storage Requirement:** Describes the percent of Founder Plants that have met Genetic Storage goals. Genetic storage of at least 50 seeds each from 50 individuals, or at least three clones each in propagation from 50 individuals, is required for each PU. If there are fewer than 50 founders for a PU, genetic storage is required from all available founders. For example, if there are at least 50 seeds from five individuals, or at least three clones in propagation from five individuals, then listed in the tables is 10%.

### 4.5 FIVE YEAR RARE PLANT PLANS

These plans are intended to include all pertinent species information for stabilization, serve as a planning document and as an updated educational reference for OANRP staff. In many cases, data or information is still being gathered and these plans will continue to be updated. A brief description of each section is given here:

- **Species Description:** The first section provides an overview of each taxon. The IP stability requirements are given, followed by a taxon description, biology, distribution, population trends, and habitat.
- **Reproductive Biology Table:** This information was summarized by OANRP based on best available data from the MIP, OIP, USFWS 5-year Status Updates, OANRP field observations and other published research. Phenology is primarily based on observations in the OANRP rare plant database. The suspected pollinator is based on casual observations, pollinator syndromes as reported in the MIP and OIP, or other published literature. The information on seeds is from data collected at the Army seed lab and from collaborative research with the Harold L. Lyon Arboretum.
- Known Distribution & Historic Collections Table: This information was selected from Bishop Museum specimen records and collections listed in published research, the Hawaii Biodiversity and Mapping Program and other collectors notes.
- **Species Occurrence Maps:** These maps display historic and current locations, MUs, landmarks and any other useful geographic data for each taxon. Other features may be used on public documents to obscure locations of rare elements.
- **Population Units:** A summary of the PUs for each taxon is provided with current management designations, action areas and management units.
- Habitat Characteristics and Associated Species: These tables summarize habitat data taken using the Hawaii Rare Plant Restoration Group's Rare Plant Monitoring Form. The data is meant to provide an assessment of the current habitat for the in situ and outplanting sites. Temperature and rainfall estimates are also included for each site when available.
- **Pictures:** These photos document habitat, habit, floral morphology and variation; and include many age classes and stages of maturing fruit and seed. This will serve as a reference for field staff making collections and searching for seedlings.
- **Taxonomic Background:** This section provides information pertaining to the history of the taxonomy of the species.
- **Population Structure & Trends:** Data from monitoring the population structure for each species is presented with a plan to establish or maintain population structure at levels that will sustain stability goals. A review of population estimates for each Population Unit (PU) is displayed in a table. Estimates come from the MIP, OIP, USFWS 5-year Status Updates and OANRP field observations. In most cases, these estimates cannot be used to represent a population trend.
- **Reintroduction Plan:** A standardized table is used to display the reintroduction plans for each PU. Every outplanting site in each PU is displayed showing the number of plants to be established, the PU stock and number of founders to be used and type and size of propagule (immature plants, seeds, etc.). Comments focus on details of propagation and planting strategies.
- Threats & Stabilization Goals Update: For each PU, the status of compliance with all stability goals is displayed in this table. All required MFS PUs are listed for each taxon. 'YES, NO or PARTIAL' are used to represent compliance with each stability goal. For population targets, whether or not each PU has enough mature plants is displayed, followed by an estimate on whether a stable population structure is present. The major threats are listed separately for each PU. The boxes are shaded to display whether each threat is present at each PU. A dark shade identifies PUs where the threat is present and the lighter boxes where the threat is not applicable. The corresponding status of threat control is listed as 'YES, NO or PARTIAL' for each PU. A summary of the status of genetic storage collections is displayed in the last column.

- Genetic Storage Section: This section provides an overview of propagation and genetic storage • issues. A standardized table is used to display information recorded for each taxon or PUs where applicable. The plan for genetic storage is displayed and discussed. In most cases, seed storage is the preferred genetic storage technique; it is the most cost-effective method, requires the least amount of maintenance once established, and captures the largest amount of genetic variability. For taxa that do not produce enough mature seed for collection and testing storage conditions, micropropagation is considered the next best genetic storage technique. The maintenance of this storage method is continual, but requires much less resources and personnel than establishing a living collection in the nursery or a garden. For those taxa that do not produce storable seed and cannot be established in micropropagation, a living collection of plants in the nursery or an inter situ site is the last preferred genetic storage option. In most cases, current research is ongoing to determine the most applicable method. For species with substantial seed storage data, a schedule may be proposed for how frequently seed bank collections will need to be refreshed to maintain genetic storage goals. This schedule is based only on storage potential for the species; other factors such as threats and plant health must be factored into this schedule to create a revised collection plan. Therefore, the frequency of refresher collections will constantly be adjusted to reflect the most current storage data. The re-collection interval is set prior to the time period in storage where a decrease in viability is detected. For example, Delissea waianaeensis shows no decrease in viability after ten years. OANRP would not have to re-collect prior to ten years as the number of viable seeds in storage would not have yet begun to decrease. The re-collection interval will be 10 years or greater (10 + yrs). If its viability declines when stored collections are tested at year 15, the interval will be set between 10 and 15 years. Further research may then be conducted to determine what specific yearly interval is most appropriate. The status of seed storage research is also displayed and discussed. Collaborative research with the USDA National Center for Genetic Resources Preservation (NCGRP) and Lyon Arboretum Seedlab is ongoing.
- Management Discussion & 5-Year Action Plan: A summary of the management approach, overall strategy and important actions for each taxon. This section displays the schedule of actions for each PU. All management is planned by 'MIP or OIP Year' and the corresponding calendar dates are listed. This table can be used to schedule the actions proposed for each species into the OANRP scheduling database. Comments in this section focus on details of certain actions or explain the phasing or timeline in some PUs.

# Plantago princeps var. princeps



Scientific name: *Plantago princeps* Cham. & Schlechtend. var. *princeps*Hawaiian name: *Ale, laukahiu kauhiwi*Family: Plantaginaceae (Plantain family)Federal status: Listed Endangered

**Requirements for Stability:** 

- 4 Populations (PU; found in two Action Areas)
- 50 reproducing individuals in each population (short-lived perennial)
- Threats controlled
- Complete genetic representation in storage

**Description and biology:** *Plantago princeps* var. *princeps* is a woody shrub, which is unusual for the genus. Most continental species in this genus are small herbs. The plant is either single stemmed or sparingly branched at the base, and attains a height range of around one foot to three feet, but is sometimes taller. The leaves are arranged in a cluster at the tip of each branch, are strap-shaped, and measure up to 20 cm (7.8 in) long. Each stem tip usually bears several erect, axillary inflorescences, each of which consists of a single stem bearing densely arranged flowers on its upper portion. The flowers and capsules are small and inconspicuous. The capsules each bear 1-3 black seeds measuring 1.5-2.1 mm (0.06-0.08 in) long.

Flowering and fruiting specimens have been collected throughout the year, and timing varies among different populations. The surface of the seed, once wet, is covered by a mucilaginous membrane (Wagner *et al.* 1990), which is theorized to cause the seeds to stick to animals (Carlquist 1974). It may also potentially aide in germination by maintaining imbibition and providing moisture. With the complete absence of ground mammals in Hawaii prior to the arrival of the Polynesians, birds, including the many now extinct flightless species, would have been the primary dispersal agents of Hawaiian *Plantagos*. Little is known about the breeding system and pollination. The longevity of individuals of this taxon is unknown, but since the plant is a small shrub, its longevity is presumed to be less than 10 years, and it is therefore a short-lived taxon for the purposes of the Makua and Oahu Implementation Plans.





**Figure 1:** Description and *ex situ* Conservation: Fruit, Seeds, Seedlings, Propagation. A) Infructescence with capsules, B) seedlings growing in growth chambers, C) plants growing in the nursery, D) a collection of seeds and capsules depicting mature seeds in the top half of ripe capsules, which are lovingly referred to as 'party hats'.

Table 1. Damas du ativa	Dialary Com	and a mark of Dland		
Table 1: Reproductive	Biology Sun	nmary of <i>Plante</i>	<i>igo princeps</i> vai	: princeps

	Observed Phenology			Reproductive Bi	ology	Seeds*		
Population Unit	Flower	Immature Fruit	Mature Fruit	Breeding System	Suspected Pollinator	Seeds / Infructescence	Dormancy	
Ekahanui	April**- May	March - May	May - June			69		
Halona	May	March – May	March – May	Hammanhuaditia	Wind?	96	Not Dormant	
North Mohiakea	Oct – Dec	Oct – Dec	Oct – Dec	Hermaphroditic	w ma ?	88		
Ohikilolo	March - Oct	Apr – Oct	Apr - Oct			28		

\*There are 1-3 seeds per fruit. Calculations are an average from all collections made in each Population Unit. \*\*Assumptions are that flowering occurs earlier, but observations have not been made every month so we are reporting based on what we have actually seen.

**Known distribution:** *Plantago princeps* var. *princeps* has been recorded from three general areas on the island of Oahu. Most of the currently known plants are scattered throughout cliffs on both the leeward and windward sides of the Waianae Mountains. There are also historical records of it from the southeastern Koolau Mountains in the valleys of Kalihi, Nuuanu, and Manoa. It had not been observed in that region for over half a century. The taxon was then discovered for the first time in the central Koolau Mountains in 2001, when plants were found at Waiawa. These plants are located a short distance to the lee of the Koolau summit ridge. Since then, a population was relocated in Nuuanu, and a large population was found near Konahuanui on the windward side of the summit. Recorded elevations for these plants in the Koolau Mountain range from 480-792 m (1,580-2,600 ft.).

Area	Year	Collector	Pop. Reference Code	Notes
Kalihi	unknown	J. Rock		
Manoa Cliff Trail	1931	H. St. John		Also 1915 J. Rock
Nuuanu Pali	1910	C.N. Forbes	NUU-A?	
Mt. Tantalus	1931	H. St. John		
Palawai	1987	J. Obata	PAL-B	Extirpated (recorded as from
				Napepeiauolelo)

Table 2: Selected Historic Collections of P. princeps var. princeps

### Map removed to protect rare resources. Available upon request

Figure 2: Map 1. Populations of *P. princeps* var. *princeps* on Oahu.

### Map removed to protect rare resources. Available upon request

Figure 3: Map 2. Populations of *P. princeps* var. *princeps* in the Northern Waianae Mountains.

### Map removed to protect rare resources. Available upon request

Figure 4: Map 3. Populations of P. princeps var. princeps in the Southern Waianae Mountains.

**Table 3:** Population Units for *P. princeps* var. *princeps*. Includes Current and Proposed Management Designations for all populations. MFS = Manage for Stability; GS = Manage for Genetic Storage. MMR = Makua Military Reservation; SBW = Schofield Barracks West Range. See Population Structure and Management Discussion sections below for discussion on proposed changes. \*Dependent on population surveys, these PU may swap designation over the next 5 years.

Population Unit	Current Management Designation	Proposed Management Designation	Action Area	Management Unit (MU)
	<u> </u>	<u> </u>		
Ekahanui	MFS	MFS	None	Ekahanui
Halona	MFS	MFS*	None	Palikea IV
North Mohiakea	MFS	MFS	SBW	Lihue
Ohikilolo	MFS	MFS	MMR	Ohikilolo
Konahuanui	No Management	GS*	None	Iolekaa to Kamooalii No MU
North Palawai	GS	GS	None	Palawai No MU
Nuuanu	No Management	GS	None	Honolulu No MU
Pahole	GS	GS	MMR	Pahole
Waiawa	No Management	GS	None	Waiawa No MU
Waieli (introduction)	GS	GS	None	Kaluaa and Waieli

**Habitat:** *Plantago princeps* var. *princeps* occurs in two extremely different types of habitat. In the Waianae Mountains the plants are found in the mesic vegetation on cliff faces, cliff ledges, and at the bases of cliffs. The majority of these plants are accessible only via rappelling. At one time, this cliff habitat was vegetated with native grasses, sedges, herbs, and shrubs, but is increasingly dominated by alien species. The southeastern Koolau Mountain Range plants grow in mesic to wet cliff habitats. The Konahuanui population, however, is mostly wet cliffs and wet forest. The Waiawa plants are situated in a wet forest area close to the Koolau summit ridge and were observed growing on a streamside embankment (Perlman pers. comm. 2000). These Koolau Mountain habitats are also becoming dominated by weeds.

