What to do when we can't bank on seeds: Applying zoo population management tools to rare plants in living collections

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Presented by Greg Mueller, Chicago Botanic Garden

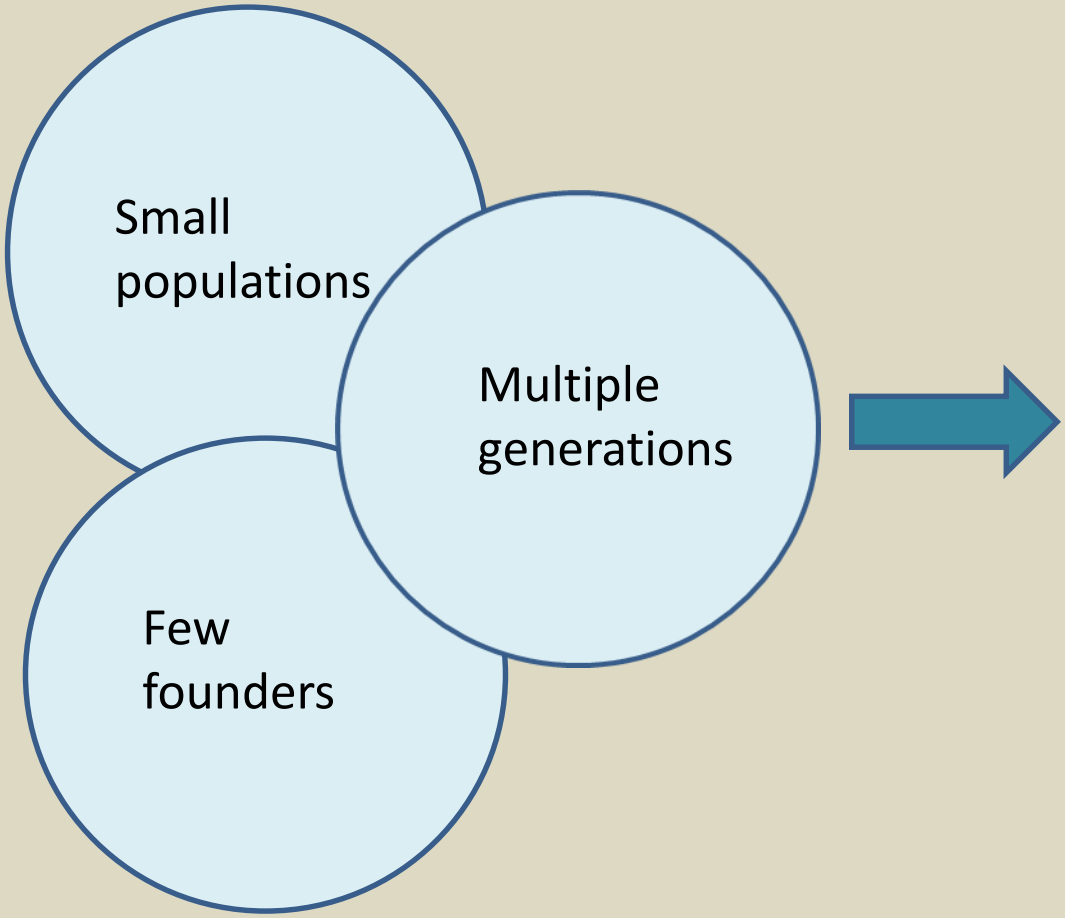
Ex situ plant conservation: GSPC Targets 3 & 8

- Seed banking is the most cost-effective and lowest risk form of ex situ conservation for threatened plants
- 1800 botanic gardens hold 2.5 million accessions, representing 18,000 taxa
- About 100 taxa exist only in botanic gardens
- Many *ex situ* collections vastly outnumber surviving wild populations
- In the US, over 80 rare taxa are being reintroduced

Exceptional Species

- Over 20% of seed plant taxa either do not produce seeds that withstand storage conditions well or produce very few seeds
- These “exceptional species” need alternative conservation methods, including tissue culture, cryopreservation, or maintenance in living collections
- Many economically important species are “exceptional” …including *Theobroma cacao*!

