

***Hibiscus waimeae* subsp. *hannerae* Management Plan**

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Introduction: *Hibiscus waimeae* subsp. *hannerae*, also known as koki'o ke'oke'o in Hawaiian, is a subspecies of hibiscus endemic to the island of Kaua'i in Hawai'i. Field work conducted from 2015 through 2017 documented that there are approximately 200 mature individuals of the taxon remaining *in situ*. It was federally listed as Endangered in 1996 (USFWS, 2023). The most recent, published formal conservation assessment, conducted using the IUCN Red List of Threatened Species criteria categorized the taxon as Critically Endangered (Tangalin and Wood, 2015). The taxon is a tree that grows to an average of 6-10 meters tall. The flowers are perfect and strongly fragrant, turning from white to pinkish-white during their one-day lifespan. Subspecies *hannerae* is confined to several northwestern valleys of Kaua'i, while the subspecies *waimeae* can be found in drier regions of the island in the Waimea Canyon and ocean-facing valleys of the western and southwestern regions of Kaua'i. The population of *H. waimeae* subsp. *hannerae* has decreased due to a number of threats including habitat degradation in the lowland rainforest habitat in which it occurs by primarily feral pigs (*Sus scrofa*) and invasive plants such as *Erigeron karvinskianum*, *Lantana camara*, *Miconia crenata*, and *Psidium guajava*. A possible loss of pollinators and consequently a genetic bottleneck are suspected to also have contributed to the decline of this subspecies (Tangalin and Wood, 2015). The taxon has been a focus for *ex situ* conservation and *in situ* restoration for a number of years. A recent genetic study revealed that although the taxon has 500-600 individuals represented *ex situ* by at least seven botanical institutions, 69% of all alleles existing *in situ* are represented across *ex situ* collections (Hoban et al., 2020). Recommendations in the most recent USFWS five-year review include to "continue to increase genetic diversity of the [taxon] by using propagules from a mixture of genetic stock to increase reproductive vigor." (USFWS, 2022).

Analytical Assumptions and Exclusions: For this subspecies the pedigree, empirical, and managed populations are all the same. This population consists of 53 individuals housed at eight institutions. To our knowledge, the managed population includes all known *ex situ* individuals. In developing the pedigree, the maternal source (dam ID) of each wild born individual was based on available accession information. Individuals with unique accession numbers were given unique wild maternal IDs (e.g., WILD1, WILD2, etc.). Individuals with shared accession numbers were given a shared wild maternal ID (e.g., WILD3, WILD3, etc.). Wild born individuals were given WILD paternal IDs. The maternal ID for captive born individuals was the accession number. The sire and dam ID was designated as unknown (UNK) if it was not known whether the individual was wild born or captive born (i.e., there was no accession information) or if an individual was captive born but the sire was unknown.

Demography: Demographic analyses were beyond the scope of the analyses based on the limited amount of demographic information documented for the subspecies.

Genetics: The pedigree indicates that the population is descended from 60 founders. The gene diversity of the *ex situ* population is 93.33%, which is equivalent to the gene diversity found in 7

- 8 individuals (FGE =7.49). The current mean kinship is 0.0667 meaning that, on average, individuals have similar kinship coefficients as do half-cousins (0.0625) or first cousins once removed (0.0625). See glossary below for definitions of terms in Table 1.

Table 1. Genetic status of ex situ population

Genetics Summary - 2023	
Founders	60
Potential (additional)	0
Living Individuals	
N Individuals	53
Living Descendants	22
% Pedigree Known	42%
% Analytic Known	100%
% Ancestry Certain	42%
Gene Diversity	
Based on Kinship Matrix	0.9333
Based on Gene Drop	0.9773
Potential	0.9773
Gene Value	—
Population Mean Kinship	0.0667
Founder Genome Equivalents	
Based on Kinship Matrix	7.49
Based on Gene Drop	22.00
Potential	22.00
Founder Genomes Surviving	22
Mean Inbreeding	0.0000
Mean Ne	8.49
Over Generations:	1.00
Current Ne	0.00

Ne/N

0.0000

Recommended breeding plan: We recommend that future management plans prioritize the breeding pairs outlined below (Table 2). Breeding decisions should seek to increase the genetic representation of genetically unique individuals (or founder genomes) that are currently under-represented *ex situ*. In ensuring equal representation of individuals, managers can reduce the amount of genetic diversity lost over time. Ideally, breeding would include few offspring per breeding event. To be included in a breeding pair, individuals must (1) be ranked as a genetically valuable individual (see MKRank below, Table 3), (2) be in a pair that results in offspring with little inbreeding (F), and (3) be in a pair that results in a positive or no change in gene diversity.

Table 2 includes the Unique ID for the first recommended parent (ID1), the location of that parent (Location1), the Unique ID for the second recommended parent (ID2), and the location of that parent (Location2), the inbreeding coefficient of the offspring of the recommended pairing (F), and the change in gene diversity of the population that results from the recommended pairing (Gene Diversity).