**Table 4:** Habitat Characteristics by Population Unit. Commas separate information by Population Reference Site. An asterisk (\*) indicates the Koolau Mountain Population Units. Average Annual Rainfall data is from the Rainfall Atlas of Hawaii (Giambelluca et al. 2013). All other data from OANRP observations.

Population Unit	Population Reference Codes	Elev. (ft.)	Slope	Canopy Cover	Topography	Aspect	Average Annual Rainfall (mm)
		Manag	e for Stabili	ty Population U	Inits		
Ekahanui	EKA-A, B, C, D (reintro <sup>¥</sup> )	2520, 2631 <sup>¥</sup>	Steep – Vertical	Intermediate	Upper Slope	NE	1217
Halona	HAL-A	2408 - 2674	Steep – Vertical	Intermediate	Upper Slope	NW	1155
Mohiakea	SBW-A	3045-3050	Steep – Vertical	Intermediate – Open	Upper Slope	N	1460
Ohikilolo	MMR-A, B	2620, 2870	Vertical, Steep	Intermediate, Open	Upper Slope	N, N- NW	1700, 1527
	·	Gen	etic Storage	<b>Population Uni</b>	its	9	
Konahuanui* (proposed)	NUU-A	1600	Steep - Vertical	Open	Mid-slope	N-NE	2258
North Palawai	PAL-A, B	2600, 2664	Moderate – Steep, Vertical	Intermediate	Mid Slope	NW, N	1158
Nuuanu*	NUU-B	1719	Moderate – Steep, Vertical	Intermediate	Mid Slope	Ν	3184
Pahole	PAH-A	2000	Steep – Vertical	Intermediate	Upper Slope	Ν	1425
Waiawa*	AWA-A	2060	Steep – Vertical	Partial – Full Sun	Gulch Bottom	N	4322
Waieli (introduction)	ELI-A	2726	Steep	Intermediate	Upper Slope	NE-E	1204

**Table 5:** List of Associated Species (six letter code = first three letters of genus, followed by first three letters of species) for each Population Unit for both canopy and understory. Some outplanting sites have yet to have the associated species recorded. Species observed by OANRP staff are listed in alphabetical order; introduced taxa precede native taxa and are underlined: <u>AbuGra, CycPar</u>

undernined. <u>11</u>	<u>buGra, CycPar</u>		
Population Unit	Population Reference Codes	Canopy	Understory
Ekahanui	EKA-A, B, C	<u>PsiCat, PsiGua</u> , <u>SchTer</u> , Antpla, ChrFor, CopFol, DioHil, DubLax, MetPol, Myrlan, Myrles, PsyMar, SopChr, Zankau	<u>Ageade, AgeRip, BleApp, MelMin, NepBro, OplHir,</u> <u>PasSub, SchTer, SonOle, UnkSpp</u> , AlySte, AspKau, AspUnis, BidTor, CarMey, CarWah, CocOrb, CypHilHil, DiaSan, EraGra, KadAcu, KadCor, LepThu, Lyshil, MicSpe, MicStr, PepTet, PlePar, RumAlb, VioChaTra
Halona	HAL-A	LanCam, MorFay, SchTer	<u>AgeAde, AgeRip, MelMin, PasSub</u> , CarMey, CarWah, EraGra, KadAcu, KadCor, PepTet, PlePar, RumAlb
Mohiakea	SBW-A	<u>PsiCat, SchTer, UnkSpp</u> , CopLon, DubLax, IleAno, LabTin, LepTam, MetPol, MetTre, PerSan, PipAlb, PitFlo, VacCal	AgeRip, BleApp, CliHir, CycPar, EriKar, KalPin, PasCon, RubArg, RubRos, VerLit, ArtAus, BidTor, BoeGra, CarMey, CarWah, DiaSan, DryUni, DubPla, EraGra, EraVar, LysHil, LytMar, MacAng, MetPol, MetRug, RumSpp, UnkSpp, VacCal,
Ohikilolo	MMR-A	<u>GreRob</u> , <u>SchTer,</u> MetPol, MetTre, MyrLes, PsyHat, SopChr	<u>AgeAde, AgeRip</u> , <u>BleApp</u> , <u>CupCar</u> , <u>FesBro</u> , <u>KalPin</u> , <u>ThuEre</u> , BidTor, CarMey, ElaPal, EupMul, KadAcu, KadCor, LysHil, MelTenf, PsyHat, PteAqu, SphChi,
Konahuanui	NUU-A	<u>CitCau</u> , MetPol, MetTre, PipAlb	<u>AgeAde, CliHir, EriKar, MelMin, OplHir, HedGar, SpaCam,</u> EraGra, MacAng, SetPar,
North Palawai	PAL-A, B	<u>PasSub, PsiCat, SchTer</u> DodVis, MetPol, PitCon, PitFlo, PsyHat,	<u>AgeAde, AgeRip, BleApp, PasSub, SchTer, UnkSpp,</u> AlySte, DubPla, EupCel, KadAff, Lyshil, RumAlb, VioChaTra
Nuuanu	NUU-B	No Data Available	
Pahole	PAH-A	<u>SchTer,</u> AlySte, BidTor, IleAno, MetPol, ScaGaua,	<u>AgeRip</u> , <u>CocGra</u> , <u>SchTer</u> , AlySte, BidTor, CarMey, CopFol, DicLin, Dodvis, KadAff, KadCor, KadDegDeg, LysHil, MetPol, MicStr, NepExaHaw, OdoChi, PsyMar, VacRet
Waiawa	AWA-A	<u>CliHir</u> , BroArg, CibCha, DicLin, DubPla, IsaDis, MetPol, PolOah, PriMar, PsySpp, SadSpp, ScaGaua, SyzSan, TreMac, UnkSpp, VacRet, WikOahOah	<u>CliHir</u> , <u>SacInd</u> , BidMac, MacAng, SelArb
Waieli	ELI-A (introduction)	<u>SchTer, TooCil,</u> CanGal, CibCha, CorFru, DioSan, FreArb, GreRob, IleAno, LabKaa, MetPol, MyrLes, PasEdu, PerSan, PipAlb, PisUmb, PitSpp, PlaSan, PsiCat, PsyMar,	<u>BleApp. BudAsi</u> , <u>CliHir</u> , <u>ConBon</u> , <u>CraCre</u> , <u>CycPar</u> , <u>EriKar</u> , <u>KalPin</u> , <u>LanCam</u> , <u>MelMin</u> , <u>OplHir</u> , <u>PasCon</u> , <u>PasSub</u> , CarMey, CarWah, CibCha, CopFol, CopLon, CorFru, DiaSan, EupMul, FreArb, KadAff, KadCor, LabSpp, LobYuc, MetPol, MicStr, MyrSpp, NepSpp, PhyDis, PipAlb,

### Photographs by Population Unit Waianae Mountains



**Figure 5:** Photographs from Ekahanui PU: A) Mature plant with fruit, B) Large, multi-headed plants (now mostly dead), C) Ripe infructescences, D) Immature, E) Habitat, F) *Achatinella mustelina* (kahuli tree snail) on *P. princeps* 



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**Figure 6:** Halona. A) Mature plant and Habitat B) Mature plant and habitat; C) female stage flowers; D) reproducing plant and habitat

Figure 7: Pahole. A) Mature plant, B) Habitat



Figure 8: Palawai. Mature plant at Palawai, PAL-A.2016 Makua and Oahu Implementation Plan Status Report



**Figure 9**: Mohiakea (Puu Kalena). A) Mature plant with view, B) Mature Plant with infructescence, C) Flowering plants, D) Habitat and *Dubautia plantaginea* comparison.

Koolau Population Units (Nuuanu and Konahuanui; no photographs from Waiawa)





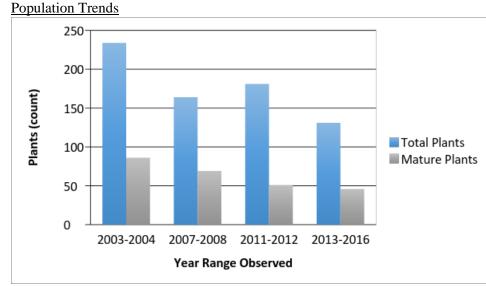
**Figure 10**: A) Nuuanu (NUU-B), B) Konahuanui (NUU-A) mature plants with ripe infructescences, C) immature plant at Konahuanui, D) habitat at Konahuanui.

**Taxonomic background:** *Plantago princeps* is endemic to the Hawaiian Islands. The species is divided into four varieties: var. *anomala* of Kauai; var. *laxiflora* of Molokai, Maui, and Hawaii; var. *longibracteata* of Kauai and the Koolau Mountains of Oahu; and var. *princeps* of both mountain ranges on Oahu. All of the varieties except var. *longibracteata* are sizable woody shrubs. In contrast, var. *longibracteata* is a small herb. When the Waianae Range plants were rediscovered in 1987, the specimens collected were identified as var. *anomala*. Only the southeastern Koolau Range plants were considered to represent var. *princeps* (Wagner *et al.* 1990). The Waianae Range plants were later reclassified as var. *princeps* (Wagner *et al.* 1999). There have been no subsequent vouchers taken from any Waianae Mountain populations. Due to this taxonomic history, vouchers would be useful from Population Units that have never been vouchered.

In 2008, a study of the molecular variation and adaptive radiation of the Hawaiian *Plantago* was published as part of a PhD dissertation at the University of Hawaii (Dunbar-Co 2008; Dunbar-Co, Wieczorek, Morden 2008). Only the Ekahanui and Waiawa populations were included in this study, which showed genetic separation from each other based on microsatellite data. While this variation is meaningful and could also suggest cryptic species, it is not enough to separate them into separate taxa, particularly in light of their morphological similarities. Phylogenetic analyses did not separate the Koolau and Waianae populations, and suggests a single dispersal event to Oahu for *P. princeps* (Stephanie Dunbar-Co, personal communication). It would however be useful to sample more populations to determine if indeed there is enough genetic separation to define into separate taxa. Additionally, there is taxonomic uncertainty regarding varieties of *P. princeps* will be considered var. *princeps*.

**Population Structure and Trends:** During the finalization of the Makua Implementation Plan, population trend data was only available for the population in the North branch of North Palawai gulch (PAL-B). When this site was discovered in 1987 by John Obata, there were approximately 20 plants. By 2003 there were only 5 plants. This site lost its last plant by 2011. In this case, the rapid decline was attributed to competition from daisy fleabane (*Erigeron karvinskianus*), a highly invasive alien plant. Currently, all populations in the Waianae Mountains are in decline and all have *E. karvinskianus* is now present at all of the sites.

Population structure for this taxon is weak, but not non-existent (Fig.11). OANRP staff have observed seedlings at eight different Population Reference Sites, in seven Population Units (PU). There were seedlings at the Waieli introduction in 2012, and have since grown into immature plants, of which eight remain (2016). The current high number of immature plants in the Waianae Mountains is primarily due to the Ekahanui population, where 90% of the total plants are immature. At these sites, there has been a large decline in mature individuals (from 46 plants in 2014 to 7 plants in 2016), but the number of small immature plants has been greater than 50 for the last six years. Unfortunately, these small immature plants (around 2-4 leaves and a couple centimeters in height) fail to transition into large immatures. It is hypothesized that they succumb to downy mildew (Peronosporaceae) or possibly slugs (see Threats section below). Substantial declines in the number of mature plants at both the Halona and Ohikilolo PUs have also been observed over the last couple years.



**Figure 11:** A) Overall counts of total plants and mature plants only for all monitored Waianae Mountain Population Units, excluding reintroductions.

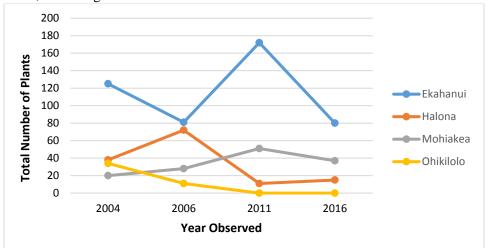


Figure 11: B) Counts for the total number of plants at the four current MFS PU (Waianae Mountains; excluding reintroductions).