Challenges with Living Collections of Plants

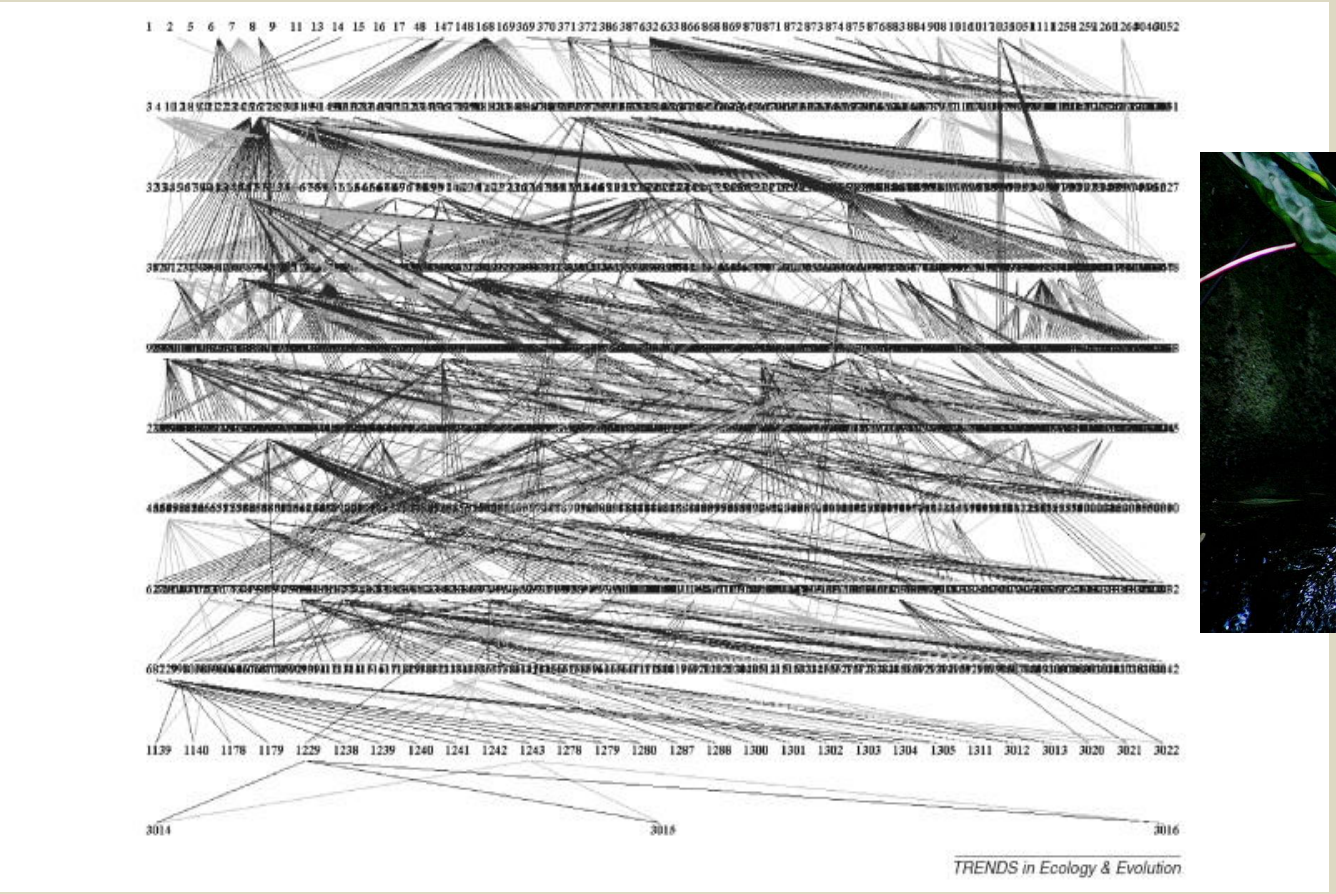


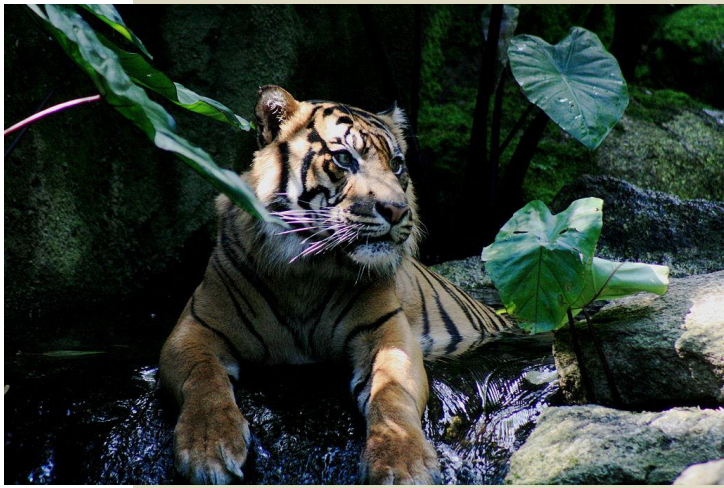
- Maintaining living plants is riskier than seed banking…they may:
  - Hybridize
  - Lose genetic diversity (drift, selection, inbreeding)
  - Adapt to cultivation
  - Acquire pathogens
  - Fail to reproduce
  - Die

Zoo Solutions

- Zoos face many of these same problems
- They rely on a metapopulation approach to cooperatively manage all ex situ individuals of a given species
- They use **International Studbooks** for each species to track pedigrees and **population management software** to determine crosses that retain as much genetic diversity as possible

Zoo Solutions





Zoo Sumatra tiger population pedigree (Pelletier et al. 2009)