Table 2. Recommended breeding pairs - not ordered by change in gene diversity

Unique ID1	Local ID	MKRank ID1	Location1	Unique ID2	Local ID2	MKRank ID2	Location 2	F	Gene Diversity (+)
HHAN_204	SMD196	1	NTBG	HHAN_193	SMD185	3	NTBG	0.0283	0.0016
HHAN_01	UNK	2	ABGMA	HHAN_183	SMD175	5	NTBG	0	0.0024
HHAN_02	UNK	4	BISH	HHAN_195	SMD187	8	NTBG	0	0.002
HHAN_03	UNK	13	DBG	HHAN_202	SMD194	11	NTBG	0	0.0011
HHAN_07	UNK	9	WA	HHAN_20	SKW148	6	NTBG	0.0121	0.0015
HHAN_202	SMD194	11	NTBG	HHAN_198	SMD190	7	NTBG	0	0.0011
HHAN_195	SMD187	8	NTBG	HHAN_204	SMD196	1	NTBG	0	0.0018
HHAN_183	SMD175	5	NTBG	HHAN_24	SKW152	14	NTBG	0	0.001
HHAN_02	UNK	4	BISH	HHAN_193	SMD185	3	NTBG	0.0454	0.0011
HHAN_198	SMD190	7	NTBG	HHAN_08	UNK	22	WA	0	0.007
HHAN_206	SMD198	17	NTBG	HHAN_183	SMD175	5	NTBG	0	0.0006

HHAN_13	UNK	16	WA	HHAN_01	UNK	2	ABGMA	0	0.0006
HHAN_34	SKW231	18	NTBG	HHAN_20	SKW148	6	NTBG	0	0.0004
HHAN_77	SKW396	19	NTBG	HHAN_08	UNK	22	WA	0	0.0003
HHAN_06	UNK	12	LNG	HHAN_202	SMD194	11	NTBG	0	0.0003

Mean kinship ranking: Below we include the mean kinship for the empirical population and their rankings. Included is the Unique ID of each individual, its current location, the LocalID or collector number, the empirical mean kinship (MK), and the rank of each individual based on empirical mean kinship (MKRank).

Table 3. Empirical mean kinship rankings

UniqueID	Location	LocalID	MK	MKRank
HHAN_204	National Tropical Botanical Garden	SMD196	0.0244	1
HHAN_01	ABGMA	UNK	0.0351	2
HHAN_193	National Tropical Botanical Garden	SMD185	0.0354	3
HHAN_02	Bishop Museum	UNK	0.0391	4
HHAN_183	National Tropical Botanical Garden	SMD175	0.0411	5
HHAN_20	National Tropical Botanical Garden	SKW148	0.0413	6
HHAN_198	National Tropical Botanical Garden	SMD190	0.0429	7
HHAN_195	National Tropical Botanical Garden	SMD187	0.0434	8
HHAN_07	MBG	UNK	0.0454	9
HHAN_05	The Huntington Library, Art Museum, and Botanical Gardens	UNK	0.0472	10
HHAN_202	National Tropical Botanical Garden	SMD194	0.0475	11
HHAN_06	Longwood Gardens	UNK	0.0502	12
HHAN_03	Denver Botanic Gardens	UNK	0.0505	13
HHAN_24	National Tropical Botanical Garden	SKW152	0.0508	14
HHAN_197	National Tropical Botanical Garden	SMD189	0.0527	15
HHAN_13	Waimea Arboretum and Botanical Garden	UNK	0.0541	16
HHAN_206	National Tropical Botanical Garden	SMD198	0.0546	17

HHAN_34	National Tropical Botanical Garden	SKW231	0.0601	18
HHAN_77	National Tropical Botanical Garden	SKW396	0.0608	19
HHAN_23	National Tropical Botanical Garden	SKW151	0.0608	20
HHAN_188	National Tropical Botanical Garden	SMD180	0.0611	21
HHAN_08	Waimea Arboretum and Botanical Garden	UNK	0.0624	22
HHAN_16	National Tropical Botanical Garden	KW17093	0.0641	23
HHAN_76	National Tropical Botanical Garden	SKW395	0.0643	24
HHAN_11	Waimea Arboretum and Botanical Garden	UNK	0.0646	25
HHAN_09	Waimea Arboretum and Botanical Garden	UNK	0.065	26
HHAN_199	National Tropical Botanical Garden	SMD191	0.0657	27
HHAN_19	National Tropical Botanical Garden	KW17096	0.067	28
HHAN_79	National Tropical Botanical Garden	SKW398	0.067	29
HHAN_196	National Tropical Botanical Garden	SMD188	0.0672	30
HHAN_80	National Tropical Botanical Garden	SKW399	0.0678	31
HHAN_190	National Tropical Botanical Garden	SMD182	0.0684	32
HHAN_189	National Tropical Botanical Garden	SMD181	0.0689	33
HHAN_10	Waimea Arboretum and Botanical Garden	UNK	0.0695	34
HHAN_12	Waimea Arboretum and Botanical Garden	UNK	0.0702	35
HHAN_17	National Tropical Botanical Garden	KW17094	0.0702	36
HHAN_81	National Tropical Botanical Garden	SKW400	0.0718	37
HHAN_194	National Tropical Botanical Garden	SMD186	0.072	38
HHAN_32	National Tropical Botanical Garden	SKW229	0.0725	39
HHAN_78	National Tropical Botanical Garden	SKW397	0.0742	40
HHAN_187	National Tropical Botanical Garden	SMD179	0.0747	41
HHAN_201	National Tropical Botanical Garden	SMD193	0.0753	42