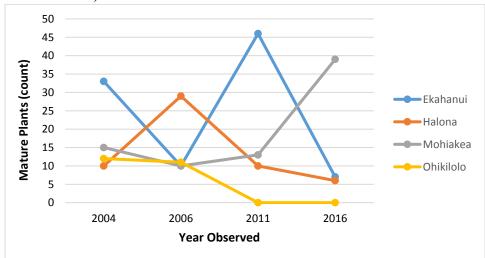


Figure 11: C) Counts of mature plants for the current four MFS PU (Waianae Mountains; excluding reintroductions).

The Koolau Mountain populations have only been monitored once by OANRP, and therefore no population trends are available for these sites. The last two seedlings at the Nuuanu (NUU-B) population were dug up and brought into cultivation to Lyon Arboretum by the Oahu Plant Extinction Prevention Program (OPEPP) since the last OANRP survey. They have since maintained representation of those two plants in cultivation, as well as have propagated and reintroduced several individuals into the Manoa Cliffs Trail Restoration Site from this collection. OPEPP also attempted to relocate the Waiawa population twice, but was unsuccessful at finding any plants. OANRP and OPEPP should conduct a thorough survey in this area one more time before establishing that this population no longer exists. The Konahuanui population needs to be re-monitored as it will be ten years since the last census, especially to verify that it is the largest population of this taxon.

**Table 6:** Summary of Population Structure of Koolau Populations of *P. princeps* var. *princeps*. Observations in 2016 conducted by OPEPP.

Population Reference Site	Year Observed	Total # of Plants	Mature Plants	Year Observed	Total # of Plants
Waiawa, AWA-A	2003	83	16	2016	0?
Konahuanui, NUU-A	2006	110	100	2016	No update
Nuuanu, NUU-B	2007	12	4	2016	0
ALL KOOLAU SITES		205	120		?

**Current status:** The known population units of *P. princeps* var. *princeps* in the Waianae Mountains total approximately 131 plants, consisting of mature and immature plants. All of the wild plants from Ohikilolo have died, and last year 57 individuals were reintroduced back into Ohikilolo at a new location. Four of these individuals matured quickly after they were planted. In 2003 there were about 35 mature plants in the Makua action area, and now, with the exception of the new Ohikilolo reintroduction, there are only nine (all located in the Pahole Natural Area Reserve). The population in the Schofield action area, however, is the only population in the Waianae Mountains not observed to be in decline. This is mostly due to the discovery of more plants in a new area adjacent to the original population site off of Puu Kalena.

### **STABILIZATION EFFORTS**

The following section uses the above information, plus additional information we have learned about this taxon, to determine appropriate stabilization efforts for the next five years (July 2016 – June 2021). The following actions are requirements for stabilization:

- 4 Populations (PU)
- 50 reproducing individuals in each population (short-lived perennial)
- Threats controlled
- Complete genetic representation in storage

**Population Units:** Four Manage for Stability Population Units (MFS PU) are required for this taxon as it is in both the Makua and Schofield Action Areas. Due to the decline in this taxon, all other populations that are not MFS PU will become Manage for Genetic Storage Population Units. Representation of these populations will be maintained and depending on how taxonomic questions are addressed and future outplanting needs, these populations may be incorporated into mixed-source reintroductions.

We also propose to carefully monitor and survey the Konahuanui PU while securing collections. In the future, it might be necessary to change one of the existing Waianae Mountain MFS PUs to a Genetic Storage PU and begin to manage the Konahuanui PU as an MFS PU. The Waianae Mountain MFS PU that would be swapped is most likely be either Halona or Ohikilolo, depending on where the next reintroduction could be located for these source populations, the success of the Ohikilolo outplanting, and the population size of the current MFS PUs.

**Outplanting considerations and plan:** *Plantago princeps* var. *princeps* is the only native *Plantago* in the Waianae Mountains. The situation is more complex in the Koolau Mountains, where, in addition to var. *princeps*, there is another

variety of *P. princeps* recorded, var. *longibracteata*. This variety is known from historical specimens collected on the windward side of the Koolau Mountains in the Kaluanui area between Punaluu Valley and Hauula. It has been recorded on wet cliffs and alongside waterfalls, but are currently known from Oahu. Additionally, there is a second native species in the Koolaus, *P. pachyphylla*, which is common in the Koolau summit areas. On Kauai, *P. princeps* var. *longibracteata* and *P. pachyphylla* form a hybrid population at the Waialeale summit (Bruegmann pers. comm. 2000). It is not yet known whether the ranges of *P. pachyphylla* or *P. princeps* var. *longibracteata* overlap that of *P. princeps* var. *princeps* in the Koolau Mountain forests, and whether any hybridization occurs or could potentially occur. No OANRP outplantings have occurred in the Koolaus, but if they are deemed necessary in the future, further study should be conducted on the distribution of *Plantago* taxa in the Koolau Mountain Range, and their potential for hybridization.

Given the extreme differences between the habitats of the Waianae and Koolau Mountain Range plants, it may not be prudent to mix the two stocks at a single outplanting site. However, depending on aspects such as the results of the molecular study, the decline of the Waianae Mountain populations, and the known and potential variation of impact of downy mildew on populations, this may need to be revisited in the future.

There have been three outplantings of *P. princeps* var. *princeps*: Waieli, above the Puu Hapapa shelf (introduction), Ekahanui (augmentation), and Ohikilolo (reintroduction). The Hapapa introduction is the oldest planting, initiated in 2007, with additional outplantings in 2009 and 2012. Survival at this site is 45%. The original plants from 2007 have mostly died, but very few of the plants from the 2009 planting have died, and now there are eight immature plants recruiting under one plant. The location of this cohort, as well as the site management (*i.e.* weed control and other threat control) could be helpful in determining suitable locations for planting in the future. Unfortunately, OANRP does not feel confident that there is more suitable habitat at Hapapa to expand this outplanting. The Ekahanui augmentation has completely failed. Eighty-nine plants were planted between 2014 and 2015. All outplants have succumbed to downy mildew, except for three plants observed in 2016; these three were all in poor health. The Ohikilolo reintroduction was initiated in 2016. Four of the 57 plants have matured. Several show signs and symptoms of downy mildew. However, in general, the plants look healthy and are growing. Propagule collections will be needed from these plants to produce additional outplants for this site and secure seeds for genetic storage.

Differences in the success of the reintroductions may not only be due to the location, but also the source population. The source population for the Waieli population is Mohiakea, which is the only wild population not in decline and anecdotally the stock least susceptible to downy mildew. On the other hand, the failed Ekahanui augmentation is from Ekahanui stock, which is the most susceptible to downy mildew in our nursery facilities. Downy mildew is one of the hypothesized leading causes of death at the Ekahanaui, and possibly Halona, populations. See the threats section for further discussion. Conducting a controlled breeding study in the nursery may be one approach to improving outplanting success. If the more downy mildew-susceptible maternal lines (source populations) are mixed with less downy mildew-susceptible maternal lines, more genetically diverse progeny may be available for reintroduction. Concerns for outbreeding depression would need to be addressed, as well as possible locations for mixed-source outplantings.

### Hapapa Introduction



Figure 12: A) a flowering plant, B) a recruited immature plant, C) several recruiting immature plants.

### Reintroduction Plan

The proposed outplanting sites for the Waianae Mountain Population Units are designed to meet the stability goal for the number of reproducing individuals, as currently none meet this goal. We plan to wait a year to see how the new Ohikilolo reintroduction performs before we finish planting into this site. We recognize that the Ekahanui augmentation will need to be replaced, but at this time we would like to discuss options and develop a plan to address downy mildew before we proceed with another planting of this stock (see Threats section below). Both the Halona and Mohiakea PUs will need outplantings to raise the number of reproducing plants to meet that stability goal. As the Mohiakea stock appears to be the

healthiest, we should pursue site selection and proceed with a single source outplanting in this PU. However, this stock may also need to be incorporated into outplantings at other PUs (see Threats and Management Discussion below). Determination of how to proceed with outplantings in the Ekahanui and Halona PUs due to the impact of downy mildew will delay these plantings. The Mohiakea population is the highest in elevation (so potentially cooler), but in a similar rainfall range with the majority of sites. It will also be important to determine the impact of drought on the ability for a plant to survive downy mildew, and choose outplanting sites accordingly.

We have proposed an outplanting for all Koolau stock, but understand at the present time that surveys are needed to determine if outplanting is necessary. The most recent estimates of population size, though somewhat dated (2007), indicate that there may be enough reproducing individuals at Konahuanui to meet stabilization goals.

**Table 7:** Current and Proposed Outplantings of *P. princeps* var. *princeps* to meet stabilization goal of 50 reproducing individuals per Population Unit (PU). The propagule type for each planting will be immature plants grown from seeds collected from wild or outplanted plants. The estimate of the number of mature plants at Konahuanui is from 2007. An asterisk (\*) indicates outplantings that have not yet been initiated. Note: We know how many mature plants are currently at population reference sites, but we recognize that the number of actively reproducing individuals (a requirement for stability) would likely be lower than the total number of mature plants.

Population Unit	Reintroduction Site(s)	Number of Plants to Outplant	Existing Mature Plants in PU	Propagule Population(s) Source	Number of Founders in Source Population.	Plant Size	Pot Size
Ekahanui	EKA-D EKA-E*	89 200	9	EKA-A, B, C (SBW-A?)	42-50		
Halona	HAL-B*	200	6	HAL-A (SBW-A?)	18-25	~20-40	4"-6"
North Mohiakea	ELI-A SBW-B*	100 200	39	SBW-A SBW-A	19-30 19-30	cm	round
Ohikilolo	MMR-B	200	4	MMR-A	12		
Konahuanui	Koolau*	TBD	TBD	ALL Koolau	ALL Koolau		

**Threats:** The primary threats to *P. princeps* var. *princeps* that were known at the time the Makua Implementation Plan was finalized (2003) included feral pigs and goats (though few goats are known in the Koolau Mountains). Feral pigs are negatively impacting the habitat in Halona, by eroding and degrading the ridge above the cliffs that the plants are found on. This disturbance includes additional weeds, rock falls, and trampling. Fencing this area to protect this cliff habitat is necessary, but may not be feasible given the terrain. OANRP will visit to determine if fencing could reduce ungulate impact. The U.S. Navy may also pursue plans to fence Halona in its entirety. If these fencing plans do not come to fruition, OANRP will work with the State of Hawaii to determine other actions to reduce ungulate presence on these ridges, such as snaring.

Landslides are a secondary threat due to the nature of cliff habitat in higher rainfall areas. A large landslide occurred below the Halona population and small slumping events also impacted the Ekahanui population.

Various alien plant species threaten *P. princeps* var. *princeps* by altering its habitat and competing with it for sunlight, moisture, nutrients, and growing space. Also, the spread of highly flammable alien grasses increases the incidence and destructiveness of wildfires. There has been little weed control around these populations as they are primarily found on vertical slopes that require rappelling to access. A review of the weeding needs should be conducted to highlight where restoration work (weeding and common native seed sow) could improve these cliff habitats.

Predation of plants and fruits by rodents and slugs has been documented, and it can be assumed that they potentially have an influence on population stability. Rats have eaten plants in Ekahanui, and slugs have been seen on seedlings. We will continue to assess how these threats are impacting population stability as we monitor the populations.