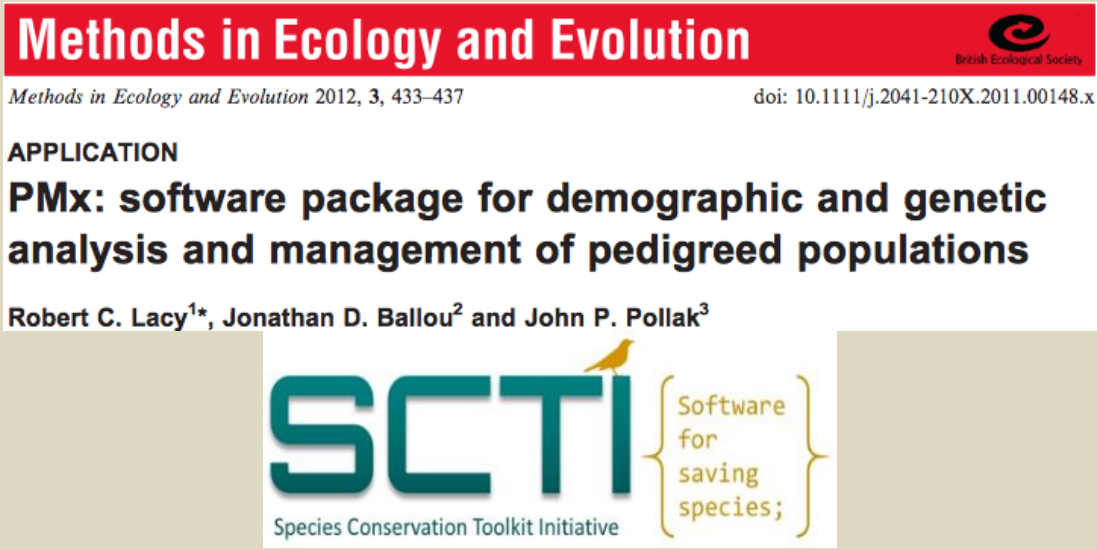
Zoos developed software to analyze pedigreed populations

PMx was developed to provide management recommendations

PMx relies on a pedigree or genetic data to make estimates of kinship (relatedness) for every individual

PMx helps minimize kinship (relatedness) to minimize loss of genetic diversity and prevent inbreeding

PMx: Pedigree analysis and management software

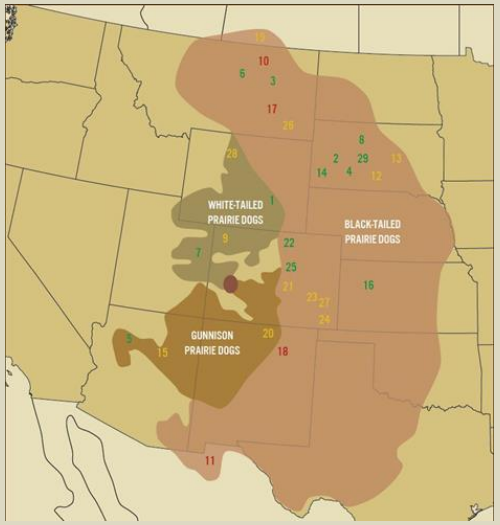


SCTI
Species Conservation Toolkit Initiative
Software for saving species;

Chicago Zoological Society
Inspiring Conservation Leadership

A Success Story: Black Footed Ferrets

- Was “Presumed Extinct”
- Then “Extinct in Wild”
  - Began with **18 individuals** (2 mothers)
    - Coordinated breeding effort with studbook
- Now “Endangered”
  - Reintroduction program resulted in more than 7,000 black-footed ferret kits births across six zoos
  - 2,600 of those animals have been reintroduced to the wild.
  - Many wild populations at 300+ black-footed ferrets and 280 in breeding facilities.



Testing Approach in Plants

- Conservation issues in living collections of plants parallel those of captive animal populations
- We have similar conservation goals
  - Maintenance of diversity
  - Producing individuals for reintroduction or display
- Are the tools Zoos have developed transferable to Botanic Gardens?

