

PERSPECTIVES IN THEORETICAL AND HAWAIIAN ETHNOBOTANY:  
BIOCULTURAL DIVERSITY IN TWO CULTIVATED PLANTS,  
**'AWA** (*Piper methysticum* G. Foster) AND **KALO** (*Colocasia esculenta* (L.) Schott)

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## Dedication

This dissertation is dedicated to the honor  
of all the *kūpuna* (elders and ancestors)  
who have worked tirelessly to provide  
that which we have today,  
and for the generations of the future  
for whom we have so much *aloha*.

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## Abstract

The discipline of ethnobotany has accumulated an abundance of data about the diversity of ecological resource-management methodologies, but has yet to do so using standard units of measure such that cross regional comparisons can be made. As a means to address this deficiency in the discipline the theoretical “Quantum Co-evolution Unit” is described and defined from the perspective of quantum ethnobotany. It recognizes that the process of co-evolution occurs in many kinds of relationships and on various scales. One example of a co-evolutionary relationship is that of a plant and a culture with which it interacts. Such relationships are dynamic and ever changing. Changes over time in the links of the relationship between biological evolution and sociocultural evolution is a co-evolutionary process. A collection of QCUs for a linked plant and human population would be its “ethnobotanical population.” This could be measured at various points in time to quantify the changing relationships between plants and people. These models set up a structure to discuss methodologies for quantifying co-evolutionary relationships such as are seen in the evolution of ethnobotanical populations. The co-evolving relationship between ‘awa (kava—*Piper methysticum*) and Hawaiian culture is used as an example to illustrate this idea.

Both biological diversity and sociocultural diversity are important factors that should be managed to strengthen resilience in social-ecological systems. Quantum ethnobotany provides theory and models to measure links between biological diversity and sociocultural diversity for comparisons across regions. The

relationships between biological and cultural diversity can be dynamic, and cyclical between processes of co-evolution and co-extinction. The ability to measure links between biological and sociocultural diversity is provided by quantum ethnobotany. This will be useful for resource managers, policy makers, stakeholders and cultural practitioners to manage both biological and cultural diversity through co-extinction cycles for the purpose of maintaining or increasing resilience in social-ecological systems.

As a means to further examine the links between biodiversity and cultural diversity the biocultural diversity of *kalo* (*Colocasia esculenta* (L.) Schott.) in the past and the present was assessed. *Kalo* once held supreme importance in Hawaiian culture, but its status has declined after experiencing a decrease in cultivation, biodiversity, and associated cultural knowledge. There was no documentation of diversity at the peak of its cultural importance. Previous estimates of biodiversity lack any sense of a methodological approach. A new attempt was made to assess levels of biodiversity around the peak cultivation period. Results were then compared to current levels. Nomenclatural synonymy and extinction have presented some challenges which made standard methods for quantifying biodiversity not viable. A set of new tools was used to sort through a master list of 676 varietal names. A comparison of what is known from the nineteenth century and modern time periods makes it apparent that changes in biodiversity, varietal prominence, ethnonomenclature, and ethnotaxonomy have occurred. This paper discusses the direction of such trends, and postulates a new estimate for *kalo* diversity at the end of the 19th century

(approximately 100 years after the assumed peak of cultivation and diversity) to be between 368-482 distinct cultivars, while only 65-73 still exist today.

In a larger perspective, taro has been transported by humans out of its natural range into new biogeographic regions due to its cultural importance, primarily as a food source. This has resulted in repeated bottlenecks and their associated genetic drift away from the parent population. Hawaiians anciently developed taro into approximately 400 culturally distinct cultivars—all of which were named and classified—making Hawai`i a diversity center in the Pacific. From the 18<sup>th</sup> to 20<sup>th</sup> centuries Hawaiian taro underwent an extinction period with loss of perhaps 85% of diversity. In this same period, immigrant cultures and researchers were bringing new cultivars as genetic founders from different regions around the Pacific and East Asia. An AFLP study of contemporary taro diversity in Hawai`i demonstrates that genetic diversity is re-expanding. This new genetic diversity opens up opportunities to both increase functional redundancy of cultural uses of taro, and to create new plant-based traditions. This study demonstrates that this has indeed taken place. The AFLP study in conjunction with an examination of the Hawaiian ethnonomenclature system of taro demonstrates that Hawaiian ethnoclassification is based on shared morphological characteristics that are not necessarily reflective of parentage or genetic relationships.

# Table of Contents

Acknowledgements.....	iii
Abstract.....	vi
List of Tables .....	xiii
List of Figures .....	xiv
Chapter 1. Introduction .....	1
1.1 A co-evolutionary relationship between cultural and biological systems.....	1
1.2 Theoretical and Quantum Ethnobotany .....	2
1.3 Selected studies in Hawaiian ethnobotany .....	3
1.4 Taro and kava in Hawaiian culture .....	5
1.5 Important questions in ethnobotany .....	7
1.6 Literature cited.....	8
Chapter 2. The Quantum Co-Evolution Unit: An Example of ‘Awa (Kava— <i>Piper methysticum</i> G. Foster) in Hawaiian Culture .....	14
2.1 Abstract.....	14
2.2 Introduction .....	14
2.3 Changing interactions between plants and people: The example of ‘Awa ( <i>Piper methysticum</i> ) in Hawaiian culture .....	15
2.4 The need to measure changing relationships between plants and people .....	17
2.5 Quantum ethnobotany and an introduction of new concepts.....	18
2.6 Quantum co-evolution units and ‘Awa ( <i>Piper methysticum</i> ): Implications for quantifying ethnobotanical evolution .....	22
2.7 Conclusions .....	24
2.8 Literature Cited .....	26
Chapter 3. Quantifying Evolution of Cultural Interactions with Plants: Implications for Managing Diversity for Resilience in Social-Ecological Systems.....	29
3.1 Abstract.....	29
3.2 Introduction .....	29
3.2.1 Ethnobotany research and ecological resource management .....	29
3.2.2 Ecosystem resilience and biodiversity .....	30

3.2.3 <i>Social-ecological systems and resilience</i> .....	30
3.2.4 <i>Sociocultural and biological evolution</i> .....	32
3.3 Quantum Ethnobotany .....	33
3.3.1 <i>In relation to complex systems theory</i> .....	33
3.3.2 <i>Quantum co-evolution units and ethnobotanical populations</i> .....	34
3.4 Evolution of cultural interactions with plants .....	35
3.4.1 <i>Changes in composition of ethnobotanical populations</i> .....	35
3.4.2 <i>Changes in composition of ethnobotanical populations over time</i> .....	37
3.5 The cylindrical process of co-evolution and co-extinction .....	39
3.5.1 <i>Co-evolutionary process of people-plant interactions: increases in biocultural diversity</i> .....	39
3.5.2 <i>Co-extinction process of people-plant interactions: Decreases in biocultural diversity</i> .....	44
3.5.3 <i>The back-loop cycle: reorganizing diversity between co-extinction and co-evolution cycles</i> .....	45
3.5.4 <i>Maintaining cultural diversity through cyclical evolutionary processes</i> .....	47
3.6 Conclusions .....	47
3.6.1 <i>Applications of quantum ethnobotany for conservation of biodiversity</i> .....	47
3.6.2 <i>Proposed hypotheses</i> .....	48
3.7 Literature Cited .....	49
CHAPTER 4. <i>Kalo</i> [Hawaiian taro, <i>Colocasia esculenta</i> (L.) Schott] Varieties: An assessment of nomenclatural synonymy and biodiversity.....	54
4.1 Abstract.....	54
4.2 Introduction .....	54
4.3 Methods.....	58
4.3.1 <i>Surveys of nomenclatural diversity</i> .....	59
4.3.2 <i>Biodiversity assessment</i> .....	62
4.3.3 <i>Rigor assessments for cited authorities</i> .....	62
4.3.4 <i>Status assessments for once-prevalent cultivars</i> .....	63
4.4 Results .....	63

4.4.1 Previous estimates of synonymy .....	63
4.4.2 Field surveys .....	64
4.4.3 Rigor of previous diversity assessments .....	65
4.4.4 Taxonomic trends.....	68
4.4.5 Trends in nomenclature .....	70
4.5 Discussion.....	71
4.5.1 Hawaiian taxonomic systems and synonymy .....	71
4.5.2 The complexity of synonymy and previous estimates of kalo diversity .....	72
4.5.3 Taxonomic and nomenclature trends .....	76
4.5.4 Nomenclature trends at the species and subspecies levels .....	76
4.5.5. The relationship of reduplication and backformation in synonymy .....	77
4.5.6 Changes in nomenclature and taxonomy .....	78
4.5.7 Decreases in traditional diversity and effects on cultural diversity .....	80
4.5.8 Increases in diversity via modern hybrids .....	81
4.6 Conclusions .....	81
4.7 Literature Cited .....	83
CHAPTER 5. Using molecular characterization of genetic variability and relationships of taro [ <i>Colocasia esculenta</i> (L.) Schott] in Hawai`i, with amplified fragment length polymorphism, to understand phylogeny, trends in ethnobotanical evolution, and ethnotaxonomy.....	88
5.1 Abstract.....	88
5.2 Introduction .....	89
5.2.1.1 Genetic diversity of taro in Hawai`i .....	89
5.2.1.2 Utilization of AFLP .....	90
5.2.2.1 Links between the genetic diversity of taro and the diversity of practices in Hawaiian culture as evidence of ethnobotanical evolution.....	92
5.2.3.1 Insights from trends in ethnonomenclature and ethnoclassification .....	93
5.3 Materials and Methods.....	94
5.3.1.1 Plant material and DNA isolation .....	94
5.3.1.2 AFLP analysis.....	97

5.3.1.3 Data analysis.....	98
5.3.2.1 Determining links between biodiversity and cultural diversity, and whether or not ethnobotanical evolution is taking place .....	98
5.3.3.1 Comparative analysis of AFLP phylogenies and Hawaiian classification .....	99
5.4 Results.....	99
5.4.1.1 The AFLP profile .....	99
5.4.1.2 Genetic relationships within and among taxa, and comparisons to assumptions of bottlenecking .....	100
5.4.2.1 Ethnobotanical profiles for extinct Hawaiian cultivars and non-Hawaiian cultivars.....	100
5.4.3.1 Comparisons between genetic phylogeny and ethnoclassification .....	101
5.5 Discussion.....	105
5.5.1.1 Genetic diversity of taro in Hawai`i reflects human migration patterns .....	105
5.5.1.2 Genetic relationships of taro in Hawai`i in light of assumptions about genetic bottlenecking .....	106
5.5.1.3 The functionality of AFLP analysis .....	107
5.5.2.1 Historic collapse and re-expansion of genetic diversity of taro in Hawai`i, with correlations to cultural diversity.....	107
5.5.3.1 Determining pathways for diversification, and whether or not Hawaiian classification of taro is founded in genetic relationships.....	109
5.6 Conclusions .....	110
5.6.1.1 Value of AFLP in research.....	110
5.6.2.1 Co-evolutionary relationship between humans and plants has affects upon plant evolution .....	110
5.6.3.1 Relationship between ethnoclassification and genotypic relationships.....	111
5.7 Literature Cited .....	112
Chapter 6. Conclusions .....	116
6.1 Summary of postulates regarding theoretical ethnobotany .....	116
6.2 Summary of hypotheses, results, and emergent theory .....	116
6.3 Implications for future research .....	120
6.4 Proposed theory regarding human interactions with plants .....	121

6.5 Literature Cited .....	121
Appendix 1. List of <i>Kalo</i> Varietal Names .....	123
Appendix 2. Lab notes for taro AFLP study.....	136
Appendix 2A: CTAB Protocol.....	142
Appendix 2B: AFLP Protocol for ABI Genetic Analyzer 3100 .....	145

## List of Tables

<b>Table 2- 1.</b> Taxa (folk or biological) and human population scales that can be measured with a Quantum Co-evolution Unit.....	20
<b>Table 2- 2.</b> The Quantum Co-evolution Units of `awa in pre-contact Hawaiian culture as gleaned from Winter (2004). .....	23
<b>Table 3- 1.</b> Respective QCU frequencies of a highly simplified and hypothetical ethnobotanical subpopulation that focuses on plants involved with religious offerings as measured between two intervals of time .....	39
<b>Table 3- 2.</b> Classifications of co-evolutionary relationships, the respective state of the ethnobotanical population, and the potential insights that can be gained for managing diversity.....	40
<b>Table 4.1.</b> Publications and manuscripts that documented <i>kalo</i> ( <i>Colocasia esculenta</i> (L.) Schott.) diversity in the Hawaiian islands.....	64
<b>Table 4.2.</b> The extant diversity of <i>kalo</i> ( <i>Colocasia esculenta</i> (L.) Schott) representing documented cultivars, 20 <sup>th</sup> century Hawaiian X Hawaiian hybrids, and undocumented cultivars *strongly-to-debatably assumed to be Hawaiian.....	65
<b>Table 4.3.</b> Cultural and scientific rigor of data sources used to qualitatively assess <i>kalo</i> ( <i>Colocasia esculenta</i> (L.) Schott.) diversity in the Hawaiian islands.....	68
<b>Table 4.4.</b> Classification of the Hawaiian <i>kalo</i> ( <i>Colocasia esculenta</i> (L.) Schott.) cultivar, 'Apuwai kea, following Berlin's (1992) general system.....	69
<b>Table 4.5.</b> Trends in nomenclature for 255 <i>kalo</i> ( <i>Colocasia esculenta</i> (L.) Schott.) sub-species.....	72

<b>Table 4.6.</b> Importance of <i>kalo</i> ( <i>Colocasia esculenta</i> (L.) Schott.) varieties based on number of citations between 1879 and 1940 verses current observations through field surveys.....	79
<b>Table 4.7.</b> <i>Kalo</i> ( <i>Colocasia esculenta</i> (L.) Schott.) color group-names and possible synonyms.....	80
<b>Table 4.8.</b> The most important <i>kalo</i> ( <i>Colocasia esculenta</i> (L.) Schott.) groups based on number of recorded sub-species names.....	81
<b>Table 5.1.</b> The most diverse groups of taro in the Hawaiian classification system from Winter (2012), with their interpreted meanings and named cultivar diversity.....	94
<b>Table 5.2.</b> <i>Colocasia</i> germplasm accessions cultivated in Hawai'i and included in the study.....	95
<b>Table 5.3.</b> Selective Primer Combinations used in selective amplification for an AFLP analysis of the genetic diversity of Hawaiian Taro ( <i>Colocasia esculenta</i> (L.) Schott).....	98
<b>Table 5.4.</b> Ethnobotanical profiles for selected cultivars (extinct Hawaiian, and introduced non-Hawaiian) of taro ( <i>Colocasia esculenta</i> ).....	101

## List of Figures

<b>Figure 2.1.</b> A hypothetical Quantum Co-evolution Unit (QCU) as it relates to the co-evolutionary relationship between plants and human cultures.. ..	199
<b>Figure 2.2.</b> An example of an ethnobotanical population.....	21
<b>Figure 2.3.</b> A hypothetical ethnobotanical subpopulation based on <i>Piper methysticum</i> in Hawaiian culture as measured between two time periods.. ..	24
<b>Figure 3.1.</b> An example of a QCU cleavage event and the subsequent substitution of a subunit via a linkage event.....	36
<b>Figure 3.2.</b> A highly simplified and hypothetical ethnobotanical subpopulation measured at two intervals in time.....	38

- Figure 3.3.** Increase in abundance of domesticates over time (After Rindos 1984, Figure 5.3) [ $\mu$  is the relative abundance of domesticates as a fraction of the total possible in the environment.  $\lambda$  Is the logarithm of the relative increase in domesticates.].....41
- Figure 3.4.** Relative contribution ( $r$ ) of varieties of domesticated plants ( $D$ ) verses wild plants ( $W$ ) over time as a function of their relative abundance ( $\mu$ ) in the environment. (Adapted from Rindos 1984, Figure 5.2).....42
- Figure 3.5.** Co-evolution of a cultivated (or otherwise managed) plant and the culture that cultivates it as depicted in an outwardly expanding spiral.....43
- Figure 3.6.** The abating spiral depicting the co-extinction of plants and the sociocultural system that manages it.....45
- Figure 4.1.** Classification of the Hawaiian kalo (*Colocasia esculenta* (L.) Schott.) cultivar, 'Apuwai kea, as an example in the context of other taxa following Berlin's (1992) system.....69
- Figure 4.2.** Two examples of the relationship between reduplication and backformation in species-level nomenclatural synonymy of *kalo* (*Colocasia esculenta* (L.) Schott.).....78
- Figure 5.1.** A rooted, phylogenetic tree of contemporarily-occurring taro (*Colocasia esculenta*) in Hawai'i using the neighbor-joining algorithm and Nei-Li distance measure, with zuiki (*C. gigantea*) as an outgroup.....102
- Figure 5.2.** A bootstrap consensus of the phylogenetic tree of contemporarily-occurring taro (*Colocasia esculenta*) in Hawai'i using the neighbor-joining algorithm and Nei-Li distance measure, with zuiki (*C. gigantea*) as an outgroup.....103
- Figure 5.3.** A rooted, phylogenetic tree of Hawaiian taro (*Colocasia esculenta*) exclusively, using the neighbor-joining algorithm and Nei-Li distance measure....104

# Chapter 1. Introduction

## 1.1 A co-evolutionary relationship between cultural and biological systems

The concept of evolution in socio-cultural systems has not been as accepted as the concept of evolution in biological systems despite having been addressed by many scholars over the last 50 years (Jantsch 1980; Kaplan and Manners 1972; Sahlins 1976; Sztompka 1994; Trigger 1998; White 1959). Several of these works stand out for their emphasis upon the environment and particular biological resources. White's law (White 1959), a cornerstone concept for the evolution of culture, implies that cultural evolution is related to changing intensities of interactions with the environment (as measured by efficiency of capturing and using environmental energy). This points to the material (i.e., plant) basis of culture as the key to understanding change through time. Conklin (1961) showed, moreover, that humans modify ecosystems in specific and patterned ways that are dynamic yet predictable. Rappaport (1984) demonstrated, in addition, that the fundamental basis for change within cultures and their environments are specific relationships that develop between people and particular plant and animal species. Many others have since addressed the question of the role of plants and animals in cultural change and evolution of human populations through time (Boyd and Richerson 1985; Harris and Hillman 1989), but surprisingly none appear to have scaled back from the big picture to ask what these fundamental measurable units may be. Richerson and Boyd (2005) acknowledge that there is a fundamental difference between the bits of culture and the genes of inheritance, and point out that the two are not inherited in the same "faithfully replicated" way. They go on to point out that:

Skeptics who distrust Darwinism are common, particularly in the social sciences. But Darwinism is not inherently an individualist, adaptationist footpad sneaking into the social sciences to explain everything by genetic reductionism. Nor does it signal a return to progressive, Eurocentric ideas of the past. A great variety of substantive theories arise when the all-important details are specified. Some models end up looking a lot like rationale choice; and in others, arbitrary cultural differences can arise from the dynamics of interacting cultural elements. (Richerson and Boyd 2005:60)

The concept of biological co-evolution has been addressed by many scholars over the past four decades, especially as relevant to the relationship between plants and

animals and to the idea that this process has been a major driver in the generation of the Earth's biological diversity (Ehrlich and Raven 1964; Herrera 1982; Janzen 1966; Rausher 2001). Scholarship has been slower to recognize the existence of a co-evolutionary relationship between plants and the cultures that interact with them. This dissertation addresses these issues by proposing both hypotheses and theory (Chapters 2 – 3), and by using both new methods (Chapter 4) and well-established methods (Chapter 5) to explore more deeply the co-evolutionary relationship between plants and cultures.

## **1.2 Theoretical and Quantum Ethnobotany**

An *a priori* review of the history of modern science suggests that scientific theory emerges as a discipline matures—when researchers add to already-relatively large bodies of data, sharpen previously blunt tools/methods, and shift from a narrow perspective to a systems perspective. As the discipline of ethnobotany has matured these three criteria have been met and theory has been produced (Prance 1987, McClatchey and Bridges 2005, Bridges and McClatchey 2009). One of the contributions of this dissertation to the discipline of ethnobotany is in this regard.

A main focus of ethnobotany is examining human interaction with the natural world (Salick *et al.* 2003; Prance *et al.* 2007). However, being a relatively young discipline, ethnobotany is still developing the rigor of well-articulated theory—a foundational aspect of any scientific field. This dissertation proposes new theory in the hopes of contributing to a stronger foundation of ethnobotany as a discipline. The theory addressed herein deals with the identification and measurement of the most fundamental units of interaction between plants and people, as well as the potential applications of such research. This new theoretical approach was dubbed, “quantum ethnobotany,” by McClatchey and Bridges (2005) in a symposium at an annual meeting of the Society for Economic Botany. The concept was born out of ideas from and conversations with other leading ethnobotanists, anthropologists, and biologists such as, Dan Moerman (former editor of *Economic Botany*); Scott Atran (Atran 1990), Brent Berlin (Berlin 1993), Michael Balick (Brosi *et al.* 2007), Hal Conklin (Conklin 1963), and others. The first publications to use the term were Winter and McClatchey (2008), and Winter and McClatchey (2009) which are chapters 2 and 3 in this dissertation.

As reflected in the coming chapters, this dissertation emphasizes the concept that humans are a part of, not separate from nature (Balée 2006), supporting the views established by Berkes and Folke (1998), and Berkes *et al.* (2003) which hold that

social and ecological systems are linked, and that delineations between social and natural systems are arbitrary and artificial. Such human-in-nature systems are referred to as “social-ecological systems”. Undoubtedly, there are countless social-ecological systems that could be chosen from around the world to model the above expressed theories. There is also a broad spectrum of intensification when it comes to human management of natural resources. In order to test the validity of the concepts and theories discussed above a small, but complete, social-ecological system would be needed. Islands are excellent examples to discuss system function for several reasons. Some islands are big enough to possess all the biological, ecological, chemical and physical processes needed for complete system study, but yet small enough that the complexity of such systems is perceivable (Fosberg 1962, Vitousek 2004).

The Hawaiian Islands group has been selected as a model in this discussion for various reasons. Being the most isolated land mass in the world it has become the textbook example for adaptive radiation (Campbell and Reece 2004, Raven *et al.* 2005) due to the amazing evolutionary events that created unique biodiversity among its flowering plant, avian, and invertebrate taxa. It also possesses an indigenous culture that has a recorded history in its own language and from its own perspective (Desha 2000, Handy and Pukui 1972, Iʻi 1959, Kamakau 1976, 1991, Kepelino 2007, Malo 1951). Furthermore, it possesses a research university (University of Hawaiʻi) and other research institutions (National Tropical Botanical Garden, B.P. Bishop Museum), all of which have multinational collaborations, and collectively produce high quality research in all relevant areas associated with this dissertation: ethnobotany, botany, biology, ecology, history, indigenous studies, etc. To explore the theories expressed above, this dissertation will focus on the evolution of components in the social-ecological system occupied and managed by the Hawaiian culture (Chapters 2 – 4).

### **1.3 Selected studies in Hawaiian ethnobotany**

The first to substantively document the relationships to, and roles of, plants in the Hawaiian culture were native Hawaiian historians of the 19<sup>th</sup> century—whose works were eventually published as books in the 20<sup>th</sup> century (Desha 2000, Iʻi 1959, Kamakau 1976, 1991, Malo 1951). In the 20<sup>th</sup> century archaeologists, anthropologists, and ethnographers contributed much to our understanding of traditional Hawaiian culture. Much of this research came out of the Bishop Museum. Terangi Hiroa (a.k.a., Peter S. Buck) served as the museum’s director from 1936-1951, and made valuable contributions to the documentation of Hawaiian material

culture (Hiroa 1957), and set the museum on a course of a strong focus in ethnography in the ensuing decades. Most notable among Bishop Museum's researchers during this period was, Mary Kawena Pukui, who surpassed all others in her attempts to document Hawaiian culture using modern scholarly approaches. Although she did not focus solely on ethnobotanical relationships, the role of plants in Hawaiian culture up to and including her lifetime was paramount; and her publications are seminal in the field of Hawaiian ethnobotany (Handy et al. 1972, Pukui 1983, Pukui and Elbert 1986).

However, while Bishop Museum ethnographers were prolifically publishing their research in the 20<sup>th</sup> century, others were diligently creating volumes of manuscripts, the vast majority of which were never published. Most notable of these were the scholarly partners of Henry E.P. Kekahuna and Theodore Kelsey who collectively produced countless manuscripts in both Hawaiian and English. Their work is scattered between the Bishop Museum Archives, the State of Hawai'i Archives, and private collections. A research associate of Kelsey eventually published two selected compilations of their work (Gutmanis 1976, 1983). The remainder of their unpublished manuscripts represents a virtual treasure trove of untapped cultural history.

It wasn't until the end of the 20<sup>th</sup> century that Hawaiian ethnobotany emerged as an area of research within a scientific discipline. Dr. Isabella Aiona Abbott was first native Hawaiian to approach documenting the relationships between Hawaiians and plants from a perspective of biological science. From a scientific standpoint her works (Abbott 1984, 1992) are among the first publications focused on Hawaiian ethnobotany. During her tenure at the University of Hawai'i at Mānoa she inspired and mentored several emerging Hawaiian ethnobotanists (Aiona 2003, Ili 2000, Napoleon 2004, Pang 2003).

However, not only researchers working within of the University of Hawai'i system were making valuable contributions to Hawaiian Ethnobotany towards the end of the 20<sup>th</sup> century and into the emerging 21<sup>st</sup> century. Paleoecology (Burney and Kikuchi 2006) and the latest carbon-dating techniques (Wilmshurst et al. 2010) have contributed to a new understanding of settlement patterns in Eastern Polynesia, including Hawai'i. Patrick Kirch, has combined the latest archaeological methods with an examination of the development of the Hawai'i civilization to reshape our understanding of ancient settlement patterns in Hawai'i at a detailed level (Kirch 2012), but while his archaeology seems sound, some Hawaiian scholars, including myself, feel that some of his conclusions (Kirch 1985) have been founded in a flawed understanding of societal structure. In other areas, some of the most cutting edge advances in our understanding of the how ancient Hawaiian society

functioned on a systems level have come from an interdisciplinary approach between archaeology, botany, and ecology. Notable among these is the Vitousek BioGeoChemistry Lab at Stanford University, and its collaborations (e.g., Ladefoged et al. 2009) which have completely reshaped our understanding of the underappreciated role that sweet potato (*Ipomoea batatas* (L.) Lam.) played in sustaining the Hawaiian civilization. Furthermore, the new technology of geospatial modeling has also given us a valuable insight into the extent and limits of intensified agriculture in Hawai'i (Burton 2011, Kurashima & Kirch 2011, Müller et al. 2010). As we move further into the 21<sup>st</sup> century, the field of Hawaiian ethnobotany will continue to utilize new, multidisciplinary methods (McElroy 2012, Wichman 2012) and the latest scientific approaches, such as the GIS technologies described above, to gain a better understanding of the dynamic relationship between the Hawaiian culture and plants of the Hawaiian Islands.

#### **1.4 Taro and kava in Hawaiian culture**

Taro (*Colocasia esculenta* (L.) Schott) and kava (*Piper methysticum* G. Foster), are important plants in the global economy. Taro was the fifth most-produced root crop in the world during 2010, with global production of 9.0 billion kilograms (Food and Agriculture Organization of the United Nations, 2010). It is one of the most important staple crops in the Pacific Islands, and is grown widely in Africa, Asia, the Caribbean, and South America (Plucknett et al. 1970). It is a non-graminaceous monocot consumed primarily for its starchy corm (Plucknett et al. 1970). In addition, taro leaves serve as a vegetable, providing good sources of dietary fiber and vitamin C (Ferguson et al., 1992). Kava, on the other hand, is important throughout Oceania for its medicinal qualities, and its active ingredients are sought after by the pharmaceutical/nutraceutical industry (Lebot et al. 1997) for processing and marketing in the United States and Europe.

These two cultivated plants are known in Hawaiian as *kalo* and *`awa* respectively, and are henceforth referred to as such. They were originally brought to the Hawaiian Islands by the ancient Polynesian founders (Abbott 1992) no later than 1,000 years B.P. (Kirch 2012). Anciently, *kalo* was the most important cultivated food plant (Handy et al. 1972). *`Awa* was the most important cultivated non-food plant due to its significance in the ancient religion, but also for its medicinal properties (Pukui n.d., Winter 2004). Because of the paramount importance of these two cultivated plants to the Hawaiian culture, these relationships serve as ideal case studies to examine ethnobotanical evolution more closely.

*Kalo* is connected to origin stories of the Hawaiian culture (Handy et al. 1972, Kame‘eleihiwa 1992) and is, therefore, often considered to be the most important crop plant from that perspective, at least on a symbolic level. On a practical level its prominence is rivaled only by ‘*uala* or sweet potato (*Ipomoea batatas* (L.) Lam.) (Handy 1940, Handy et al. 1972, Ladefoged et al. 2009). This is true especially in the era prior to contact with foreigners in 1778 (Handy 1940, Handy et al. 1972, Kamakau 1972, 1976, Kame‘eleihiwa 1992). Contemporarily, *kalo* continues to be held in the utmost level of respect among many staunch cultural practitioners, although its general prominence in the Hawaiian culture as a whole is no doubt less than in previous generations. This is due, in large part, to changes in land tenure and socioeconomic systems which resulted in a loss of cultural identity (Kame‘eleihiwa 1992, Trask 1999). Because of the glaring losses of both biodiversity and associated cultural knowledge it is important to gain a more refined understanding of *kalo* diversity at its height of diversity for several reasons, not the least of which includes quantifying its importance to the Hawaiian culture and gaining insight into the horticultural skills of the ancient Hawaiians.

‘*Awa* is traditionally consumed as a ritual or ceremonial drink made by Pacific island peoples, but has recently gained global interest for its medicinal and psychoactive properties. It is traditionally prepared by straining its ground-up roots in water (Lebot et al. 1997). At the height of Hawaiian civilization, ‘*awa* had gained such prominence within the culture that it was used in almost all aspects of life ranging from religious, medicinal, social, to political roles. However, beginning in the 1820s with the arrival and influence of Christian missionaries, usage of ‘*awa* began to decline because of its strong relationship with the traditional religious practices that were labeled “pagan.” Concurrently, as the traditional political system broke down through the process of colonization by foreign powers, usage of ‘*awa* in political gatherings waned. As colonization continued, the knowledge of traditional herbal healers, along with the plants that they used, including ‘*awa*, were discredited. During this same period, the social drink of the Hawaiian people shifted from ‘*awa* to alcohol. Eventually disappearing from everyday use, ‘*awa* was recognized by only a handful of individuals in a few abandoned and isolated patches deep in the mountains—remnants from ancient plantings (Winter 2004).