There are additional threats to this taxon that were not described in the Makua Implementation Plan. These include climate change and downy mildew. Climate change has been anecdotally described as an impact to these cliff habitats, in conjunction with weeds, as populations have gotten drier and weedier over time in consecutive visits. This is despite a very low Climate Change Vulnerability Score of 0.28 (Fortini et al. 2013; 0 = not vulnerable to climate change, 1 =extremely vulnerable), which is likely due to the mid elevation wet forests location of the Koolau Mountain populations. Downy mildew, first observed when this taxon was propagated in the nursery, has since been seen at several wild populations and at outplantings. Downy mildew has been seen at all of the outplantings, and is assumed to be the leading cause of death in the Ekahanui augmentation. Downy mildew has also been seen at the wild populations in Ekahanui, Halona, and may have possibly been the cause of the extirpation of the Ohikilolo population. There appears to be variation among populations in susceptibility to downy mildew. In the nursery, the Ekahanui stock appears most susceptible, typically dying once infected. The Mohiakea stock is the least susceptible, and the Ohikilolo stock moderately susceptible. Both the Palawai and Halona stock has yet to be propagated *ex situ*. In the nursery, and typically during cooler rainy winter months, if plants show signs of downy mildew, they typically die within two weeks, depending on the stock. Treating them with fungicide after symptoms appear will typically only prevent a small percentage of them from dying. Plants kept on the mist bench have not shown symptoms, even without preventative fungicides. However, certain stocks have stayed off the mist bench and have not shown symptoms, again without preventative fungicides. More propagation is needed to determine best practices. However, preventative fungicides and leaving plants on the mist bench should be further explored as ways to prevent death by downy mildew in the nursery. It is uncertain at this time if and why misting of plants reduces the downy mildew symptoms, as this has also been observed to possible reduce symptoms of other mildews. This should be explored further. Currently, there are no techniques to treat plants that show signs of downy mildew in the field. It is believed that the mildew is always present, but it is only when it appears heavily on the leaves that it becomes virulent. OANRP will submit leaf samples to Dr. Anthony Amend at the University of Hawaii Botany Department in attempt to identify the downy mildew.

Progress on threat control efforts are summarized below (Table 8).



Figure 13: Downy mildew on wild plants at Ekahanui.

	PU Stability T	MU Threat Control					Genetic Storage		
Population Units	Has the Stability Target for mature plants been met?	Does population structure support long- term population stability?	C	Weeds	Rodents	Fire	Slug	Downy Mildew	Are Genetic Storage goals met?
Manage for Stabi	lity Population U	Units							
Ekahanui	No	No	Yes	Partial (0%)	Yes	No	No	No	No
Halona	No	No	No	No	No	No	No	No	No
North Mohiakea	No	No	Yes	No	No	No	No	No	No
Ohikilolo	No	No	Yes	Yes	No	No	No	No	No
Konahuanui	Yes*	No	No	No	No	No	No	No	No
Genetic Storage I	Population Units	(Waianae Mounta	uins)						
North Palawai	No	No	No	No	No	No	No	No	No
Pahole	No	No	Yes	No	No	No	No	No	No
Waieli	No	No	Yes	Yes	No	No	No	No	No
Genetic Storage I	Population Units	(Koolau Mountain	ns)						
Nuuanu	No	No	No	No	No	No	No	No	No
Waiawa	No	No	No	No	No	No	No	No	No

**Table 8:** Progress on threat control efforts for *P. princeps* var. *princeps*. \* indicates OANRP uncertainty in reaching this goal due to the lack of current census data (based on population estimates from 2007).

Grey Shading = threat to taxon within Population Unit. No shading = absence of threat to taxon. Ungulate Managed = culmination of cattle, goats, and pig threats. Yes = All Population Reference Sites within Population Units have threat controlled. No = All Population Reference Sites within Population Units have no threat control. Partial% = percent of mature plants in Population Unit that have threat controlled. Partial 100% = all Population Reference Sites within Population Units have the threat partially controlled. Partial 0% = Threat partially controlled, but not around any mature plants.

### Genetic Storage Plan

**Table 9:** Action plan for how to maintain genetic storage representation, and provide propagules for reintroduction, for *P. princeps* var. *princeps*.

is used to meet	What is the source for the propagules?	What is the Genetic Storage Method used to meet the goal?	What is the proposed re- collection interval for seed storage?	Is seed storage testing ongoing?	Plan for maintaining genetic storage
Seeds	<i>in situ &amp;</i> outplantings	Collecting infructescences	15 years	Yes	Collect seeds and maintain reintroductions for re- collecting

- It will be important to act quickly to collect from as many wild plants as possible while they persist given the rapid decline observed.
- Seeds need to be collected when they are dry. If it has been very wet weather just prior to a planned collection time, the collection should be reconsidered. Seeds that have been enclosed in their mucilaginous coat for several days may have imbibed enough water to initiate germination. Seeds that have begun to germinate cannot be stored in long term seed banking.

<u>Management Highlights</u>: The following key actions, in conjunction with the timeline below (Table 10), highlight the management direction for *P. princeps* var. *princeps* over the next five years:

- Pursue researchers to resolve taxonomic issues, including vouchers and molecular studies
- Pursue an *ex situ* controlled breeding study to determine if:
  - breeding plants from other populations with stock from Mohiakea produces offspring that are less susceptible to downy mildew
  - o hybridization with other *Plantago* taxa is a concern (in conjunction with molecular studies)
  - o Pursue researchers or staff to conduct studies
- Use results from the controlled breeding study and *in situ* monitoring to finalize timeline, stock, and locations for the next Waianae reintroductions
- Monitor and collect from Koolau Mountain populations, determine appropriate and feasible threat control needs and whether or not a reintroduction is needed
- Monitor and collect from Waieli and Ohikilolo reintroductions
- Revise Management Designations for populations as described above:
  - o Change all No Management PUs to Manage for Genetic Storage
  - After 2017 surveys and monitoring, decide if to designate the Konahuanui PU as Manage for Stability and to change one (and which one) Waianae Manage for Stability PU to Manage for Genetic Storage.
- Evaluate the ungulate impact and threat control Halona PU
- Evaluate the need, technique, and capacity for restoration of the cliff habitats to combat weeds and the effects of climate change
- Submit samples of infected material to Dr. Anthony Amend at the University of Hawaii Botany Department to identify the downy mildew
- Coordinate with OPEPP and Lyon Arboretum regarding propagules of new Manage for Genetic Storage Populations
- Coordinate with OPEPP regarding management actions for Koolau Population Units, as they have worked on these in the past.

Proposed A	Proposed Actions for the following years:						
PU	July 2016 – June 2017	July 2017 – June 2018	July 2018 – June 2019	July 2019 - June 2020	July 2020- June 2021		
Ekahanui	<ul> <li>Monitor</li> <li>Collect</li> <li>Breeding Study</li> </ul>	<ul> <li>Monitor</li> <li>Collect</li> <li>Breeding Study</li> </ul>	<ul><li>Monitor</li><li>Collect</li><li>Threat Control</li></ul>	<ul><li>Monitor</li><li>Collect</li><li>Outplant?</li></ul>	<ul> <li>Monitor</li> <li>Collect</li> <li>Outplant</li> <li>Threat Control</li> </ul>		
Halona	Assess threat control (fence?)	<ul> <li>Monitor</li> <li>Collect</li> <li>Implement additional threat control</li> </ul>	<ul><li>Monitor</li><li>Collect</li></ul>	<ul><li>Outplant?</li><li>Monitor</li><li>Collect</li></ul>	<ul><li>Monitor</li><li>Collect</li><li>Outplant?</li></ul>		
North Mohiakea	<ul> <li>Monitor</li> <li>Collect</li> <li>Evaluate Threat Control Actions</li> </ul>	<ul> <li>Monitor</li> <li>Collect</li> <li>Scope Outplanting site(s)</li> </ul>	<ul> <li>Monitor</li> <li>Collect</li> <li>Implement additional threat control if needed</li> </ul>	<ul><li>Monitor</li><li>Collect</li><li>Outplant</li></ul>	<ul><li>Monitor</li><li>Collect</li><li>Outplant</li></ul>		
Ohikilolo	<ul> <li>Monitor</li> <li>Collect</li> <li>Determine Threat Control Actions</li> </ul>	<ul> <li>Monitor</li> <li>Collect</li> <li>Implement Threat Control</li> </ul>	<ul> <li>Monitor</li> <li>Collect</li> <li>Implement Threat Control</li> <li>Outplant</li> </ul>	<ul> <li>Monitor</li> <li>Collect</li> <li>Implement Threat Control</li> </ul>	<ul> <li>Monitor</li> <li>Collect</li> <li>Implement Threat Control</li> <li>Outplant</li> </ul>		
Konahuanui		<ul><li>Monitor</li><li>Collect</li></ul>	Determine     Threat Control     Actions	<ul><li>Monitor</li><li>Collect</li></ul>	• Determine if need to scope an outplanting site		
Palawai	<ul><li>Monitor</li><li>Collect</li></ul>	<ul><li>Monitor</li><li>Collect</li></ul>	<ul><li>Monitor</li><li>Collect</li></ul>	<ul><li>Monitor</li><li>Collect</li></ul>	<ul><li>Monitor</li><li>Collect</li></ul>		
Nuuanu	• Survey	<ul><li>Monitor</li><li>Collect</li></ul>		<ul><li>Monitor</li><li>Collect</li></ul>			
Waiawa	<ul><li>Survey</li><li>Monitor</li><li>Collect</li></ul>		<ul><li>Monitor</li><li>Collect</li></ul>		<ul><li>Monitor</li><li>Collect</li></ul>		

### Table 10: Notes for key actions for Manage for Stability Population Units (MFS PU).

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### Cyanea superba subsp. superba

- Scientific name: Cyanea superba (Cham.) A. Gray subsp. superba
- Hawaiian name: Haha, ohawai
- Family: Campanulaceae (Bellflower family)
- Federal status: Listed endangered September 11, 1991
- MIP Requirements for Stability
- 4 Population Units (PU) (extirpated in the wild)
- 50 reproducing individuals in each PU (long-lived perennial with a history of precipitous decline, extremely low genetic variability)
- Stable population structure
- Threats controlled
- Complete genetic representation of all PUs in storage
- **Description and biology:** *Cyanea superba* subsp. *superba* (here after *C. superba*) is a tree 4-6 m tall with a single major stem, or occasionally two or more major stems arising from the base of the plant. Two of the basal-branching plants formerly growing at Kahanahaiki each had about 8-10 major stems (Lau pers. comm. 2000). The taxon's leaves measure 0.5-1.0 m long, and are clustered at the stem tips. The inflorescences hang below the leaves, and terminate in a cluster of 5-15 flowers. The corollas are whitish to cream, curved, and measure 5.5-8.8 cm long. The berries are yellow to orange, egg-shaped, and measure 16-22 mm long (excerpt from MIT 2003).
- This taxon flowers from September through October. It was probably originally pollinated by nectar-feeding birds, as is thought for *Cyanea* species in general, with their long tubular flowers. *C. superba* is capable of self-pollination and can be autogamous, as evidenced by the production of fertile seeds in the Kahanahaiki population unit in years when only a single plant had flowered (Pender et al. 2013). Fruit-eating birds presumably dispersed the seeds. Based on growth rates and the size of mature plants, *C. superba* may live for up to 20 years or more (Lau pers. comm. 2000).

modified from: Makua Implementation Team (MIT). 2003.