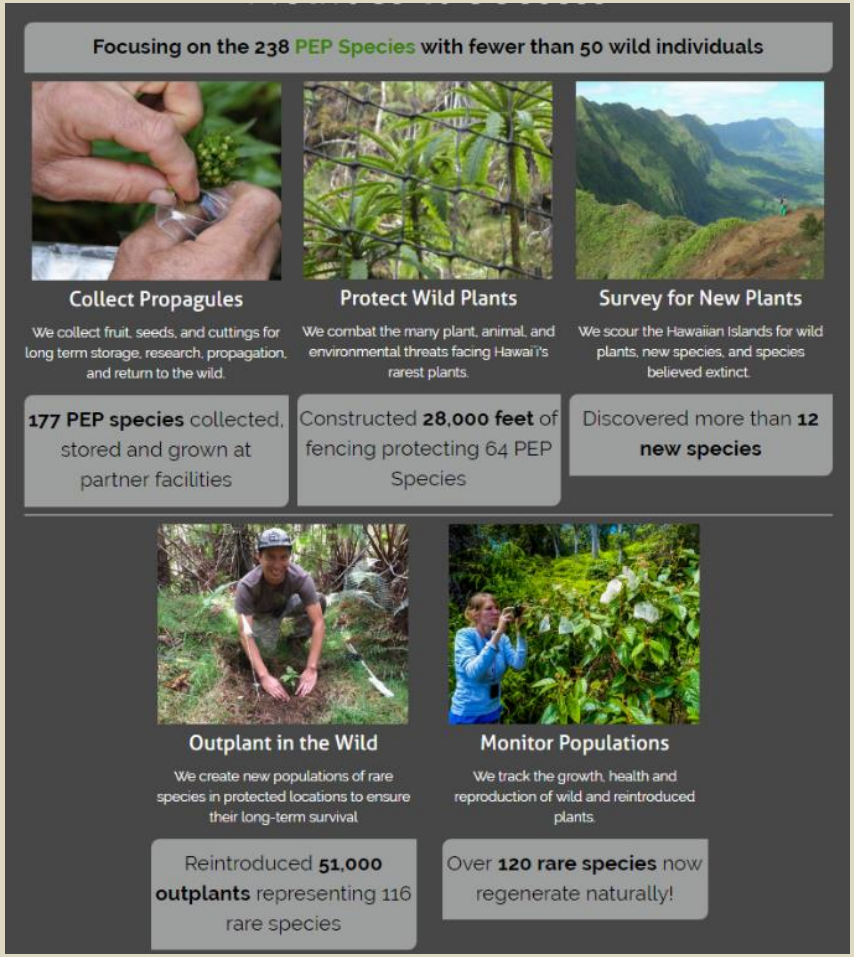
Target Plant Taxa

Plant species that could benefit from the Zoo protocol

- Species at critically low numbers
- Exceptional Species (which do not seedbank)
- Species maintained ex situ as living plants
- Species with short life cycle

In Hawaii alone there a 238 species with fewer than 50 individuals.

- Many of which could benefit from this approach



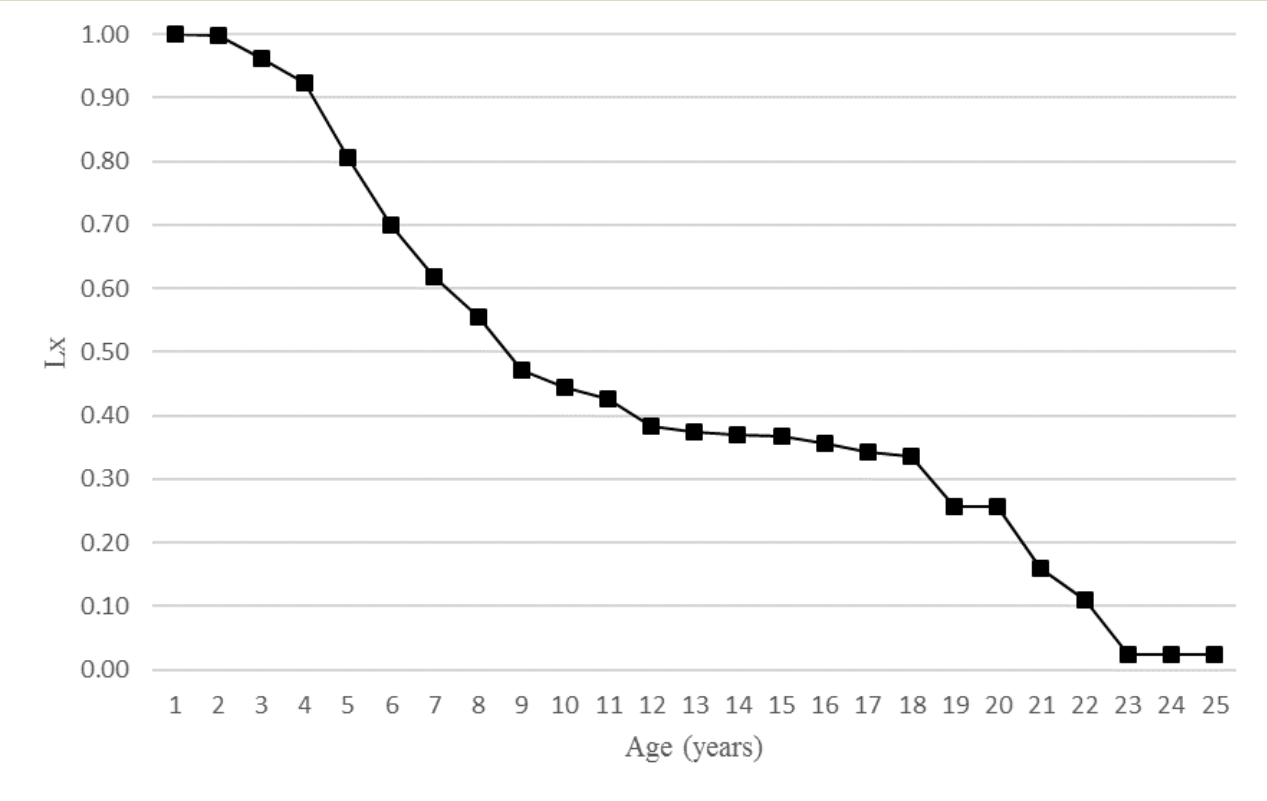
Many target taxa in Hawaii's PEP (Plant Extinction Prevention) program