Due to changes in land tenure, socioeconomic systems, religion, politics and more the prominence of the role that these plants play in Hawaiian culture has been dynamic and changing in the past 200 years—including a period of waning and a period of resurgence (Kanahele 1979). This dissertation uses these two plants to explore methods for quantifying co-evolving relationships between people and

plants on various levels in order to gain a more complete understanding of Hawai`i's biocultural history.

### **1.5 Important questions in ethnobotany**

The questions asked and addressed in this dissertation are done so at the relatively small-scale perspective of Hawaiian ethnobotany, but are related to some large, over-arching questions in the discipline that remain to be resolved adequately. The source of the questions that I ask herein stem from personal questions I had many years ago of myself, my identity, and the context of the quickly-evolving traditions that I grew up in during my formative years.

I grew up in Hawai`i, was educated in an institutionalized, American system. This included being taught by an approach to curriculum, in schools and on field trips (to museums and botanical gardens), that always referred to the Hawaiian culture in the past tense, as if it no longer existed. One could only speculate as to the reason for this, but its affects were profound. As traditions started to be reclaimed by cultural practitioners during the Hawaiian renaissance from the 1970s-1990s there was a tendency for both Hawaiians and non-Hawaiians alike to label practices as either "authentic" or "inauthentic". I was a part of that. But I began to ask questions like, "Is there such a thing as an authentic practice?" and "Is it ok if traditions evolve?" The journey I took to answer these questions for myself led me down a path in academia, and this dissertation is the result. Some of the questions that this dissertation attempts to address are the following:

- What is ethnobotanical evolution, and how can it be measured?
- How do cultures and plants reciprocally influence each other's evolutionary trajectory?
- If a human population enters into a co-evolutionary relationship with a plant population in a period of diversification, then is it destined to be tied to a co-extinction process in a time of decline?
- If the links between biodiversity and cultural diversity are understood, then what are the applications for resource managers and policy makers who aim to preserve and perpetuate biocultural diversity?
- Are there patterns to synonymy in ethnonomenclature systems?
- If a biogeographic region is recognized as a center of diversity for a particular cultivated species, how many taxa are recognized by that culture?
- How closely does a culture's cognition of biodiversity reflect actual genetic relationships between biological taxa?

The above bullets represent the kinds of questions that the discipline of ethnobotany aims to address. In the following chapters of this dissertation, they will be addressed using models and examples from Hawaiian ethnobotany.

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## Chapter 2. The Quantum Co-Evolution Unit: An Example of 'Awa (Kava—*Piper methysticum* G. Foster) in Hawaiian Culture<sup>1</sup>

### 2.1 Abstract

The process of co-evolution occurs in many kinds of relationships and on various scales. One example of a co-evolutionary relationship is that of a plant and a culture with which it interacts. Such relationships are dynamic and ever changing. Researchers have discussed this concept and its implications for decades, yet no quantifiable unit or standardized scale has been accepted with which to measure this change. The theoretical “quantum co-evolution unit” (QCU) is proposed as the smallest measurable scale of interactions between plants and people. A collection of QCUs for a linked plant and human population would be its “ethnobotanical population.” This could be measured at various points in time to quantify the changing relationships between plants and people. These models set up a structure to discuss methodologies for quantifying co-evolutionary relationships such as are seen in the evolution of ethnobotanical populations. The co-evolving relationship between 'awa (kava—*Piper methysticum*) and Hawaiian culture is used as an example to illustrate this idea.

### 2.2 Introduction

As discussed in Chapter 1, the co-evolutionary process in biology has long been recognized, but examining aspects of human culture as the co-evolutionary partner of biological taxa has yet to happen on a quantifiable level. Chapter 2 focuses on the ethnobotanical interactions between a plant (as a member of a taxon that may be a species, variety, population, or other) and a person (as a member of a human culture). In the course of this chapter we will introduce the following three concepts into the theoretical realm of quantum ethnobotany:

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<sup>1</sup> This chapter has been published in a peer review journal: Winter K. & McClatchey W. 2009. The Quantum Co-evolution Unit: An Example of 'Awa (*Piper methysticum* G. Foster) in Hawai'i. *Economic Botany* 63:4, 353-362.

1. The Quantum Co-Evolution Unit (QCU),
2. Quantum Co-Evolution Unit profiles (QCU profiles), and
3. Ethnobotanical populations and ethnobotanical subpopulations.

In order to illustrate these three concepts this chapter discusses a specific example of the relationship between plants (kava, *Piper methysticum* G. Foster) and people (native Hawaiians) in contemporary Hawaiian communities. Within this structure, we produce a set of hypotheses that we hope will point to future theoretical and applied research.

### **2.3 Changing interactions between plants and people: The example of 'Awa (*Piper methysticum*) in Hawaiian culture**

Hawaiian civilization began to change drastically after contact with foreign powers—starting with Captain Cook in 1778. These early European explorers, along with the whalers who soon followed, introduced and promoted alcohol consumption. This practice quickly spread and undoubtedly affected the culture in major ways. Cultural erosion continued with the arrival of missionaries in 1820, and accelerated with the subsequent colonization process that followed until the culture was completely overwhelmed—signaled by the overthrow of the Hawaiian Kingdom in 1893 (Beamer 2008). Dramatic population decline occurred throughout this period due to introduced diseases; upwards of 90% of the population perished (Stannard 1994). This, along with colonizing powers' use of cultural, economic, and linguistic genocide as intentional and unintentional tools to further their agenda—taking over Hawai'i—led to an assimilation and identity shift of the Hawaiian people into American culture (Beamer 2008; Kame'eleihiwa 1992; Stannard 1994; Trask 1999). The result was an extreme loss of cultural knowledge and practices. At the same time, Handy et al. (1972) report that a precipitous decline in the diversity of cultivated plants was occurring simultaneously.

'Awa (*Piper methysticum*, also known by its Polynesian cognate, "kava," or its popular English cognate "kavakava") is traditionally consumed as a ritual or ceremonial drink made by Pacific island peoples, but has recently gained global interest for its medicinal and psychoactive properties. It is traditionally prepared by straining its ground-up roots in water (Lebot et al. 1997). 'Awa was originally brought to Hawai'i approximately 1,500 years ago as a part of the botanical toolkit of the long-distance voyaging Polynesian peoples. At the height of Hawaiian civilization, 'awa had gained such prominence within the culture that it was used in almost all aspects of life ranging from religious, medicinal, social, to political roles: It

was the most important non-food crop in ancient Hawai'i. However, beginning in the 1820s with the arrival and influence of Christian missionaries, usage of 'awa began to decline because of its strong relationship with the traditional religious practices that were labeled "pagan." Concurrently, as the traditional political system broke down through the process of colonization by foreign powers, usage of 'awa in political gatherings waned. As colonization continued, the knowledge of traditional herbal healers, along with the plants that they used, including 'awa, were discredited. During this same period, the social drink of the Hawaiian people shifted from 'awa to alcohol. Eventually disappearing from everyday use, 'awa was recognized by only a handful of individuals in a few abandoned and isolated patches deep in the mountains—remnants from ancient plantings (Winter 2004).

With few exceptions, the 1940s marked the end of 'awa traditional usage that had been passed down from generation to generation since pre-contact times (Winter 2004). In the 1970s, a renaissance of Hawaiian culture began. Many of the traditions that were close to extinction were revived by younger generations (Kanahēle 1979). Some examples of these are *hula* (traditional dance form), *mele* (music), *oli* (chanting), various other art forms (carving, felt production, and others), long-distance canoe voyaging, Hawaiian language, and more. During this time, the practice of drinking 'awa was revived as well. Although its revival was slower than some of the other ancient practices, in time it would become a symbol for the re-emergence of Native Hawaiian cultural traditions (Winter 2004).

During the Hawaiian renaissance, 'awa drinking was initially limited to those involved with the revival of long distance canoe voyaging. This was likely because members of the voyaging community were, at first, the only group of cultural practitioners with a practical use for 'awa. 'Awa was still the social drink of the South Pacific Polynesians with whom these canoe voyagers had relatively frequent contact. It was therefore important that they understand its usage. The practice slowly spread to a closely affiliated group, those involved with the revival of the Hawaiian martial art, *lua*. The practice of drinking 'awa did not even begin to re-enter mainstream Hawaiian culture until the advent of 'awa bars in the late 1990s. By that time, the practices associated with the drinking of 'awa were very different from those of traditional 'awa drinkers at the height of Hawaiian civilization. Now, although 'awa is often referred to as a sacred plant, many of the old rituals associated with 'awa drinking are no longer practiced by mainstream 'awa drinkers (Winter 2004).

A major reason for changes in 'awa traditions was that mainstream 'awa use emanated from 'awa bars—an unaffiliated group of businesses emerging around the turn of the 21st century—which had not previously existed in Hawai'i. By the time

'*awa* bars began to open in Hawai'i, there was a three-to-four generation gap between those associated with drinking in '*awa* bars and their ancestors who had interacted with '*awa* for at least 1,000 years. The first '*awa* bars were opened with little knowledge and/or recognition of Hawaiian traditions associated with '*awa*. As a result, the revival of '*awa* usage failed to accompany a complete revival of its associated cultural practices within these commercial bar settings. Instead, original Hawaiian practices were supplanted by other Polynesian traditions and western alcohol consumption practices (Winter 2004).

The relationship between plants and people, as evidenced in the case of '*awa* and the Hawaiian people, is in a continual state of adaptation and change. Ethnobotanists need to be able to take a quantitative "snapshot" of this relationship at different points in time in order to measure the level and the trajectory of change. The following theoretical discussion is directed towards that end.

#### **2.4 The need to measure changing relationships between plants and people**

Garibaldi and Turner (2004) introduced the concept of "cultural keystone species" (emphasis by present authors) as "the culturally salient species that shape in a major way the cultural identity of a people." Brosi et al. (2007) similarly introduced the concept of "cultural keystone practices," (emphasis by present authors). These represent practices that are so intrinsic to the culture that if they were to disappear, the culture would be irreversibly altered to a point where its survival would be put into question. Many of these cultural keystone practices have a material (i.e., plant) base. In these cases, the associated plants would be cultural keystone species. Such a relationship would indicate that particular plant-people relationships within a human population are particularly important and/or influential for the health of both the ecosystem and the culture. Through the process of globalization, many cultural keystone species and cultural keystone practices are starting to change or even disappear. This may be related to the global decline in cultural and/or biological diversity. The ability of ethnobotanists to measure these changes will help us better understand the trajectory of change and therefore enable us to mitigate its damaging effects on both biological and cultural diversity. The concepts of quantum ethnobotany (see discussion below) could be used to analyze and address this issue.

Another important justification for measuring the changes in the relationships between plants and people is because this relationship is at the foundation of

human interaction with the natural world. It is the manner in which humankind interacts with the natural world—as our population increases exponentially—that will determine our success or failure in generations to come. Indeed, one of the major roles of ethnobotany and the related fields of ethnobiology and ethnoecology is to translate the sustainable interactions of various traditional cultures with their environment for a global palate (Prance et al. 2007; Salick et al. 2003). There exists a great need for measuring and quantifying the changing relationships between people and plants (on a small scale) and cultures and their environments (on a large scale) if we are to make inferences about how these interactions will affect the health of our planet in the future.

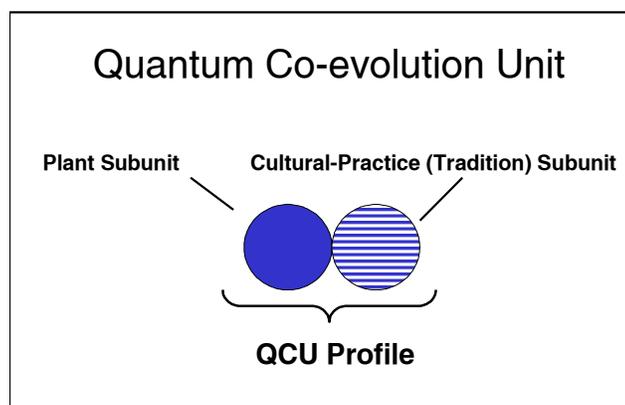
## **2.5 Quantum ethnobotany and an introduction of new concepts**

Ethnobotany is the study of the relationships between plants and people. These relationships are co-evolutionary in the sense that each affects the other's evolutionary trajectory. Quantum ethnobotany is the study of the most basic or fundamental interactions between plants and humans (McClatchey and Bridges 2005). We propose a concept for the smallest discrete unit with which to measure ethnobotanical relationships. We will refer to this as a “Quantum Co-Evolution Unit,” or “QCU.” In the context of ethnobotany, a QCU is made up of two subunits: A “plant subunit” and a “cultural-practice sub-unit” (or “tradition subunit”) (see Figure 2.1). In order to be a functioning and measurable (quantifiable) QCU, both subunits must be present and interacting with one another.

We also introduce the term, “Quantum Co-Evolution Unit profile,” or “QCU profile,” which would indicate a description of the specific relationship between the individual subunits in a complete QCU. An example of a QCU profile in the field of ethnobotany would be, “drinking Echinacea as a medicinal tea.” This would be a QCU that is composed of the plant subunit of “Echinacea sp.” coupled with the cultural-practice subunit of “drinking a medicinal tea.”

It is important to note that there are differing scales with which to measure a QCU (see Table 2.1). Some examples of these as they relate to ethnobotany are as follows:

1. A particular plant taxon at the level of genus or higher and an individual (or family's) traditions associated with it (QCU profile; e.g., conifers and Christmas decorating traditions),



**Figure 2.1.** A hypothetical Quantum Co-evolution Unit (QCU) as it relates to the co-evolutionary relationship between plants and human cultures. It is composed of two subunits: the plant subunit, and the cultural-practice (or tradition) subunit. The complete unit of the QCU is referred to and described by its QCU profile.

2. A particular plant species and an individual's (or family's) traditions associated with it (QCU profile; e.g., *Ginkgo biloba* and herbal-medicine traditions), and
3. A particular plant cultivar and an individual's (or family's) interactions with that particular cultivar (as opposed to other cultivars of the same species) (QCU profile; e.g., usage of Riesling grapes by certain German communities to produce ice wine).

Each subunit of a QCU has a set of intrinsic properties. "Plant subunits" have physical, chemical, genetic, behavioral, and emergent properties that contribute to the nature of the plant and its potential for interaction with other organisms and the abiotic environment. These intrinsic properties define a set of limits as well as opportunities for the plant. "Cultural-practice subunits" are similar in that they also have intrinsic properties including rules, history, timing, and emergent implications that contribute to the nature of the practice and its potential for being used within a human community. The intrinsic properties define a set of limits as well as opportunities for the practice. Each plant and cultural practice, therefore, is not able to participate within the full range of possibilities for all plants or all cultural practices but is limited in scope.

The respective subunits within a QCU create opportunities for and produce constraints on each other. Initially it would seem that the limitations would restrict the interaction to ways in which the plant and practice intersect in potential. However, it is likely that through the interactions of the otherwise separate subunits, additional synergistic properties may emerge that were not possible prior to the coupling of the two subunits. It is these emergent features that give rise to

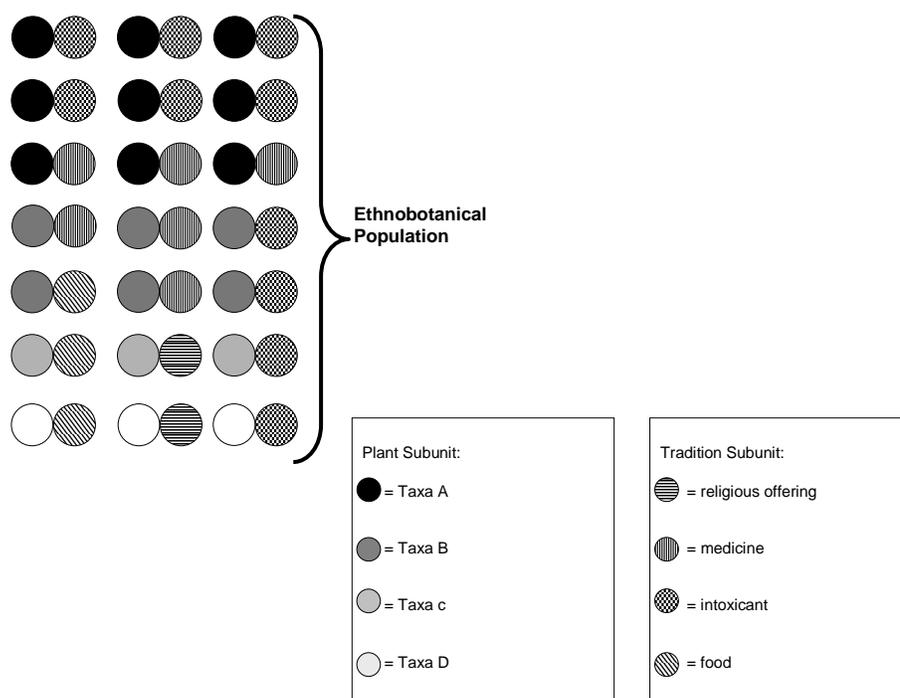
much of the diversity and complexity of human culture—from the mundane to the complex—such as emergent properties seen in religious and/or philosophical interactions with plants (QCU profile; e.g., the Christian practice of drinking red wine as a symbolic gesture of partaking in the blood of Jesus Christ).

**Table 2.1.** Taxa (folk or biological) and human population scales that can be measured with a Quantum Co-evolution Unit. Any combination of the plant subunit and cultural-practice subunit could be used as a particular scale. However, for the purposes of applied research a consistent scale should be used between comparative analyses.

Possible Plant Subunit Scales	Possible Cultural-Practice Subunit Scales
Individual	Individual
Community	Family
Population	Community
Ecosystem	Population
Biome	Culture
Variety/Cultivar/Landrace	Human Race ( <i>Homo sapiens</i> )
Species	
Genus	
Life form	

All QCUs—including their individual subunits—found in a particular human population can be understood as comprising an “ethnobotanical population” (Figure 2.2). Hundreds of published papers have produced lists of plants and their usages. Were these converted into an intersecting matrix, each positive intersection between a plant and a specific usage tradition would represent respective QCUs (the description of this intersection being its “QCU profile”) and the total matrix the ethnobotanical population. It is doubtful that any publication reports all known human interactions with plants in any one community. Therefore, complete descriptions of ethnobotanical populations do not exist and instead are represented by data sets that approach some percentage of the total population. Additionally, because cultural interactions with plants are dynamic, the total population is difficult to determine and, if once determined, at some point in time would certainly

change. It is also important to note that the most accurate studies of ethnobotanical populations would include not only all QCU profiles for a particular human population, but also the frequencies that the QCU occur within that population.



**Figure 2.2.** An example of an ethnobotanical population. A hypothetical collection of Quantum Co-evolution Units represented within a human community showing proportionality and frequency of various QCUs in relation to one another. Over various points in time the ethnobotanical population of a culture could be sampled. Changes could be observed and further quantified. Such changes could include the adoption of new QCUs into the profile, deletion of QCUs from the population, and changes in individual QCU frequency within the population.

QCUs and ethnobotanical populations also can be measured on different geographical and temporal scales. All useful plants everywhere in the world and in every society, by nature of being useful, are at some point in time part of a two subunit system and therefore part of QCUs. But because humans and human culture have not always existed but have evolved, their ethnobotanical populations—including all ethnobotanical subpopulations—have also evolved (as QCUs and their relative frequencies within the ethnobotanical population have changed over time). Thus, at some point in time, there was an instance at the genesis of any particular plant-person relationship that actually consisted of only one plant and one human, as in the first person who discovered the usefulness of a particular plant. Over time, the scale of the relationship changed as knowledge was shared across human

populations and plants were propagated and distributed (or otherwise accessed) for use by more and more people.

Richerson and Boyd (2005) produced a logical framework for discussion of how natural selection acts on transmission of cultural variation. Their reasoning may be extended with any or all of the following conditions being met in order for evolution to occur within an ethnobotanical population and/or subpopulation (e.g., QCU frequency or proportionality changes over time).

- Particular QCUs have increased in frequency because of selection,
- Particular QCUs have decreased in frequency because of selection, and/or
- One or more QCU(s) have been added or lost through events homologous to evolution (mutation, extinction, others).
- One or more QCU subunit(s) has changed (i.e., replacement of a lost or abandoned plant or tradition subunit) resulting in the creation of a new QCU.

“Selection” in the above context refers to any form of non-random selection, such as natural or artificial selection. That said, it is important to recognize that the above events are happening within cultural settings and not those of true natural selection. However, people are also excellent models of non-random selectors and have been used as examples of evolution by Darwin and others. The foundational logic is the same in the evolution of biological species and ethnobotanical populations. If none of the above were to happen between intervals of time, then an ethnobotanical population would be considered static and non-evolving.

## **2.6 Quantum co-evolution units and `Awa (*Piper methysticum*): Implications for quantifying ethnobotanical evolution**

Surveys of quantification methodologies for the field of ethnobotany (Hoffman and Gallaher 2007) reveal that the discipline has yet to adopt a unit of measure for change over time. Based on the discussion above, the need to quantify ethnobotanical change, or evolution, is apparent. The theoretical concepts of “Quantum Co-Evolution Units” (QCUs) and “ethnobotanical populations” may provide a structure for better understanding which methodologies should be developed to quantify ethnobotanical evolution.

For the purposes of applied research, a particular subset of the total ethnobotanical population, which we introduce as an “ethnobotanical subpopulation,” could be separated out for specific analysis. An ethnobotanical subpopulation could be determined by identifying a particular QCU subunit and all of its associated

corresponding subunits. For the interests of this paper, we will look at the example of *awa* on the scale of genus (*awa/Piper methysticum*) and culture (Native Hawaiian culture) to address how changes in this relationship can be measured over time.

A list of cultural interactions with *awa* in the pre-contact era (Winter 2004) illustrates the QCU profiles within this relationship (each row being its own QCU profile) (Table 2.2). Once QCU profiles such as these have been identified, a survey can be done to determine relative frequencies within this ethnobotanical subpopulation.

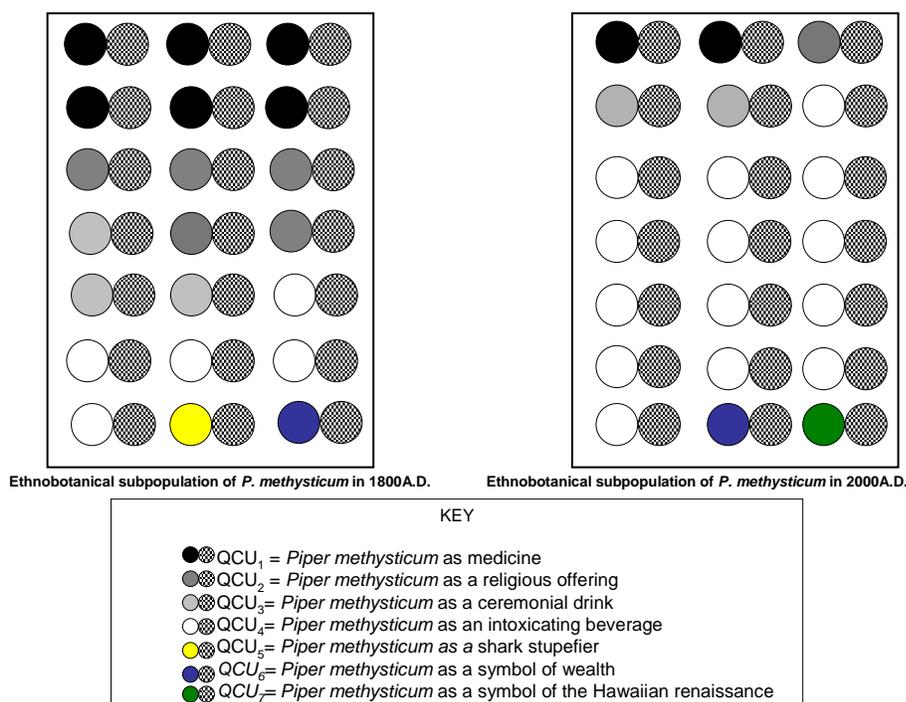
**Table 2.2.** The Quantum Co-evolution Units of *awa* in pre-contact Hawaiian culture as gleaned from Winter (2004). Each row in the table would represent a complete Quantum Co-evolution Unit and would be referred to as a particular “QCU profile.”

QCU plant subunit	QCU cultural-practice subunit
<i>awa (Piper methysticum)</i>	Medicine
<i>awa (Piper methysticum)</i>	Religious offering
<i>awa (Piper methysticum)</i>	Ceremonial drink
<i>awa (Piper methysticum)</i>	Intoxicating beverage
<i>awa (Piper methysticum)</i>	Shark stupefier
<i>awa (Piper methysticum)</i>	Symbol of hospitality and wealth

Using the approach in Table 2-2, the measurement of an “ethnobotanical subpopulation” for *awa* in Hawaiian culture can be made at various points in time. For the purposes of quantification, it would be impossible to assess an ethnobotanical subpopulation for any particular point in the past. However, knowledge of past practices could be used as a baseline to help guide researchers in more accurately documenting the existence of respective QCU profiles. Once the QCU profiles are determined on a given scale and an ethnobotanical subpopulation is quantified, the changes in ethnobotanical subpopulations (including QCU proportionality and frequency as well as changes in QCU profiles) through time could also be measured.

Because of the drastic loss and subsequent revival of the traditions of *awa* in Hawaiian culture (indicating change in QCU frequency within this ethnobotanical subpopulation over time), and all of the changes to traditions that have occurred in the past 200 years (i.e., addition, loss, or change of respective QCU profiles), it is clear that the ethnobotanical subpopulation of *awa* and Hawaiian culture has

evolved during this time period. It is quite likely that if an ethnobotanical subpopulation survey had been measured at 50-year intervals for the past two centuries, changes in QCU profile and frequency could be quantified, and changes observed would represent quantification of ethnobotanical evolution. However, the above-described ethnobotanical subpopulations are hypothetical; no such survey has ever been carried out.



**Figure 2.3.** A hypothetical ethnobotanical subpopulation based on *Piper methysticum* in Hawaiian culture as measured between two time periods. A hypothetical example of how ethnobotanical (sub)populations can change over time in regards to QCU relative frequency. Note not only the change in relative frequency, but the addition and subtraction of QCUs as well.

As an example, based on the authors' field observations of 'awa traditions in Hawai'i, the practice of using 'awa to stupefy sharks is a QCU profile that has either been lost or has, at the least, gone dormant within the culture. Also, if compiling an ethnobotanical subpopulation of this type today, we would have to create at least one new QCU profile that did not exist in pre-contact times. This QCU profile would be that of 'awa and the associated practice of imbibing 'awa to symbolically assert oneself as a cultural practitioner associated with the Hawaiian renaissance. Furthermore, if such an ethnobotanical subpopulation were continued for a few more generations, we might discover that more QCU profiles have been dropped, others gained (or revived), and some changed. It is quite likely that between the points in time when ethnobotanical populations were measured, not only would the composition of QCU profiles within the ethnobotanical population change, but the

proportionality and frequency of the various QCU profiles in relation to one another would have also changed (Figure 2.3). Our ability to measure this change could be the first step in quantifying ethnobotanical evolution.

## 2.7 Conclusions

Quantum Co-Evolution Units (QCUs) and ethnobotanical populations are quantifiable. We propose six hypotheses about human interactions with plants on the basis of the above discussion.

1. Human relationships with plants are co-evolutionary. They consist fundamentally of two parts: A plant subunit and a cultural-practice subunit. Together they comprise the smallest measurable (quantum) unit through which changes in human interaction with plants can be evaluated over time.
2. Quantum Co-Evolution Units can be measured on differing scales depending on the taxon of the plant and the unit of human population with which it interacts.
3. There is a set of dynamic Quantum Co-Evolution Units within each culture that comprise an ethnobotanical population for that culture.
4. Cultural practices without an associated plant and plants without an associated cultural practice cannot be investigated by researchers using the approaches of quantum ethnobotany as they lack one of the needed subunits.
5. Ethnobotanical populations, comprising the total amount of Quantum Co-Evolution Units within a human population, can be measured at various points in time.
6. Changes in proportionality and frequency of Quantum Co-Evolution Units (QCUs) to one another within the ethnobotanical population, as well as additions, subtractions, and changes to and of QCUs in the ethnobotanical population between various points in time, can also be quantified. Such a methodology could be used to demonstrate ethnobotanical evolution.

If supported by future research, these hypotheses may help further the field of theoretical ethnobotany. They may also have implications in the applied fields of ethnobotany, ethnobiology, ethnoecology, evolutionary biology, conservation biology, restoration ecology, and any other field where plant and people interactions play a major role.

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## **Chapter 3. Quantifying Evolution of Cultural Interactions with Plants: Implications for Managing Diversity for Resilience in Social-Ecological Systems<sup>2</sup>**

### **3.1 Abstract**

The discipline of ethnobotany has gathered an abundance of data about the diversity of ecological resource management methodologies, but has yet to do so using standard units of measure such that cross regional comparisons can be made. Both biological diversity and sociocultural diversity are important factors to manage for resilience in social-ecological systems. Sociocultural evolution has strong links to biological evolution. Quantum ethnobotany provides theory and models to measure links between biological diversity and sociocultural diversity for comparisons across regions. Links between biological and cultural diversity are dynamic relationships cycling between processes of co-evolution and co-extinction. The ability to measure links between biological and sociocultural diversity is provided by quantum ethnobotany. This will be useful for resource managers, policy makers, stakeholders and cultural practitioners to manage both biological and cultural diversity through co-extinction cycles for the purpose of maintaining or increasing resilience in social-ecological systems.

### **3.2 Introduction**

#### ***3.2.1 Ethnobotany research and ecological resource management***

Human interaction with the natural world is a main focus of ethnobotany (Salick et al. 2003; Prance et al. 2007). Ethnobotanists have long been generating data about the relationships between sociocultural systems and ecosystems from different locations around the world; however, these data have rarely been produced using standardized methods or converted into common units so that true comparisons could be accomplished on regional or global scales. Reasons for avoiding regional or

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global studies include fundamental cultural differences, floristic and ecological differences, perceptions of cultural authenticity, longevity of plant-cultural interactions, and inabilities to see common threads across cultural experiences with plants. This chapter sets out a theoretical model grounded in resilience (Holling 1973), social-ecological systems (Berkes and Folke 1998), and quantum ethnobotany (McClatchey and Bridges 2005). Quantum Co-evolution Units (Chapter 2) will be used to address evolution of fundamental interactions between human cultures (as the basis for sociocultural systems) and plants (as the basis for ecosystems). The purpose of this chapter is to better understand the cyclical processes of co-evolution and co-extinction involved in human interaction with the natural world; and how knowledge of these processes can serve modern resource managers, policy makers, stake holders, and cultural practitioners.