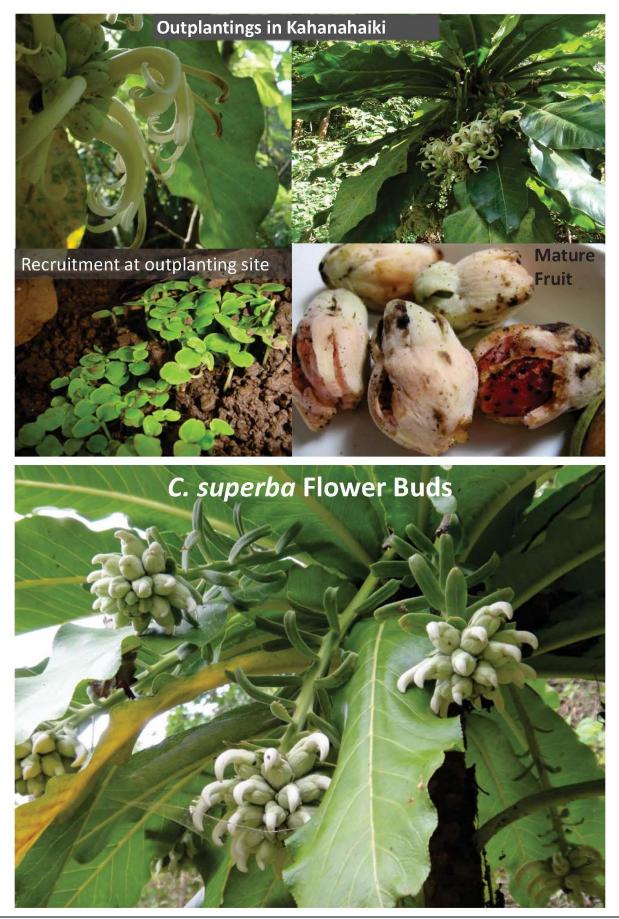
# **Reproductive Biology Table**

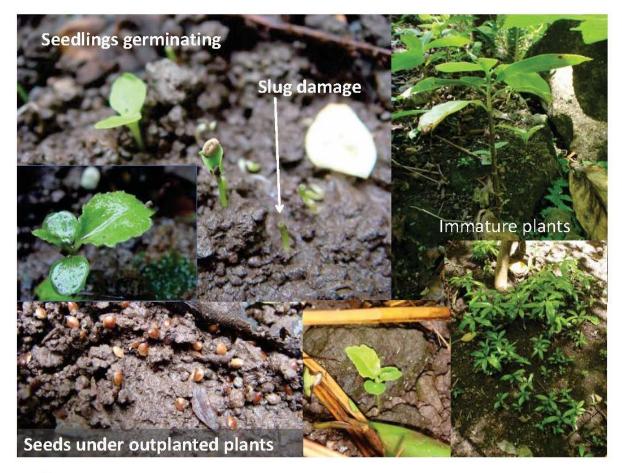
	Observed Phenology			Reproduct	Reproductive Biology		Seeds	
Population Unit	Flower	lmmature Fruit	Mature Fruit	Breeding System	Suspected Pollinator	Average # Per Fruit	Dormancy	
ALL	Sept-Oct	Oct-Jan	Oct-Jan	Hermaph- roditic	Bird*	112 ± 80	Not Dormant	

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# Cyanea superba subsp. superba

- **Known distribution:** The few documented locations for *C. superba* are all in the northern Waianae Mountains. These locations are the eastern slope of Mt. Kaala, Makaleha Valley, Pahole Gulch, and Kahanahaiki Valley. After the original collections prior to 1870, no plants were known until its rediscovery in 1971 in Pahole. The Kahanahaiki site was discovered in 1987. By 1991, a total of less than 20 plants were known from Pahole and Kahanahaiki. The Pahole plants were gone by 1994 and the last Kahanahaiki plant died in 2002.
- Habitat: The historic sites in Kahanahaiki and Pahole, are on the lower to upper gulch slopes. These slopes are fairly steep. The vegetation at these sites consists of mesic forest comprised of a mix of various native and alien tree species.
- **Taxonomic background:** *Cyanea superba* is endemic to Oahu. It is comprised of two subspecies: subsp. *superba* of the northern Waianaes, and subsp. *regina* of the southeastern Koolau Mountains. *Cyanea superba* subsp. *regina* was last recorded in 1960. In 1913, Joseph Rock wrote in <u>The Indigenous Trees of the Hawaiian Islands</u>, "The queen of all is the lobeliaceous *Cyanea superba* var. *regina*, an exceedingly beautiful plant found only on Oahu, in the gulches of Wailupe and Niu, and in Makaleha of the Kaala range."
- **Population trends:** Populations of *C. superba* subsp. *superba* have plummeted over the last three decades. The decline of the Pahole population was especially steep. The population was discovered in the 1970's. In 1978, 36 mature plants, 10 saplings, and six seedlings were reported. By 1989 the number had declined to 10-12 plants. The site was then fenced to protect the plants from feral pigs. Despite of the protection offered by the fence, the last Pahole plant died in 1994. The last wild tree in Kahanahaiki died in 2002. It is possible that the last wild plants died of a fungal disease but we do not know this with any certainty, or if they died of another cause, including old age, and the fungal infection was secondary. We have noticed tip wilt and possible fungal infections in some of the outplanted plants to date but do not understand if it is the same cause of death. This tip wilt and consequent death has also been seen in *C. grimesiana* subsp. *obatae*.

modified from: Makua Implementation Team (MIT). 2003.

# Selected Historic Collections of *C. superba* subsp. *superba*

Area	Year	Collector	Population Unit & Notes		
Mt. Kaala 01 Mar 1870		Hillebrand, W.B.	"East Slope of Mt. Kaala" (Schofield)		
Pahole Gulch	21 Mar 1971	Yamashita, G. Montgomery, S.L. Obata, J. Carson, H. Carson, Mrs. H.	Wild Pahole Gulch Site Found with 36 mature plants, 10 saplings, and six seedlings		
Kahanahaiki 13 Sep 1987		Perlman, S.P. Obata, J.	Wild Kahanahaiki Site (MMR-A) Found with 12-15 plants		

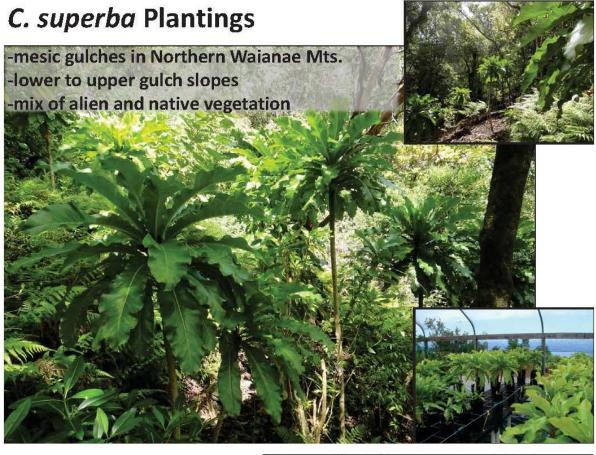


Data compiled from Bishop Museum Herbarium Records provided by Bishop Museum, 2015.

# Possible Fungal Infections of Wild C. superba



2016 Makua and Oahu Implementation Plan Status Report

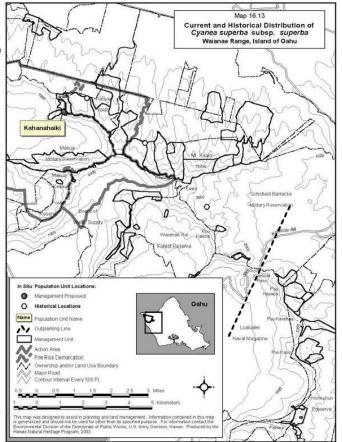


#### **Outplanting considerations:**

"Based on current and historical records of C. superba locations, under natural conditions it would be normal for the taxon to be growing with other species of Cyanea and with species of the related genera Delissea and Clermontia. It is common to find several Cyanea species and Cyanea relatives growing together, yet to date there is no evidence of hybridization occurring between species of Cyanea or between a Cyanea and a Cyanea relative. Consequently, concerns are minimal with respect to the possibility of inadvertently allowing unnatural hybridization to occur through the outplanting of C. superba subsp. superba. Cyanea superba subsp. superba has been documented only in the northern part of the Waianae Mountains, and not in the southern part. The southern Waianae Mountains are therefore not considered part of the taxon's natural range (MIP 2003)".

In the MIP, an outplanting line was drawn through the central Waianae Mountains limiting proposed reintroductions to the areas north of the line.

However, this restriction should be re-evaluated to allow for the possibility of managed translocations (assisted migration) of taxon to higher elevations or wetter habitats due to the impacts of climate change.



### **Species Occurrence**

(# in circles = # mature plants)

#### Map removed to protect rare resources. Available upon request

### **Habitat Characteristics**

Population Unit	Population Reference Codes	Elev. (m)	Slope	Canopy Cover	Topography	Aspect	Average Annual Rainfall (mm)
Kahanahaiki <sup>1</sup>	MMR-A MMR-E MMR-H	623 585-607 580-597	Moderate (10-45)	Intermediate Intermediate Closed	Upper Slope Mid Slope Gulch Bottom	North	1359.8 1347.0 1357.8
Makaha <sup>2</sup>	MAK-A	700	Moderate (10-45)	Intermediate	Mid Slope	North - Northeast	1748.5
Manuwai <sup>2</sup>	ANU-A	506	Moderate (10-45)	Intermediate	Gulch Bottom	Northeast	1406.0
Pahole to Kapuna <sup>1</sup>	PAH-A	543-561	Moderate (10-45)	Intermediate	Gulch Bottom	Northwest	1410.1 1457.7
Puu Palikea <sup>2*</sup>	PAK-A	853	Moderate (10-45)	Closed	Mid Slope	South East	1165

• Information was compiled from OANRP's HRPRG Rare Plant Monitoring forms & GIS data.

• Rainfall data complied from Rainfall Atlas of HI (Giambelluca et. al. 2013). Rainfall data is not precipitation and does NOT take into account cloud/mist "fog drip" moisture. Averages from 1977-2007.

• All Population Units are existing reintroductions<sup>1</sup> or introductions<sup>2</sup>.

• \*Not a current PU but 20 plants were planted here by The Nature Conservancy in 2005.

### **Associated Species**

Population Unit	Population Code(s)	Canopy	Understory
Kahanahaiki	MMR-E MMR-H	AcaKoa, <u>AleMol</u> , AlySte, AntPla, BobBre, <u>BudAsi</u> , CarMey, CarWah, ChaTom, CibCha, CopFol, <u>CorFru</u> , CyrDen, DioHil, DioSan, EupCel, FreArb, <u>GreRob</u> , GynTri, KadAff, MetPol, MyrLes, NesSan, PipAlb, PisBru, PisSan, PisUmb, PlaSan, <u>PsiCat</u> , <u>PsiGua</u>	AcaKoa, <u>AleMol</u> , AlySte, <u>AngEve</u> , AspExc, AspKau, AspMac, <u>BleApp</u> , CarMey, CarWah, ChaTom, CibCha, <u>CliHir</u> , CocOrb, CopFol, <u>CorFru, CycDen</u> , <u>CycPar</u> , DiaSan, EupMul, FluNeo, KadAff, <u>LanCam</u> , MicStr, NepExaHaw, OdoChi, <u>OpIHir</u> , <u>PasCon</u> , PipAlb, PlaSan, <u>PsiCat</u>
Makaha	MAK-A	AcaKoa, <u>AleMol</u> , AntPla, ClaSan, <u>CofAra</u> , DioHil, DioSan, ElaBif, GynTri, HibArnArn, MetPol, MyrLes, NesSan, PanBee, PipAlb, PisBru, PisSan, PisUmb, PlaSan, <u>PsiCat</u> , <u>PsiGua</u> , PsyOdo, SapOah, <u>SchTer</u> , StrPen, SyzCum, <u>TooCil</u> , UreGla, XylHaw	AlySte, AspNid, BidTor, <u>BleApp</u> , <u>BudAsi</u> , CarMey, CarWah, ChaObo, ChePla, <u>CofAra</u> , <u>ConBon</u> , CopFol, <u>CorFru, CycPar</u> , DooKun, DubPla, EraGra, EupMul, HibArnArn, <u>KalPin</u> , <u>LanCam</u> , LepTam, LysHil, MelMak, MicSpe, MicStr, NesSan, <u>PasEdu</u> , PipAlb, PolPelPel
Manuwai	ANU-A	No Mon	itoring Data
Pahole to Kapuna	РАН-А	AcaKoa, <u>AleMol</u> , AntPla, ChaTom, CibSpp, DioHil, DioSan, DioSpp, <u>FraUhd</u> , HibArnArn, MetPol, MyrLes, NesSan, PipAlb, PisBru, PisUmb, PlaSan, <u>PsiCat, PsiGua</u> , PsyMar, <u>SchTer</u> , UreGla, XylHaw	<u>AdiHis</u> , AlySte, <u>BleApp</u> , <u>BudAsi</u> , CarWah, <u>CliHir</u> , ColOpp, CopFol, <u>CycDen</u> , <u>CycPar</u> , DelWai, DioSan, DooKun, DryFus, DrySan, <u>FraUhd</u> , MetPol, MicStr, <u>NepBro</u> , NepExaHaw, <u>OplHir</u> , <u>PasCon</u> , <u>RubRos</u> , <u>SolAme</u>
Puu Palikea*	РАК-А	AlySte, CibGla, <u>CryJap</u> , CyrGar, ElaBif, IleAno, KadAff, LabKaa, MetPol, <u>MorFay</u> , PipAlb, PitSpp, PlaSan, <u>PsiGua</u> , PsyHat, UreGla, XylHaw	AlySte, AspAcu, AspMac, <u>BleApp</u> , <u>BudAsi</u> , ChaTom, CibGla, <u>CliHir</u> , <u>CycDen</u> , <u>CycPar</u> , CyrWai, DipSan, EupMul, FreArb, KadAff, KadCor, MicStr, PepMem, PerSan, PipAlb, PisSpp, <u>RubRos</u> , <u>SchTer</u> , SelArb, TecGau, TouLat, WikOahOah

\*Not a current PU but TNC planted 20 plants there in 2005.