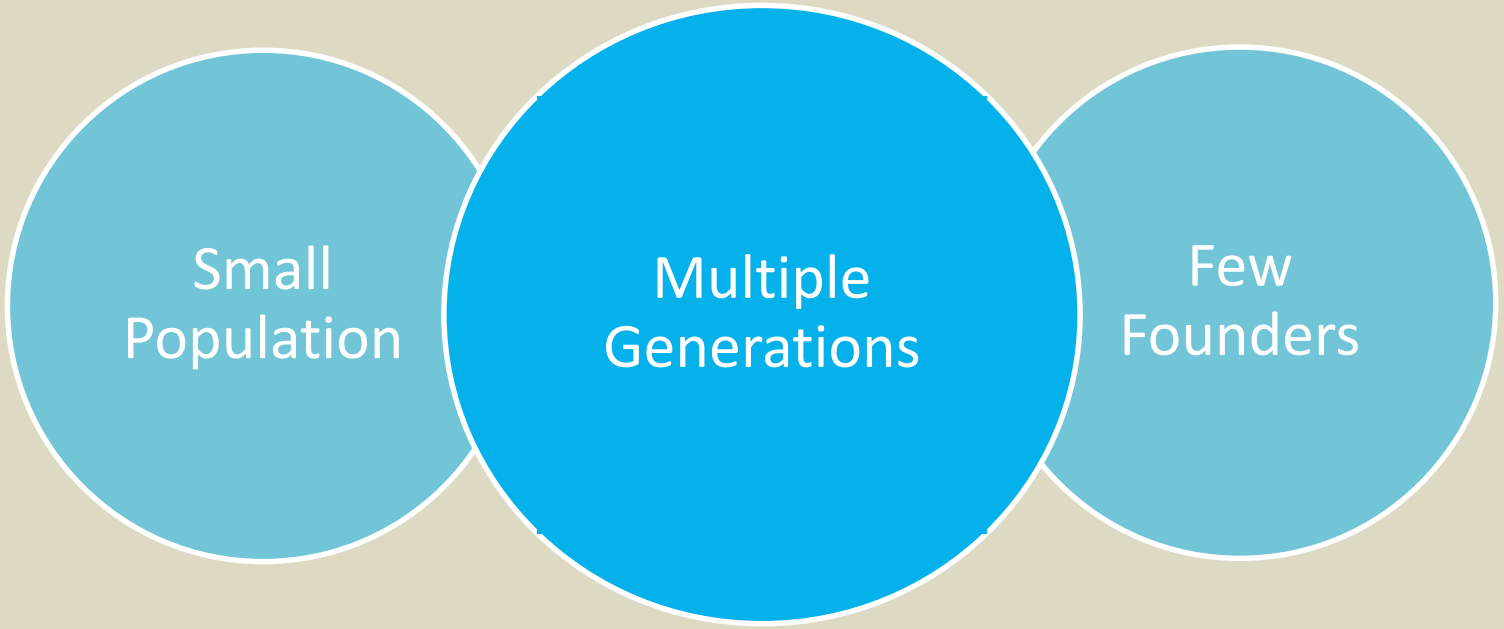
Case Study: *Brighamia insignis*

- Endemic to island of Kauai in Hawaii
- Functionally extinct in the wild
  - Hurricanes and goats have removed all but one plant
- Cultivated in at least 57 botanic gardens
  - Thanks to collectors from NTBG
  - 300-400 individuals in collections (we have data for)
- Seed can be banked for only a short period before seeds lose viability
- Short lived in cultivation
  - 10-20 year life cycle
  - Seed set is poor

Brighamia Results

- 50% of plants live 8 years, all dead by 23 years





Loss of genetic diversity will increase with the number of generations held ex situ.

What can botanic gardens do to preserve the widest possible genetic diversity?