### ***3.2.2 Ecosystem resilience and biodiversity***

Ecosystem resilience (Hollings 1973; Resilience Alliance 2002) is a measure of a system's relative ability to absorb disturbance without changing to a different state, such as a different biological community with different ecosystem services (Folke et al. 2004). Biological diversity has been shown to be a key factor in ecosystem resilience (Holling 1996; Walker et al. 2004) because it plays a major role in renewing and reorganizing ecosystems after disturbance, and it helps to maintain desired states of dynamic ecosystem regimes in the face of uncertainty and surprise (Folk et al. 2004).

Loss of biodiversity is of serious concern for all ecosystems (i.e., not just rainforests) because it leads to compromises in resilience and productivity of these systems. Furthermore humans (i.e., sociocultural practices) play a central role in either degrading or maintaining high levels of biodiversity, a key factor for system resilience (Berkes et al. 1995; Berkes and Folke 1998; Folke et al. 1998; Berkes 1999; Davidson-Hunt and Berkes 2003; Colding et al. 2003, Folke et al. 2003, 2004).

### ***3.2.3 Social-ecological systems and resilience***

There are three things that must be understood about social-ecological systems and resilience:

1. Humans are a part of ecosystems and cannot be separated out when developing management practices,
2. Humans can increase biodiversity,
3. Resilience depends on both biological AND cultural diversity.

Each of these points will be elaborated on below.

For the purposes of biodiversity conservation there is a need to understand how human-nature interactions affect biodiversity (either positively or negatively). The discipline of ethnobotany, focusing on the juncture of the biological and the sociocultural world, can provide research theory and tools with which to guide ecological resource management that will be mutually beneficial to both the ecological and sociocultural sides of these linked systems (Prance et al. 2007). This chapter emphasizes the concept that humans are a part of, not separate from nature (Balée 2006), supporting the views of Berkes and Folke (1998), and Berkes et al. (2003) which hold that social and ecological systems are linked, and that delineations between social and natural systems are arbitrary and artificial. Such human-in-nature systems are referred to as “social-ecological systems” (Berkes and Folke 1998; Berkes et al. 2003).

The concept of the importance of biodiversity for system resilience has been applied to social-ecological systems (Berkes and Folke 1998; Berkes et al. 2003). Negative effects of sociocultural interactions with ecosystems on biological diversity have been well documented (Hooper et al. 2005). However, research has also shown that there are strong links forged between biological and cultural diversity (Gadgil 1987; Moore et al. 2002; Maffi 2005). Furthermore, particular traditional ecological management systems actually increase biodiversity (Posey 1985; Lewis 1989; Berkes et al. 1995; Folke et al. 1998; Berkes et al. 2003; Balée 2006). As more research emerges we may see that instances of sociocultural interactions with ecosystems enhancing biodiversity may not be a rare occurrence. Research focusing on the process by which particular social-ecological management systems increase biodiversity is needed. Understanding the initiation and intensification of the relationships between people and plants within social-ecological systems may reveal insights that will help us to manage biodiversity and therefore resilience in these systems.

The idea of the importance of diversity in system resilience can be applied, not only to the biological side of the social-ecological system equation, but to the sociocultural side as well through historical ecology (Balée 2006). As witnessed in the loss of languages on the planet, cultures are going extinct at an alarming rate. Nearly 90% of existing languages are projected to be extinct by the end of this century (Nettle and Romaine 2000). With these extinctions varying world views and practices associated with interactions with the natural world will also be lost. Some of these world views and human-nature interactions undoubtedly are associated with practices that enhance biological diversity. In all areas of the world

there exists a need to quantify these interactions for comparative analyses—before they are lost to time—as it is likely they include practices associated with increasing biodiversity. It is of vital importance that as these data are collected the studies are done in such ways as to be compared across space and time with other social-ecological systems.

### ***3.2.4 Sociocultural and biological evolution***

Sociocultural evolution (Trigger 1998) has been a contentious issue because some researchers have elected to equate cultural evolution with “cultural progress.” Throughout this chapter “evolution” and “cultural evolution” are being equated with “cultural change” and NOT with any sort of evaluation of the quality of that change. The approach is taken that all cultures are equally evolved but on different trajectories.

Human interactions with the natural world are not static, but rather ever evolving. White’s law (White 1959), as a cornerstone concept for the evolution of culture, implies that cultural evolution is related to changing intensities of interactions with the environment (as measured by efficiency of capturing and using environmental energy). Research has demonstrated patterned evolutionary relationships between humans and specific ecosystems (Conklin 1963), animals (Rappaport 1984), plants (Harris and Hillman 1989), and nature (i.e., ecosystems) and other complex systems (Boyd and Richerson 1985; Norgaard 1994). There has even been a question of which partner is driving the relationship (Pollan 2002). In all likelihood the evolutionary relationship is co-evolving, with no driver, and the intensity of the relationship can be measured as it changes over time.

Berkes et al. (2003) allude to the idea that understanding co-evolutionary processes of social-ecological systems is paramount to human survival on the planet:

“In the present era of the human-dominated biosphere, co-evolution now takes place also at the planetary level and at a much more rapid and unpredictable pace than previously in human history... Facing complex co-evolving systems for sustainability requires the ability to cope with, adapt to, and shape change without losing options for future adaptability.” (Berkes et al. 2003: 353)

However, despite the many calls to the importance of understanding co-evolutionary processes and their trajectories, researchers have yet to scale back from the larger picture to propose distinct units with which this co-evolution/extinction could be quantified. That is, until the emergence of quantum

ethnobotany. This chapter will focus on the cyclical, but sometimes also finite, processes of co-evolution and co-extinction in social-ecological systems, and put forth theoretical concepts about quantifying these processes. As will be presented below, the new field of quantum ethnobotany can demonstrate not only this, but also quantify how the links between biological and cultural diversity affect the resilience of social-ecological systems. However, perhaps more importantly, quantum ethnobotany can demonstrate how these relationships evolve over time. This can give researchers and resource managers better ideas about the trajectory of evolution and its implications for social-ecological system resilience. Moreover, quantum ethnobotany provides theory and the models to collect and analyze these data in ways that are comparable across space and time. These should serve as simple models that are economical, clear, and able to detect useful generalizations in the midst of the complexity of human behavior (Richerson and Boyd 2005: 95).

This chapter proposes the idea that the tools of quantum ethnobotany can be used to better understand how the trajectories of co-evolving relationships between plants and people are affecting diversity on both ecological and sociocultural levels, both of which are major factors in social-ecological system resilience. The ability to do this may enable resource managers, policy makers and others to not only mitigate potential threats to diversity, but also promote management methodologies that enhance diversity. The following discussion will cover the concepts of quantum ethnobotany, evolving interactions between plants and people, and the cyclical processes of co-evolution and co-extinction; then concludes with a set of hypotheses relating to the theories expressed in this chapter.

### **3.3 Quantum Ethnobotany**

#### ***3.3.1 In relation to complex systems theory***

Quantum ethnobotany is a theoretical field that attempts to identify fundamental measurable units of interaction between people and plants (McClatchey and Bridges 2005). As discussed in Chapter 2, the quantum units are scalable from the most basic (minimum) of interactions (one person and one plant) to very complex relationships (all of humanity and all plants interacting with humanity). Quantum ethnobotany specifically addresses hypotheses about potential for survival in environments based on implementation of different botanical and cultural tool kits. Quantum ethnobotany has not yet, however, addressed the origin and continuing change of the interactive relationships that form the basis of the quanta (units of plants and people) being studied. This chapter aims to address continuing change

(i.e., the cyclical co-evolution and co-extinction processes) within the complexity of social-ecological systems.

Complexity theory has addressed many relevant areas to social-ecological systems such as organizational and management studies (Anderson 1999), the management of ecological systems (Janssen 2002), landscape ecology (Green et al. 2006), and anthropology (Hannerz 1992). Nowotny (2005) points out that it is the emergent properties that come about due to an interface of two otherwise separate properties that gives rise to complexity. While complex systems often have synergistic affects whereabouts the whole is greater than the sum of the individual parts, there is value in understanding the most basal components of this complexity. Quantum ethnobotany examines the interface between the biological and the sociocultural, as well as the emergent properties of these interactions at the most fundamental level. Doing so could shed light onto the how the building blocks of social-ecological systems contribute to the complexity of such systems.

### ***3.3.2 Quantum co-evolution units and ethnobotanical populations***

As discussed in Chapter 2, the Quantum Co-evolution Unit (QCU) is the smallest unit through which interactions between human cultures and plants can be measured; and, as discussed below, is the unit used to measure ethnobotanical evolution. We assert that the most basic of human interactions with plants are those between a person (as a member of a human culture) and a plant (as a member of a taxon that may be a species, landrace, population, etc.) (Figure 2.1). A description of any people-plant relationship would be a “QCU profile” (Chapter 2). An example of a QCU profile would be ‘giving red roses on St. Valentine’s Day’ – the particular plant taxa being a specific color of rose (*Rosa* spp.) and an individual’s (or society’s) associated tradition of giving them to loved ones annually on February 14<sup>th</sup>. All useful plants everywhere in the world and in every society by the nature of being useful are at some time part of a two subunit system and therefore can be described and quantified as QCUs. Each subunit has a set of intrinsic properties that define its set of limits and opportunities for interactions. It is the emergent properties of interactions within QCUs, and the complexity of QCU populations that likely gives rise to much of the complexity of human culture. These concepts are more fully discussed in Chapter 2.

All QCUs and their individual subunits found in a particular social-ecological system can be understood as comprising an “ethnobotanical population” (Figure 2.2). An ability to quantify an ethnobotanical population at various points in time will help us to measure changes in QCU frequency over time (see discussion below). As

discussed in the previous chapter, subpopulations can also be used to analyze select subsets of the larger population.

Quantum ethnobotany provides the tools for understanding the dynamics and evolution of ethnobotanical populations which can be key in maintaining both biological and cultural diversity—and-therefore resilience—in social-ecological systems. This is not only true for understanding human *pressures* on biodiversity, but as will be illustrated in the following section, perhaps more importantly for understanding human *promotion* of biodiversity.

### **3.4 Evolution of cultural interactions with plants**

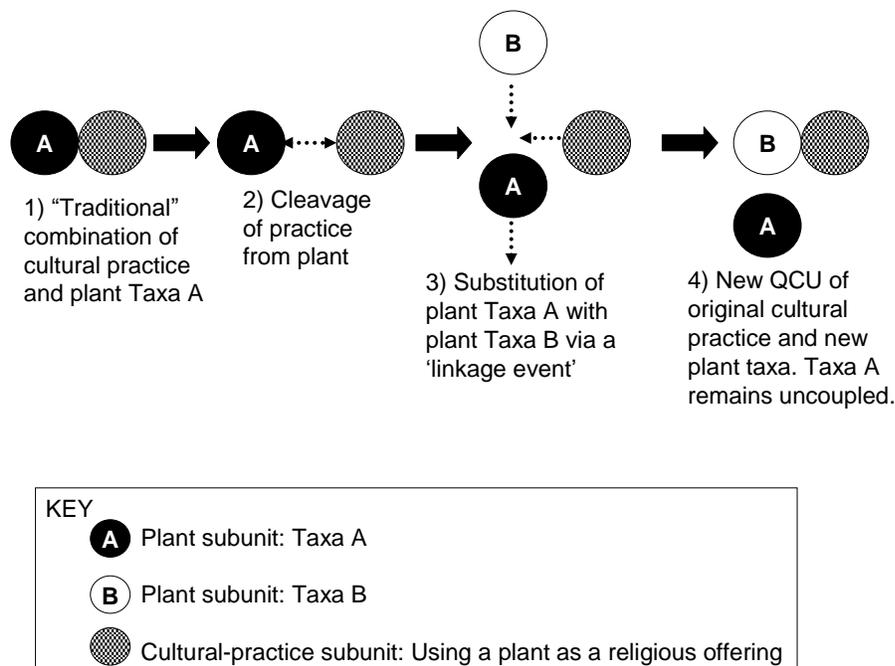
#### ***3.4.1 Changes in composition of ethnobotanical populations***

Humans, and not plants, determine if a relationship between plants and cultural practices is developed, maintained, changed, or abandoned. Humans, either voluntarily or involuntarily, determine if the interaction exists at all, or can link other subunits together to form new QCU in a population or can separate subunits to lose QCU in a population. If separated, the individual subunits can exist, but without the interaction, may in time cease to exist or change in ways that are possible because of the loss of constraints of the previously corresponding subunit. However, sociocultural interactions with plants change as a result of changes in plant genetics (e.g., phenotypic expression) over time. As a result we see that this co-evolutionary process has no real driver.

This chapter refers to the joining of two otherwise unconnected subunits into a QCU as a “linking event.” Linking events are a major driver of changes in composition of ethnobotanical populations as they are adding diversity to the said population. Such events also play a key role in understanding the co-evolution process between cultures and plants.

We refer to the breakup of a QCU into two disjointed subunits as a “cleavage event.” Cleavage events may be temporary. If one subunit of a QCU is lost, people may attempt to replace it by finding a corresponding subunit (e.g., if a specific plant is lost a replacement may be sought, or conversely if a specific traditional practice is lost a replacement may be sought or developed) (Figure 3.1). Cleavage events may also be permanent. If unable to find a corresponding subunit, the remaining plant or tradition may eventually be lost (i.e., die out), leading to an extinction event of that QCU. Cleavage events are also a major driver in changes to composition of

ethnobotanical populations, and play an important role in understanding the co-extinction process between cultures and plants.



**Figure 3.7.** An example of a QCU cleavage event and the subsequent substitution of a subunit via a linkage event. Parameter setting conditions such as environment, available biological diversity and available cultural diversity (see below section "The backloop cycle") are determining factors in both cleavage events and linkage events. In this example through a change in the parameter setting condition of religion the practice of using a particular plant taxa as an offering remains the same, but the taxa offered has changed. The process is: 1) Under a certain set of parameter setting conditions a particular QCU exists and contributes particular emergent properties to social-ecological system complexity; 2) A variable in the parameter setting conditions changes (religion in this example) inducing a cleavage event which separates the plant subunit from the cultural-practice subunit; 3) The same change in a parameter setting condition (i.e., religion) that caused the cleavage event induces a linkage event which rejoins the cultural-practice subunit to a different plant subunit creating a new QCU; 4) The new QCU linkage produces different emergent properties which contribute in different ways to social-ecological system complexity than the original QCU. The survival of the original, now de-coupled, plant (Taxa A) subunit is now in question. Its long-term survival (i.e., maintenance of genetic integrity) may be dependent on its ability to re-couple to a cultural-practice subunit. If able to survive on its own it will no longer have the constraints of the previously associated cultural-practice subunit, and may be set on a new evolutionary course not previously possible.

### ***3.4.2 Changes in composition of ethnobotanical populations over time***

Understanding the processes involved with evolution of ethnobotanical populations will be key in developing management strategies for resilient social-ecological systems across a range of scales. A large part of this depends on the ability to quantify changes in ethnobotanical populations (i.e., cultural relationship to plants) over time.

The ethnobotanical state of a social-ecological system (its ethnobotanical population) can be measured at various points in time. If an ethnobotanical population is measured at different points in time and is found to have changed, then the magnitude of the change may be measured. Biological evolution is traditionally discussed as change in allele frequency over time. Likewise, ethnobotanical evolution may be discussed as a process of co-evolution as a change in the QCU frequency within an 'ethnobotanical population' over time (changes in: allele frequency of plants, and/or cultural practices or traditions). In the following the discussion of perspectives of human-plant co-evolution with limited analogy to genetic evolution will proceed. Within this structure, this chapter produces a set of hypotheses that we hope will point to future theoretical ethnobotany and applied conservation research.

Richerson and Boyd (2005) produced a logical framework for discussion of how natural selection acts on transmission of cultural variation. Their reasoning may be extended with any or all of the following conditions being met in order for evolution to occur within an ethnobotanical population and/or subpopulation (e.g., QCU frequency or proportionality changes over time).

- Particular QCUs have increased in frequency because of selection.
- Particular QCUs have decreased in frequency because of selection.
- One or more QCU(s) have been added or lost through events homologous to those involved in the process of biological evolution (mutation, extinction, etc.).
- One or more QCU subunit(s) has changed (i.e., replacement of a lost or abandoned plant or tradition subunit) resulting in the creation of a new QCU.

Recognition that the above events are happening within cultural settings and not those of true natural selection is important. However, people are also excellent models of non-random selectors and therefore have been used as examples of evolution by Darwin and others. The foundational logic is the same in evolution of biological species and ethnobotanical populations. If none of the above conditions

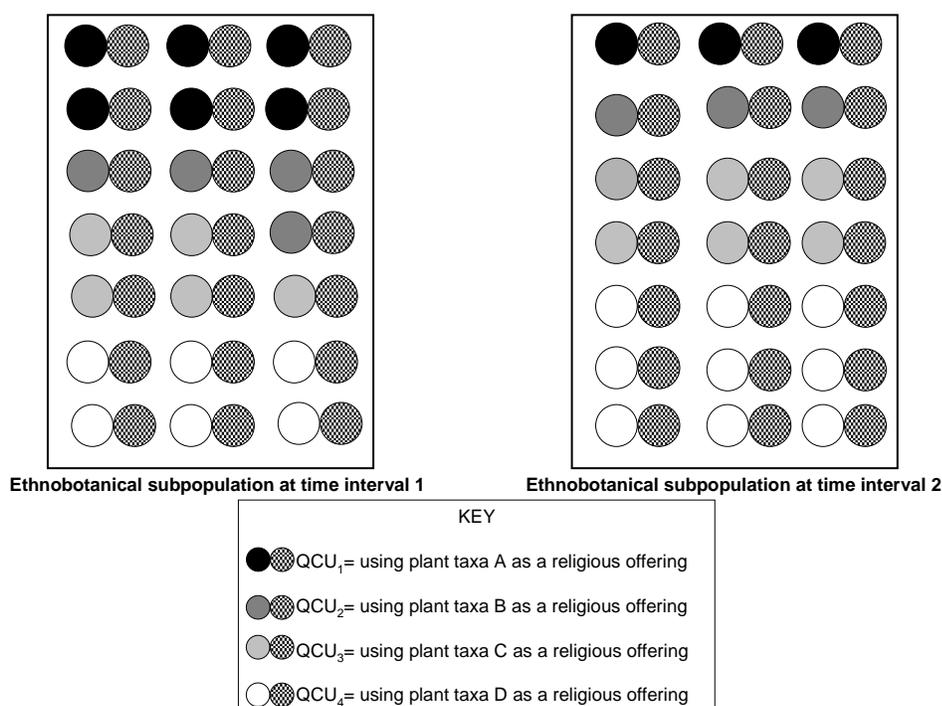
were to happen between intervals of time then an ethnobotanical population would be considered static and non-evolving.

Based on the above discussion the following equation is proposed to calculate QCU frequencies within ethnobotanical populations, where the value for the ethnobotanical population will always be 1:

$$\Sigma = QCU_1/QCU_{total} + QCU_2/QCU_{total} + \dots + QCU_n/QCU_{total}$$

$n$ =the number of QCUs in an ethnobotanical (sub)population

For the purposes of illustration Figure 3.2 depicts a highly simplified and hypothetical ethnobotanical subpopulation focusing on plants within a social-ecological system that are used as religious offerings. A survey of the subpopulation was taken at two intervals of time. If the above formula is applied to this subpopulation it can be demonstrated that between the two intervals of time the respective QCU frequencies within the subpopulation have indeed changed (Table 3.1), indicating that ethnobotanical evolution has taken place.



**Figure 3.8.** A highly simplified and hypothetical ethnobotanical subpopulation measured at two intervals in time. This ethnobotanical subpopulation focuses on the cultural practice of using plants as a religious offering and measures all of the plant taxa linked with that practice. Between the two intervals in time that this ethnobotanical subpopulation was measured changes in frequency can be observed (Table 3.1) which would indicate that evolution within this subpopulation has taken place.

**Table 3.2.** Respective QCU frequencies of a highly simplified and hypothetical ethnobotanical subpopulation that focuses on plants involved with religious offerings as measured between two intervals of time. Each calculation represents the frequency of respective QCUs in an ethnobotanical subpopulation at a particular time. The frequencies have changed between intervals of time which indicates that ethnobotanical evolution has taken place.

	QCU <sub>1</sub>	QCU <sub>2</sub>	QCU <sub>3</sub>	QCU <sub>4</sub>
<b>Time interval 1</b>	0.2875	0.1905	0.2381	0.2875
<b>Time interval 2</b>	0.1429	0.1429	0.2875	0.4286

Although there are many aspects of evolution in ethnobotanical (i.e., QCU) populations that may be explored, one is particularly germane for addressing systematic data collection across different regions for conservation and management of biodiversity. As human interactions with plants intensify researchers commonly observe that plant biodiversity also increases (Lunt and Spooner 2005; Sheuyange et al. 2005). This may subsequently result in a diversification of traditions—hence co-evolution (see discussion below)—a portion of which are associated with maintaining or further increasing this biodiversity. These relationships, therefore, warrant the attention of conservation biologists, resource managers and policy makers (Meffe et al. 2002; Cook et al. 2004). Quantum ethnobotany provides the model for measuring the above.

It is critical to understand how people-plant interactions intensify over time, becoming more complex and interdependent. Such relationships are likely to be similar to that which Berkes et al. (2003) described about social-ecological systems: they are either more resilient if complexity is maintained, or more brittle as a result of homogeneity. In relation to this idea quantum ethnobotany sets a model to identify and measure the linkages between plants and people as relates to social-ecological system resilience.

### **3.5 The cylindrical process of co-evolution and co-extinction**

#### ***3.5.1 Co-evolutionary process of people-plant interactions: increases in biocultural diversity***

This chapter contends that within social-ecological systems people-plant relationships are continually changing, but in a manner in which they influence each other's evolutionary trajectory. Changes in plant genetics (e.g., phenotypic variations) will change both the opportunities for and constraints upon interactions

with people (i.e., cultures). Likewise, changes in culture (e.g., cultural priorities) will affect which phenotypes are managed and how, in essence influencing the trajectory of plant evolution. Thus this relationship is co-evolutionary.

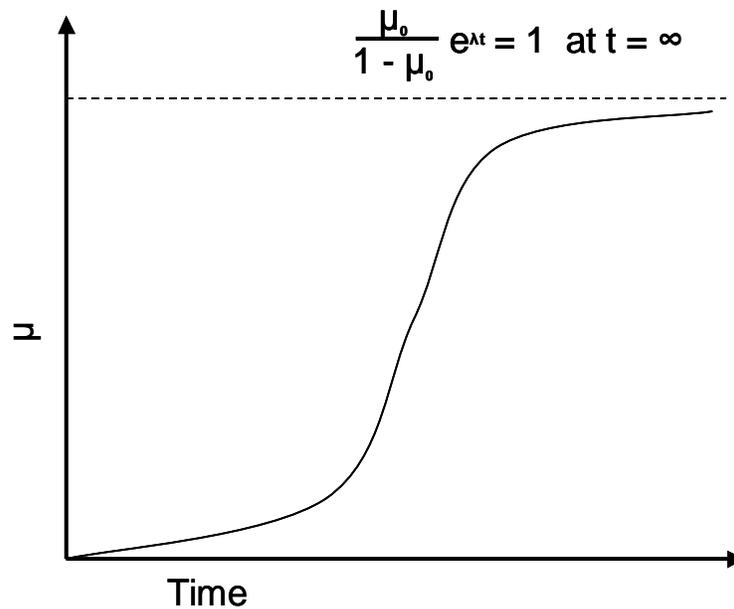
This chapter further submits that there are three classes of co-evolutionary relationships in the people-plant context: non-intensifying co-evolution, intensifying co-evolution, and deteriorating co-evolution (or co-extinction)—all of which are important to understand for conservation of biodiversity. The trajectory of an ethnobotanical population can be an indicator to aid in the classification of co-evolutionary relationships, and the kinds of insights that can be gained through observation (Table 3.2). Understanding the intricacies of a population in a state of expansion such as in an ‘intensifying co-evolutionary relationship’ would be important for understanding socioculturally driven increases in biodiversity, and is therefore the class of relationship that we will focus on in this section.

**Table 3.2.** Classifications of co-evolutionary relationships, the respective state of the ethnobotanical population, and the potential insights that can be gained for managing diversity.

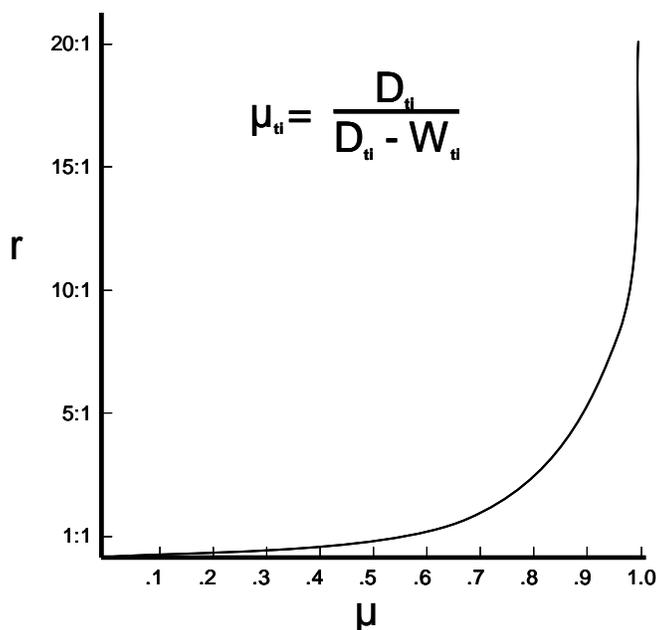
<b>Classification</b>	<b>State of ethnobotanical population</b>	<b>Insights gained</b>
Intensifying co-evolution	‘Linkage events’ > ‘cleavage events’ (i.e., growing)	Management practices that lead to increases in diversity
Non-intensifying co-evolution	‘Linkage events’ $\approx$ ‘cleavage events’ (i.e., relatively stable)	Management practices that maintain diversity
Deteriorating co-evolution (or co-extinction)	‘Linkage events’ < ‘cleavage events’ (i.e., shrinking)	Management practices that lead to decreases in diversity

Sociocultural systems have the ability to not only increase biodiversity through management of natural systems, but also through the process of intensification, such as through agriculture (Balée 2006). The number of plant varieties and landraces recognized by a culture demonstrates the relative importance of that plant to the culture (Rindos 1984). This is especially true for domesticated plants. In a broad sense several researchers have addressed the ideas of how and why plants came to be managed by people (e.g., agriculture) and how this relationship

intensified (Sauer 1952; Böserup 1965; Rindos 1984; Rindos 1989; Zohary 1989). An important question is therefore: How is an increase in recognition of plant diversity correlated with cultural importance? An important model of this process was proposed by Rindos (1984) in which he hypothesized that the intensification of agriculture associated with increasing numbers of varieties of domesticated plants provides opportunities for population increases, and reduced dependency upon less predictable wild plant resources. His model (Figures 3.3, 3.4) implies that the rate of change over time in the system is most dramatic in cultures that are fully dependent upon agriculture and have intensified their utilization of specific crops to include many varieties and landraces of the specific species that they utilize.



**Figure 3.9.** Increase in abundance of domesticates over time (After Rindos 1984, Figure 5.3) [ $\mu$  is the relative abundance of domesticates as a fraction of the total possible in the environment.  $\lambda$  is the logarithm of the relative increase in domesticates.]

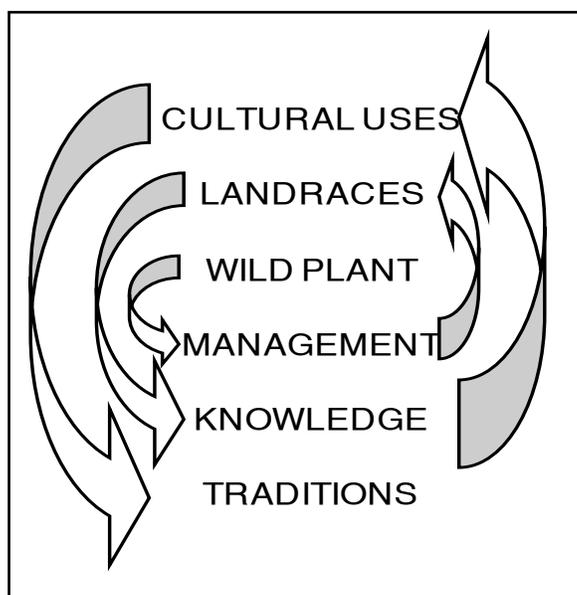


**Figure 3.10.** Relative contribution ( $r$ ) of varieties of domesticated plants ( $D$ ) versus wild plants ( $W$ ) over time as a function of their relative abundance ( $\mu$ ) in the environment. (Adapted from Rindos 1984, Figure 5.2)

A better understanding of this trend can be seen by taking a closer look at the developmental process of plant management (e.g., cultivation) and the effects that subsequent diversification of a cultivated (or otherwise managed) plant has on the evolution of human culture (Figure 3.5). Quantum ethnobotany scales down to the most basic level of people-plant interactions, and provides the models to quantify and analyze these changes.

As seen through the lens of quantum ethnobotany the process of intensifying co-evolution between people and plants results in a simultaneous increase in both biological and cultural diversity. The research of Berkes and Folke (1998) and Berkes et al. (2003) would suggest that such increases in diversity are related to social-ecological system resilience. As illustrated in Figure 3.5, on the sociocultural side an intensification of management leads to an increase in knowledge, which leads to an increase in practices, which leads to an increase in traditions. The ability for this to happen, however, hinges on increases in plant biodiversity along all levels of the process, as it increases the potential for human interaction (see discussion below). The process of co-evolution between people and plants in social-ecological systems can, under certain circumstances, lead to an intensification of this relationship which in turn increases diversity on both the biological and the sociocultural sides of the system. This process results in a complex and diverse

relationship between people and plants that would likely have a high level of system resilience. Quantum ethnobotany provides the model, using QCUs, to better understand this process.



**Figure 3.11.** Co-evolution of a cultivated (or otherwise managed) plant and the culture that cultivates it as depicted in an outwardly expanding spiral. This involves a six step process which depends on linkage events along the entire cycle: 1) A wild plant is determined to be useful in some way; 2) The plant is brought into a management system (e.g., agriculture); 3) Landraces are developed through selection; 4) Knowledge about each specific landrace is accumulated; 5) Diversification of associated cultural practices takes place; 6) These practices get passed on as traditions which will influence the evolution of the culture.