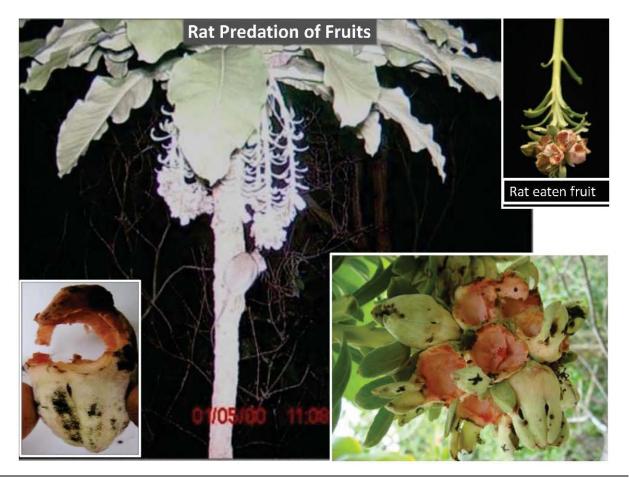
Species are listed in alphabetical order as observed by OANRP; introduced taxa are underlined: AbuGra, CycPar

#### THREATS

- Major threats to *C. superba* include feral pigs, which degrade the taxon's habitat and harm the plants by feeding, trampling and/or uprooting them. Alien plants alter its habitat and competing with it for sunlight, moisture, nutrients, and growing space. Also, the spread of highly flammable alien grasses increases the incidence and destructiveness of wildfires. Rats predate plant parts and fruits and introduced slugs and snails threaten the taxon by feeding on its leaves, stems, and seedlings (Joe & Daehler 2008, OANRP pers. obs.). Fungal pathogens may also be a substantial threat to this taxon.
- Over the past 10 years, enormous progress has been made in refining and increasing the effectiveness and efficiency in threat control techniques. All plantings are contained in fences to control ungulates, have partial to full rodent control where reproducing plants are present, and receive slug control where rare native snails are absent.
- The long-billed, nectar-feeding native Hawaiian birds, which are the presumed pollinators of C. superba, have been almost totally eliminated from the taxon's historic range in Waianae Mountains. Studies at Kahanahaiki (Pender et al. 2013) found no native avian pollinators visiting flowers and non-native birds only nectar-robbing, making any cross-pollination unlikely. The loss of the taxon's normal pollinating vectors may lead to decreases in the level of outcrossing in this species, which is presumed to be bird-pollinated, despite the taxon's capability of autogamy. An increase in inbreeding could therefore potentially lead to an expression of inbreeding depression (IBD) in successive generations. Genetic analysis of all of the available wild and cultivated stocks of C. superba, which included, at most, representation from four wild plants from one, or possibly both, populations, has shown that the genetic variability within the taxon is already extremely low (Morden pers. comm. 2000). Over the past five years, OANRP has noticed a substantial portion of the fruit produced do not contain any seeds. These fruits are typically found at the bottom of an infructescence which flowers last, potentially indicating that it could be tied to resource allocation by the maternal plant. Additional reasons for low to no seed set likely stem from a lack of a pollinator (pollen limitation) and/or possibly IBD. The observation of fruit with no seeds on the same infructescence as fruit with dozens of seeds suggests the former may be more likely. Potential means of overcoming IBD include human-mediated outcrossing among maternal lines and potentially incorporating additional genetic material into C. superba subsp. superba by hybridizing it with closely related species of Cyanea, including C. superba subsp. regina of the Koolau Mountains, if the subspecies is ever rediscovered. The most morphologically-similar species, Cyanea procera, has only one remaining plant in the wild, and could also be a potential recipient of genetic rescue via hybridization with C. superba. This may be something to explore further down the road if IBD is identified and limits population stability. Prior to such extreme actions, OANRP would like to identify if there are other potentially suitable habitats outside of historic range that would have pollinators and fruit dispersers present to serve as new outplanting sites.



- Recent studies by OANRP show seeds that remain in senesced fruits rapidly lose 50% of their viability. No avian dispersal was reported in studies of the Kahanahaiki plants or in monitoring by OANRP since 1997, meaning that at least half the seeds not predated by rats are lost as the fruit senesces on the plant. Due to these factors, recruitment and survival of new plants in restoration sites has been minimal and insufficient to meet MIP stabilization goals. OANRP would like to identify effective pollinators and dispersers and investigate whether or not there are other sites on Oahu where pollinators and fruit dispersers are more abundant. It is uncertain if there are any effective pollinators of *C. superba* on Oahu. We are currently uncertain of any sites on Oahu that have more avian dispersers as we do not know what species, native or alien, disperse *C. superba* fruit. Seedlings at Puu Palikea, however, have recruited throughout the Management Unit as opposed to only beneath the mature trees as in the other PUs. Therefore, some ambient (natural) dispersal is suspected, but the disperser is unknown. Recruitment has only been beneath the mature trees at the Kahanahaiki and Pahole to Kapuna PUs. There are a few seedlings that have recruited ~30m away from mature trees in Makaha PU.
- Climate change: Shifts in climatic conditions, including drought and increased temperatures, could potentially reduce recruitment for this taxon endemic to mesic forests. We are not sure if seedlings fail to establish and grow due to droughts, though we see the majority of seedlings in the spring as compared to later in the year. A trend of increasing lethal water stress has been observed on Maui for the Haleakala silversword (*Argyoxyiphium sandwicense* subsp. *macrocephalum*) and attributed to climate change (Krushelnycky et al. 2012). Conversely, it is possible that drier conditions may have fewer slugs, and we are uncertain if seedling-stage failure is due to a combination of slugs and drought. However, while we can control for slugs in sites (when no rare snails are present), we cannot combat climate change in existing sites. *C. superba* has a climate change vulnerability index of 0.936 (on a scale of 0-1, with 1 being the most vulnerable). It is the 4<sup>th</sup> highest value for IP taxa in the Waianae Mountains. 1<sup>st</sup> *K. parvula* (0.96); 2<sup>nd</sup> *P. kaalaensis* (0.946); 3<sup>rd</sup> *S. obovata* (0.944; Fortini et al. 2013).



### **Stabilization Goals Status (2016)**

**MIP Requirements for Stability** 

- 4 Population Units (PU)
- Threats controlled
- 50 reproducing individuals in each PU
- Stable population structure
- Complete genetic representation of all PUs in storage

MFS Population Units	PU Stab	ility Target	MU Threat Control					Genetic Storage
	50 Repro- ducing Plants?	Does the PU have stable population structure?	Ungulates	Weeds	Rodents	Fire	Slugs	Are there enough propagules in Genetic Storage?
Kahanahaiki	YES	NO	YES	PARTIAL 78%	YES	NO	Partial 45%**	
Manuwai	NO*	NO	YES	YES	NO*	NO	NO*	
Makaha	Partial 54%	NO	YES	NO	YES	NO	NO**	YES
Pahole to Kapuna	YES	NO	YES	PARTIAL 71%	PARTIAL 60%	NO	NO***	

\* No mature, reproducing plants currently at this site (142 immature outplants planted in 2013)

\*\* Slug control is limited due to the presence of rare native snails

\*\*\*Slug control had been monthly from Jan 2014 to April 2015

### **Genetic Storage Plan**

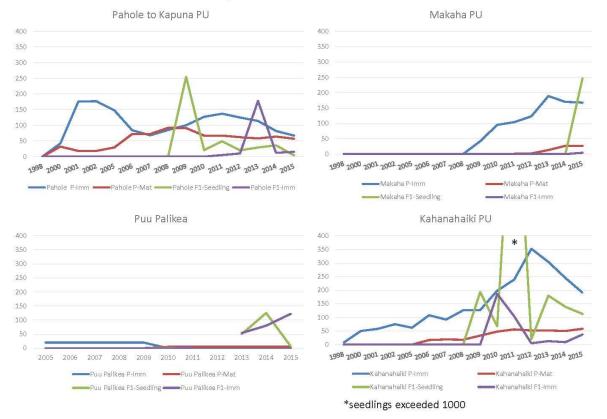
	What is the source for the propagules?	Method used to	What is the proposed re- collection interval for seed storage?	Is seed storage testing ongoing?	Plan for maintaining genetic storage.
Seeds	Reintroductions & Introductions	Seed banking (5C or -80C / 20% RH)	> 10 years	Yes	Collections will be made from outplantings as needed.

#### Genetic Storage Plan Comments:

- There were propagules from 3 founders from the wild Kahanahaiki population at the first outplanting, but seeds were
  never collected from one founder line before the outplanted progeny died. An additional "founder" was established via
  propagules of unknown provenance that we believed matriculated from the same Kahanahaiki population due to
  genetic analysis, but this remains uncertain. We currently track and maintain representation from these 3 founder lines.
- Collection procedures changed in December 2006 to harvest mature fruit directly from the trees to improve longevity of seeds in storage (as opposed to waiting for fruit to drop from the infructescence). Thorough storage testing began in January 2007 with seeds from this new harvest method. Five year results do not indicate a decline in viability.
- Seeds are currently banked at 5C, however, moving collections to -80C could significantly increase the longevity of these
  collections. Research conducted at the National Center for Genetic Resources Preservation suggests -80C storage slows
  the aging process (and therefore extends their storage life) more than 5C.
- As no wild plants remain, collections are made from outplantings, including reintroductions and introductions.
   Founders are tracked and collections made appropriately and currently refreshed every 10 years (at current 5C storage).
   The re-collection interval could be extended if seeds are stored at -80C. Collections were refreshed in Dec 2015.
- When new recruits become reproductive, collections will be made to represent these individuals as there are only 3
  maternal lines, the recruits have undergone selective pressures at the seed, seedling, and immature stages and could
  contribute to maximizing the amount of genetic diversity in the taxon. Additional maternal lines could also greatly
  improve the efficiency of hand-pollination efforts to promote increased genetic diversity.

### **Population Structure**

- There are currently 184 mature *C. superba*. Seedlings or immature plants were never observed at the Kahanahaiki wild site during the entire time they were monitored (1995-2002). Seedlings and immature plants were reported when the Pahole site was first discovered in 1971, but were gone by 1994. 1997-2002 was a well-recorded drought for much of the state and could have negatively impacted this population in mesic forest.
- In 2009, regeneration of seedlings under outplanted individuals was first observed. Over the 2009-2010 winter, over 500 seedlings were found under 8 separate plants in the Kahanahaiki PU. There were 21 mature trees at this time. An additional 300 seedlings were observed under two plants in the Pahole to Kapuna PU. Lastly, one immature plant was observed under a plant at Puu Palikea that had been planted by The Nature Conservancy. In total, seedlings were observed at 6 of the 12 outplanting sites (found within two PU and the Puu Palikea planting) with mature plants. Over the 2010-2011 winter season, there over 1,000 seedlings counted in the Kahanahaiki PU and 50 in the Pahole to Kapuna PU.
- In the years since 2011, there has been a decline in the number of seedlings (except for the newly established Makaha introduction which saw its first seedlings in 2015). New seedlings are observed each spring and several dozen have survived for more than six years as immature plants. None of these F1 plants have matured yet. Immature plants that naturally recruited are now present at 8 different outplanting sites in the Kahanahaiki PU (4 sites), Pahole to Kapuna PU (2 sites), Makaha PU, and Puu Palikea.
- Besides collections of fruit made for genetic storage and propagation, all other fruit has been left to mature on the plants. The fruit not eaten by rats was left to senesce and fall below the plants where new regeneration has been observed. Fruit at most PUs have been somewhat regularly dispersed by OANRP staff while conducting work in the area via smearing fruits across various substrates, and more thoroughly cleaning fruits to disperse seeds at Puu Palikea.
- The next slide graphically displays population trends at the three Manage Reintroduction for Stability PU that currently have reproducing plants (outplants have yet to flower at the most recent Manuwai PU), and the Puu Palikea outplanting. Please note that in the current absence of mature recruits, we are focused on quantifying the presence of naturally-occurring immature recruits (not planted) that have grown past the seedling stage. This refers to the purple line in the graphs. The other colors include immature outplants (blue), mature outplants (red), and recruits in the seedling stage (green). The lack of consistent monitoring at Puu Palikea is due to the fact that this is not a Manage for Stability Reintroduction at this time for OANRP.