HHAN_184	National Tropical Botanical Garden	SMD176	0.0753	43
HHAN_207	National Tropical Botanical Garden	SMD199	0.0755	44
HHAN_82	National Tropical Botanical Garden	SKW401	0.076	45
HHAN_31	National Tropical Botanical Garden	SKW228	0.0762	46
HHAN_200	National Tropical Botanical Garden	SMD192	0.0776	47
HHAN_203	National Tropical Botanical Garden	SMD195	0.0778	48
HHAN_185	National Tropical Botanical Garden	SMD177	0.078	49
HHAN_191	National Tropical Botanical Garden	SMD183	0.0781	50
HHAN_205	National Tropical Botanical Garden	SMD197	0.0789	51
HHAN_186	National Tropical Botanical Garden	SMD178	0.0843	52
HHAN_192	National Tropical Botanical Garden	SMD184	0.0864	53

Glossary:

PMx definition of a founder - PMx considers a founder to be an individual with Wild/Wild parentage that has living descendants in the selected population

Potential Founder - living individuals that have no living relatives in the population but have the potential to reproduce and contribute to the population

Living Descendants - The number of living individuals descended from founders. This may be a fractional number because PMx normally excludes all individuals with unknown parents from genetic analyses, and includes only those parts of individuals that can be traced back to known founders. For example, an individual with a known dam but an unknown sire will be tallied as half an individual in genetic analyses

N Individuals - Number of living individuals in the currently selected population for genetic analysis

% Ancestry Certain - The percentage of the living individuals' pedigree that can be completely identified (exact identity of both parents is known) and traceable back to known founders. Individuals that are 100% Certain do not have any MULTs or UNKs in their pedigree. Certainty represents a higher degree of knowledge than Known and therefore is always less than or equal to Known.

Gene Diversity - Proportional gene diversity (as a proportion of the source population) is the probability that two alleles from the same locus sampled at random from the population are not identical by descent from a common ancestor. Gene diversity is the heterozygosity of founder alleles ($= 1 - \text{inbreeding}$) expected in progeny produced by random mating

Gene Value - Gene value is the gene diversity of the living individuals weighted for the reproductive value of individuals. Gene value is the heterozygosity of founder alleles expected in progeny produced by random mating and if each individual were to produce the number of offspring expected from its age-based reproductive value

Gene Diversity (Based on Kinship Matrix) - Gene diversity calculated from the kinship matrix as $1 - m_k$, where m_k is the average mean kinship in the population

Gene Diversity (Based on Gene Drop) - Gene diversity (GD) calculated from gene drop simulations. Each founder is assigned two unique alleles that pass stochastically through the pedigree. Final GD is calculated from the final distribution of alleles and is based on the number of alleles and the evenness of allele frequencies

Potential Gene Diversity - Potential GD of the population if optimal reproduction of potential founders were to be achieved (calculated as $1 - (1/2FGE_{\text{potential}})$, where $FGE_{\text{potential}}$ is the population's potential founder genome equivalent).

Gene Value - Gene value of the current living population and is the gene diversity of the population weighted for the reproductive value of individuals

Population Mean Kinship - Average mean kinship value of individuals in the current living population

Founder Genome Equivalents - The number of unrelated individuals (founders) that would represent the same amount of gene diversity as does the population of currently living individuals. A population's FGE becomes smaller than the actual number of population founders over time, as founder representations vary and gene diversity is lost from the population

Founder Genome Equivalent (Potential) - Maximum FGE that can be attained (if all founder allele retentions are set to their potential retention). Provides an upper limit to what might be achieved through perfect genetic management but is not usually realistically achievable

Founder Genomes Surviving - The sum of allelic retentions of the individual founders (i.e., the product of the mean Genomes allelic retention and the number of founders)

Mean Ne - Average effective population size of the selected population over the analysis time span. Given in relation to the number of Generations over which N_e is calculated

Current Ne - Effective population size of the selected living population, based on the number of living males and females that have produced offspring

Ne/N - Ratio of the effective population size to census size of living, captive-born individuals

Mean Inbreeding - The average of the inbreeding coefficients among the living individuals, weighted by the % known for each individual. The mean inbreeding coefficient of a population will be the proportional decrease in observed heterozygosity relative to the expected heterozygosity of the founder population.

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