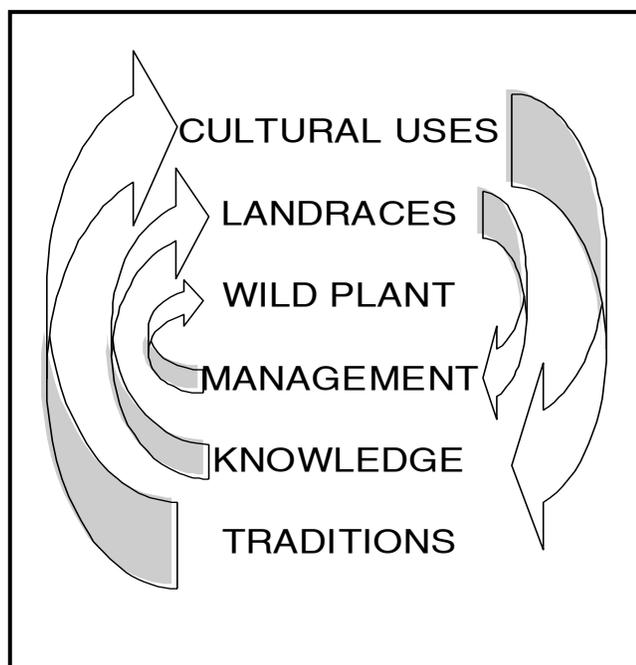
The ‘intensified co-evolution’ of people-plant relationships can be understood as a series of ‘linkage events.’ According to quantum ethnobotany theory as landraces are developed via agriculture or other management systems the only way that it can be maintained (i.e., survive) for the long term while keeping its genetic integrity is to be connected to a particular cultural practice via a linkage event. As management is intensified and more landraces are developed and recognized there will be more opportunities for linkage events. Particular taxa that become culturally important will continue to diversify and gain more associated practices and traditions via further linkage events, a process that will likely result in the taxa becoming more and more important to the culture. This process is readily apparent in agricultural systems, but can be applied to many other natural resource management methodologies. Through this model we can see both how biodiversity is linked to cultural diversity, as well as the reasoning behind the idea that recognized diversity is directly related to cultural importance.

### ***3.5.2 Co-extinction process of people-plant interactions: Decreases in biocultural diversity***

Holling (1986) points out that ecosystems are dynamic and go through regular, non-linear cycles of organization, collapse and renewal. This also applies to social-ecological systems (Berkes et al. 2003). The process of co-extinction (Figure 3-6) would be that “collapse” process that they referred to (the co-evolution process would relate to the ‘organization’ phase). This may be brought on for reasons such as cultural colonization (see below section). The process of co-extinction is very much the reverse of the process of co-evolution. This may happen rapidly or slowly, and may very well lessen resilience of social-ecological systems. Ebenman and Jonsson (2005) have shown that owing to interdependencies among species in ecological communities, the loss of one species can trigger a cascade of secondary extinctions with potentially dramatic effects on the functioning and stability of the community. We contend that the same is true for not only the ecological side of social-ecological systems, but for the sociocultural side as well. Furthermore this concept can be applied to linked biological-cultural relationships. Understanding this process is vital to preserving biodiversity as these relationships break down.

In the broad sense co-extinction of linked biological-cultural diversity is very much the opposite of the process described by Rindos (1984) and would be the inverse of the process illustrated in Figures 3.3 and 3.4. This would imply that if a culture loses domesticates then this loss will be rapid when they are most dependent upon them. If this process of intensification is reversed, then it appears that the earliest and latest parts (Steps 1 and 6 in Fig. 3.6) of the cycle are slow and the middle parts (Steps 2 through 5) are rapid as defined by the steep slope depicted in Figure 3.3. The implications for cultures with intensified agricultural or other resource management traditions that are faced with changes are profound. It appears likely that changes will happen rapidly to both components of social-ecological systems.

In the terms of quantum ethnobotany theory, this process can be scaled down to analyze how it operates on the most basic level. Just as an intensifying co-evolution process can be understood in terms of ‘linkage events,’ an abating co-extinction process can be understood in terms of ‘cleavage events.’ Cleavage events break linkages that are key to connecting cultural and biological diversity. A better understanding of this process can help resource managers to maintain biodiversity and enable it to persist through the cycle until it can be reorganized back into the social-ecological system.



**Figure 3.12.** The abating spiral depicting the co-extinction of plants and the sociocultural system that manages it. A six step process that involves cleavage events along the whole process: 1) A tradition associated with a plant use/management is no longer passed down to the younger generations; 2) Cultural interactions with particular landraces are discontinued; 3) In the absence of cultural-biological interactions with particular landraces knowledge about them will be lost; 4) Without having either knowledge or practices associated with landraces, plant diversity will be lost; 5) In the absence of management a plant will revert to wild plant; 6) The plant will remain wild – with greatly reduced biodiversity – and will continue as such until it is rejoined to a sociocultural system via a linkage event.

### ***3.5.3 The back-loop cycle: reorganizing diversity between co-extinction and co-evolution cycles***

People-plant relationships have been noted to go through processes of growth, dismantling and back into regrowth (Winter 2004; Chapter 2). Using ecological models (Holling 1986) and quantum ethnobotanical models (McClatchey and Bridges 2005) a better understanding of these processes at the most fundamental level can be gained.

Holling (1986) articulated that ecological processes are a cyclical rotation between three phases: organization, collapse and renewal. The renewal (sometimes referred to as the ‘reorganization’) phase is important because that is the phase in which novelty and innovation occur (Holling 1986; Holling et al. 1995). Folke et al. (2004) point out that biodiversity is such an important factor in ecosystem resilience because it plays a major role in renewing and reorganizing ecosystems after

disturbance. There are two important components of the renewal phase which involve the 'release' and 'reorganization' of elemental building blocks of larger systems. Such events correspond with periods of change which are collectively referred to as the 'backloop phase.' Backloop phases are the most neglected and least understood in conventional resource management (Berkes et al. 2003).

Berkes et al. (2003) state that sociocultural systems follow the same cyclical processes described by Holling (1986). This chapter contends that people-plant relationships of social-ecological systems also cycle between processes of co-evolution, co-extinction, back into co-evolution and so on. This process can also be understood in the terms of Holling (1986) described above. The co-evolutionary process can be related to the phases 'organization.' The co-extinction process can be related to 'collapse' phase. The process by which a co-extinction phase cycles back into a co-evolution phase would be analogous with the 'renewal' phase. This chapter further contends that the success of renewal cycles in social-ecological systems are dependent upon the diversity of linked sociocultural-biological relationships. Models provided by quantum ethnobotany allow us to understand what is happening in these processes on the most fundamental level.

Both the co-evolution and co-extinction processes have been discussed above. But how does a co-extinction cycle loop back into a co-evolution cycle? And why, as observed by Winter (2004), are people-plant relationships different at the end of two respective co-evolutionary cycles (as separated by a co-extinction cycle)? Insight may be gained by observing what happens to quantum co-evolution units as they cycle back and forth between co-evolution and co-extinction.

Linking events associated with an intensifying co-evolution process occur in a particular order, and under a certain set of parameter setting conditions. This plays a role in which subunits are linked and when. Examples of parameter setting conditions would be ranges of environment, available biological diversity, and available cultural diversity. The order of linking events work in concert with parameter setting conditions to set a trajectory of co-evolution.

Cleavage events associated with an abating co-extinction process also occur in a particular order, but not necessarily in exactly the reverse order as they were linked. Cleavage events may initiate because the system is being operated under a different set of parameter setting conditions than the set associated with the previous co-evolution process. As these subunits are being separated this new set of conditions will determine which subunits survive long enough to be available for future linking events, and which subunits go extinct—forever taking them out of the pool of possible future linking events.

When an altogether new set of parameter setting conditions come to pass this may induce another co-evolution cycle. This different set of conditions will influence which subunits are involved in a new series of linking events. It is important to note that not the same set of existing QCU will be at the foundation of this new co-evolution cycle as the previous co-evolution cycle. Furthermore, the new set of conditions may yield new subunits previously unavailable in the pool for potential linking events. As the co-evolutionary process continues some of the original subunits that are remaining in a pool of unlinked subunits may be relinked, but not necessarily in the same order as they were lost. This, in conjunction with linking events creating entirely new QCU, will change the structure of the ethnobotanical population and therefore affect trajectory of co-evolution. This is the reason why ethnobotanical populations are most likely to never be the same after going through a co-extinction process, even if it goes back through another co-evolution cycle.

#### ***3.5.4 Maintaining cultural diversity through cyclical evolutionary processes***

While much of the research and theoretical discussion of resilience in social-ecological systems has focused on the importance of biodiversity (Berkes and Folke 1998; Berkes et al. 2003; Walker et al. 2006), it is likely that cultural diversity, as well as linked biocultural diversity, is just as important. This is especially probable when we consider that it is through the broad spectrum of cultural practices that we see management strategies develop that either increase, maintain or decrease biodiversity. This applies to both intra- and inter-cultural diversity within ecosystems. There exists a need to not only manage biodiversity, but also cultural (i.e., tradition/practice) diversity for understanding and maintaining—not to mention the potential to increase—social-ecosystem resilience.

Quantum ethnobotany provides the tools to analyze specific cultural interactions with specific taxa of interest. This, when compared to studies on that taxa's health in a social-ecological system, could give us better understanding of how a particular spectrum of cultural practices affect taxa over time. Quantum ethnobotany could potentially contribute to the answers that resource managers are seeking when making decisions regarding the health and resilience of social-ecological systems.

### **3.6 Conclusions**

#### ***3.6.1 Applications of quantum ethnobotany for conservation of biodiversity***

Before an understanding can be reached as to what is being lost there needs to be an understanding of what exists. Biodiversity is often quantified in social-ecological

systems, however the links between cultural practices and biological taxa have yet to be quantified in such a way as would lend to comparisons across regions and disciplines. The theoretical model this chapter has presented here provides an actual measure of culture and cultural change as it relates to biodiversity and changes in biodiversity. It will help to better understand and manage the cultural practices that both promote and threaten biodiversity. It may also provide a way to early-on identify stress/pressure factors within sociocultural systems, and parts of culture that are under pressure to change and those that are not. Despite what we have said above it is important to keep in mind that social-ecological systems are exceedingly complex. What we are proposing to measure is the minimum of change in order to detect useful generalizations within the complexity of social-ecological systems. Because of expected synergy within complex systems any actual evolutionary change will no doubt be greater than that of the sum of the parts we propose to measure.

### ***3.6.2 Proposed hypotheses***

Just as humans have been directing the evolution of plants through management practices and selection since before the advent of agriculture, humans can also influence the evolution of culture by selecting for cultural practices. For cultures that have lost plant or cultural practice diversity, either may be recreated, but it is difficult to determine if the newly linked QCU is the same or different from those of the past. It also may not matter.

This chapter proposes several hypotheses about human interactions with plants on the basis of the above discussion:

1. Quantum co-evolution units can be used to measure how specific sociocultural practices influence biodiversity within a social-ecological system.
2. There is a set of criteria that can be used to test whether an ethnobotanical population is evolving. If any/all of the criteria are met then the population is evolving. If none are met the population is static. The criteria are:
  - a. Particular QCUs have increased in frequency because of selection.
  - b. Particular QCUs have decreased in frequency because of selection.
  - c. One or more QCU(s) have been added or lost through events homologous to those involved in the process of biological evolution (mutation, extinction, etc.).

- d. One or more QCU subunit(s) has changed (i.e., replacement of a lost or abandoned plant or tradition subunit) resulting in the creation of a new QCU.
3. Co-evolution and co-extinction of plant-culture relationships are cyclical processes and quantum ethnobotany can be used to understand how these affect the trajectory of evolution in ethnobotanical populations.
4. Re-emerging cultures may resurrect traditional recognition of plant diversity and create or borrow practices in order to restore (redevelop) relationships with plants, and therefore social-ecological system resilience.

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## CHAPTER 4. *Kalo* [Hawaiian taro, *Colocasia esculenta* (L.) Schott] Varieties: An assessment of nomenclatural synonymy and biodiversity<sup>3</sup>

### 4.1 Abstract

The prominence of *kalo* (*Colocasia esculenta* (L.) Schott.) in Hawaiian culture has declined after experiencing a decrease in cultivation, biodiversity and associated cultural knowledge. There was no documentation of diversity at its height. Previous estimates of biodiversity lack any sense of a methodological approach. A new attempt was made to assess levels of biodiversity around the peak cultivation period. Results were then compared to current levels. Nomenclatural synonymy and extinction have presented some challenges which made standard methods for quantifying biodiversity not viable. A set of new tools was used to sort through a master list of 676 varietal names. A comparison of what is known from the nineteenth century and modern time periods makes it apparent that changes in biodiversity, varietal prominence, ethnonomenclature, and ethnotaxonomy have occurred. This paper discusses the direction of such trends, and postulates a new estimate for *kalo* diversity at the end of the 19th century (approximately 100 years after the assumed peak of cultivation and diversity) to be between 368-482 distinct cultivars, while only 65-73 still exist today.

### 4.2 Introduction

Taro, *Colocasia esculenta* (L.) Schott, has been historically one of the most important crop plants in the tropical Pacific with distribution reaching as far east as the islands of Polynesia. The global trends in loss of both biodiversity and associated cultural knowledge have not escaped this species throughout its range. The most remote island group of Polynesia is Hawai‘i, where taro is known as *kalo*.

*Kalo* is connected to origin stories of the Hawaiian culture (Handy et al. 1972, Kame‘eleihiwa 1990) and is, therefore, often considered to be the most important crop plant from that perspective, at least on a symbolic level. On a practical level its

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prominence is rivaled only by ‘uala or sweet potato (*Ipomoea batatas* (L.) Lam.) (Handy 1940, Handy et al. 1972, Ladefoged et al. 2009). This is true especially in the era prior to contact with foreigners in 1778 (Handy 1940, Handy et al. 1972, Kamakau 1972, 1976, Kame‘eleihiwa 1990, MacCaughey & Emerson 1913, Malo 2006). Contemporarily, *kalo* continues to be held in the utmost level of respect among many staunch cultural practitioners, although its general prominence in the Hawaiian culture as a whole is no doubt less than has been in previous generations. This is due, in large part, to various reasons such as changes in land tenure and socio-economic systems which, in many cases, resulted in a loss of cultural identity (Kame‘eleihiwa 1992, Trask 1999). Because of the glaring losses of both biodiversity and associated cultural knowledge it is important to gain a more refined understanding of *kalo* diversity at its height for several reasons, not the least of which includes quantifying its importance to the Hawaiian culture and gaining insight into the horticultural skills of the ancient Hawaiians.

The intensity of *kalo* cultivation has changed over time. *Kalo* was originally brought to Hawai‘i in Polynesian voyaging canoes (Abbott 1995) at least 1,000 years ago (Burney & Kikuchi 2006), and the intensity of its cultivation undoubtedly increased as the Hawaiian population did. It is assumed that this period of intensified cultivation was accompanied by a corresponding increase in intraspecies diversity (i.e., cultivar diversity) via somatic mutations (Handy et al. 1972) and possibly even cross-breeding (Handy et al. 1972, Irwin et al. 1998). Cultivation is assumed to have reached its peak at the height of population density around the time of contact with foreign cultures in 1778. At that time the Hawaiian population was estimated to be approximately 1 million people. This period was followed by a 90% population collapse due to introduced diseases (Stannard 1989). The intensity of *kalo* cultivation decreased dramatically during this same period (Anonymous 1879, Cho et al. 2007, Handy et al. 1972, Ladefoged et al. 2009, Müller et al. 2010). During this post peak-cultivation period, several authors attempted to record *kalo* diversity (Handy 1940, Henriques n.d., Iokepa & Kekahuna n.d., Kalaniana‘ole n.d., MacCaughey & Emerson 1913, 1914, Rooke n.d., Whitney et al. 1939, Wight n.d., Wilder 1934), likely in reaction to the extinction crisis that they were witnessing. All of these authors were documenting biodiversity in the late 19<sup>th</sup> and early 20<sup>th</sup> centuries during the period of rapid diversity loss (see below), so in analyzing the works of these authors we could, at best, only gauge post peak-diversity levels.

The terms “cultivar” and “variety” are used throughout this paper. The word “cultivar” is used to describe an anthropomorphically recognized taxa that is genetically distinct from other taxa. The word “variety” is used to describe the name

of a particular cultivar. It is important to note that a single cultivar can have several varietal names that are associated with it. These are considered to be synonyms.

A good measure of a crop's importance to any particular culture is the number of cultivars which are managed by that culture (Rhindos 1984). *Kalo* is no exception to this rule as the number of recorded varieties far surpasses that of any of the other crop-plants cultivated by the ancient Hawaiians (e.g., sweet potato (*I. batatas*), bananas (*Musa acuminata* X *balbisiana* Colla.), breadfruit (*Artocarpus altilis* (Parkinson) Fosberg), yams (*Dioscorea* spp.), and arrowroot (*Tacca leontopetaloides* (L.) Kuntze) (Handy 1940)). A major challenge in assessing biodiversity is the fact that there has been a substantial time-gap between the height of *kalo* cultivation in Hawai'i — when the highest levels of biodiversity are assumed to have existed — and the current era. This has been due, in large part, to a dramatic decrease in both area of *kalo* cultivation and the number of *kalo* farmers. In the course of this time gap most of the varieties have apparently gone extinct. Further complicating the situation is the concurrent decrease in associated cultural knowledge that has accompanied the decline in cultivation. This is perhaps best illustrated in the shift away from Hawaiian as the only language towards English as the only language that accompanied this period (Schütz 1994), and which undoubtedly had a negative influence on the general understanding of the ethnonomenclature and ethnoclassification systems of the Hawaiian language. This knowledge vacuum regarding the traditional names and relationships of the various *kalo* cultivars was eventually filled by horticulturalist-based views on the matter. Since neither the knowledge of traditional taxonomy, nor the high biodiversity levels of *kalo* exists today, there is no standardized way to assess the former levels of biodiversity. Furthermore, there does not seem to be more than a few farmers alive today in all of Hawai'i who represent an unbroken link in the once widespread tradition of maintaining multiple (more than a dozen) cultivars on a family farm.

A major reason for this paucity of traditional farmers is the fact that, for several decades during the last half of the 19<sup>th</sup> century and the first half of the 20<sup>th</sup> century, much of *kalo* farming was done by Chinese and Japanese immigrants rather than Hawaiians (Cho et al. 2007, MacCaughy & Emerson 1913). Even today much of the *kalo* which is currently under cultivation is farmed by the descendants of Chinese, Japanese, and Filipino immigrant laborers; as well as first generation immigrants from the Philippines and Micronesia. In many cases contemporary Hawaiian *kalo* farmers, at some point in the last 150 years, have had at least a single generational break in tradition between themselves and the Hawaiian farmers of old. Because of this, and the now-prolific horticulturist-based views on *kalo* diversity, the general

understanding of *kalo* diversity (i.e., nomenclature and taxonomy) held by contemporary farmers has likely changed in the past 150 years.

In spite of previous attempts to document *kalo* diversity there is still a lack of certainty about the true levels at, or near, the height of cultivation in Hawai‘i. This lack of certainty stems from the information sources either lacking a strong understanding of traditional Hawaiian nomenclature and taxonomy, scientific rigor in their diversity assessments, or both. In order to more accurately assess the true diversity of *kalo* at or near the height of cultivation in Hawai‘i, this study attempts to marry a firm understanding of traditional Hawaiian nomenclature and taxonomy, with a more rigorous assessment of the entire collection of names and descriptions amassed from several different sources.

A major contributing factor for our lack of understanding about previous levels of diversity, aside from the extinction crisis which started in the 19<sup>th</sup> century as described above, is the complexity of synonymy that exists in the body of recorded varietal names. Synonymy is the application of more than one name to a single taxon. Synonymy is known to exist in the Hawaiian language, and has been well documented broadly—in both the lexicon (Pukui & Elbert 1979, 1986) and taxonomic systems (Abbott 1995), and for *kalo* specifically (Chun 1994, 1998, Handy 1940, Iokepa & Kekahuna n.d., MacCaughey & Emerson n.d., 1913, Pukui & Elbert 1986, Wilder 1934, field notes). In a review of the above sources it is apparent that there are two general classes of synonymy in the Hawaiian lexicon — linguistically-based synonymy and classification-based synonymy. Linguistically-based synonymy occurs when different regions which follow the same classification system have differing pronunciations of the same taxon. Such differences correspond to either vowel or consonant deletions, additions or substitutions. As an example, the variety name, *Mākohi*, was also called *Mākohe*, *Mōkohi*, *Mōkohe* and *Mākihi* (Handy 1940, MacCaughey & Emerson n.d., 1913, Pukui & Elbert 1986) in other areas that followed the same classification system. On the other hand, classification-based synonyms occur when one taxon is found in multiple areas, some of which follow differing classification systems. The same taxon, *Mākohi*, was known as ‘*Ele‘ele mākoko* (Iokepa & Kekahuna n.d.) in another classification system, and also as *Maka‘ōpio* (Chun 1994) in yet another. This recognition has influenced the methodological approach, data analysis and conclusions of this chapter.

This chapter is not meant to be the “end all, be all” definitive authority on *kalo* diversity. It is merely an attempt to add data and analysis to the contemporary body of knowledge, and to reawaken the discussion of *kalo* diversity. Recent research has acknowledged the formerly high levels of *kalo* diversity and its subsequent decline

(Cho et al. 2007), but new data and analysis have not been contributed to the debate since Whitney et al. (1939) and Handy (1940).

Although it is not dealt with substantively in this manuscript, it is important to note that many varieties had (and some continue to have) unique and specific traditions associated with them. Examples of these are the now-extinct *Hoene*, which was used medicinally for enemas; and currently-rare '*Apuwai*, whose cup-shaped leaves catch rainwater that is used in ceremonies. Once varieties go extinct, so too die the cultural practices which required those specific taxa for implementation (Chapter 2). Further research may be able to quantify the cultural loss that has accompanied a loss of cultivars.

### 4.3 Methods

This study has three specific assumptions that are based in the idea that there are direct relationships both between biodiversity and linguistic diversity (Nettle & Romaine 2000), and also between biodiversity and cultural diversity (Winter and McClatchey 2008 & 2009). They are as follows:

1. *Kalo* biodiversity and associated cultural knowledge is linked, and this relationship has generally experienced both a coupled increase and a coupled decrease in the past 1,000 years,
2. The height of *kalo* diversity and associated cultural knowledge existed at the height of *kalo* cultivation in Hawai'i, and
3. The height of *kalo* cultivation existed at the point of contact with foreign cultures in 1778.

Since this study attempts to assess a level of intraspecies diversity that no longer exists, readily accepted methodologies for assessing diversity (i.e., field surveys and free-listing exercises to reveal relative abundance/varietal richness resulting in an index of diversity,) are not viable methods. In order to measure change over time, *kalo* diversity has been measured at two points in time: 1) The era ranging from the late 19<sup>th</sup> to early 20<sup>th</sup> centuries (citations ranging from 1879-1940) which represents a period of diversity decline after the presumed height of *kalo* cultivation, via an assessment of nomenclatural diversity; and 2) Contemporarily, via biodiversity assessments. Comparisons between these estimates were then made.

### 4.3.1 Surveys of nomenclatural diversity

Nomenclatural diversity assessments were done beginning with a review of published and archival resources in Hawaiian and English. Published and archival materials were qualitatively analyzed in two areas: adherence to trends in Hawaiian nomenclature and taxonomic systems, and adherence to the scientific method. Trends in ethnonomenclature and ethnotaxonomy were identified by Berlin (1992) and have been demonstrated to be applicable to Proto Oceanic ethnonomenclature and ethnoclassification systems (Evans 2008) which are the evolutionary precursor to Hawaiian nomenclature and classification systems. In Hawaiian systems, Berlin's (1992) trends have been substantiated by the descriptions of both Hawaiian plant experts (i.e., *kahuna lā 'au lapa 'au* or highly skilled herbal healers) (Chun 1994, 1998, Gon 2008) and botanists studying Hawaiian nomenclature systems (St. John 1982). These trends are later used in assessing the quality of citations (see below section, Rigor Assessments for Cited Authorities).

The trends seen documented in sources regarding synonymy in the Hawaiian lexicon (Abbott 1995, Chun 1994, 1998, Handy 1940, Iokepa & Kekahuna n.d., MacCaughey & Emerson n.d., 1913, Pukui & Elbert 1986, Wilder 1934, field notes) were used in assessing synonymy within the nomenclature of *kalo*. An attempt was made to sort through the collection of *kalo* names and descriptions to come to both a liberal and a conservative estimate of the number of distinct cultivars in the post-contact cultivation period, as this is the era when the documentation occurred. The first twenty-three rules (see below), which primarily deal with linguistically-based synonymy in nature, are applied to find the highest end of the estimated range (i.e., a liberal estimate of *kalo* diversity). Rule 23 is derived from the presence of documented varietal names corresponding to proper names of *kalo*-based characters in stories such as the names seen in "The Story of Big Taro and Little Taro" (Anonymous 1861, Nohokuaaina 1867). An additional twelve rules, which relate to adherence to trends in ethnonomenclature and ethnoclassification, are applied to find the lowest end of the estimated range (i.e., a conservative estimate of *kalo* diversity).

Synonymy or other erroneous nomenclature was identified by the presence of the below described conditions. All of the conditions are founded in already documented synonymy (Abbott 1995, Chun 1994, Handy 1940, Iokepa & Kekahuna n.d., MacCaughey & Emerson 1913, 1914, Pukui & Elbert 1979, 1986, Whitney et al. 1939, Wilder 1934), and then applied broadly. If any of these conditions were met the names were either consolidated in or eliminated from the master list. It is important to note that although these synonyms may initially appear questionable, due to their possible meaning-altering appearance, several of these are previously

documented synonyms (i.e., *Kū 'oho* and *Kū 'ohu*), and therefore may represent examples of linguistic engineering for ease of pronunciation.

Vowel substitutions and additions:

1. o/u substitutions (example: *Kū 'oho* / *Kū 'ohu*).
2. a/o substitutions (example: *Hāakea* / *Hāokea*).
3. a/e substitutions (example: *'Ie 'ie* / *'Ia 'ia*).
4. e/i substitutions (example: *Wehiwa* / *Wehewa*).
5. "a" additions on the end of a word (example: *Papakole* / *Papakolea*).
6. "o" additions at the end of a word (example: *Māna* / *Māna-o*).

Consonant substitutions and deletions:

7. n/l substitutions (example: *Māna 'owene* / *Māna 'uwele*).
8. ' /m/n substitutions (example: *Manauea* / *Mamauea* / *Ma 'auea*).
9. "w" deletions (example: *Pualu* / *Puwalu*).

Various classes of reduplication:

10. Reduplication of first vowel (example: *'Apu* / *'A 'apu*).
11. Reduplication of last vowel (example: *Māi 'i* / *Māi 'i 'i*).
12. Full reduplication of both species names (example: *Pia* / *Piapia*), and subspecies epithets (example: *'ula* / *'ula 'ula*).
13. Partial reduplications (example: *Hāpu 'u* / *Hāpu 'upu 'u*).
14. Prefix reduplications (example: *Wehiwa* / *Wewehiwa*).

Backformation:

15. Contractions (example: *'Ele 'ele* / *'E 'ele*).

Miscellaneous:

16. Prefix additions of *hā-* on subspecies epithets (example: *hā 'ula 'ula* / *'ula 'ula*).
17. Similar color descriptors unless indicated as separate cultivars (example: *kea* / *ke 'oke 'o*, *melemele* / *lenalena*, *'ula 'ula* / *lehua*).
18. *Maoli* as type specimen in group, and a *kalo* species with no subspecies epithet associated with it (example: *Hāpu 'upu 'u maoli* / *Hāpu 'upu 'u*).
19. Documented synonymy for group names where subspecies epithets are the same (example: *Piko 'ele 'ele* / *Haehae 'ele 'ele* / *Uaua 'ele 'ele*).
20. Obvious mis-spellings or transcription errors (example: *Haehae ke 'oko 'a* / *H. ke 'oke 'o*).
21. Similar meanings in subspecies epithets using synonymous words/terms (example: *Lehua kū-i-ka-wao* / *Lehua kū-kuahiwi*).
22. Names documented as being assigned to post-contact introductions (example: *Pākē*, *'Iliuaua*).

23. Seemingly proper names (example: *Kalo nui e / Kalo iki e*).

Further consolidations for a conservative estimate, founded in adherence to trends in ethnonomenclature and ethnoclassification, were done if the following conditions occurred:

24. The name is poetic (example: *Ka 'awelu-i-ka-pali-o-Awakea*).
25. The name is a trinomial.
26. The name is seemingly vulgar (examples: *He- 'owā-hulu-nui, Heu-ā-lehu, Heu- 'ele, Pani-kohe*).
27. Group names exist that are noted as synonyms for other crops (example: *Manini / Koa 'e, Mākea / Mahakea*).
28. *'Ula 'ula* as a group name is a synonym for the *Kūmū* group.
29. *'Ele 'ele* and *Nohu* as group-names are synonyms for the *Naioea* group.
30. *'Ala* as a group name is a synonym for the *Kāi* group.
31. A lack of documentation of the word "*maoli*" (meaning "type-specimen") in the subspecies epithet (example: for the *'Apuwai* group three names are collectively recorded - *'Apuwai, 'A. ke 'oke 'o* and *'A. 'ula 'ula*. These will be counted as two cultivars instead of three. For the *Hāpu 'u* group six names are collectively documented - *Hāpu 'u maoli, H. 'ele 'ele, H. kea, H. lenalena, H. nūkea, and H. 'ula 'ula*. These were counted as six cultivars).
32. The name is a monomial which is also the name of an *ahupua 'a* (traditional land-division unit). Such names may have been given to honor the cultivar's place of origin when cultivars were transferred in the absence of knowledge about their original name.
33. Species-level names listed as monomials are sub-species epithets noted elsewhere in a binomial (*Lauloa papamū / Papamū*).
34. Species-subspecies nomenclatural reciprocations (*'Ula 'ula poni, Poni 'ula 'ula*).
35. The various monomials which vaguely indicate either a black-colored or striped petiole are considered synonyms for other varieties accounted for elsewhere in the list (*Pōpolo, Hiwa; Kāni 'o, Ni 'o*).

Data on the occurrence of documented synonymy for each respective source were also recorded. The percent age of synonymy was calculated for each source using the following equation:

$$ns / nt = \text{synonymy percentage,}$$

where,  $n_s$  is the number of varietal names documented as having a synonym, and  $n_t$  is the total number of varietal names documented.

#### **4.3.2 Biodiversity assessment**

*Kalo* collections of botanical gardens, agriculture stations, and private individuals/families (including both commercial and subsistence farms) were surveyed on the islands of Kaua‘i, O‘ahu, Moloka‘i, Maui, and Hawai‘i between the years of 2003-2012. Accession information for living specimens was reviewed (if available), and the specimens' morphological traits were compared against the descriptions of Whitney et al. (1939) to confirm accuracy. Through the generosity of these entities and individuals, collections were made for propagation on the island of Kaua‘i where a common garden experiment was conducted under a nursery setting between 2011-2012. The plants in the experiment were verified using Whitney et al. (1939), and also by recognized community experts on *kalo* diversity (Jerry Konanui & Penny Levin). Voucher specimens have been prepared of these varieties that have been deposited in the National Tropical Botanical Garden herbarium (PTBG), Kalaheo, Kaua‘i, Hawai‘i.