#### **Population Structure**

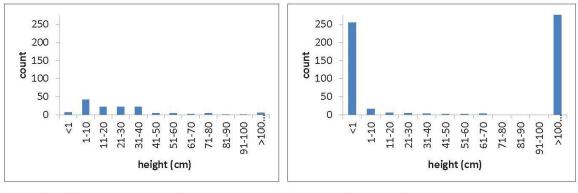
Population Unit	F1 total (2015)	•	predominant F1 size class	outplant total (2014)	total mature outplants (2014)	Year of 1 <sup>st</sup> planting	total outplants matured in last 10 years	ratio of F1s to mature outplants (2014)	ratio of F1s to outplants matured in last 10 years
Kahanahaiki	289	0.5-68	<1cm (88%)	295	50	1998	90	5.8	3.2
Manuwai	Outplar	nts have	yet to mature	– recent re	introductio	n			
Pahole to Kapuna	19	0.5- ca. 20	>1cm (79%)	425	95	2001	175	0.2	0.1
Makaha	250	0.5- ca. 20	<1cm (98%)	215	27	2009	27	9.3	9.3
Palikea	129	0.5-93	>1cm (95%)	20	5	2005	ca. 10	25.8	ca. 12.9

#### Counts of F1 plants in relation to numbers of outplants\*

\*Outplant data sourced from 2014 monitoring, as not all population reference sites were monitored in 2015. Anecdotally, seedlings at Makaha are now predominantly >1cm.

OANRP investigated variation in recruitment among PUs. To document the number of F1 plants and their heights, surveys at Kahanahaiki (MMR-D, MMR-E, MMR-F, MMR-H) and Puu Palikea (PAK-A) were conducted in April 2015. Kahanahaiki had more F1s in total, but Puu Palikea had a much higher ratio of F1s to mature outplants (see below). Most F1s at Kahanahaiki were <1 cm tall, while most at Palikea were >1 cm tall. While seeds may successfully germinate at Kahanahaiki, survival appears to be relatively poor. Recruitment at Kahanahaiki was primarily located below mature plants. Fruit that is not predated by rats is likely senescing, falling to the ground and having little to no recruitment. By comparison, at Palikea, recruitment often occurred distant from the few mature plants present. Given the results of the laboratory germination trial and F1 surveys at Kahanahaiki and Palikea, the relatively limited recruitment observed at Kahanahaiki (despite prolific production of viable seed), may be influenced by a lack of dispersers and/or habitat.

#### Comparison of Size Distributions of all Plants at Kahanahaiki and Puu Palikea

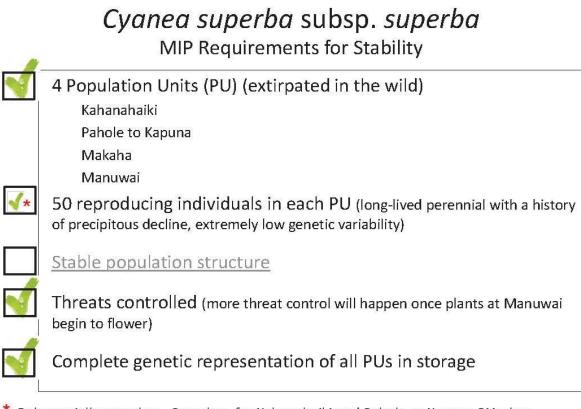


Size distribution of *Cyanea superba* subsp. *superba* at Palikea MU (n = 135), April 2015

Size distribution of *Cyanea superba* subsp*. superba* at Kahanahaiki MU (n = 539), April 2015

Plants measured from the ground to apical meristem. All plants <100 cm are F1s. All plants >100cm are outplants.

Note the small amount of recruits that are greater than 1cm at the Kahanahaiki PU. We consider plants less than 1cm to be that year's seedlings. There are more immature plants at Puu Palikea despite fewer mature outplants.



\* Only partially complete. Complete for Kahanahaiki and Pahole to Kapuna PUs, but Manuwai and Makaha PUs are too young & the majority of outplants have yet to mature

# 🗖 Management Discussion

- Outplantings of plants grown from the last remaining (only available) three founders from the wild Kahanahaiki site were established at Kahanahaiki in 1998 and at Pahole in 1999. Between 1997 and 2014, OANRP propagated nearly 1750 plants and outplanted over 1400 into 13 sites in the Kahanahaiki, Kapuna, Pahole, Makaha and Manuwai MUs. In addition, outplantings were conducted by NARS in Upper Kapuna and at Puu Palikea and Kaluaa by The Nature Conservancy. Survival across all sites is currently 54%. Currently there are 186 mature *C. superba* at eight different population sites across three PUs with recruitment (to the size class of small immature).
- Plants are outplanted as immature. Of the 303 plants to mature, plants took on average five years after outplanting to mature. Of the 117 outplants that have matured and died, they lived for an average of nine years total. The remaining 186 outplants that have matured are still alive.
- Despite observing recruitment, and a transition from seedlings to immature plants, none of these immature plants have grown very large or matured. Currently, the only obstacle to obtaining stability for this taxon is to establish stable population structure.

### Management Discussion

In order to establish restoration sites that become stable, new efforts to incorporate the limiting factors identified in the threats section are needed. Restoration factors that could be addressed include the following:

1) Habitat site selection (large scale and micro-site locations): OANRP proposes selecting one or two new introduction sites and several seed sow sites to identify sites and micro-site conditions that promote recruitment and stage class transitions to immature and mature plants. New sites that would be considered as managed relocations should be considered to combat climate change.

2) **Overcoming inbreeding and lack of pollinators:** OANRP could conduct hand-pollinations to breed between different maternal lines within or among taxa, and use these progeny to establish new trial outplantings sites. We could also pollinate to increase seed set. OANRP could conduct pollinator observations to determine if certain sites have more visitation than others, or if areas have more potential pollinators than others.

3) **Fruit Dispersal**: OANRP could support ongoing fruit disperser research (identify species and quantify fruit dispersal). identify dispersers and quantify fruit dispersal among existing sites and propose new sites with more dispersers present. OANRP could conduct artificial (human-assisted) fruit dispersal.

4) The number of outplants per population site and unit. OANRP could consider increasing the number of outplants used to establish a new site or supplement an old site to promote recruitment.

5) **Threat Control**: OANRP to review all ongoing threat control efficacy to determine if increased efforts could have a positive effect on recruitment.

Manage For Stability Population Units	Fenced?	Easy Rodent Control? <sup>1</sup>	Easy slug control? <sup>2</sup>	Pollinators Present?	Dispersers Present?	Managed relocation?
Kahanahaiki	Yes	Ongoing	Ongoing (Partially)	No	No	No
Makaha	Yes	Ongoing	No	?	Yes?	No
Manuwai	Yes	Yes	Yes	?	?	No
Pahole to Kapuna	Yes	Yes	Yes	?	No	No
Proposed Alt	ernate Popu	lation Units (to c	hoose 1-2 for	planting and 1	-2 for trial see	d sows)
Central & East Makaleha	Maybe	To be conside of Hawaii DO		ure depending o	on fence constr	uction (State
		Ul Hawan DO	FAW)			
Kaluaa	Yes	Yes	Maybe	?	?	No
	Yes Yes		and the second se	?	?	No Yes
Kaluaa		Yes	Maybe			
Kaluaa Lihue (Haleauau)	Yes	Yes Yes	Maybe Yes	?	?	Yes
Kaluaa Lihue (Haleauau) Lower Opaeula	Yes Yes	Yes Yes No Ongoing	Maybe Yes No	3 5 5	? ?	Yes Yes

#### **Current & Proposed Alternate Population Units**

<sup>1</sup> "Yes" means we can maintain rodent grid monthly. "No" means quarterly baiting.

<sup>2</sup> "Yes" indicates Sluggo can be applied monthly. "Maybe" indicates surveys for rare snails are needed. "Partially" indicates that some slug control cannot happen in that PU due to the presence of rare snails. For Makaha, "No" means there are native snails present and slug control cannot occur. For Lower Opaeula, "No" means that it is unlikely to maintain monthly applications of Sluggo.

## 1) Habitat Selection: Managed Relocation

- The outplanting line, which only permitted plantings north of Kolekole Pass, has been eliminated.
- Choosing reintroduction sites based on their Reintroduction Site Ranking Score from the 2003 Implementation Plan has been put on hold until a new scoring system can be devised that includes more threats to each species, especially climate change.
- Climate change: *C. superba* has a climate change vulnerability index of 0.936 (on a scale of 0-1, with 1 being the most vulnerable). It is the 6<sup>th</sup> highest value for IP taxa in the Waianae Mountains. 1<sup>st</sup> *D. herbstobatae* (0.986); 2<sup>nd</sup> Sanicula mariversa (0.968); 3<sup>rd</sup> *K. parvula* (0.96); 4<sup>th</sup> *P. kaalaensis* (0.946); 5<sup>th</sup> *S. obovata* (0.944; Fortini et al. 2013). OANRP should execute sound judgement in evaluating the need to choose introduction sites higher in elevation or wetter in climate for species with high vulnerability scores.

The Rare Plant Implementation Team has eliminated the MIT 2003 outplanting line and has chosen Palikea as a 5<sup>th</sup> Managed Reintroduction for Stability Population Unit (MRFS PU). Palikea will be considered a Managed Relocation. Five Manage for Stability PUs will remain until Palikea is established, at which point all management will stop for the Pahole MFS PU.

# 1) Habitat Selection: Microsite (Seed Sow Trials)

- *In situ* field seed sow trials could aide in identifying small-scale sites ideal for seedling recruitment and successful establishment of small to immature plants.
- Compare recruitment among existing and proposed PUs as well as among various microsite conditions within PU sites.
- Conduct seed sow trials along rainfall/precipitation gradients; consider purchasing data loggers & weather stations that track precipitation and humidity.
- There is a possibility that the seeds used in these trials could produce mature trees. The amount of seed sown, along with the average number of seeds produced per mature plant, will help address the number of outplants needed that could produce stable population structure at these sites.

OANRP proposes to initiate seed sow experiments at Lihue (upon permission from the Army) and Lower Opaeula to determine their potential as future introduction sites. OANRP will also conduct seed sow trials at Makaha and Palikea to investigate effects of environmental conditions on seedling establishment.

### **UPDATED Population Units**

Site	OANRP Proposed Actions
Lihue (Haleauau)	Ask Army if we can plant/seed sow into Schofield as plants not present in this Action Area and look for potential sites; conduct seed sow if allowed
Kahanahaiki*	Continue as Manage Reintroduction for Stability PU (MRFS)
Makaha*	Continue as MRFS; conduct seed sow trial
Pahole to Kapuna*	Continue as MRFS (consider dropping status if/when a new PU established with completed outplanting)
Kaluaa	Look for sites to establish a trial planting, seed sow, or introduction further up the gulch from the current planting
Lower Opaeula	Conduct a seed sow (with minimal or no slug control)
Puu Palikea*	Augment existing introduction; make a new MRFS; conduct floral visitor & disperser observations (cameras); conduct seed sow trial
Manuwai*	Continue as MRFS; wait for plants to flower

\*Current Manage Reintroduction for Stability Population Units

NOTE: The content on this slide and the following slides is based on discussions from the 2016 Rare Plant Implementation Team meeting.