Brighamia insiginis Approach

Genotype every individual

- Inbreeding levels
- Kinship
- Track paternity…

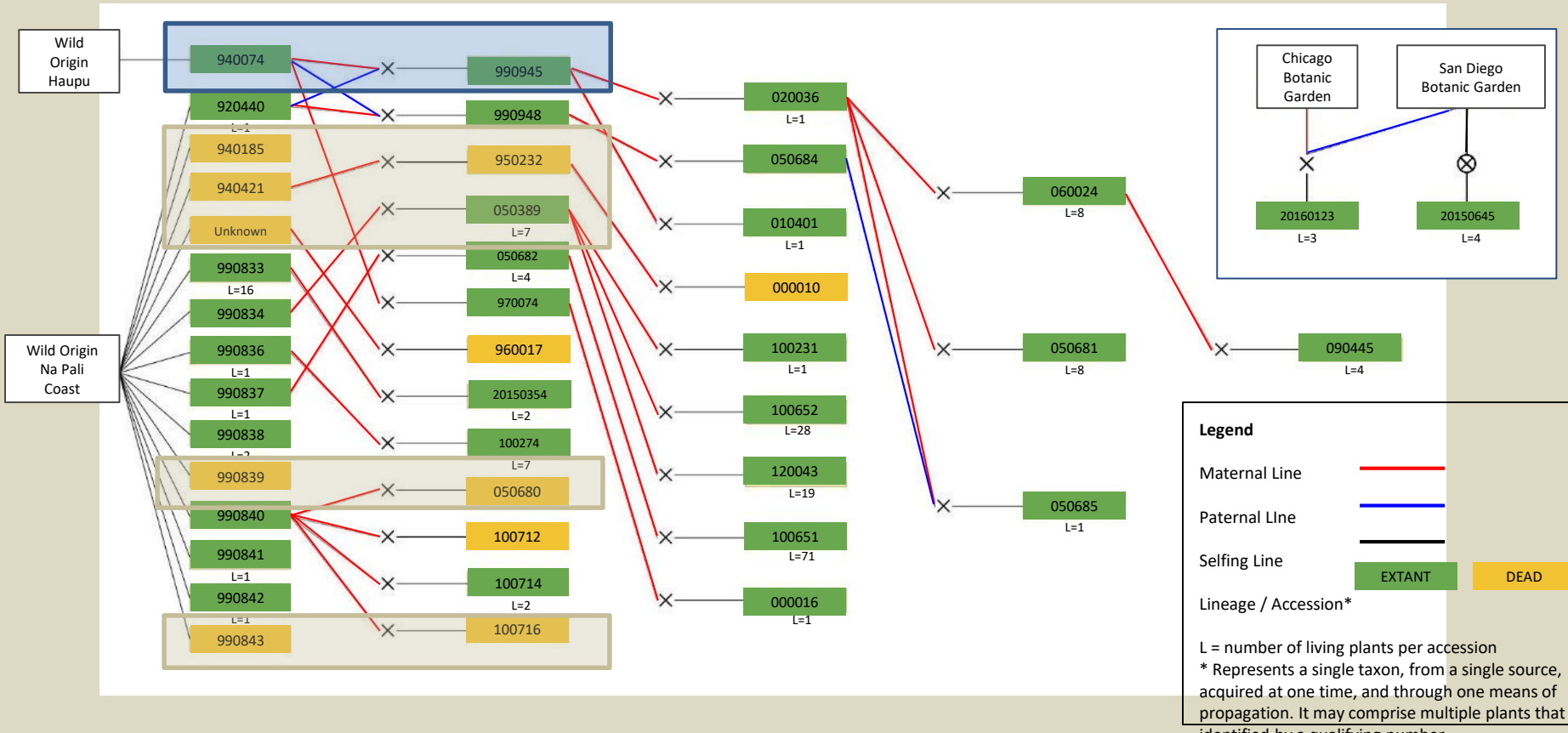
PMx to manage collections

- Identify genetically valuable (unique) individuals
- Calculate size needed to maintain diversity (given demographic factors)
- Ideal crosses to increase diversity
- Size and number to use in reintroduction

Brighamia Pedigree

- **Step 1:** Create a "studbook" with every individual in the managed population
  - There are approximately 1,400 individuals in the pedigree, but only 300 – 400 living individuals
- **Step 2:** Add probabilities for multiple possible parents
- **Step 3:** Add genetic data to make empirical estimates of kinship
  - Genetic data for 244 living individuals

Pedigree of *Brighamia insignis* accessions at National Tropical Botanic Garden



Results: Population Summary

	Pedigree only	[redacted]
population statistics		
Founders	24	
Potential founders	3	
Living animals	403	
Living descendants	193.25	
Percent pedigree known	48%	
Percent pedigree certain	48%	
genetic statistics		
percent analytic known	N/A	
Gene diversity	0.79	
Gene value	0.74	
Founder genome equivalents	2.34	
Founder genome surviving	17.1	
Mean inbreeding	0.16	
population mean kinship	0.21	
Ne/N	0.03	