#### **4.3.3 Rigor assessments for cited authorities**

Qualitative cultural and scientific rigor assessments were conducted. Cultural rigor was weighed against adherence to trends in Hawaiian nomenclature systems. Scientific rigor was weighed against whether or not data collection was qualitative or quantitative, and if comparative analysis was done.

Cultural rigor was assessed and given designations by the following criteria:

- High – Authors have produced other manuscripts and publications regarding ethnonomenclature and ethnoclassification.
- Medium – Authors did not produce other manuscripts and publications regarding ethnonomenclature and ethnoclassification, but data produced follows the trends articulated by Berlin (1992).
- Low – Authors did not produce other manuscripts and publications regarding ethnonomenclature and ethnoclassification, and data produced is in conflict with the trends articulated by Berlin (1992).

Scientific rigor was assessed and given designations by the following criteria:

- High – Collected quantitative data regarding morphology in a common garden experiment, and did comparative analysis of this data.
- Medium – Collected qualitative data regarding morphology in a common garden experiment, and did comparative analysis of this data.
- Low – Collected qualitative data regarding morphology from different locations and environments for comparative analysis.
- Non-existent – No evidence of either data collection regarding morphology or comparative analysis.

#### ***4.3.4 Status assessments for once-prevalent cultivars***

Status assessments for once-prevalent cultivars were done based on the above described field observations. Field status was assessed and given designations by the following criteria:

- Common – Cultivar was documented in commercial farms, family farms/gardens, and botanical collections.
- Occasional – Cultivar was not documented in commercial farms, but was documented in family farms/gardens, and botanical collections.
- Rare – Cultivar was neither documented in commercial farms nor family farms/gardens, but was documented in botanical collections.
- Extinct – Cultivar was not documented in commercial farms or family farms/gardens, and there is no evidence of it existing in botanical collections.

## **4.4 Results**

### ***4.4.1 Previous estimates of synonymy***

Fourteen documents (including published and unpublished archival-manuscripts) relating to names and descriptions of *kalo* diversity were assessed. These were written in Hawaiian and English. The document dates ranged from 1879-1986. The most recent of these (Pukui and Elbert 1986) was a compilation of previous works, not a representation of field work. Future mention of these document dates will

refer to 1879-1940 which represents the range of time that field data was collected. Five of the manuscripts have no date associated with them, but judging from their presence in the Bishop Museum Archives they are likely from the same period. From the lists within these, a total of 669 varietal names of *kalo* were accumulated. Seven more were added from oral history documents representing a total of 676 names. Typographical errors and non-Hawaiian varieties (i.e., Asian and South Pacific cultivars that were given adoptive Hawaiian-names) were removed reducing the list to 598 varietal names. Linguistically-based synonyms were then consolidated to 512 varietal names (Appendix 1) that were used for further analysis including a calculation of synonymy. The manuscripts documented synonymy with a range from 0 - 80% (Table 4.1).

**Table 4.1.** Publications and manuscripts that documented *kalo* (*Colocasia esculenta* (L.) Schott.) diversity in the Hawaiian islands. Wight (n.d.) was used as a reference by Handy (1940) to make his list of varietal names. This work has been missing from the Bishop Museum Archives since the 1980s and was therefore unavailable for analysis.

<b>Authority</b>	<b>Language: Hawaiian or English</b>	<b>Number of listed cultivar</b>	<b>Synonymy Percentage</b>
Henriques n.d.	Hawaiian	34	0%
Iokepa and Kekahuna n.d.	Both	92	26%
Kalaniana'ole n.d.	English	26	0%
Rooke n.d.	English	42	0%
Wight n.d.	N/A*	N/A*	N/A*
Anonymous 1879	Hawaiian	37	0%
MacCaughy and Emerson 1913	English	272	20%
MacCaughy and Emerson 1914	English	298	08%
Fornander 1919	Both	30	0%
Wilder 1934, n.d.	English	95	35%
Whitney et al., 1939	English	141	80%
Handy 1940	English	354	12%
Pukui and Elbert 1986	English	235	25%
Chun 1994	Both	31	29%

#### **4.4.2 Field surveys**

Field surveys of botanical gardens, agricultural stations, and individual collections (both commercial and subsistence farms) throughout Hawai'i have determined that there has indeed been a decline in *kalo* diversity, even in the recent historical

period. At least 63 of the heritage *kalo* cultivars documented in Whitney et al. (1939) remain. In addition to these, nine more have been added to the list of confirmed Hawaiian varieties. These include two Hawaiian cultivars were added via hybridization in the 20<sup>th</sup> century (Maui *Lehua* which is a cross between *Lehua maoli* and *Pi'iali'i*, and *Pili-aloha* which is a cross between *Kāi kea* and *Moi*), one, *Moi 'ula*, which was documented by other sources (Handy 1940, Iokepa & Kekahuna n.d., Pukui & Elbert 1986, Wilder 1934), and one previously undocumented cultivar, *Pi'ikea*. The remaining five (*'Elepaio hāuliuli*, *Kāi KBS*, *Lauloa koukou'ai*, *Makalau*, and *Niumalu*) do not match with documented descriptions, but are debatably Hawaiian. A possible rediscovery of *Lauloa 'ōni'oni'o* via somatic mutation (not yet confirmed) brings the total number of remaining Hawaiian cultivars to 73 (Table 4-2). Outside of the Hawaiian cultivars, at least 71 cultivars have been introduced from around the Pacific and Asia. In addition to these, literally several thousand cultivars have been created since the 1980s via both officially sanctioned and unofficially sanctioned hybridization experiments (including hybrid crosses) conducted by University of Hawai'i researchers and affiliates using Hawaiian and non-Hawaiian cultivars. Countless numbers of these have been distributed to farmers, but there has not been rigorous documentation about their morphology and distribution. Due to the nature of this complexity, an analysis of these hybrids was excluded from this study.

Field surveys to assess collections of botanical gardens and University of Hawai'i agriculture stations took place on the islands of Kaua'i, O'ahu, Moloka'i, Maui and Hawai'i which represented broad ranges in environmental conditions. These surveys occurred between 2003 and 2012. Accession information has revealed that the vast majority of these collections trace back to the original collections used to produce the Whitney et al. (1939) publication.

#### **4.4.3 Rigor of previous diversity assessments**

All of the sources documenting *kalo* diversity were qualified as to the rigor of their cultural knowledge as indicated by their adherence to trends in Hawaiian nomenclature systems, and scientific rigor as indicated by scientifically-based data collection and analysis (Table 4.3). No source ranked high in both categories.

**Table 4.2.** The extant diversity of kalo (*Colocasia esculenta* (L.) Schott) representing documented cultivars, 20<sup>th</sup> century Hawaiian X Hawaiian hybrids, and undocumented cultivars \*strongly-to-debatably assumed to be Hawaiian.

<b>Cultivar Name</b>	<b>Documented in Whitney et al. 1939</b>	<b>Modern Hybrid</b>	<b>Not in Whitney et al. 1939, but assumed to be Hawaiian</b>
<i>‘Āpi‘i / Moana</i>	1	0	0
<i>‘Apowale</i>	1	0	0
<i>‘Apu</i>	1	0	0
<i>‘Apuwai</i>	1	0	0
<i>‘Āweuweu / ‘Āhē</i>	1	0	0
<i>‘Ele‘ele lauloa</i>	1	0	0
<i>‘Ele‘ele mākoko</i>	1	0	0
<i>‘Ele‘ele naioea</i>	1	0	0
<i>‘Elepaio (kea)*</i>	1	0	0
<i>‘Elepaio hāuliuli</i>	0	0	1
<i>Hāokea</i>	1	0	0
<i>Hāpu‘u</i>	1	0	0
<i>Kāi ‘ala</i>	1	0	0
<i>Kāi kea</i>	1	0	0
<i>Kāi uliuli</i>	1	0	0
Kāi unknown (a.k.a., Kaua‘i Branch Station)	0	0	1
<i>Kalalau</i>	1	0	0
<i>Kūmu ‘ele‘ele</i>	1	0	0
<i>Kū‘oho*</i>	1	0	0
<i>Lauloa ‘ele‘ele</i>	1	0	0
<i>Lauloa ke‘oke‘o</i>	1	0	0
<i>Lauloa ‘ōni‘oni‘o**</i>	0	0	1
<i>Lauloa palakea</i>	1	0	0
<i>Lauloa papamu</i>	1	0	0
<i>Lauloa pakakea-ula</i>	1	0	0
<i>Lauloa koukou‘ai</i>	0	0	1
<i>Lehua ‘āpi‘i</i>	1	0	0
<i>Lehua maoli</i>	1	0	0
<i>Lehua palai‘i</i>	1	0	0
<i>Lihilihimolina*</i>	1	0	0
<i>Maea</i>	1	0	0
<i>Makalau</i>	0	0	1
<i>Mana ‘ele‘ele</i>	1	0	0

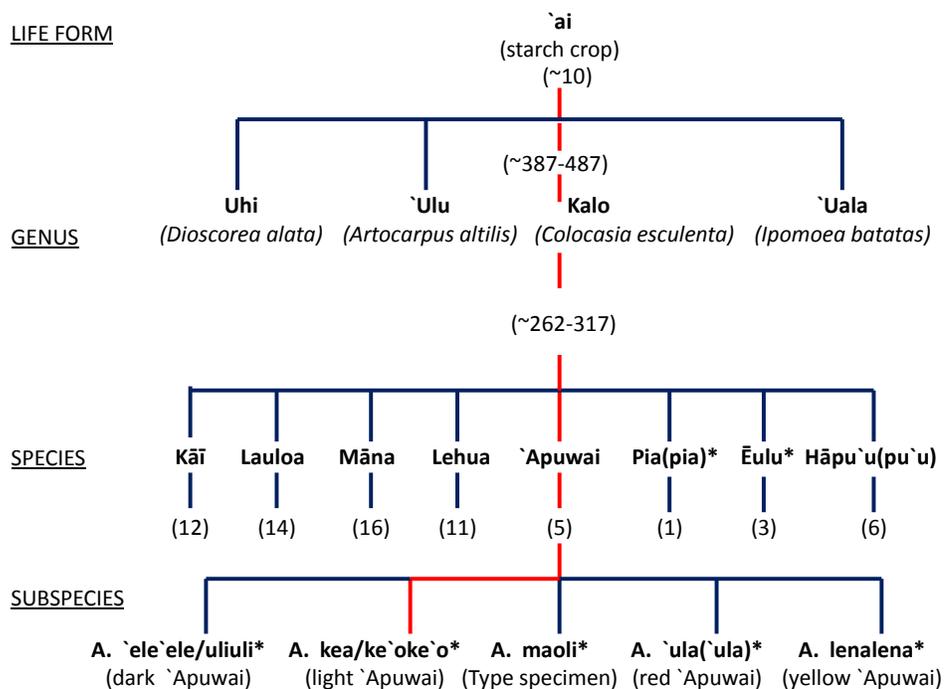
<i>Mana ke'oke'o</i>	1	0	0
<i>Mana lauloa</i>	1	0	0
<i>Mana 'oko'a</i>	1	0	0
<i>Mana 'ōpelu</i>	1	0	0
<i>Mana 'ula'ula</i>	1	0	0
<i>Mana uliuli</i>	1	0	0
<i>Mana 'ulu</i>	1	0	0
<i>Mana-piko</i>	1	0	0
<i>Manini kea</i>	1	0	0
<i>Manini 'ōpelu</i>	1	0	0
<i>Manini 'owali</i>	1	0	0
<i>Manini uliuli</i>	1	0	0
<i>Maui Lehua</i>	0	1	0
<i>Moi (kea)</i>	1	0	0
<i>Moi 'ula'ula</i>	0	0	1
<i>Nāwao</i>	1	0	0
<i>Nihopu'u</i>	1	0	0
<i>Niumalu</i>	0	0	1
<i>'Oene</i>	1	0	0
<i>'Ohe</i>	1	0	0
<i>'O'opukai</i>	1	0	0
<i>Pa'akai</i>	1	0	0
<i>Papapueo</i>	1	0	0
<i>Pi'iali'i</i>	1	0	0
<i>Pi'ikea</i>	0	0	1
<i>Piko 'ele'ele</i>	1	0	0
<i>Piko kea</i>	1	0	0
<i>Piko ke'oke'o</i>	1	0	0
<i>Piko uaua</i>	1	0	0
<i>Piko 'ula'ula</i>	1	0	0
<i>Piko uliuli</i>	1	0	0
<i>Pili-aloha</i>	0	1	0
<i>Pololū</i>	1	0	0
<i>'Uahi-a-pele</i>	1	0	0
<i>'Ula'ula kūmū</i>	1	0	0
<i>'Ula'ula moano</i>	1	0	0
<i>'Ula'ula poni</i>	1	0	0
<i>Waiākea</i>	1	0	0
<i>Wailana</i>	1	0	0
<i>Wehiwa</i>	1	0	0
<b>TOTALS:</b>	<b>63</b>	<b>2</b>	<b>8</b>

**Table 4.3.** Cultural and scientific rigor of data sources used to qualitatively assess *kalo* (*Colocasia esculenta* (L.) Schott.) diversity in the Hawaiian islands. Cultural rigor was weighed against adherence to trends in Hawaiian nomenclature systems. Scientific rigor was weighed against the scientifically-based data collection and analysis. Wight (n.d.) was used as a reference by Handy (1940) to make his list of varietal names. This work has been missing from the Bishop Museum Archives since the 1980s and was therefore unavailable for analysis.

Authority	Cultural rigor	Scientific rigor
Henriques n.d.	Low	Non-existent
Iokepa and Kekahuna n.d.	High	Medium
Kalaniana'ole n.d.	Medium	Low
Rooke n.d.	Medium	Non-existent
Wight n.d.	N/A*	N/A*
Anonymous 1879	Medium	Low
MacCaughey and Emerson 1913	Medium	Low
MacCaughey and Emerson 1914	Medium	Low
Fornander 1919	High	Low
Wilder 1934, n.d.	Medium	Medium
Whitney et al., 1939	Low	High
Handy 1940	High	Medium
Pukui and Elbert 1986	Medium	Low
Chun 1994	High	Low

#### 4.4.4 Taxonomic trends

Traditional Hawaiian nomenclature and taxonomy as gleaned from the works produced in collaboration with recognized Hawaiian plant experts (i.e., those designated as having “high” cultural rigor in Table 4.3) conformed to the trends in ethnobiological classification as articulated by Berlin (1992). This is illustrated by Figure 4.1 as a contextual example, and in Table 4.4 as an isolated example.



**Figure 4.1.** Classification of the Hawaiian *kalo* (*Colocasia esculenta* (L.) Schott.) cultivar, ‘*Apuwai kea*, as an example in the context of other taxa following Berlin’s (1992) system. The bracketed numbers for the generic level represent the documented diversity found in Handy (1940) except for *kalo* that is from this research. The bracketed numbers for the specific level represent former, not current, diversity. The current diversity is lower at the specific level due to extinction. Generic and specific taxa were chosen to demonstrate the range of diversity. Presumed extinct taxa noted with \*.

**Table 4.4.** Classification of the Hawaiian *kalo* (*Colocasia esculenta* (L.) Schott.) cultivar, ‘*Apuwai kea*, following Berlin’s (1992) general system.

<b>Berlin’s Taxonomic Hierarchy</b>	<b>Hawaiian Example</b>
Kingdom	<i>Lā`au</i>
Life Form	<i>`Ai / Meakanu</i>
Genus	<i>Kalo</i>
Species	<i>`Apuwai</i>
Sub-species	<i>Kea</i>

#### 4.4.5 Trends in nomenclature

One of the trends seen in the data is that many of the species-level names fall into one of the following categories.

Fish names (at least 26 species-level names)

- ‘*Akilolo* – yellow-tail wrasse (*Coris gaimard* Quoy & Gaimard, 1824)
- *Kūmū* – whitesaddle goatfish (*Parupeneus porphyreus* Jenkins, 1903)
- *Manini* – convict tang (*Acanthurus triostegus* L., 1758)

Morphological trait (at least 17 species-level names)

- ‘*Apuwai* – “cup-shaped leaf”
- *Lauloa* – “long leaf”
- *Māna* – “branching corm”

Forest plants (at least 15 species-level names)

- ‘*Ahakea* – *Bohea elatior* Gaudich., a hardwood tree
- *Hāpu ‘u(pu ‘u)* – *Cibotium* spp., a tree fern
- *Lehua* – *Metrosideros collina* (J.R. Forst. & G. Forst.) subsp. *polymorpha* (Gaudich.) Rock, a hardwood tree

Shared names with other crops varieties (at least 15 species-level names)

- *Kawelo* – species-level name for a sweet potato (*I. batatas*) variety
- *Loha* – species-level name for a banana (*M. acuminata* X *balbisiana*) variety
- *Mahakea* – species-level name for an ‘*awa* (*Piper methysticum* G. Forst.) variety

Group characteristic (at least 10 species-level names)

- ‘*Ala* – “fragrant when cooked”
- ‘*Apowale* – “easy to harvest”
- *Uaua* – “hard to harvest”

Seemingly esoteric (at least 9 species-level names)

- *Mākohi* – ambiguous meaning
- *Naioea* – unknown meaning
- *Pi ‘iali ‘i* – “ascending royalty”

Shared name with an ahupua‘a or land division (at least 5 species-level names)

- *Kahalu ‘u* – an *ahupua ‘a* name found on two islands (O‘ahu and Hawai‘i)
- *Kalalau* – an *ahupua ‘a* on the island of Kaua‘i
- *Pololū* – an *ahupua ‘a* on the island of Hawai‘i

Bird names (at least 5 species-level names)

- ‘*Elepaio* – monarch flycatcher (*Chasiempis* spp.)
- *Koa‘e* – tropic bird (*Phaethon* spp.)
- *Pueo* – Hawaiian short-eared owl (*Asio flammeus sandwichensis* A. Bloxam, 1827)

Predominant petiole color (at least 5 species-level names)

- ‘*Ele‘ele* – “blackish”
- *Poni* – “purplish”
- ‘*Ula‘ula* – “reddish”

Genus-level names of other Polynesian-introduced plants (at least 4 species-level names)

- ‘*Ohe* – Polynesian bamboo (*Schizostachyum glaucifolium* (Rupr.) Munro)
- ‘*Ōlena* – turmeric (*Curcuma longa* L.)
- *Pia(pia)* – arrowroot (*T. leontopetaloides*)

Religious connotations (at least 4 species-level names)

- *Pā‘ū-o-Hi‘iaka* – “skirt-of-Hi‘iaka”
- *Piko-o-Wākea* – “navel-of-Wākea”
- ‘*Uahi-a-Pele* – “smoke-of-Pele”

A trend in subspecies-level nomenclature is that they mostly correspond to petiole color. Of the 295 subspecies epithets in the data (Appendix 1), 178 or 60% are undoubtedly given their epithet designations due to their petiole coloration (Table 4.5).

## 4.5 Discussion

### 4.5.1 Hawaiian taxonomic systems and synonymy

In the context of traditional Hawaiian nomenclature systems, synonymy—especially among the islands—was prevalent for useful plants both wild and cultivated (Abbott 1995, Chun 1994, Pukui & Elbert 1986). Before the time of Kamehameha’s consolidation of the five kingdoms into one (i.e., the Kingdom of Hawai‘i, around the turn of the 19<sup>th</sup> century) under his rule, each kingdom had their own systems to understand their world — including calendars and taxonomic systems (Handy et al. 1972). Despite differing levels of endemism (for both wild and cultivated plants) on each island, each island seemed to be dealing with the same core set of useful wild and cultivated plants. However, since each kingdom had its own system for understanding and working with the natural world, many plants had not only

different names, but also different relationships (i.e., classifications) within taxonomic systems on the different islands (i.e., former kingdoms). The data collected and analyzed in this paper originated in different classification systems, documentation of which is spotty at best. This has added to the complexity of data analysis.

**Table 4.5.** Trends in nomenclature for 255 *kalo* (*Colocasia esculenta* (L.) Schott.) sub-species.

Subspecies Epitaph Designation	Amount Documented
Often ambiguous, singly-occurring epitaphs	57
Type specimen ( <i>maoli</i> ), assumed	46
Blackish petiole ( <i>ʻeleʻele</i> , <i>hiwa</i> , <i>uliuli</i> , <i>hāuliuli</i> )	39
Whitish petiole ( <i>keʻokeʻo</i> , <i>kea</i> , <i>hakeʻokeʻo</i> )	37
Reddish petiole ( <i>ʻulaʻula</i> , <i>lehua</i> , <i>hāʻula</i> , <i>hāʻulaʻula</i> , <i>koko</i> )	33
Yellowish petiole ( <i>lenalena</i> , <i>lena</i> , <i>hālenalena</i> , <i>ʻulu</i> , <i>ʻoene</i> )	11
Purplish petiole ( <i>poni</i> )	5
Leaf cut to sinus ( <i>piko</i> )	4
Streaked/blotched petiole ( <i>manini</i> , <i>ʻōniʻoniʻo</i> , <i>hāniʻoniʻo</i> )	3
Type specimen ( <i>maoli</i> ), specified	3
Red vascular hub on leaf ( <i>piko ʻula</i> )	3
Mackerel scad - <i>Decapterus</i> spp. ( <i>ōpelu</i> )	3
Branching corm ( <i>māna</i> )	3
Streaked-sunset colored petiole ( <i>welowelolā</i> , <i>welowelokā</i> )	2
Unknown meaning ( <i>ʻāniholoa</i> )	2
Ambiguous meaning ( <i>pipika</i> )	2
Long leaf ( <i>lauloa</i> )	2
TOTALS	255

#### **4.5.2 The complexity of synonymy and previous estimates of *kalo* diversity**

Like the taxonomic system for native plants, some *kalo* varieties have different names on different islands. They also may be positioned differently within each island's respective taxonomic system (Handy 1940, Handy et al. 1972, MacCaughy & Emerson 1913).

The amount of synonymy within the collection of varietal names can cause much confusion. Collectors sometimes noted synonymy, but undoubtedly were not able to document every case. Furthermore, several contradictions exist in records of

synonymy and descriptions. As an example the variety, *Pi 'iali 'i*, is listed by Wilder (1934) as having the following synonyms: *'Ahapi 'i*, *Moi 'ula*, and *Mākohe*. Of these, *Mākohe* is listed as a synonym for *Mākohi* by MacCaughey and Emerson (1913, 1914). *Mākohi* is listed as a synonym for *'Ele 'ele mākoko* by Iokepa and Kekahuna (n.d.), but Whitney et al. (1939) have treated *'Ele 'ele mākoko* and *Pi 'iali 'i* as completely different and unrelated cultivars. Furthermore, *Mākohi* is listed as a synonym for *Maka 'ōpio* by Chun (1994), but Whitney et al. (1939) treats *Maka 'ōpio* as a synonym for *Piko 'ele 'ele*, *Haehae 'ele 'ele*, *Hele-mauna*, and *Ipu-o-lono* — each of which is listed as further synonyms for yet other varieties by the other authors, and the complexity continues on. Because of the contradictions that exist between the authors, the synonymy that they document was left out of the analysis of data accumulated here (Appendix 3), and linguistics and ethnoclassification became the primary tools for analysis.

Previous authors expressed different thoughts and conclusions on the matter of synonymy. Handy (1940) did not think that *kalo* had the same high level of synonymy as *'uala* (sweet potato):

“...I am inclined to think that there is far less duplication of names than might be expected... The taros are more uniform in coloring than the sweet potatoes. Their cultivation was an all-year science, while sweet potato planting was spasmodic. The taro farmer was a systematic gardener, the sweet potato planter a casual farmer. Furthermore, taros are less easily transported for replanting than sweet potatoes. Hence there was great exactitude in nomenclature and less renaming—when the original name was unknown or forgotten—by the giving of a new descriptive name or the name of the person who brought in a new variety or created one from bud mutations.” (Handy 1940)

Handy (1940) did not, however, state an estimated level of synonymy. Both MacCaughey and Emerson (1913), and Whitney et al. (1939) agreed that about 50% of collected varietal names could be considered synonyms (this will be referred to as the “50% Rule” from hereafter). However, this 50% Rule that they put forward seems to have no foundation as the estimate since it is not reflected in an analysis of their own data (Table 4.1), and they provided no other reasons for the estimate. In fact, an analysis of the data collected by the various authors reveals that they documented synonymy ranging from eight (8) to 80% (Table 4.1). This range is too broad to pin down even an approximation of actual synonymy that existed at or near the height of cultivation, and therefore further analysis was needed.

Previous works relating to the subject were assessed in an attempt to sort through the inconsistent nomenclature, synonymy, and taxonomic treatments. One problem is that several of the early authors (Anonymous 1879, Henriques n.d., Iokepa & Kekahuna n.d., Kalaniana'ole n.d., MacCaughey & Emerson 1913, 1914, Rooke n.d.) did not use a standardized method for collection or description. These sources range from only a collection of names with no descriptions, to a list of either vague or at best non-scientifically substantiated descriptions. It wasn't until Wilder (1934) and Handy (1940) that attempts were made to approach documenting diversity in a scientific manner. However, the data that they produced was qualitative and the rigor of their approach is questionable. Finally, Whitney et al. (1939) attempted to compile a comprehensive collection of names and scientifically quantified descriptions which were based on a common garden experiment. However, Whitney et al. (1939) state, regarding their study, that many of the Hawaiian *kalo* cultivars had already gone extinct or were so rare that they were not located. Whitney et al. (1939) tried to sort through synonymy via their common garden studies. Their conclusions lumped together several varieties that previous authors noted as being clearly distinct by using characteristics that do not change based on environmental conditions (e.g., petiole-base color). Specifically, Whitney et al.'s (1939) taxonomic treatment was in contradiction to those of Handy (1940), Wilder (1934) and Anonymous (1879) whose descriptions note greater differences between some of the cultivars, that Whitney et al. (1939) lumped together, than can be attributed to environmental influences (Winter, Field Notes 2003-2012). It is likely that the high level of synonymy (80%) reported by Whitney et al. (1939) is not representative of the level of synonymy that existed at the height of cultivation. This figure is in fact substantially higher than any of the other authors who documented synonymy (6-26%), as well as Whitney et al.'s (1939) own stated estimate of 50%.

MacCaughey and Emerson (1913) were the first to put forward an estimate of *kalo* diversity at the height of cultivation. They asserted (presumably using their 50% synonymy estimate) that there may have been between 150-175 cultivars represented by approximately half of the names they collected. Handy et al. (1972) represented perhaps the most thorough examination of *kalo* ethnobotany, and estimated that there were once at least 300 cultivars. They qualify that there was a relatively small number of culturally salient cultivars that could be found throughout the Hawaiian islands, and further attribute the remainder of the diversity to the high number of localized cultivars occurring as endemics within various districts. Despite these estimates, there has been some hesitation to accept either of them because of the complex synonymy found within traditional *kalo* taxonomy. As a result of this hesitation there is no general agreement on even an

approximate number of cultivars at the height of *kalo* diversity, although the diversity is generally qualified as high.

Iokepa and Kekahuna (n.d.) documented 92 varietal names that are the most accurate collection in terms of adherence to Hawaiian binomial-nomenclature trends. Iokepa was a lifelong *kalo* farmer, and well-respected elder of his area while Kekahuna was a learned Hawaiian who dedicated his life to documenting Hawaiian knowledge systems. Kekahuna's method was to seek out the most well-respected elders in a given area and document their knowledge (Kekahuna 1956). An analysis of Iokepa and Kekahuna's varietal list shows 26% synonymy. The sample size (92) and the level of cultural knowledge attributed to both Iokepa and Kekahuna points to a high level of accuracy. It is likely that 26% represents the best approximation for the actual synonymy level that existed at or near the height of *kalo* cultivation and, therefore, will henceforth be called the "26% Rule".

Secondary to the Iokepa and Kekahuna (n.d.) manuscript, is the work of Chun (1994). It also represents a high level of cultural knowledge in that the manuscript was a collaborative effort among *kahuna lā 'au lapa 'au* (highly skilled herbal healers) who are considered plant experts within the culture. Chun (1994) does not contain a comprehensive list of *kalo* varieties, but rather a list of 31 varieties that are used medicinally. Chun (1994) is considered secondary to Iokepa and Kekahuna (n.d.) because the report is of a relatively small subset of varieties. However, Chun (1994) does support Iokepa and Kekahuna's (n.d.) binomial nomenclature system, as well as synonymy level (29%), which will henceforth be called the "29% Rule".

Part of the discrepancy between the three rules may stem from the sources accounting for different classes of synonymy. Perhaps, the 50% Rule proposed by MacCaughey and Emerson (1939) and Whitney et al. (1939) stems from adding both linguistically-based and classification-based synonyms together. Either way, there is no data that support this estimate so it will not be used in further analysis. The other two rules—26% based on Iokepa and Kekahuna (n.d.), and 29% based on Chun (1994)—clearly only relate to classification-based synonymy. When the 26% Rule and the 29% Rule were applied to the list of 512 linguistically-consolidated names, an initial estimate was obtained which produced a possible range of 364-379 distinct cultivars. This maybe a good starting point for estimating a conservative and liberal range of *kalo* diversity; however this is just a cursory analysis. Therefore, this work puts forth a more systematic approach using linguistic tools, in concert with trends in ethnonomenclature and ethnoclassification (Berlin 1992), to assess diversity.

### **4.5.3 Taxonomic and nomenclature trends**

As Berlin (1992) points out, human cultures have systemized ways of classifying the biological world around them. Hawaiian culture is no exception. The system laid out by Berlin seems to describe well the way in which Hawaiians apply classification, taxonomy and nomenclature. Hawaiian nomenclature at the finest level is mostly binomial. Names applied to *kalo* cultivars in Hawai‘i correspond to Berlin (1992) (Table 4.4). About 92% of the 652 recorded *kalo* varietal names are binomials (or monomials that fit into a binomial system). This certainly confirms to the binomial trend seen in publications produced in collaboration with herbal healers (Chun 1994, 1998, Gon 2008), and recognized by botanists who studied Hawaiian nomenclature systems (St. John 1982). Only two (2)% (14 names) of the entire collection are trinomials. There are three likely explanations for these trinomials:

1. They are names that were incorrectly recorded.
2. They reflect the extreme importance of *kalo* to Hawaiian culture resulting in development of a taxonomic level below subspecies.
3. They are a part of a taxonomic system created by scientists who did not adhere to Hawaiian nomenclature traditions.

Seven of 14 (50%) of the recorded trinomials are found in (Whitney et al. 1939). The others could be either misspellings, or debatable binomials. Whitney et al. (1939) admittedly created a system that achieved their goal of a dichotomous key, and this could have included creating the trinomials seen in the *lau loa* group (Appendix 3).