### 2) Investigations into potential inbreeding depression

- As stated in the MIP, and with additional information gathered on this taxon and other species of *Cyanea*, inbreeding depression is a concern in this species due to:
  - Presumed bird-pollinated outcrossing species with no observations of pollination (except insect floral visitation); suggests high rates of selfing
  - Only three maternal lines from a single population available for outplantings
  - A rough approximate of half of the developing fruit do not contain any seeds; which could be a sign of pollen limitation or inbreeding depression (or resource allocation)
- Given the very limited amount of genetic variation among the remaining lines, it will likely take many generations of human-mediated outcrossing to increase genetic variation with little return.
- If inbreeding depression is limiting the taxon's ability to survive in the wild, it may be necessary to study strategies for increasing the genetic variability of the taxon.

The Rare Plant Implementation Team has decided that it is unreasonable to handpollinate tall trees and unlikely that birds will be observed effectively pollinating C. superba. Therefore, OANRP may only decide to place game cameras at Palikea during flowering to observe visitation.

#### 3) Fruit Dispersal: Cyanea superba Germination Trial

Limited dispersal and recruitment of *Cyanea superba* subsp. *superba* occurs at reintroduced populations, with the majority of fruits either depredated by rats (seeds are destroyed), or rotting on the plant and falling to the ground with limited subsequent seed germination and seedling survival, despite having typically high seed germination rates in fresh mature fruit (Pender et al. 2013, OANRP 2015a, 2015b, M. Akamine pers. obs.). A laboratory trial was completed in 2015 that explored two questions to gain a preliminary understanding of recruitment limitations and factors affecting seed sow success. Do seeds from senesced *C. superba* subsp. *superba* fruit have reduced viability as compared with those from fresh mature fruit? If seed sowing is used to sustain populations, does total removal of fruit pulp promote higher germination rates? The trial examined: 1) *C. superba* subsp. *superba* seed germination rates of senesced fruit in comparison with fresh material as a means of exploring the ability of seeds from senesced fruit to germinate upon falling on the ground vs. those from fresh fruit; and 2) germination rates of *C. superba* subsp. *superba* seeds with and without pulp extract to examine the effects of fruit pulp on germination during seed sow efforts.

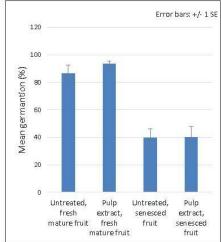
Results: Seeds were approximately 50% less viable from senesced vs. fresh C. superba subsp. superba fruit regardless of treatment (untreated seeds: t = 6.659, p < 0.001; seeds with pulp extract: t = 5.077, p < 0.001). The reduced germination in seeds from senesced fruit limits recruitment potential in the absence of dispersers, as fresh mature fruits that are not consumed by dispersers will senesce and fall to the ground, and subsequently have reduced potential for germination. Fruit extract had no effect on germination of seeds for either senesced (t = 0.022, p = 0.982) or fresh mature fruit (t = 1.075, p = 0.296).

Seed viability from senesced and fresh mature fruits, with and without fruit pulp extract. Fruits from 10 individuals were used for each of four treatments (with a minimum of 1 fruit per plant per treatment): untreated seeds of senesced fruits, seeds of senesced fruits with pulp extract, untreated seeds of fresh fruits, and seeds of fresh fruits with pulp extract. The degree of senescence was not quantified, but was estimated to be less than 1 week following peak maturation. Number of seeds sown per sample ranged from 22 to 200 (mean = 88.4, SD = 37.4, 3534 total seeds sown).

- A follow up trial began in Dec 2015 to explore two questions to gain a more precise understanding of recruitment limitations in association with fruit senescence. 1) What is the rate of decline in seed germination as *C. superba* subsp. *superba* fruits senesce, and 2) at what point are seeds no longer viable? This laboratory trial explores the ability of seeds from senesced fruit to germinate over time upon falling on the ground.
- Mature fruits were collected from infructescences (not from the ground) from at least 5 individuals, to include a total of 24 fruits. Fruits were cleaned and stored individually at ambient room temperature at the OANRP Seed Laboratory. Seeds from 4 randomly chosen fruits were sown twice a week for 3 weeks, beginning on the collection date, for a total of 6 viability assay dates. Seeds were sown on agar in Petri dishes, to include 50 seeds per fruit (fruits typically contain 100-200 seeds each). Petri dishes will be stored in a Percival Controlled Environment Chamber (with diurnal light and temperature settings matching average monthly temperatures for the Nike missile installation at Pahole, at approximately 2100 feet elevation), and examined weekly for germination for a total of 10 weeks.

Photos: clockwise fresh fruit, removing seeds from fruit (top), seedlings germinating in the OANRP Seed Lab (bottom).







# 3) Quantifying *In Situ* Fruit Dispersal & Understanding Effects of Limited Dispersal

OANRP will continue to support research of M.S. student Sean McDonald who is documenting dispersers and quantifying presence and amount of fruit dispersal at most C. superba PUs, as well as testing different ways to increase dispersal. We will also report on our research for effects of senescing fruit on seed quality. Together, we will provide management recommendations for increasing fruit dispersal if it is possible.

#### 4) Increasing # Mature Plants at Reintroductions

- It is possible that the number of reproducing plants required for stability will not yield a stable population, despite maximum feasible threat control efforts.
- To obtain stable populations, we need to see replacement of existing mature outplants with naturally recruiting mature trees.
- Increasing the number of mature outplants at a reintroduction site will result in a larger *in situ* seed bank and hopefully increase the number of plants recruiting and surviving through each size class and outcompete the present threats.
- Allee effects may also contribute to population stability by increasing the attractiveness of the population to pollinators and dispersers.

OANRP proposes to continue to establish outplantings with enough plants to reach 50 mature outplants for each PU, while site selection and fruit dispersal studies are ongoing. If seed sow trials are able to produce a mature tree, this information will be applied to adjust our outplanting goals (how many trees to plant) to maintain 50 mature naturally recruited trees and stable population structure. While there was excitement about planting more plants, OANRP will wait until they have more information to direct the effort.

### 5) Threat Control

- All plantings are contained in fences to control ungulates, have some weeding
  ongoing, partial to maximum rodent control where reproducing plants are
  present, and receive slug control when the absence of rare native snails is known.
- Current PUs with all threats mitigated to a feasible extent still have not produced enough recruitment to yield stable populations.
- This level of threat control alone (ungulates, rodents, slugs, weeds) is not likely to produce stable populations at existing sites with the current number of plants.
- Pursuing ways to mitigate for additional threats (lack of pollinators and dispersers and climate change) may help in establishing stable populations.

#### OANRP proposes to maintain and optimize existing threat mitigation and research and develop new efforts to deal with effects of climate change (PU and microsite selection) and lack of pollination and fruit dispersal.

## **Monitoring Plan**

• These existing and any new outplanting sites in MFS PUs will be monitored annually in Quarter 2, using the Hawaii Rare Plant Restoration Group (HRPRG) Rare Plant Monitoring Form (RPMF):

- Kahanahaiki- MMR-E, MMR-H
- o Makaha- MAK-A
- o Manuwai- ANU-A
- Palikea PAK-A
- All other existing sites will be monitored bi-annually. The RPMF will be used to record population structure, age class, reproductive status, and vigor of all accessible plants. If there is any threat to the health and safety of plants due to repeated monitoring and/or tagging, reductions in the number of tagged individuals will be made so that no harm is done to the plants. This monitoring data will serve to document the populations at the remaining sites to guide threat management and genetic storage needs.
- Seedlings will be monitored to track survivorship and growth.
- From most recent veg monitoring, MU native understory/canopy median % cover is 25/15 at Kiki, 7.5/25 at Makaha, 7.5/15 at Kapuna (no data for Pahole), 7.5/20 for Manuwai, 7.5/25 for Kaluaa, 35/25 for Palikea.

Population Unit	Reintroduction Site(s)	Number of Plants to be planted*	Propagule Type	Propagule Population(s) Source	Number of Founders in Source Population	Plant Size	Pot Size
	MMR-E	100 (166%)					
Kahanahaiki	MMR-H	100 (166%)					
	MMR- B, D, F, G	0					
Pahole to Kapuna	Pahole: PAH-A	100 (137%)					
Pahole to Kapuna	Pahole: PAH-B	0	Immature	MMR-A		-A .5-1 gallon	
Pahole to Kapuna	Kapuna: KAP- A,B	0	Plants				
Makaha	MAK- A	150		3 fou	nders		
Manuwai	ANU-A	150					
Puu Palikea	PAK-A***	150					
Lihue	SBW-A		Seed Sow			т	BD
Lower Opaeula	OPA-A		Seed sow			Т	BD

## **Reintroduction Plan**

\*Number of plants to be planted: The target number for each site is listed followed by a percentage for sites with existing plantings. The total number planted (adjusted for time to mature after planting) was divided by the number of these plants that are mature. The percentage displayed is the multiplier needed to compensate for the survivorship of mature plants calculated for each site. The target number is multiplied by this percentage to get the number of plants to be planted. For sites with no existing or recently planted plants, the baseline 150 plants (50 of each of the three founders) will be planted initially, and more will be added if needed.

### **Reintroduction Plan Comments**

- The reintroduction of *C. superba* into Kahanahaiki was the first endangered species in the nation to be reintroduced onto Army managed lands.
- The MMR-B, D, F, G sites in Kahanahaiki are no longer supplemented due to poor performance when compared with MMR-E and MMR-H.
- The sites in the Pahole to Kapuna PU at KAP-A, KAP-B and PAH-B are Oahu NARS reintroductions planted from 1995 to 1999. They are monitored and collected from but not extensively managed.
- Reintroduction protocols for *C. superba* are well developed and will be followed for future reintroductions. The propagule type, plant and pot size are standardized.
- The average time that plants take to mature is five years after outplanting.
- The Palikea site had only 20 plants planted in 2005 but will receive ~150 new plants in 2017. All other MFS PU sites will be supplemented as needed.

### **5 Year Action Plan**

	Proposed Actions for the following years:								
Population Unit	MIP YEAR 12 October 1, 2015 – September 31, 2016	MIP YEAR 13 October 1, 2016 – September 31, 2017	MIP YEAR 14 October 1, 2017 – September 31, 2018	MIP YEAR 15 October 1, 2018 – September 31, 2019	MIP YEAR 16 October 1, 2019 – September 31, 2020				
Kahanahaiki	•Monitor & Collect •Rat Control •Slug control (partial) •Weed Control	•Monitor & Collect •Rat Control •Slug control (partial) •Weed Control	•Monitor & Collect •Rat Control •Slug control (partial) •Weed Control	•Monitor & Collect •Rat Control •Slug control (partial) •Weed Control	•Monitor & Collect •Rat Control •Slug control (partial) •Weed Control				
Manuwai	•Monitor	•Monitor •Rat Control •Slug Control •Weed Control	•Monitor & Collect •Rat Control •Slug control •Weed Control	•Monitor & Collect •Rat Control •Slug control •Weed Control	•Monitor & Collect •Rat Control •Slug control •Weed Control				
Makaha	•Monitor & Collect •Rat Control •Weed Control	•Monitor & Collect •Rat Control •Weed Control •Seed Sow	•Monitor & Collect •Rat Control •Weed Control	•Monitor & Collect •Rat Control •Weed Control	•Monitor & Collect •Rat Control •Weed Control				
Pahole to Kapuna	•Monitor	•Monitor	•Monitor	•Monitor	•Monitor				
Kaluaa	•Find potential seed sow sites		•Seed Sow	•Monitor	•Monitor				
Lower Opaeula	•Find potential seed sow sites	•Seed Sow	•Monitor	•Monitor	•Monitor				
Lihue	•Find potential seed sow sites	•Seed Sow	•Monitor	•Monitor	•Monitor				
Puu Palikea	•Find Outplanting Site	•Seed Sow •Rat Control •Slug Control	<ul> <li>Introduce</li> <li>Rat Control</li> <li>Slug Control</li> <li>Weed Control</li> </ul>	•Monitor •Rat Control •Slug control •Weed Control	•Monitor •Rat Control •Slug control •Weed Control				

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# Mahalo