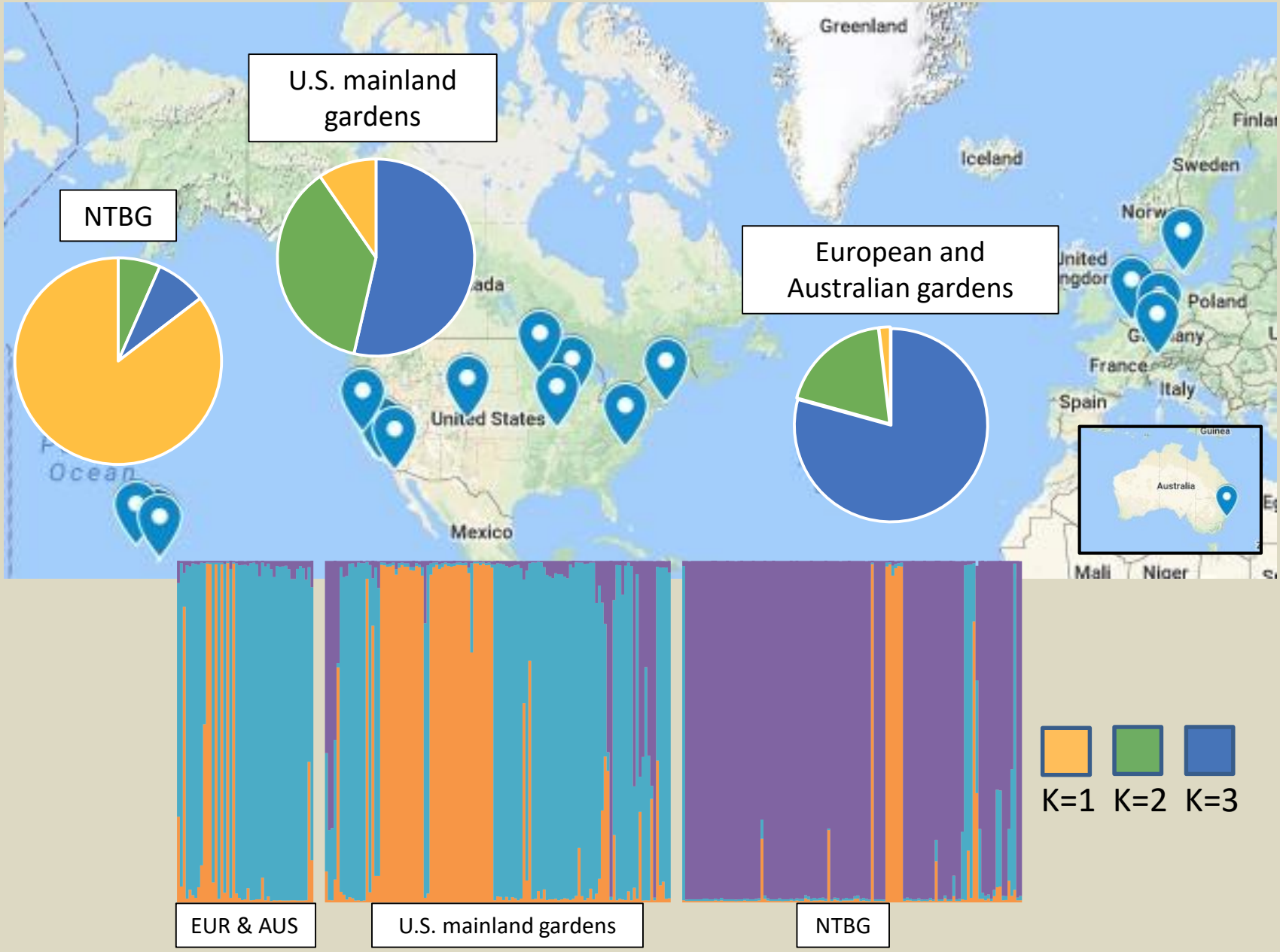
Results: Population Summary

	Pedigree only		genotypic only
population statistics			
Founders	24		N/A
Potential founders	3		N/A
Living animals	403		244
Living descendants	193.25		63.75
Percent pedigree known	48%		27%
Percent pedigree certain	48%		27%
genetic statistics			
percent analytic known	N/A		70%
Gene diversity	0.79		0.46
Gene value	0.74		0.51
Founder genome equivalents	2.34		0.93
Founder genome surviving	17.1		9.29
Mean inbreeding	0.16		0.63
population mean kinship	0.21		0.54
Ne/N	0.03		0.04

Results: Population Summary

	Pedigree only	pedigree with genotypic	genotypic only
population statistics			
Founders	24	N/A	N/A
Potential founders	3	N/A	N/A
Living animals	403	403	244
Living descendants	193.25	193.25	63.75
Percent pedigree known	48%	48%	27%
Percent pedigree certain	48%	48%	27%
genetic statistics			
percent analytic known	N/A	68%	70%
Gene diversity	0.79	0.66	0.46
Gene value	0.74	0.60	0.51
Founder genome equivalents	2.34	1.49	0.93
Founder genome surviving	17.1	14.01	9.29
Mean inbreeding	0.16	0.36	0.63
population mean kinship	0.21	0.34	0.54
Ne/N	0.03	0.03	0.04