The remaining 5% of names are questionable, poetic names that stem mostly from Henriques (n.d.). These are likely to be proper names of *kalo*-based characters in stories rather than cultivar names.

### **4.5.4 Nomenclature trends at the species and subspecies levels**

Different trends emerged regarding naming at both the species and subspecies level. Species-level names include monomials with no clear relationship to other cultivars, and the group name for a set of binomials. In regards to monomials, many have no documented meaning or obvious source. These, perhaps, represent taxa that were named after people (Handy 1940). For the remainder of the monomials, a few have religious connotations indicating a possible role in ceremony. The remaining monomials indicate a strong tendency for either naming after a morphological

resemblance to something in nature (reef fish, forest plant, forest bird, or other cultivated plants). If the name is binomial there is a pattern of the group name (i.e., species-level name for a set of subspecies) being given due to common morphological trait (such as branching-corm, or long-leaf) or other characteristic of the group (such as fragrant-when-cooked, or predominantly-black petiole).

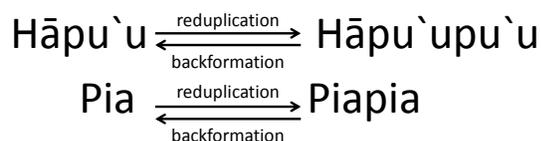
In examining the subspecies epithets a different trend emerges. The majority (60%) of the subspecies epithets are color designations. This indicates that the ancient Hawaiians taxonomically grouped *kalo* together according to a common morphological trait, and separated out individuals that were different color variants of that common trait. About 16% of subspecies are either specifically indicated with the epithet, *maoli*, or otherwise assumed to be classified as such. “*Maoli*,” is defined in Hawaiian as “Native, indigenous, aborigine, genuine, true, real, or actual” (Pukui & Elbert 1986:240). In effect, this is a Hawaiian term for the original or type specimen. The data reflects that subspecies outside of this type specimen are generally distinguished from one another by the predominant petiole coloring. This might indicate that the type-specimen is the original form from which somatic mutations produced subspecies that retain the general morphology, but differ in petiole color. If so, then the most common mutation colors are (in descending order) blackish, whitish, reddish, yellowish, and purplish (Table 4.5). Since it is likely that many have gone extinct, and the remaining cultivars represent a severe genetic bottleneck, so we can only speculate about whether or not the data reflect a genetic reality.

#### **4.5.5. The relationship of reduplication and backformation in synonymy**

While this paper identifies synonymy trends in data, it is beyond the scope to identify the linguistic foundations of this synonymy (such as various kinds of vowel substitutions and consonant substitutions). However, the relationship between reduplications or backformations is one form of synonymy that may be explored since it directly relates to physical morphologies differentiating taxa (Figure 4.2). The question naturally arises, which came first the reduplication or the backformation (and how does this relate to why these taxa were named as such). Ross (2008) observed that, in Proto Oceanic (an evolutionary precursor to Hawaiian), reduplications were common when naming a plant that resembles another in some way, but is not the original. In Figure 4.2, *Hāpu ‘u* and *Hāpu ‘upu ‘u* are treated as synonyms. *Hāpu ‘u* is a native tree fern (*Cibotium* spp.). It has a dark trunk and green fronds. The *kalo* of the same name (*kalo Hāpu ‘u*) has dark petioles and green leaves. The *kalo* was named in reference to the tree fern. If the trends of Proto Oceanic carry forward into the Hawaiian language, then the original name

given to the *kalo* would have been *Hāpu`upu`u* as it resembles, but is clearly not the tree fern. Over time, presumably for ease of pronunciation, a backformation likely took place, making *Hāpu`u* a synonym for the original, *Hāpu`upu`u*. The second example in Figure 4.2, *Pia* and *Piapia* cannot be confirmed since *Piapia* is extinct and unavailable for morphological analysis. However, if the above example is correct then *Piapia* might have been a *kalo* with a taste/consistency (or other character) that resembled *Pia* (*T. leontopetaloides*), and was therefore called *Piapia*. Over time, presumably for ease of pronunciation, a backformation took place, making *Pia* a synonym for the original form, *Piapia*.

**Figure 4.2.** Two examples of the relationship between reduplication and backformation in species-level nomenclatural synonymy of *kalo* (*Colocasia esculenta* (L.) Schott.).



#### 4.5.6 Changes in nomenclature and taxonomy

Given the language shift in Hawai‘i which went from Hawaiian as the only language, to Hawaiian as a second language, and going as far as English as the only language for the majority of Hawaiians, it can be predicted that there would also be associated shifts in nomenclature and taxonomy of biological taxa. There is evidence to support this. All indications are that Hawaiians used a binomial system until the mid-20<sup>th</sup> century when a horticulturalist-based understanding of *kalo* diversity began to fill the knowledge void left as more and more Hawaiian-speaking farmers passed away, and their knowledge of Hawaiian nomenclature and taxonomy was lost to time.

Whitney et al. (1939) is currently the authoritative work used in Hawai‘i botanical gardens and agriculture stations to distinguish the cultivars in their collections. Historically these gardens and stations, not Hawaiian-speaking farmers, have been sources of planting material used by farmers desiring to enhance the diversity within their collections. Because of this, nomenclature and taxonomic systems currently used in Hawai‘i have been heavily influenced by Whitney et al. (1939). An example of their influence is the proliferation of trinomials that were previously rare or nonexistent. The results of this study propose that trinomials represent only

2% of all the varietal names collected, but are currently applied to 11% of extant varieties. Whitney et al. (1939) are the first to report trinomials with the *Lauloa palakea* group (*L. p. 'ele'ele*, *L. p. ke'oke'o*, *L. p. papamū*, and *L. p. 'ula*) and the variety *Piko lehua 'āpi'i*. They alluded to the challenges they had in creating their taxonomic treatment, and indicated that they created trinomials as a solution. Their treatment lists binomial synonyms for all but one of these trinomials, and it is likely that these binomials were the actual names used by Hawaiians before 1939.

Whitney et al.'s (1939) influence over contemporary taxonomy and nomenclature is further evidenced in an examination of other varietal names. For example, the variety name *Maka 'ōpio* was once commonly used, as is evident from the high number of sources that documented its presence (Table 4.6). However, it is treated as a secondary synonym to *Piko 'ele'ele* (Whitney et al. 1939) which was documented by only five sources. Evidence suggests that the varietal name, *Maka 'ōpio*, was once more commonly used than *Piko 'ele'ele*, but today *Piko 'ele'ele* is the only name that is used and *Maka 'ōpio* has fallen out of use. From the above examples it is seen that Whitney et al. (1939) created a uniform nomenclature and taxonomic system that worked for their purposes, and it has had an influence on the nomenclature and taxonomic system used in Hawai'i today.

**Table 4.6.** Importance of *kalo* (*Colocasia esculenta* (L.) Schott.) varieties based on number of citations between 1879 and 1940 verses current observations through field surveys. Only varieties with nine or more citations were compared. Documented diversity excludes synonyms.

Variety Name	Number of References*	Current Status**
<i>Hā-o-kea / Hā-a-kea</i>	12	Occasional
<i>Ipu-o-Lono</i>	11	Rare
<i>'U(w)ahi-a-Pele</i>	11	Rare
<i>Nohu</i>	10	Name no longer used
<i>Kāi koi</i>	10	Extinct
<i>Mākohi</i>	10	Name no longer used
<i>Kāi kea / K. ke'oke'o</i>	9	Rare
<i>'Elepaio</i>	9	Rare
<i>Maka`ōpio</i>	9	Name no longer used
<i>Māna `ele'ele</i>	9	Rare
<i>Pi`iali`i</i>	9	Occasional

Aside from the changes in nomenclature and taxonomy articulated above there is evidence that other changes may have occurred. One piece of evidence comes from reviewing nomenclature for other Hawaiian crop plants with relatively high levels of recognized diversity—such as sweet potato, bananas, and ‘*awa* (Handy 1940, Kalaniana‘ole n.d.). Although these crops do have species-level taxa that are named after colors (eg., ‘*awa Hiwa*, *mai ‘a ‘Ele ‘ele*), these species names represent the finest level of diversity associated with those genera, and do not correspond with group-names at the species levels. However, within the review of *kalo* group-names (Appendix 3), a few colors—‘*Ele ‘ele* (blackish), ‘*Ula ‘ula* (red-dish), and *Poni* (purplish)—are seen. This deviation from the nomenclatural trend for Hawaiian crop plants makes these groups intriguing. Synonyms within the system (Table 4.7) imply traditions that are more consistent with the trends of other Hawaiian crop plants. These names were lumped together for the “conservative estimate” of *kalo* diversity. The color synonyms may be evidence that higher *kalo* diversity persisted into the period when the Hawaiian language, and thus knowledge of traditional nomenclature, was in decline. It is possible that these color group-names represent an evolution of *kalo* nomenclature corresponding to the 19<sup>th</sup> century, although there is not enough evidence to definitively determine the validity of this.

**Table 4-7.** *Kalo* (*Colocasia esculenta* (L.) Schott.) color group-names and possible synonyms.

Color group-name	Possible group-name synonym
‘ <i>Ele ‘ele</i>	<i>Naioea</i> or <i>Nohu</i>
‘ <i>Ula ‘ula</i>	<i>Kūmū</i>

#### **4.5.7 Decreases in traditional diversity and effects on cultural diversity**

No individual cultivars documented previously (1879-1940) as common are still common today (Table 4.6). Reduction of diversity since this period is further evidenced by examining the most diverse subspecies-groups of the past and comparing them with contemporary levels of diversity. Diversity within these groups have declined drastically (Table 4.8). Since individual cultivars and subspecies-groups have specific, and sometimes unique, traditions associated with them, it can be predicted that relative *kalo*-based traditions will shift over time with shifting levels of biodiversity. For example, the variety, *Nohu*, was once one of the most common in the period from 1879-1940 (Table 4.6). We could assume that specific traditions associated with that variety were also common. Today, *Nohu* is a

varietal name that is not even used anymore which is an indication that specific traditions associated with it are no longer practiced. If this trend were to be applied at the scale of interactions between all of *kalo* diversity and all of Hawaiian cultural-practices, it is predicted that shifts in biodiversity and cultural diversity will be linked. Such coupled changes relating to shifts in biodiversity and associated traditions are examples of ethnobiological evolution (Winter & McClatchey 2008, 2009). This presents an opportunity for further research.

**Table 4.8.** The most important *kalo* (*Colocasia esculenta* (L.) Schott.) groups based on number of recorded sub-species names. Only groups with six or more cultivars are listed. Reported diversity excludes synonyms. Extant diversity is based on positive identification in current field surveys.

<b>Kalo Group</b>	<b>Documented Diversity</b>	<b>Extant Diversity</b>
<i>Māna</i>	16	5
<i>Lauloa</i>	13	5
<i>Lehua</i>	11	4
<i>Kāi / `Ala</i>	12	4
<i>Manini</i>	9	5
<i>Piko / Haehae / Uaua</i>	7	6
<i>Hā-o-kea</i>	6	1
<i>Hāpu`u(pu`u)</i>	6	1
<i>Kūmū / `Ula`ula</i>	6	3
<i>Naioea / Nohu / Eleele</i>	6	2

#### **4.5.8 Increases in diversity via modern hybrids**

Thousands of modern cultivars have been created via hybridization between Hawaiian and other Pacific and Asian cultivars since the 1980s (Cho et al. 2007, Quero-García et al. 2009). Lack of rigorous morphological documentation of these hybrids and their distributions is currently confusing identification, and stimulating controversy about whether or not these new hybrids are beneficial contributions to Hawaiian culture.

## **4.6 Conclusions**

The data collection activities reported in the literature (1879-1940) and by the author were conducted in reaction to declining *kalo* diversity and associated cultural knowledge. Sorting through synonymy is complex, which is why there has

been no consensus about levels of biodiversity at or near the period of peak of cultivation. The methods used to sort through synonymy presented in this paper are useful in estimating, but may not be enough for an accurate picture of the past due to insufficient positive records of cultivar names. Genetic tools could more accurately sort through the cultivars, however, since the majority has apparently gone extinct, the methods used in this study may represent the best that we have. Estimates of a conservative and liberal range for former levels of *kalo* diversity were determined using two different methods. The first method, using synonymy percentages calculated by analyzing the most valid sources, provided a cursory estimate of 364-379 cultivars. This was followed by a more methodological approach of sorting through both linguistically-based and classification-based synonyms resulted in an estimate of 368-482 cultivars. It is important to note that there is only a difference of four between the two different methodologies of conservatively estimating *kalo* diversity, which is cross validation that these estimates may be accurate. This far surpasses previous estimates of 100-150 (MacCaughy & Emerson's 1913), and supports Handy et al.'s (1972) estimate of at least 300. The above estimates may still be low, however, due to missing information. The above estimate is of the level of biodiversity at the end of the 19<sup>th</sup> century. Assuming that the height of *kalo* diversity was approximately 100 years earlier, and given all the circumstances, it seems safe to assume that levels at the height of diversity at the end of the 18<sup>th</sup> century were even higher.

The high number of cultivars far surpasses those of other crop plants and reinforces the idea that *kalo* was indeed the most important crop plant in Hawaiian culture. The only other crop which rivals this level of diversity is the sweet potato (*I. batatas*). Emerging research (Ladefoged et al. 2009) is showing that sweet potatoes may have had more prominence within ancient Hawaiian culture at the point of contact in 1778 than most contemporary scholars have thought. Further research could apply the methods articulated here to a collection of sweet potato names.

Field surveys have confirmed that there are only between 65-73 *kalo* cultivars remaining in Hawai'i. Using the most conservative estimate above these field surveys indicate that there has been approximately an 80% extinction rate since the late 19<sup>th</sup> century. Due to the extent with which various individuals and institutions have worked to collect all remaining *kalo* diversity for the past century, it is broadly assumed that all other cultivars have been lost to extinction. However, extinction of *kalo* cultivars may not be a permanent status. Another team of researchers has located the previously undocumented, *Pi'ikea*; and reportedly rediscovered the cultivar, *Lauloa 'ōni'oni'o*, via a somatic mutation. This rediscovery is not yet confirmed, but it does give hope that some of the lost cultivars can be regained.

An interesting and unforeseen outcome of this research is that we now also have insight into which individual cultivars and which taxonomic groups of *kalo* were the most important at or near the period of peak cultivation. None of these individual cultivars or taxonomic groups have the same status today as they once did. All have experienced a drastic decline, and other commercial cultivars have taken their place. Given that each heritage cultivar has specific associated traditions relating to cultivation, preparation and ceremony; and that there has been both a decline in diversity, and a shift away from formerly important cultivars; then the role of *kalo* in Hawaiian culture has likely changed over time, and is an example of ethnobiological evolution. Such a phenomenon presents an interesting area for further research. It may be possible to quantify the amount of cultural loss that has been associated with a decline in *kalo* diversity.

Whitney et al.'s (1939) research has been an instrumental component to scientifically documenting and understanding *kalo* diversity, but the data and analysis provided by this paper demonstrates that they did change varietal nomenclature (i.e., creating trinomials) to create a taxonomic treatment and subsequent dichotomous key that worked for their purposes. Their nomenclatural and taxonomic treatment of *kalo* is founded in, but has deviated from, an originally Hawaiian world view of *kalo*. Furthermore, due to a shift away from the Hawaiian language by the general population there is not a firm understanding of nomenclature and classification within the Hawaiian lexicon. This, coupled with the contemporary reliance on Whitney et al. (1939) in understanding *kalo* diversity seems to be the largest factor for the contemporary shift away from an unadulterated Hawaiian nomenclature and taxonomic system. The most accurate example of Hawaiian nomenclature and taxonomy is seen the work of Iokepa and Kekahuna (n.d.) and Chun (1994).

It is entirely possible that yet more data may be uncovered relating to *kalo* diversity which may cause us to rethink the conclusions of this chapter. The hope is that for now, the additional data, new forms of analysis, and conclusions of this paper will respectfully reawaken a debate about *kalo* diversity and its role in the evolving Hawaiian culture.

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## **CHAPTER 5. Using molecular characterization of genetic variability and relationships of taro [*Colocasia esculenta* (L.) Schott] in Hawai`i, with amplified fragment length polymorphism, to understand phylogeny, trends in ethnobotanical evolution, and ethnotaxonomy**

### **5.1 Abstract**

Taro has been transported by humans out of its pre-domesticated range into new biogeographic regions due to its cultural importance, primarily as a food source. This process has involved repeated population bottlenecking events and genetic drift away from the parent populations. In Hawai`i, taro has come to play a salient cultural role beyond its function as a food source. Hawaiians anciently developed taro into approximately 400 culturally distinct cultivars—all of which were named and classified—making Hawai`i a diversity center outside of taro's pre-domesticated range. This biological diversity of the species is linked to cultural diversity within the native Hawaiian population via a co-evolutionary relationship. From the 18<sup>th</sup> to 20<sup>th</sup> centuries Hawaiian taro underwent an extinction period with loss of perhaps 85% of diversity within the lineages originally brought by Polynesian migrations. Because this diversity was developed via a co-evolutionary relationship between a plant species and human a culture, this loss in biodiversity led to a corresponded loss in cultural diversity. In this same period, immigrant cultures and researchers were bringing cultivars new to the Hawaiian Islands as genetic founders from different regions around the Pacific and East Asia. An AFLP study of contemporary taro diversity in Hawai`i demonstrates that diversity is genetically re-expanding. Although this re-expansion of taro diversity was not due to a co-evolutionary relationship between the Hawaiian culture and taro as it was during the initial round of diversification, the existence of this relationship has opened up an opportunity for a re-expansion of cultural diversity as well. This has occurred and constitutes evolution of an ethnobotanical population. The AFLP study also demonstrates that there may be broader genetic diversity of taro in Polynesia than in its pre-domesticated range, which is counter to assumptions of repeated bottlenecking events. Furthermore, the data indicate that Hawaiian ethnoclassification is based on shared morphological characteristics that are not necessarily reflective of parentage or genetic relationships.

## 5.2 Introduction

### *5.2.1.1 Genetic diversity of taro in Hawai`i*

Previous genetic studies have indicated that much of the genetic base of taro, outside of its core-diversity center in South East Asia, is narrow (Lebot and Aradhya 1991). This is presumably due to human dispersal of limited taro lineages beyond its pre-domesticated range resulting in repeated genetic bottlenecks. Disease outbreaks that selectively remove certain lineages have magnified this effect, especially in Oceania (Rao et al. 2010).

Hawai`i represents the northeastern-most spread of taro in the pre-colonial period. Polynesians came to Hawai`i (in Eastern/Remote Oceania) no later than 1,000 years ago (Burney and Kikuchi 2006, Kirch 2012), and brought taro aboard their long-distance voyaging canoes due to its cultural importance in religious, culinary, and other traditions (Abbott 1995). An intensified focus on this crop in particular played an important role in the structure of the Hawaiian civilization (Kame`eleihiwa 1992), and its ability to sustain a large population (Stannard 1989). Recent research indicates that despite the human-induced bottlenecks of this crop, the ancient Hawaiians developed their foundational taro diversity into what became approximately 400 distinct cultivars—at the point of contact with colonial powers in 1778—making it a diversity center in the Eastern Pacific (Chapter 4, Winter 2012). This was achieved via both hybridization (Handy et al. 1972) and somatic mutations (Whitney et al. 1939). Today, that diversity has dropped to approximately 70 distinct cultivars (Chapter 4, Winter 2012).

There are several unknowns about ancient taro diversity in Hawai`i such as how diverse the founding gene pool was, how many biogeographic island groups contributed to the founding diversity, how many cultivars were developed via cross-breeding, and how many were developed from somatic mutations. Patterns in ethnonomenclature and ethnoclassification indicate that either a high percentage of diversity stemmed from somatic mutations (Chapter 4, Winter 2012); or at least that morphological features were of prominent importance in naming them, and this is the general consensus among taro farmers. However, the ancient process of ethnonomenclature is poorly understood. Another unknown is how closely a Hawaiian cognition of taro diversity adheres to actual parentage and genetic relationships. A study of the remaining genetic diversity of Hawaiian taro may help to answer some of these questions.

The diversity of the taro gene pool in Hawai`i has been dynamic in the past two centuries. A collapse of the Hawaiian population, changes in land tenure and economic system, and newly-introduced taro diseases were major contributing

factors to an extinction crisis that befell the Hawaiian taro cultivars (Cho et al. 2007). Perhaps as much as 85% of Hawaiian taro diversity was lost in this period (Chapter 4, Winter 2012). Concurrently, immigrant labor was brought to Hawai`i to work on sugarcane and pineapple plantations. Many of these thousands of immigrant workers came from biogeographic areas where taro thrives, and came from cultures—Chinese, Japanese, and Filipino (Daws 1968)—that had long ago incorporated taro into their culinary and other traditions. In this same period, Chinese, Japanese, and Filipino taro cultivars began to appear in Hawai`i (Staples and Herbst 2005), and were undoubtedly brought by members of these cultures to continue their traditions in their new home. Following the formal incorporation of Hawai`i into the United States via statehood in 1959, America’s geopolitical relationships with other Pacific Island nations opened the door for further immigration from countries such as (Western) Sāmoa, American Sāmoa, and the Federated States of Micronesia. In the 20<sup>th</sup> century in particular, taro cultivars from around the Pacific started to appear in Hawai`i as well. However, it wasn’t just the immigrants who were contributing new cultivars to the gene pool in this period. Researchers were bringing in cultivars to increase diversity (Wilder n.d., Whitney et al. 1939), and to do hybridization experiments (Trujillo et al. 2002, Miyasaki et al. 2012). Hawaiian taro diversity was decreasing through extinction, while new cultivars were being brought in by different waves of migrating cultures. These individuals represented founders from different biogeographic areas around the Pacific and East Asia, and have contributed to the taro gene pool found in Hawai`i today. Because of this history, a hypothesis is that the taro gene pool in Hawai`i was decreasing within the lineages originally brought by ancestral Polynesians, while increasing overall.

#### ***5.2.1.2 Utilization of AFLP***

Various molecular approaches have been used to analyze the phylogenetic relationships of taro in Hawai`i. Lebot and Aradhya (1991) conducted an isozyme analysis, and found no variation between 343 Hawaiian accessions, despite distinct morphological differences. Random amplification of polymorphic DNA (RAPD) analysis of taro varieties by Irwin et al. (1998) divided 23 Hawaiian accessions—including commercial varieties—into three major groups, with the majority of Hawaiian accessions being closely related with about 80% similarity. Microsatellite techniques have been used in studies of taro in Hawai`i (James et al. 2012) and the Pacific Island nation of Vanuatu (Sardos et al. 2011). However, there is an appeal to use amplified fragment length polymorphism (AFLP) for phylogenetic studies because the technique generates large numbers of markers, and surveys the entire

genome rather than just a single gene or a small number of genes (as with isozymes, RAPDs, and microsatellites). On top of that it is relatively inexpensive, and generates many potential phylogenetic markers (Althoff et al. 2007). AFLP protocols have been demonstrated to be a viable way to analyze intraspecific genetic diversity of cultivated plants, even in bottlenecked populations (Aradhya et al. 2004, Aradhya et al. 2012). This has already been done with taro, and bottlenecking patterns associated with human-taro co-migration have been demonstrated. Kreike et al. (2004) used AFLP protocols with three primer combinations during selective amplification to demonstrate bottlenecking and genetic drift of taro diversity (using genetic distance values) according to biogeographic region from taro's core center of diversity in SE Asia out into Western/Near Oceania. No AFLP studies on taro growing in Hawai'i (located in Eastern/Far Oceania) have been done. AFLP was therefore selected as a method for study of taro diversity in Hawai'i for the cumulative benefits of the approach described above. Furthermore, the results of Kreike et al. (2004) serve as a foundation for understanding the relationships between genetic diversity of different biogeographic regions. The expectation was that human-induced bottlenecking of genetic diversity in taro would be revealed.

As with all forms of genetic approaches to phylogenetics, AFLP does, however, have some drawbacks (Althoff et al. 2007). Currently, AFLPs are analyzed in predominately two ways, by using parsimony on the presence and absence of fragments, or converting the AFLP fragment patterns into a distance measure and using minimum evolution or neighbor joining search algorithms to determine the best phylogenetic tree. Both approaches assume that fragments of the same size are homologous across all the taxa analyzed. Althoff et al. (2007) clearly demonstrate that these assumptions are violated for some taxonomic comparisons. This will have ramifications in phylogenies produced via this analysis.

There are various ways to analyze AFLP data. Depending on a population's evolutionary history, some forms of analysis more accurately reflect genetic relationships than others. Gene flow and genetic drift shape intraspecific population structure, and the resulting genetic relationships can be decidedly nonhierarchical (Kalinowski 2009). In their study of SE Asia and Melanesian taro, Kreike et al. (2004) conducted a cluster analysis using the unweighted pair group method with arithmetic mean (UPGMA). UPGMA assumes a constant evolutionary rate, therefore all samples from the populations cluster into nodes that are equally different from each other. This assumption can severely distort genetic relationships between populations. Branch lengths in neighbor-joining trees are more flexible, and can faithfully depict genetic structure for some populations that have an isolation-by-

distance population structure. Unlike UPGMA, neighbor-joining has the advantage that it does not assume that all lineages evolve at the same rate (Kalinowsky 2009). This was taken into account in this study.

### ***5.2.2.1 Links between the genetic diversity of taro and the diversity of practices in Hawaiian culture as evidence of ethnobotanical evolution***

This dissertation contends that evolution can be defined by the change in frequency of the smallest measurable units of study over time. For biological evolution those units would be alleles. For cultural evolution those units would be practices. For ethnobotanical evolution those units would be quantum co-evolution units (Chapter 2, 3; Winter and McClatchey 2008, 2009). This dissertation also contends that ethnobotanical evolution is a co-evolutionary process where both increases and decreases in biocultural diversity are directly correlated (Chapter 2, 3; Winter and McClatchey 2008, 2009). An exploration of the biocultural diversity of taro in Hawai'i is a way to examine ethnobotanical evolution.

Ancient Hawaiian intensification of taro cultivation resulted in new named cultivars (i.e., promoting an increase of biodiversity), via hybridization and somatic mutations. Each named cultivar had associated cultural knowledge—such as ideal growing conditions, particular phenological expressions that were beneficial for particular practices or functions within the culture (culinary, medicinal, ceremonial, etc.), etc.—and this knowledge was passed on through the generations (Chapter 4, Winter 2012) in a cycle leading to increases in biocultural diversity (Chapter 3, Figure 3.6, Winter and McClatchey 2008).

In the 19<sup>th</sup> and 20<sup>th</sup> centuries as Hawaiian taro diversity was being lost, there was a concurrent loss in biocultural diversity (Chapter 4, Winter 2012). An example of this was the cultivar, '*Hoene*,' whose corm had a unique shape and was specifically used in medicinal treatments as an enema. This cultivar is now extinct. This loss of biodiversity resulted in a corresponding loss in cultural diversity. '*Hoene*' is a name that is no longer actively used in the lexicon, and the practice of using taro as an enema is no longer in existence.

In the above described period associated with losses in Hawaiian taro diversity there was also an increase in diversity on another level with the introduction of non-Hawaiian taro cultivars. However, unlike the initial round of diversification that was driven by a co-evolutionary process between taro and the Hawaiian culture, this increase in diversity was driven by multiple founders being brought by other cultures from what is presumed to be different genetic lineages. Given this, a hypothesis is that when biodiversity of a cultivated plant is increased via

subsequent founders rather than being produced via co-evolution, then there will not necessarily be a correlated increase in cultural diversity, although the opportunity for cultural diversification is there.

### ***5.2.3.1 Insights from trends in ethnonomenclature and ethnoclassification***

Winter (2012, Chapter 4) compiled a list of 598 names of Hawaiian taro varieties representing both extant and extinct cultivars. Linguistic trends in ethnoclassification were used to produce an estimate of taro diversity in Hawai`i in the late-1800s of 368-482 culturally distinct cultivars. A majority (69%) of these are morphotype groups which conform to the trend of being grouped together nomenclaturally by shared morphological traits (Table 5.1), with differentiation in the group being determined by differences in coloration, mostly of the petiole. The remaining 31% have no easily identifiable ethnonomenclatural relationship to one another or any of the other cultivars. One possible assessment of this trend is that the 69% of the diversity was produced via somatic mutations—hence the coloration variation on an otherwise conceptually-consistent morphological form, and the 31% was produced via hybridization between cultivars. However, these are both assumptions. At the end of an extinction crisis, in which perhaps as much as 85% of taro diversity has been lost along with associated cultural knowledge, it is difficult to address these assumptions with any level of certainty. Genetics may be the only tool available to shed some light into this mystery.

Berlin (1992) documented that ethnoclassification does not always closely correspond to actual genetic relationships because cultures are often cognitively dealing with a subset of actual diversity of any given group. It is not known how closely Hawaiian ethnotaxonomy mirrors actual parentage and genetic relationships of Hawaiian taro. There are two hypotheses relating to this. One is that a particular ethnotaxonomy is founded in parentage and actual genetic relationships of taro. Another is that a particular ethnotaxonomy is founded in shared morphological traits that may or may not represent actual genetic relationships (Ellen & Burnham 1979). An aim of this study is to gain insight as to which hypothesis is supported by genetic evidence. An AFLP study on taro cultivars of Vanuatu (Quero-Garcia et al. 2004) revealed that genetic clusters do not contain any particular group of morphotypes. This situation has already been observed in other clonal crops such as plantains (Crouch et al. 2000) and yams (Malapa et al. 2003). If the data on Hawaiian taro follows this trend, then that would be evidence in support of the latter hypothesis.

**Table 5.1.** The most diverse groups of taro in the Hawaiian classification system from Winter (2012), with their interpreted meanings and named cultivar diversity.

Hawaiian taro group name	Interpreted meaning	Reported diversity	Extant
<i>Māna</i>	“branching corm”	16	5
<i>Lauloa</i>	“elongated leaf”	13	5
<i>ʻAla</i>	“fragrant when cooked”	12	4
<i>Lehua</i>	“reddish <i>poi</i> ”	11	4
<i>Manini</i>	“striped petiole”	9	5
<i>Piko/Haehae</i>	“leaf lobes cut to vascular hub”	7	6
<i>Hā-o-kea</i>	“light-colored petiole”	6	1
<i>Hāpu`u(pu`u)</i>	“resembling a tree fern”	6	1
<i>Kūmū/ʻUla`ula</i>	“reddish petiole”	6	3
<i>Naioea/Nohu/ʻEle`ele</i>	“darkish petiole”	6	2

## 5.3 Materials and Methods

### 5.3.1.1 Plant material and DNA isolation

One hundred and three DNA samples were collected. These included one sample of *C. gigantea* as an outgroup and 102 of *C. esculenta*. This consisted of 73 Hawaiian cultivars (both positively identified and presumably undocumented cultivars), 21 cultivars from the South Pacific and West Pacific, two cultivars from China, three cultivars from Japan, one cultivar from the Philippines, and three cultivars of unknown origin. Field collections of taro occurred in the Hawaiian Islands between 2009 – 2012. Propagation material of Hawaiian and culturally important cultivars were collected, and subsequently grown under common garden conditions at Limahuli Garden and Preserve on the island of Kaua`i. Cultivar identification was verified using Whitney et al. (1939). DNA material was collected from these samples. Additional DNA samples representing individuals from the South Pacific, West Pacific, and Asia were collected from the Kaua`i Agricultural Research Station where the taro was grown under common garden conditions. Accession information was used to verify the identity of these cultivars. Voucher specimens and photographs were also taken for each sample. These were submitted to the Pacific Tropical Botanical Garden herbarium (Table 5.2).

**Table 5.2.** *Colocasia* germplasm accessions cultivated in Hawai'i and included in the study.

<b>Collector ID Number</b>	<b>Cultivar Name</b>
KBW001	<i>Tsurunoko</i>
KBW002	<i>Akado</i>
KBW003	<i>Miyako</i>
KBW004	<i>ʻIliuaua</i>
KBW005	<i>Bun long</i>
KBW006	<i>ʻĀweu</i>
KBW008	<i>Mana ʻulu</i>
KBW009	<i>Mana ʻopelu</i>
KBW011	<i>Mana uliuli</i>
KBW012	<i>Mana ʻulaʻula</i>
KBW013	<i>Mana lauloa</i>
KBW014	<i>Mana keʻokeʻo</i>
KBW016	<i>Lehua ʻāpiʻi</i>
KBW017	<i>Piko ʻulaʻula</i>
KBW018	<i>Piko kea</i>
KBW019	<i>Piko keʻokeʻo</i>
KBW020	<i>Piko uaua</i>
KBW021	<i>Piko uliuli</i>
KBW022	<i>Piko ʻeleʻele</i>
KBW023	<i>ʻElepaio kea</i>
KBW024	<i>Uahiapele</i>
KBW025	<i>Manapiko</i>
KBW026	<i>Veo (Tahitian)</i>
KBW027	<i>Kai uliuli</i>
KBW028	<i>Kai ʻala</i>
KBW029	<i>Kai kea</i>
KBW030	<i>ʻApu wai</i>
KBW031	<i>ʻApu</i>
KBW032	<i>Piʻialiʻi</i>
KBW033	<i>Paʻakai</i>
KBW034	<i>Moana / ʻĀpiʻi</i>
KBW035	<i>Akuugawai</i>
KBW036	<i>Lauloa ʻeleʻele</i>
KBW037	<i>ʻEleʻele lauloa</i>
KBW038	<i>Lauloa palakea</i>
KBW040	<i>Lauloa papamū</i>
KBW042	<i>Lauloa keʻokeʻo</i>
KBW043	<i>ʻEleʻele mākokō</i>

KBW044	<i>'Ele`ele naioea</i>
KBW045A	<i>Manini `owali (UH)</i>
KBW045B	<i>Manini `owali (ʻōma`o)</i>
KBW046	<i>Kūmū `ele`ele</i>
KBW047	<i>Nāwao</i>
KBW048	<i>'Ula`ula kūmū</i>
KBW049	<i>'Ula`ula poni</i>
KBW050	<i>'Ula`ula moano</i>
KBW052	<i>'O`opukai</i>
KBW053	<i>Manini uliuli</i>
KBW054	<i>Manini kea</i>
KBW055	<i>Manini toretore</i>
KBW058	<i>Nihopu`u</i>
KBW059	<i>Manini `ōpelu</i>
KBW062	<i>'Ohe</i>
KBW063	<i>Lehua maoli</i>
KBW064	<i>Lehua ke`oke`o</i>
KBW065	<i>Lehua `ele`ele</i>
KBW066	<i>Lehua palai`i</i>
KBW067	<i>'Apowale</i>
KBW068	<i>Wehiwa</i>
KBW069	<i>Papapueo</i>
KBW070	<i>Kū`oho</i>
KBW072	<i>Maea</i>
KBW073	<i>Hā-o-kea</i>
KBW074	<i>Kalalau</i>
KBW075	<i>Hāpu`u</i>
KBW078	<i>Lihilihi molina / Mokuokeawe</i>
KBW079	<i>Mana `ele`ele</i>
KBW080	<i>Mana `oko`a</i>
KBW081	<i>Moi (White moi)</i>
KBW084	<i>Pololū</i>
KBW085	<i>Maui Lehua</i>
KBW086	<i>Red moi</i>
KBW087	<i>Kai KBS</i>
KBW088	<i>Makalau</i>
KBW089	<i>Keone</i>
KBW 131	<i>Fa`ifa`ausi I</i>
KBW 132	<i>Fa`ifa`ausi II</i>
KBW 901	<i>'Elepaio uliuli</i>
KBW 902	<i>Pilialoha</i>

KBW 903	<i>Pi`ikea</i>
KBW 904	Banana Creek
KBW 905	Napali 3.5-mile
KBW 906	<i>Lauloa koukou`ai</i>

DNA was collected from young leaves. 2x3 inch leaf samples were placed in envelopes with desiccant. Dried tissue was first homogenized using a GenoGrinder 2000 (OPS Diagnostics). Total DNA was extracted by following the standard CTAB method (Doyle and Doyle 1987). Nucleic acid was precipitated with 300  $\mu$ L of isopropanol and centrifuged at low speed to pelletize the DNA. The pellet was washed twice with 75% ethanol containing 10mM ammonium acetate, air dried, dissolved in 300 $\mu$ L of TE buffer, and treated with RNase A (Qiagen) at the rate of 10 $\mu$ g/ml at 37°C for 1 hour.

### **5.3.1.2 AFLP analysis**

In anticipation of an even more bottlenecked population than the one that Kreike et al. (2004) studied, an AFLP study was performed using twenty-one primer combinations in the selective amplification, instead of three, as a means to provide more polymorphic markers for analysis. Details of AFLP assay, adapter and primer sequences, PCR conditions for preselective and selective amplifications, and selective primer designation were according to Vos et al. (1995). Genomic DNA was restricted with *EcoRI/MseI* enzyme combinations, double-stranded adapters specific to each site were ligated, and preselective amplification was performed with primers complementary to the adapters with an extra selective base on each primer (*EcoRI-A/MseI-C*). Selective amplification was carried out with the 21 primer combinations involving four *MseI* (M) and six *EcoRI* (E) primers. In each of 7 sets of combinations, one *MseI* primer served as the unlabeled primer paired with 3 different *EcoRI* primers labeled with either 6FAM, HEX, or NED fluorescent dye in one PCR assay (Table 5.3). Fragments were resolved using capillary electrophoresis on an ABI 3130xl Genetic Analyzer with the Data Collection software, version 3.0 (Applied Biosystems). AFLP fragment analysis was performed with GeneMapper, version 4.0.

**Table 5.3.** Selective Primer Combinations used in selective amplification for an AFLP analysis of the genetic diversity of Hawaiian taro (*Colocasia esculenta* (L.) Schott).

<b>Selective Amplification Primer Combinations</b>				
<u>Set</u>	<u>MseI</u>	<u>EcoRI-6FAM</u>	<u>EcoRI-HEX</u>	<u>EcoRI-NED</u>
1	D1-CAA	DB09-ACT	DG11-AGG	DY13-AGC
2	D2-CTT	DB09-ACT	DG11-AGG	DY13-AGC
3	D1-CAA	DB10-ACA	DG12-ACG	DY14-ACC
4	D2-CTT	DB10-ACA	DG12-ACG	DY14-ACC
5	D3-CTC	DB09-ACT	DG11-AGG	DY13-AGC
6	D3-CTC	DB10-ACA	DG12-ACG	DY14-ACC
7	D4-CAG	DB10-ACA	DG12-ACG	DY14-ACC

### **5.3.1.3 Data analysis**

AFLP fragment data from 21 different primer combinations was assembled into a binary data matrix (1=presence, 0=absence). This binary matrix was prepared for import into the software program PAUP\*, Version 4 (Swofford 2003). Genetic relationships within and among taxa were computed based on the proportion of fragments shared between two accession for all possible pair-wise comparisons using Nei and Li distance (Nei and Li 1979). The resulting matrix was subjected to a cluster analysis (CA) following two different algorithms— neighbor-joining (NJ) and the unweighted pair group method with arithmetic mean averages (UPGMA)—for comparative analysis. Bootstrap analysis (100 replicates) was performed to assess the relative support for different groups.

### **5.3.2.1 Determining links between biodiversity and cultural diversity, and whether or not ethnobotanical evolution is taking place**

Quantum co-evolution units (QCUs) were identified by determining ethnobotanical profiles for selected, extinct Hawaiian cultivars; as well as for each of the non-Hawaiian cultivars in this study. These profiles were then evaluated with the following criteria to determine whether or not ethnobotanical evolution is taking place (Winter 2008, Chapter 3):

- Particular QCUs have increased in frequency because of selection.
- Particular QCUs have decreased in frequency because of selection.

- One or more QCU(s) have been added or lost through events homologous to those involved in the process of biological evolution (mutation, extinction, etc.).
- One or more QCU subunit(s) has changed (i.e., replacement of a lost or abandoned plant or tradition subunit) resulting in the creation of a new QCU.

If any of these criteria are met then it would be determined that ethnobotanical evolution has taken place. Any new QCUs not previously in existence would be evidence that cultural diversity is re-entering into a diversification cycle. Calculations to determine the trajectory of evolution were not a part of this study.

### ***5.3.3.1 Comparative analysis of AFLP phylogenies and Hawaiian classification***

The named and classified groups in the Hawaiian cognition of taro diversity, demonstrated by Winter (2012), were reviewed. The two most diverse of these, 'Māna' and 'Lauloa' respectively, were chosen for comparative analysis with the phylogenetic trees which were produced from the AFLP analysis. If members of these groups cluster together in the phylogenetic tree, then that would be evidence supporting the hypothesis that Hawaiian ethnotaxonomy mirrors actual parentage and genetic relationships. If, however, members of these groups did not cluster together, then that would be evidence supporting the hypothesis that Hawaiian ethnotaxonomy is founded more in shared morphological traits than in actual genetic relationships.

## **5.4 Results**

### ***5.4.1.1 The AFLP profile***

The twenty-one primer combinations used to assay the 103 accessions representing two species of *Colocasia* (including bottlenecked populations of *C. esculenta*) that were either anciently or historically brought to Hawai'i revealed a total of 697 polymorphic fragments. The number of polymorphic fragments ranged from 8 for the primer combination D6-CTA + (6FAM)DB09-ACT to 65 for D6-CTA + (NED)DY13-AGC with an average of 33.19 fragments/primer combination. There was polymorphism within and among species with several species-specific fragments. The genetic data supports the phenotypic observations that the cultivars used in this study are distinct from one another.

#### **5.4.1.2 Genetic relationships within and among taxa, and comparisons to assumptions of bottlenecking**

The pair-wise genetic distances among all samples computed based on the proportion of shared fragments ranged from 0.0169 between ‘*Wehiwa*’ (Hawai`i) and ‘*Maea*’ (Hawai`i) to 0.2963 between ‘*Apu wai*’ (Hawai`i) and ‘*Akuugawai*’ (South Pacific), with an overall mean distance of 0.0709 indicating considerable similarity within and between species.

A rooted, phylogenetic tree of contemporarily-occurring taro (*C. esculenta*) in Hawai`i using the neighbor-joining algorithm and Nei-Li distance measure, with zuiki (*C. gigantea*) as an outgroup. Clusters dominated by cultivars from biogeographic regions were labeled (Figure 5.1).

A bootstrap consensus was performed on the neighbor-joining tree described above. Bootstrap values ranged from 53 – 100 (Figure 5.2).

#### **5.4.2.1 Ethnobotanical profiles for extinct Hawaiian cultivars and non-Hawaiian cultivars**

The extinction of Hawaiian cultivars represents the loss of associated ethnobotanical profiles, and extinction of associated QCUs in the ethnobotanical population. While extinctions, like observed with ‘*Kāi koi*,’ represent decreases in functional redundancy (i.e., other cultivars can be used to make *poi*); extinctions, like those observed with ‘*Hoene*,’ represent losses in functional diversity within the ethnobotanical population.

Conversely, the addition of non-Hawaiian cultivars represents additional ethnobotanical profiles and associated QCUs. Most of these QCUs have increased functional redundancy in the context of the roles that they play in the ethnobotanical population. However, one QCU—using the Chinese cultivar ‘*Bun Long Woo*’ to make taro chips—represents an entirely new ethnobotanical profile use since European contact, and represents the addition of functional diversity component in the ethnobotanical population.

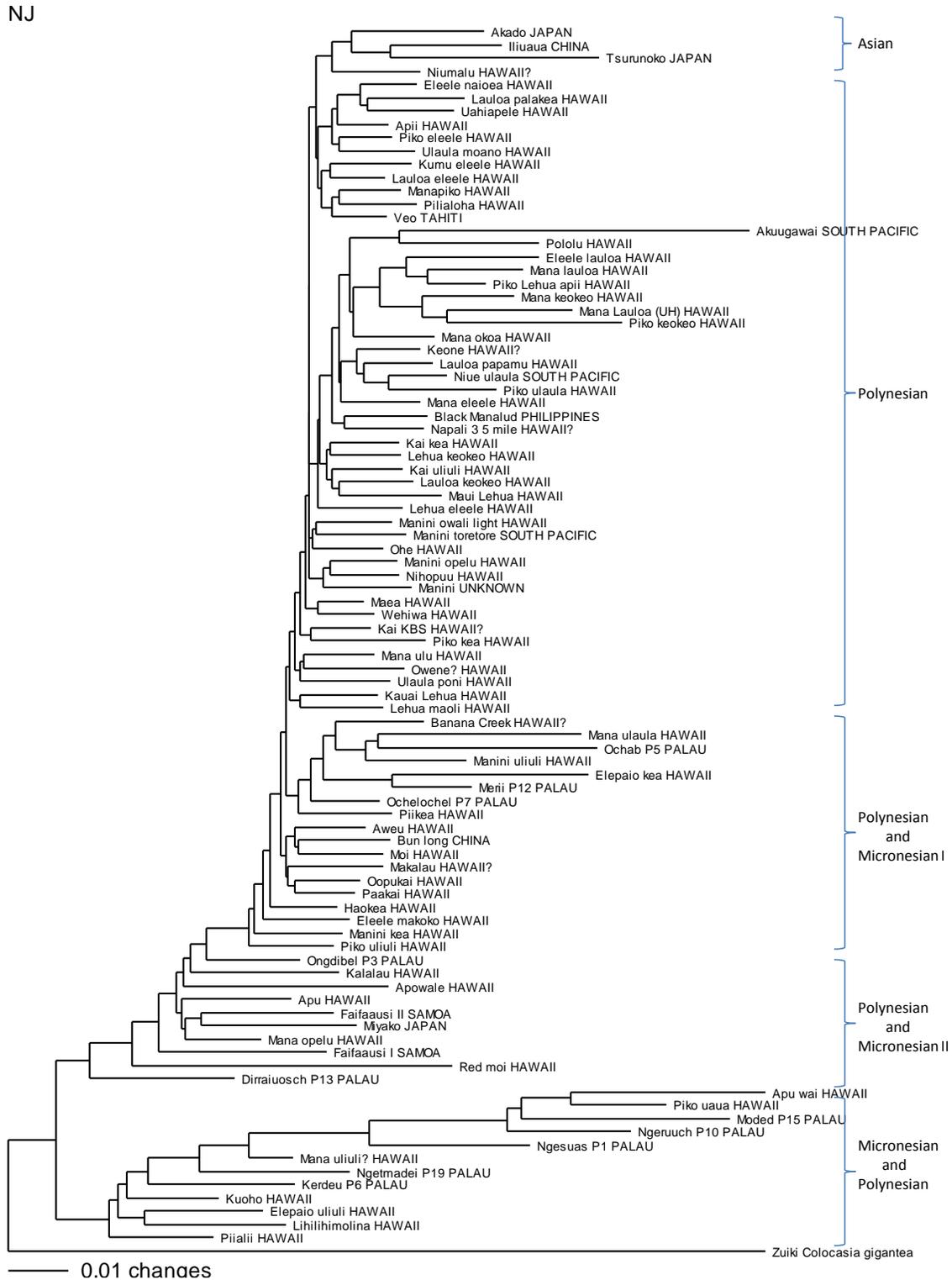
**Table 5.4.** Ethnobotanical profiles for selected cultivars (extinct Hawaiian, and introduced non-Hawaiian) of taro (*Colocasia esculenta*). From these ethnobotanical profiles, Quantum Co-evolution Units can be made for quantitative analysis. The italicized words are Hawaiian terms for different culinary preparations of taro.

<b>Plant Subunit (Source)</b>	<b>Practice Subunit</b>
' <i>Kāi koi</i> ' (Hawai`i, extinct)	Food: <i>Poi</i> taro
' <i>Hoene</i> ' (Hawai`i, extinct)	Medicine: Enema
' <i>Bun long woo</i> ' (China)	Food: Starchy corm for taro chips
' <i>Bun long woo</i> ' (China)	Food: Leafy green for <i>laulau</i>
' <i>Iliuaua</i> ' (China)	Food: Leafy green for <i>laulau</i>
' <i>Niumalu</i> ' (possibly Asia)	Food: Leafy green for <i>laulau</i>
' <i>Ngeruuch</i> ' or 'P10' (Palau)	Food: Table taro
' <i>Ngeruuch</i> ' or 'P10' (Palau)	Food: <i>Poi</i> taro
' <i>Fa`ifa`ausi</i> ' (Sāmoa)	Food: Table taro
' <i>Veo</i> ' (Tahiti)	Food: Table taro

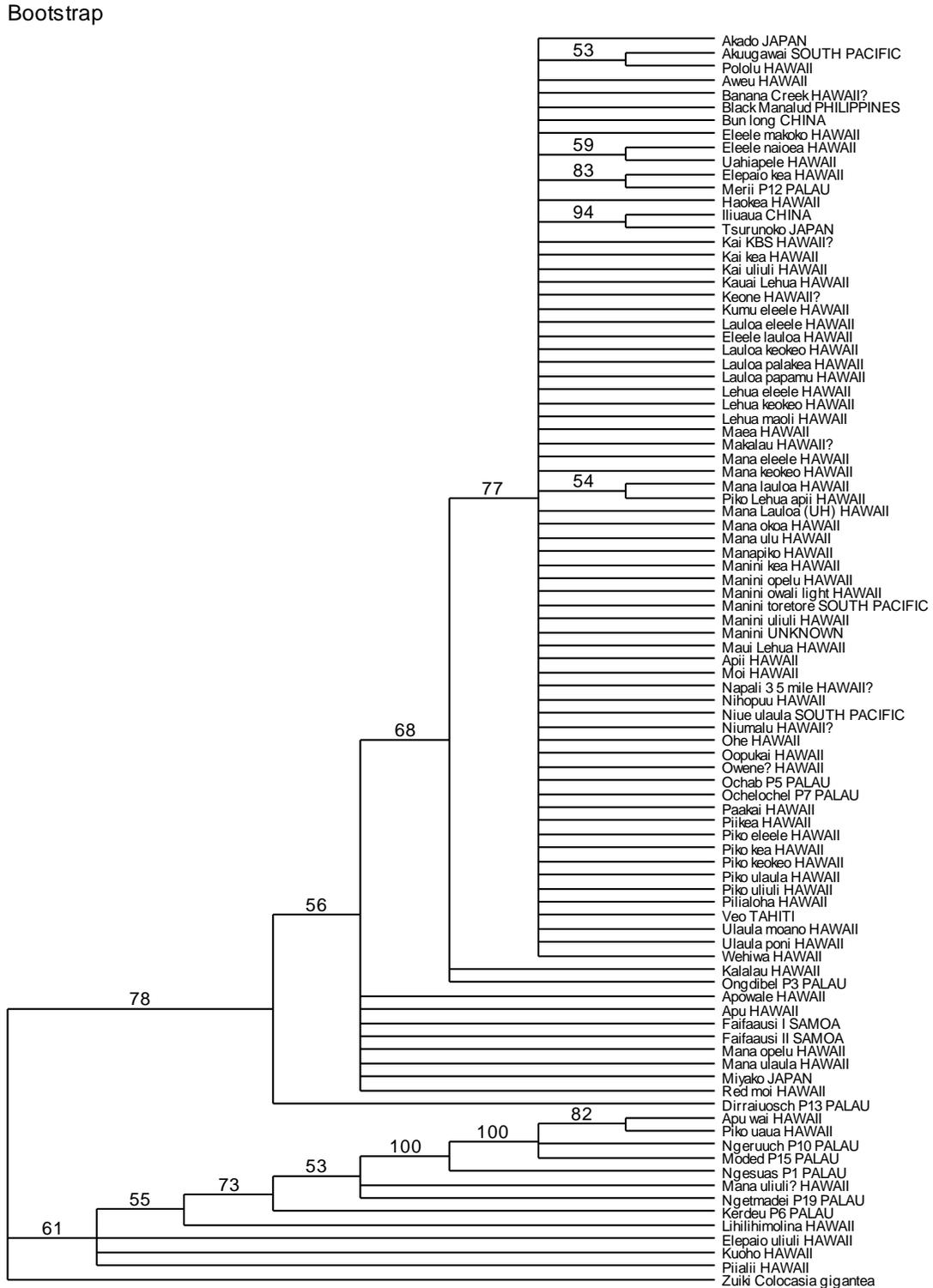
#### **5.4.3.1 Comparisons between genetic phylogeny and ethnoclassification**

A rooted, phylogenetic tree of Hawaiian taro (*C. esculenta*) exclusively was produced, using the neighbor-joining algorithm and Nei-Li distance measure. The members of the two most-diverse groups according to Hawaiian ethnoclassification, '*Māna*' and '*Lauloa*' (Winter 2012), are marked with red and green arrows respectively (Figure 3). In comparing the placement of members of these two groups of ethnoclassified taro with their corresponding placement in the phylogenetic tree, it is seen that the members do not cluster together. The '*Lauloa*' members are found among three closely related clusters. The '*Māna*' members are dispersed throughout the tree in seven different clusters.

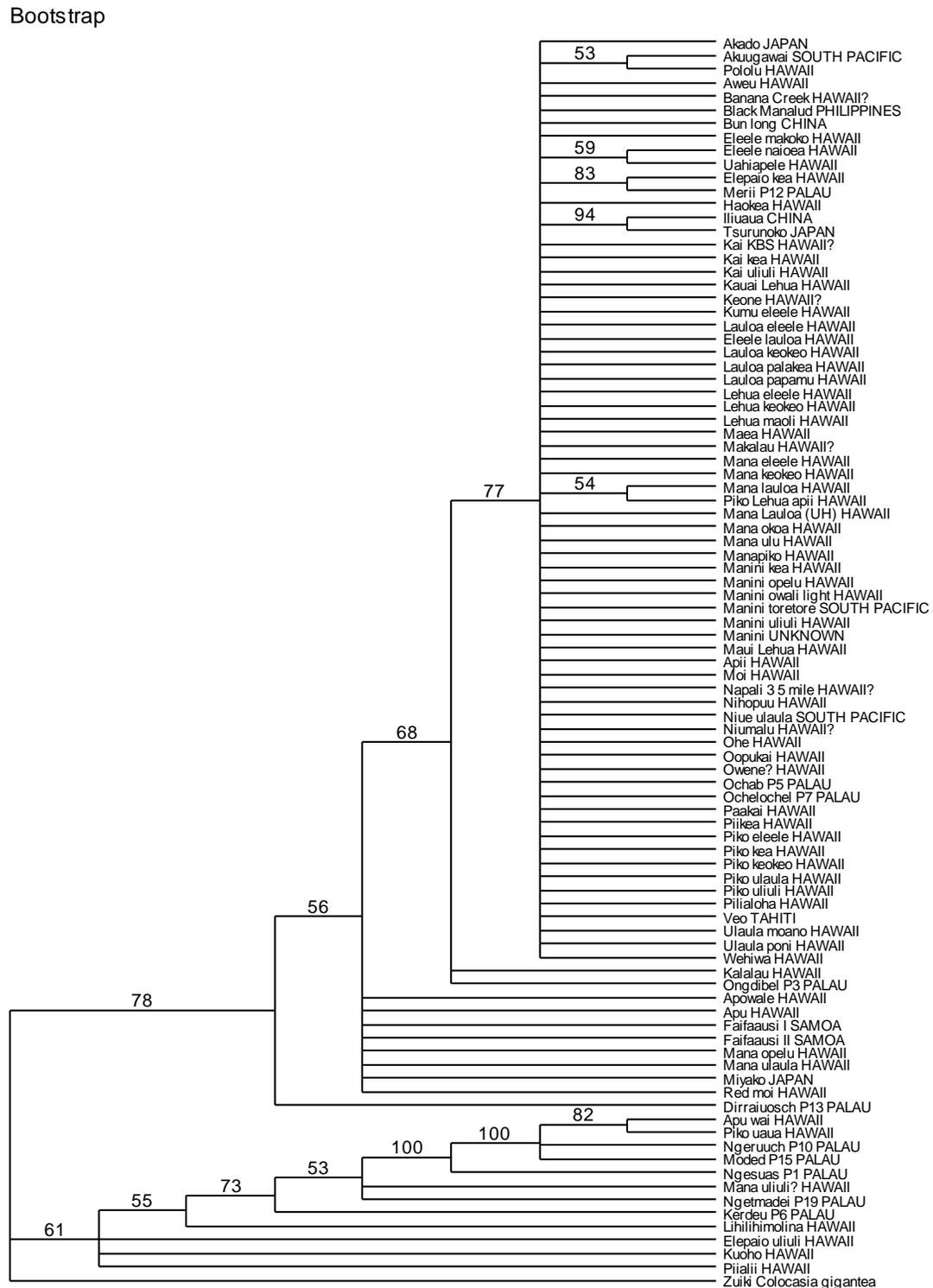
**Figure 5.1.** A rooted, phylogenetic tree of contemporarily-occurring taro (*Colocasia esculenta*) in Hawai'i using the neighbor-joining algorithm and Nei-Li distance measure, with zuiki (*C. gigantea*) as an outgroup. Clusters with associated biogeographic regions are indicated by brackets on the right.



**Figure 5.2.** A bootstrap consensus of the phylogenetic tree of contemporarily-occurring taro (*Colocasia esculenta*) in Hawai'i using the neighbor-joining algorithm and Nei-Li distance measure, with zuiki (*C. gigantea*) as an outgroup.



**Figure 5.3.** A rooted, phylogenetic tree of Hawaiian taro (*Colocasia esculenta*) exclusively, using the neighbor-joining algorithm and Nei-Li distance measure. The members of the two most-diverse groups according to Hawaiian ethnoclassification, 'Māna' and 'Lauloa' (Winter 2012), are marked with red and green arrows respectively.



## 5.5 Discussion

### 5.5.1.1 Genetic diversity of taro in Hawai`i reflects human migration patterns

Gene flow and genetic drift shape intraspecific population structure, and the resulting genetic relationships can be decidedly nonhierarchical. This is certainly true for taro diversity currently occurring in Hawai`i due to an unknown number of founder events from an unknown number of biogeographic regions/island-groups, cross-breeding and somatic mutations that shifted the gene pool, and the resultant genetic relationships among taro in this group. Phylogenetic trees made using two different algorithms for comparative analysis—neighbor-joining and UPGMA—showed similar results. Given the evolutionary history of taro in Hawai`i, the neighbor-joining tree is proposed to be a more accurate representation of genetic relationships within this group (Figure 1).

Both the neighbor-joining and UPGMA cluster analysis revealed distinct, well-supported clusters corresponding to the species (in all analyses *C. gigantea* separated out remotely), and to the biogeographic origins of the different *C. esculenta* cultivars with a few notable exceptions. The clustering in the phylogeny reflects patterns in ancient and historical human migration, which were the vector for transport of taro throughout Asia and into the Pacific. The repeated bottlenecking events corresponding to domestication and subsequent transport of taro is demonstrated by the results of this study. This is observed most readily in the Polynesian group where Hawaiian, Tahitian, Samoan, and other South Pacific taro cluster closely together with relatively little differentiation. Micronesian taro clustered separately, but were interspersed with some Hawaiian representatives. This indicates a shared, but distinctly shifting diversion into another group. Most of the Asian taro clustered in a completely separate group, which is consistent with historical evidence of these being brought into Hawai`i in the last two hundred years from distant biogeographic regions. An interesting thing to note about the Asian group is the occurrence of the cultivar, ‘*Niumalu*.’ ‘*Niumalu*’ was collected by staff at the Wailua Agricultural Station (on Kaua`i) from an old Chinese farmer in 1989 who lived in an area called Niumalu. The name and origin of this cultivar were unknown so it was henceforth referred to by the name of the place it was collected. There has been speculation about whether or not ‘*Niumalu*’ is an undocumented Hawaiian cultivar that was adopted by an old Chinese farmer, or an undocumented Chinese cultivar that is a historical relic of the immigration of Chinese to Hawai`i in the 19<sup>th</sup> century. The occurrence of ‘*Niumalu*’ in the Asian group is evidence that the latter hypothesis is supported. On a related note, it is important to point out that a well-known Chinese cultivar, ‘*Bun long woo*’, grouped in a Hawaiian cluster more closely

related to the Micronesian group; and a Japanese cultivar, '*Miyako*', grouped with a Polynesian cluster close to the Micronesian cluster. Both are surprising placements within the tree. Assuming that Kreike et al.'s (2004) conclusions are correct that Asian and Pacific Island taro are in completely different groups, this could be evidence of a flaw in this study or of movement of Polynesian/Micronesian cultivars into Japan or Micronesia. However, an ISSR-PCR study of this group (James et al. 2012) also had '*Bun long woo*' grouping among Hawaiian cultivars ('*Miyako*' was not included in their study).

#### ***5.5.1.2 Genetic relationships of taro in Hawai`i in light of assumptions about genetic bottlenecks***

The pair-wise genetic distance values for taro reported in the Kreike et al. (2004) study ranged from 0.10 for the Asian center of diversity, to 0.02 for the island groups in the Western/Near Oceania (i.e., The Philippines and Indonesia). In this study the minimum distance between two of the Hawaiian taro was 0.02, and the largest was between a Hawaiian taro and a South Polynesian taro at 0.30. This, was unexpectedly large, and may indicate that Hawaiian taro is not measurably any less diverse than taro in the island groups of Western/Near Oceania. This evidence supports the notion that ancient Hawaiians maximized the genetic diversity of the taro they cultivated (via hybridization and somatic mutations), and that Hawai`i was indeed a center of diversity.