Genetic Diversity of *Brighamia* specimens housed in botanic gardens



Results: Selecting breeding pairs and removals

Breeding pairs

						genotypic
rank	parent A	Location A	parent B	Location B	F	dGD
1	915152-A	WAF	995620/1	SSZ	-	0.017
2	915152-A	WAF	995620/1	SSZ	-	0.033
3	unknown.252	UCB	120177/0	SSZ	0.13	0.044
4	unknown.252	UCB	120177/0	SSZ	0.13	0.053
5	unknown.253	UCB	160538/a	SSZ	0.08	0.061
6	unknown.253	UCB	160538/a	SSZ	0.08	0.069
7	unknown.166	SDB	160538/c	SSZ	0.13	0.073
8	unknown.166	SDB	160538/c	SSZ	0.13	0.077
9	995620/1	SSZ	915152-A	WAF	-	0.087
10	unknown.174	SDB	160538/0	SSZ	-	0.090
11	unknown.184	SDB	160538/0	SSZ	-	0.093
12	120177/0	SSZ	unknown.252	UCB	0.13	0.098
13	160538/a	SSZ	unknown.253	UCB	0.08	0.102
14	19790159	MEI	995620/0	SSZ	-	0.102
15	19790159	MEI	995620/0	SSZ	-	0.101

Removals

		genotypic
individual	location	mk
1113	USA-NTB	0.73
090445.003	USA-NTB	0.63
090445.004	USA-NTB	0.63
060024.030	USA-NTB	0.61
020036.004	USA-NTB	0.59
020036.003	USA-NTB	0.58
100651.001	USA-NTB	0.57
100651.002	USA-NTB	0.56
100651.003	USA-NTB	0.56
100651.008	USA-NTB	0.56
100651.009	USA-NTB	0.56
100651.012	USA-NTB	0.56
100651.016	USA-NTB	0.56
100651.021	USA-NTB	0.56
100651.024	USA-NTB	0.56

Problems with use of PMx with plants

- Not all aspects of software work with non-dioecious species
- Demographic models based on an age-based approach – size or stage based models are more appropriate for most plant species
- We are working with Bob Lacy and Jon Ballou to update software to accommodate a variety of breeding systems and new demography models

Caveats for use of PMx with plants

- BG's typically track accessions, not individuals
- Lack of "studbooks" and shared plant records across botanic gardens
- Magnitude of need (50,000 plants vs. 1000 animals)

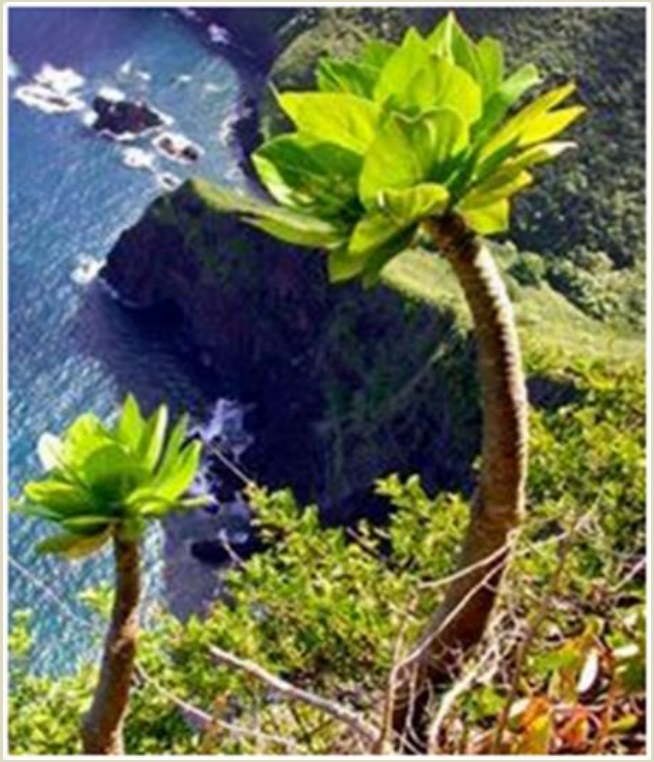
Recommendations for managing living collections of species of interest

- Track individuals not accessions
- Modify BGCI's Plant Search to enable tracking of individuals and sharing these data among botanic gardens
- Develop coordinated programs among botanic gardens to optimize management
- Refine methods to store and ship viable pollen
- Genotype founders if resources are not available to genotype other individuals

Conclusions

- Potential management improvements are significant!
- Some individuals are more valuable than others, and should be prioritized for a breeding program
- Some accessions are overrepresented, so we could transfer or remove individual plants
- Results were different based on the type of data we used…it's important to understand how the types and quality of data influence recommendations in PMx

Hopefully One Day



Successful genetic management ex situ will lead to more resilient restorations in situ, for *Brighamia insignis* and many plant other species!

The Project Team

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CHICAGO BOTANIC GARDEN

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