High genetic distance between two Polynesian taro cultivars from different island groups is as much as three times as great as the diversity in its native range in Asia. This may be a result of the extreme nature of intensification that occurred in different Polynesian island groups, and these values may be evidence of a considerable amount of genetic drift in multiple trajectories. In SE Asia, the core center of taro diversity, taro is not an intensified crop. For whatever co-evolutionary relationship exists, it is unlikely that humans have had a major influence on taro genetics. However, in Polynesia taro was intensified to a level not observed anywhere else in the world, and this was happening concurrently in several different island groups, each of which had different founding diversity. The results of this study indicate that even though genetic diversity in an island group is relatively narrow and has drifted, the cumulative difference between two populations—that have been intensified and subsequently diversified—could extend a broader range than is exhibited in SE Asia. This is evidence that the co-evolutionary relationship between human cultures and *C. esculenta* has caused a larger amount of genetic diversity than existed in the pre-domestication period.

This is not surprising given the amounts of data demonstrating the phenomenon of co-evolution between other animal species and plants.

### ***5.5.1.3 The functionality of AFLP analysis***

AFLP analysis is useful in understanding the intraspecific genetic diversity of taro—even among the closely-related Polynesian taro varieties. Given the closely-related nature of bottlenecked populations, using different AFLP restriction enzymes or other marker types like SNPs may be useful for future research.

The neighbor-joining algorithm provides a more accurate depiction of the genetic relationships of taro than the UPGMA given taro's non-hierarchical evolutionary history. The neighbor-joining analysis does mirror patterns of human migration as the Polynesian taros generally clustered closely together, and taros of other biogeographic regions generally separated out as different groups. The cultivar of speculative origins, '*Niumalu*', grouped with other Asian taro and, given its known history, is likely of Asian origin.

The data and analysis of this study indicates that Hawaiian taro morphotypes do not correspond with genetic lineage, and thus supports similar results found in studies on Vanuatu taro (Quero-Garcia et al. 2004). Therefore, this study supports the hypothesis that a Hawaiian cognition and classification of taro, originally described by Winter (2012), is based on morphology regardless of parentage and actual genetic relationships. However, due to the closely related nature of the Polynesian taro cultivars, future research using different approaches may reveal more about the nature of these relationships. Focusing on documented somatic mutations and analyzing the genetic relationship to their parent may provide useful clues as to the best ways to approach future research.

### ***5.5.2.1 Historic collapse and re-expansion of genetic diversity of taro in Hawai'i, with correlations to cultural diversity***

Each of the phenotypically distinct cultivars collected for this study were determined to be genetically distinct from one another using AFLP analysis. There is no way to sample from extinct cultivars to provide genetic evidence about a collapse in taro diversity over the last 200 or so years. However, ethnonomenclature and ethnotaxonomy, which this study supports as accurately differentiating genetic diversity, has provided evidence that genetic diversity has historically collapsed (Winter 2012, Chapter 4). Any associated cultural practices have also been lost.

The AFLP results of this study do demonstrate that the cultivars brought from Asia and other Oceanic Islands in the last two centuries are genetically distinct from the Hawaiian cultivars. The presence of these cultivars constitutes a re-expansion of genetic diversity within *C. esculenta* in Hawai'i. Given this, there is at least the foundation for an opportunity of a re-expansion of cultural diversity. It is assumed that the founders from Asia and other Oceanic islands in this study contain many genes that are already present in Hawaiian taro, but also includes genetic diversity that the Hawaiian culture has never had the opportunity to interact with. With this assumption it is possible to imagine that since many of these genes in the founder cultivars already exist in Hawaiian taro, then the cultural practices founded in this diversity will produce functionally redundant QCU. However, the flip side of that assumption is that new genes (i.e., genes previously not present in Hawaiian cultivars) provide a foundation that creates an opportunity for the creation of QCU that never before existed as well.

The results of this study support the notion described above. There is close similarity in the genetics between cultivars of non-Hawaiian and Hawaiian taro, and many of them have settled in already occupied cultural niches—like using taro as food in the form of “table taro” or “cooked leafy greens” (Table 5.4)—and therefore constitute functional redundancy in that regard. However, the non-Hawaiian taros also represent genes that Hawaiians have never had the opportunity to interact with, which opens up an opportunity for taro to fill a social-ecological niche that it has never filled before. This is seen in the Chinese cultivar ‘*Bun Long Woo*.’ The consistency of its corm is light and flaky, which is a characteristic that was probably selected against during or even prior to the Polynesian diasporas. A prized taro phenotype amongst Hawaiians is a dense, gummy corm (referred to in Hawaiian as, “*ulika*”), as this is the corm type that makes the best quality *poi* (a staple dish of the Hawaiian culture). The light and flaky corm of ‘*Bun Long Woo*’ produces an inferior, almost unpalatable *poi*. However, the same qualities that make it inferior as a “*poi taro*” make it ideal as a source for taro chips, where thinly sliced cross sections of corm are deep fried in oil.

Given both the losses and additions of ethnobotanical profiles and corresponding QCU (Table 5.4), the criteria has been met to demonstrate that ethnobotanical evolution has occurred.

### **5.5.3.1 Determining pathways for diversification, and whether or not Hawaiian classification of taro is founded in genetic relationships**

Linguistic data and analysis has given insights into ethnonomenclatural trends and the ethnoclassification system of Hawaiian taro (Chapter 4, Winter 2012), but given the massive amounts of loss in Hawaiian taro diversity there was not enough evidence to determine how much of the diversity stemmed from cross-breeding and how much stemmed from somatic mutations. It was hoped that this study would give some insight into this, but the results of this study were not able to address that question.

An analysis of the Hawaiian taro exclusively was also done to elucidate the correlation between ethnoclassification of taro cultivars and their actual genetic relationships. The neighbor-joining analysis was chosen (Figure 5.2) for the same reasons articulated above. Placement of members of the most diverse morphotype groups were noted within the data set. According to Hawaiian ethnoclassification, these are the morphotype groups ‘*Māna*’ and ‘*Lauloa*’ are respectively (Winter 2012, Table 5.1). They are both named after a salient morphological trait shared by members of the group. “*Māna*,” refers to the branching-corm shared by all members of the group. Members are differentiated by color. As examples, ‘*Māna`ula`ula*’ is a taro with a branching corm and a reddish petiole (“`*ula`ula*” means “reddish”); and ‘*Māna`ulu*’ is a taro with a branching corm that is unusually yellow when cooked which resembles the color of a cooked `ulu (“`*ulu*” means “breadfruit” or *Artocarpus altilis* (Park.) Fosberg). “*Lauloa*,” refers to the elongated leaf shared by all members of the group. Members of this group are also differentiated by color. As examples, ‘*Lauloa`ele`ele*’ is a taro with an elongated leaf and a blackish petiole (“`*ele`ele*” means “blackish”); and ‘*Lauloa`ke`oke`o*’ is a taro with an elongated leaf and a light-colored petiole (“`*ke`oke`o*” means “light green to whitish”). In comparing the placement of members of these two groups within the phylogenetic tree it was determined that there is no correlation between Hawaiian ethnoclassification and actual genetic relationships of Hawaiian taro.

University of Hawai`i taro breeders have observed that crosses between cultivars results in progeny with morphological traits not possessed by either parent. These range from coloration and length of the petiole, shape and coloration of the leaf, shape and coloration of the corm, presence or absence of stolons, and more (Yamakawa, pers. comm., 2012; Miyasaka, pers. comm., 2013). An assumption of this study is that somatic mutations produce progeny that are genetically relatively close to the parent (as opposed to cross-breeding which mixes the genes of two parents), and if the majority of Hawaiian taro diversity was produced via somatic mutations, then the expectation is that these would cluster together in the

phylogenetic tree according to morphotype. However, this was not observed in the tree produced from the data in this study. Clusters are not associated with morphotype which follows the trend established by Quero-Garcia et al. (2004). This is evident when looking at members of the morphotype groups 'Mana' and 'Lauloa' which are interspersed throughout the tree.

It is possible, however, that the methods of this study were insufficient to accurately depicting the genetic relationships of Hawaiian taro. James et al. (2012) did observe some, but not exclusive, clustering according to morphotype, especially in the morphotype groups 'Māna' and 'Manini.' Future research may be able to more accurately differentiate between this closely-related group for more insight.

## **5.6 Conclusions**

### ***5.6.1.1 Value of AFLP in research***

The use of AFLP analysis to develop phylogenetic trees remains a viable method to assess the genetic relationships within and between species. It has advantages over other methods in that it selects over the entire genome, and it is relatively inexpensive. AFLP is useful in detecting polymorphic DNA fragments, and when choosing an algorithmic analysis that is appropriate to a population's evolutionary history, can prove to be a useful tool in producing an accurate portrayal of a population's genetic relationships. However, AFLP analysis does have some assumptions that are occasionally violated, which will affect the accuracy of phylogenetic trees produced from such an approach. Combining AFLP with other genetic approaches, in concert with phenotypic-based evidence is the best way to get an accurate depiction of genetic relationships between plants.

### ***5.6.2.1 Co-evolutionary relationship between humans and plants has affects upon plant evolution***

Ancient human migrations, from one biogeographic region to another, included the transport of cultivated plants to not only ensure the survival of the group, but also to ensure the survival of their cultural identity. Although the actual histories of such migrations are coded within oral traditions, muddled, or even lost, the genetics of the plants that they brought with them can elucidate the paths that these migrations took.

Human selection in plants can erode genetic diversity and variability. With plant species that have been transported beyond their naturally-occurring range the

repeated bottlenecks, founder effects, and resultant genetic drift has likely resulted in a purging of alleles. This translates into decreases in genetic diversity within intraspecific populations, even if phenotypic diversity remains high. However, the process of intensification in cultivation—including selection of cross-bred and somatically mutated progeny—can push the evolution of a cultivated species into a trajectory that it would not go into were it not for the co-evolutionary relationship that it developed with a particular human population. The cultivars of *C. esculenta* in Polynesia are an example of this phenomenon. Since different cultures in different biogeographic regions select for different traits, and certain subsets of those are chosen for transport beyond the original biogeographic region, *C. esculenta* DNA provides genetic evidence for human-induced shifts in the evolutionary trajectory of plant species. When this phenomenon occurs in different biogeographic areas the genetic diversity of that area might be narrow, but when compared to another region of intensified diversification the genetic differences can be greater than existed in the pre-domesticated population.

Due to the co-evolutionary nature of the relationship between *C. esculenta* and human cultures, the increase in genetic diversity has resulted in an increase in cultural diversity; and decreases in diversity on either side is directly correlated with decrease in diversity on the other side. The approach and methods of this study have demonstrated evolution within an ethnobotanical population.

#### ***5.6.3.1 Relationship between ethnoclassification and genotypic relationships***

Classification systems in biological sciences are heavily founded in lineage and genotypic relationships. Ethnoclassification systems employed by indigenous cultures do not necessarily put the same emphasis on genetic relationships. A contributing factor to this is that cultures are not always dealing with an entire set of genetic relationships, but rather a subset. Ethnoclassification of cultivated plants deals with a culture's process of cognitively classifying a population that represent descendants of an unknown number of founder events (in generations past from different biogeographic regions), and that includes members that were produced from both hybridization and somatic mutations. In such cases, ethnoclassification systems may default to be founded in the shared morphologies between phenotypic expressions, even if they are analogous, rather than the genotypic relationships.

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## Chapter 6. Conclusions

### 6.1 Summary of postulates regarding theoretical ethnobotany

While botanists work within the paradigm of ecosystems (Raven et al. 2005), and anthropologists work within the paradigm of socio-cultural systems (Nanda and Warms 2010), ethnobotanists work within the paradigm of social-ecological systems (Berkes and Folke 1998, Berkes et al. 2003). From within this paradigm of linked, biocultural relationships the relatively-new theoretical perspective of “quantum ethnobotany” has emerged (McClatchey and Bridges 2005). This dissertation stands on that foundation, and has proposed both hypotheses and theory in an attempt to approach quantifying the co-evolutionary relationship between plants (as the major ecological feature) and people. The contributions of this dissertation towards that end are the following:

- A unit of measure for quantum ethnobotany, the “quantum co-evolution unit” or “QCU,” has been identified and defined (see Chapter 2). The QCU can be used as a way to evaluate changes in human interaction with plants over time, and therefore can be used to quantify ethnobotanical evolution.
- A population of QCUs is under the same evolutionary pressures as a population of alleles such as deletion, substitution, mutation, bottlenecking, and drift; and therefore, ethnobotanical evolution is defined as changes in QCU frequency over time.
- QCUs are scalable and, when used in the context of a population, allow scientists to examine plant-people relationships through the lens of population dynamics.
- Because of the strong links between biodiversity and cultural diversity (Maffi & Woodley 2010), quantum ethnobotany has applications for resource-managers and policy-makers who endeavor to maintain and promote biocultural diversity within their respective systems.

### 6.2 Summary of hypotheses, results, and emergent theory

From the standpoint of quantum ethnobotany ten hypotheses were proposed about the co-evolutionary nature of the relationship between plants and people:

1. Human relationships with plants are co-evolutionary. They consist fundamentally of two parts: A plant subunit and a cultural-practice subunit. Together they comprise the smallest measurable (quantum) unit through which changes in human interaction with plants can be evaluated over time.
2. Quantum Co-Evolution Units can be measured on differing scales depending on the taxon of the plant and the unit of human population with which it interacts.
3. There is a set of dynamic Quantum Co-Evolution Units within each culture that comprise an ethnobotanical population for that culture.
4. Cultural practices without an associated plant and plants without an associated cultural practice cannot be investigated by researchers using the approaches of quantum ethnobotany as they lack one of the needed subunits.
5. Ethnobotanical populations, comprising the total amount of Quantum Co-Evolution Units within a human population, can be measured at various points in time.
6. Changes in proportionality and frequency of Quantum Co-Evolution Units (QCU) to one another within the ethnobotanical population, as well as additions, subtractions, and changes to and of QCU in the ethnobotanical population between various points in time, can also be quantified. Such a methodology could be used to demonstrate ethnobotanical evolution.
7. Quantum co-evolution units can be used to measure how specific sociocultural practices influence biodiversity within a social-ecological system.
8. There is a set of criteria that can be used to test whether an ethnobotanical population is evolving. If any/all of the criteria are met then the population is evolving. If none are met the population is static. The criteria are:
  - a. Particular QCU have increased in frequency because of selection.
  - b. Particular QCU have decreased in frequency because of selection.
  - c. One or more QCU(s) have been added or lost through events homologous to those involved in the process of biological evolution (mutation, extinction, etc.).

- d. One or more QCU subunit(s) has changed (i.e., replacement of a lost or abandoned plant or tradition subunit) resulting in the creation of a new QCU.
9. Co-evolution and co-extinction of plant-culture relationships are cyclical processes and quantum ethnobotany can be used to understand how these affect the trajectory of evolution in ethnobotanical populations.
  10. Re-emerging cultures may resurrect traditional recognition of plant diversity and create or borrow practices in order to restore (redevelop) relationships with plants, and therefore social-ecological system resilience.

Of these ten hypotheses, this dissertation focused on addressing three—Hypotheses 1, 2, 6, and 8. It did so by examining the relationship-scale of a plant species and a human culture, specifically *Colocasia esculenta* (taro) and the Hawaiian culture. As a means to test these hypotheses, new methods were created and implemented, and well-established methods were followed. The new methods include never-before attempted approaches—using linguistics—to understand and estimate a range of diversity for an important cultivated plant (taro, *C. esculenta*) in a period of time two centuries earlier. The well-established methods include utilizing amplified fragment length polymorphism (AFLP) to better understand how co-migration patterns between humans and taro have influenced the genetics of Hawaiian taro. The methods, data, and analysis do provide evidence supporting the hypotheses. The specific results and emergent theory that this dissertation contributes to science are the following:

Result 1: Linguistics indicates that Hawaiians, anciently, developed and managed approximately 400 distinct taxa of taro at the height of cultivation, indicating that Hawai`i was once a center of diversity beyond the natural range for the species.

Emergent theory 1: Linguistics has useful tools for estimating ancient plant diversity in intensively managed plant populations.

Result 2: Field observations juxtaposed with this new understanding, of previous levels of diversity, indicate that as much as 85% of Hawaiian taro diversity has been lost to extinction in the past 200 years.

Emergent theory 2: Pre-colonial plant diversity may be estimated through combining linguistic and genetic information about current plant populations.

Result 3: Trends in Hawaiian ethnoclassification of taro clearly demonstrate a tendency for grouping taros according to morphotypes, and differentiating within the group primarily based on salient color differences.

Emergent theory 3: Ethnoclassifications and folk taxonomies include different sorting criteria at different ranks. A single explanation for classification and naming is not likely to explain all levels of recognized diversity.

Result 4: Genetics can be used to demonstrate that the history of human-taro co-migration has influenced the evolutionary trajectory of taro.

Emergent theory 4: Plant-human co-evolution is strongly influenced by co-migration patterns that result in bottlenecks and drift (both genetic and cultural).

Result 5: An introduction of new taro cultivars has opened up opportunities for an increase functional redundancy and to create new taro-based traditions to emerge.

Emergent theory 5: Plant-based traditions are founded in plant genetics, and increasing diversity of plant genetics results in an increase in cultural diversity.

Result 6: The criteria for demonstrating evolution of an ethnobotanical population has been met via a study on the diversity of taro cultivated in Hawai`i, and associated traditions; and how these have changed over time.

Emergent theory 6: The relationship between people and plants is an evolving one, and this evolution can be demonstrated.

The overall conclusion of the research produced in this dissertation is that the Hawaiian culture has a long-standing co-evolutionary relationship with taro (*C. esculenta*) which has taken each a coupled co-evolutionary trajectory that is

different than can be observed in other biogeographic regions even if a similar co-evolutionary relationship exists. Furthermore, quantum ethnobotany provides the lens through which such relationships can be examined. The co-evolutionary relationship between taro and Hawaiian culture has had an influence of the evolutionary trajectory of each, such that if the relationship didn't exist neither population would have the diversity—genetic and cultural respectively—that it has today.

### **6.3 Implications for future research**

The hypotheses and theories proposed in this dissertation open up opportunities for future research. An area of research that wasn't addressed methodologically in this dissertation is a study to establish a base-line assessment of a broad ethnobotanical population (i.e., a population of QCU's associated with more than one biological species). Once a baseline assessment is conducted, that opens up an opportunity for research on the future of specific plant-human interactions. If there are subsequent demonstrable changes observed that are consistent with predictions, then that would not only represent quantified evidence in support of the theory of ethnobotanical evolution, but allow for an understanding of the trajectory of evolution. Another aspect of research that was not addressed in this dissertation is a closer examination of how the respective properties of a particular plant species occurring in different biogeographic regions have influenced the evolutionary trajectory of the cultures in those regions.

Genetic approaches have aided our understanding of how the co-evolutionary relationship between taro and people has contributed to genetic drift in *C. esculenta*. This was done using taro growing in Hawai'i as an example. If done on a larger scale to include several biogeographic regions (i.e., Oceania, Asia, and Africa), and done using the same methods, human influence on genetic drift away from the core center of diversity can be calculated. Such an approach can also be done with other cultivated plant species that have likewise been incorporated into a co-evolutionary relationship with humans, and subsequently transported beyond its natural range. However, the best molecular methods for doing this will vary from species to species, and would need to be determined on a species by species basis. The best way to do this for taro remains to be determined, but this dissertation has contributed towards figuring that out.

## 6.4 Proposed theory regarding human interactions with plants

There is a broad, over-arching theory about human interactions with plants that this dissertation touches on. The theory is that *Homo sapiens* are among the many species in the kingdom Animalia that have entered into co-evolutionary relationships with plants (Pollan 2002). Such co-evolutionary relationships have concurrently influenced the evolutionary trajectory of both the plants and human cultures. Gaining a better understanding of this is not only a worthy academic pursuit, but may be essential to the survival of our species on this planet (Prance et al. 2007).

## 6.5 Literature Cited

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## Appendix 1. List of *Kalo* Varietal Names

The edited list of names was compiled from an original list of 677 *kalo* varietal names which reflects non-contradicted synonymy as well as color synonymy. Typographical errors have been eliminated. A “1” in the adjacent column indicates that it was included in the conservative diversity estimate, the liberal estimate, or both.

Species (Group) Name	Subspecies Epithet	Estimate	
		Cons.	Lib.
A'a / A'e / A'ea'e		1	1
'A'ala-piko		0	1
'A'apu / 'Apu / 'A'apo / 'Apo / 'Apua / 'O'apu	[maoli]	0	1
	'ele'ele / hiwa	1	1
	ke'oke'o	1	1
	lehua / 'ula'ula	1	1
	lenalena	1	1
	wai	0	0
'Ahapii		0	1
'Āhē / 'Ēhē	[maoli]	0	1
	'ele'ele	1	1
	ke'oke'o	1	1
	lenalena	1	1
	'ula'ula	1	1
'Ahu'ula		0	1
'Aiwi-kea		0	1
Akaka*		1	1
Akiahiale*		1	1
'Akilolo		1	1
'Akoki / 'Akohi		1	1
Akole ka uula*		0	1
'Ala	[maoli]	0	1
	'ele'ele	0	1
	kea / ke'oke'o	0	1
	'ōpelu	1	1
	o-Puna	1	1
	pipika	1	1
Alele*		1	1
Aneli'i*		1	1
'Ānunu		1	1
A-'o'ia-o-Kalalau		0	1

'Ape	[maoli]	0	0
	kea	0	0
	poni	0	0
'Āpi'i / 'Āpi'ipi'i	[maoli]	0	1
	kea	1	1
	lehua / 'ula'ula / 'ula	0	1
'Apuwai / 'Apowai	[maoli]	0	1
	'ele'ele / uliuli	1	1
	kea / ke'oke'o	1	1
	lenalena	1	1
	'ula'ula / 'ula	1	1
'Apowale / 'Apuwale	[maoli]	0	1
	'ele'ele / uliuli	1	1
	kea / ke'oke'o	1	1
	lenalena	1	1
	'ula'ula / 'ula	1	1
Auau leo (lio) nui*		0	1
Awa-a-puhi*		1	1
'Āweo / 'Āweu / 'Āweoweo / 'Āweuweu		1	1
'Ele'ele / 'E'ele	[maoli]	0	1
	hiwa	1	1
	hiwapa'a	1	1
	lauoa	1	1
	maka'ōpio	1	1
	mākoko	1	1
	naioea	1	1
Elekai*		1	1
'Elepaio		1	1
'Eleua		1	1
Ēulu / Ēula	[maoli]	0	1
	ke'oke'o	1	1
	kohu-uauahi	1	1
Ha'o	[maoli]	0	1
	'ele'ele	1	1
Ha'aha'a		1	1
Haehae	[maoli]	0	0
	'ele'ele	0	0
	ke'oke'o	0	0
	'ula'ula	0	0
Hahu-ko-kai		0	1

Hale-o-Lono		1	1
Hāloa		0	1
Hāokea / Hāakea / Hāa'ākea / Hā'awikea / 'Ahakea	[maoli]	0	1
	hā'ula'ula	1	1
	hāuliuli	1	1
	ke'oke'o	1	1
	māna	1	1
	piko	1	1
	piko-'ula	1	1
Haole luahine		0	1
Hāpu'u / Hāpu'upu'u	[maoli] / maoli	0	1
	'ele'ele / hāuliuli / uliuli	1	1
	kea / ke'oke'o	1	1
	lenalena / lena	1	1
	kūkea / nūkea	1	1
	'ula'ula	1	1
He'e		1	1
Heilia*		1	1
Hekili		1	1
Hele-mauna		0	1
He-nele		0	1
He-`owā-hulunui		0	1
Heu-ā-lehu		0	1
Heu-'ele		0	1
He'ula		0	1
Hinale / Hinalea		1	1
Hinali'i		1	1
Hinapū		1	1
Hinu-kalo		1	1
Hinu-pua`a		1	1
Hiwa		0	1
Hoeke*		1	1
Hoene	[maoli]	0	1
	black	1	1
	red	1	1
Hōkeo / Hōkea / Hākeo		1	1
Hona		1	1
Hō'ole-(i)-nā-wao		0	1
Hō'ole-ke-kalo-po'o- honu(e)*		0	1
Houa*		1	1

Hualani		1	1
Huamoa		1	1
Huli-pū-loa		0	1
Humuhumu		1	1
'Ie'ie / 'Ia'ia	[maoli]	0	1
	'ili'ā*	1	1
Iheihe		1	1
Iheihei lei		1	1
'Ihi-lani		1	1
'I'i		0	1
'Ili'ā / `Ili`a`a		1	1
'Iliuaua		0	0
Ipu-o-Lono	[maoli]	0	1
	kea / ke'oke'o	1	1
	'ula'ula	1	1
	piko-'ula	1	1
Kapu'ukōnane		0	1
Kaawelu-i-ka-pali-o- 'Awakea		0	1
Kaena-ke-kanaka*		0	1
Kahalu'u		1	1
Kahu-kō-kai / Kahe-kō- kai		0	1
Kāi	maoli	1	1
	'ala	1	1
	'āweuweu	1	1
	'ele'ele / uliuli	1	1
	ho'ōkia*	1	1
	kea / ke'oke'o	1	1
	koi	1	1
	koi-welawela	1	1
	nenene / nenenene	1	1
	pala	1	1
	'ula'ula	1	1
	welowelokā (likely a typo for, welowelolā)	1	1
Kaimoi*		1	1
Kaina-i-ke-kanaka*		0	1
Kaina-i-ke-kaua*		0	1
Ka-io-aweawe*		0	1
Kala	ni'o	1	1
	poni	1	1

Kalani		1	1
Kalalama makahi*		0	1
Kalalau		1	1
Kalani pili		1	1
Kalo-i-kū		0	1
Kalola		0	0
Kāmau		1	1
Kanaio		0	0
Kanawao		1	1
Kaneli`i		1	1
Kāni`o		0	1
Kapai-o-ākea*		0	1
Kapala		1	1
Kapalili		1	1
Kapuhili		1	1
Kauanio		1	1
Kaue*		1	1
Kauhaikalehuhoole*		0	1
Kawale uua*		1	1
Kawele ole*		0	1
Kawelo		1	1
Kepoe		1	1
Ki`i hekekē		1	1
Kīkī panapala / K. palapala		1	1
Kīkī`i		1	1
Kili `o`opu		1	1
Ko`i aweawe*		1	1
Koa`e	[maoli]	0	1
	`ele`ele	0	1
	ke`oke`o	0	1
	`ula`ula	0	1
Kohikū*		1	1
Kokoko-he-uhi*		0	1
Ko`okā		1	1
Kū loa		1	1
Kuamu		1	1
Kuapapa		1	1
Kueha*		1	1
Kukai`iole [sic]		1	1
Kumakau*		1	1

Kumu	[maoli]	0	1
	'ele'ele	1	1
	kea / ke'oke'o	1	1
	kūloa	1	1
	poni	1	1
	'ula'ula	1	1
	welowelolā	1	1
Kū'oho / Kū'ohu	[maoli]	0	1
	hai*	1	1
	piko	1	1
Kūpala		1	1
La'o uaua		1	1
La'aloa / Ola'aloa		1	1
Laholoa		0	1
La'i-o-kona		1	1
Lapa		1	1
Lau 'ape		1	1
Lau kapalili		0	1
Laukōnā		1	1
Lau lele		1	1
Lauloa	[maoli]	0	1
	'ele'ele / hā'ele'ele / palakea-'ele'ele	1	1
	'ele'ele-ōma'o	0	1
	'ele'ele-'ula	0	1
	ke'oke'o / hāke'oke'o / kea	1	1
	koko / hā'ula / 'ula'ula	1	1
	uliuli / hāuliuli	1	1
	manini / 'ōni'oni'o / palakea-ke'oke'o	1	1
	palakea	1	1
	palakea-'ula	1	1
	pāna'e*	1	1
	papamū / palakea-papamū	1	1
	piko-'ula	1	1
	poni	1	1
	Launui	[maoli]	0
pa'akai		1	1
Lehua	maoli	1	1
	aola*	1	1
	'āpi'i	1	1
	'ele'ele / hāuliuli	1	1
	hō'ole*	1	1

	ke'oke'o	1	1
	kū-i-ka-wao / kū-kuahiwi	1	1
	lenalena	1	1
	'ōni'oni'o	1	1
	palai'i	1	1
	'ula'ula	1	1
Lele		1	1
Le`o		1	1
Lī'apu		1	1
Lihilihi ke'oke'o		0	1
Lihilihi-molina	[maoli]	0	1
	ke'oke'o	1	1
	'ele'ele	1	1
Liko-lehua		1	1
Līī-lehua		1	1
Loha / Lola		1	1
Mā'au(w)ea / Mā'au(w)eo / Mānau(w)ea / Mānau(w)eo / Māmau(w)ea / Māmau(w)eo	[maoli]	0	1
	hāuliuli	1	1
	'ula	1	1
Ma'awe		1	1
Maea		1	1
Mahaha*	[maoli]		1
	ke'oke'o	1	1
	'ula'ula	1	1
Mahai		1	1
Mahakea / Mākea		1	2
Mahamaha ke'oke'o		1	1
Māhuna		1	1
Mai ahua*		1	1
Mā'i'i / Mā'i'i'i		1	1
Mā'i'o		1	1
Maka'iole		1	1
Maka 'ōpio / M. 'ōpi'i		1	1
Maka ua		1	1
Maka weo		1	1
Mākihi / Mākohe / Mākohi / Mōkohe / Mōkohi	[maoli]	0	1
	'ula'ula	1	1
	'ele'ele	1	1
Mākoko		0	1

Mākole		1	1
Mākūkū		1	1
Malihini-a-ka-wai		0	1
Māmane		1	1
Māmanu*		1	1
Māna / Māna-o-	[maoli]	0	1
	‘āniholoa*	1	1
	‘ele‘ele / uliuli	1	1
	‘ula‘ula / hā‘ula‘ula	1	1
	hua	1	1
	kea / ke‘oke‘o	1	1
	lauoa	1	1
	‘ulu / lenalena / melemele / ‘owene / ‘uwele	1	1
	‘ohe	1	1
	‘oko‘a	1	1
	‘ōpelu	1	1
	piko	1	1
	pipika	1	1
	uauahi	1	1
	uhapua*	1	1
	wai / wai-ke-ohe	1	1
	weo / wea	1	1
Mana-piko		1	1
Mānawai ākea*		1	1
Manini	[maoli]	0	1
	‘ele‘ele	1	1
	hākikokiko	1	1
	uliuli / hāuliuli	1	1
	kākau*	1	1
	kea	1	1
	lau-kikokiko	1	1
	‘ōpelu	1	1
	‘owali	1	1
	‘ula	1	1
	Manouulu*		0
Manu		1	1
Manuia*		1	1
Manulele		1	1
Maua	melemele	1	1
	ulu	1	1

Mauna		1	1
Melemele		1	1
Mimi'iole		0	1
Moa		1	1
Moana / Moano		0	1
Mōhihi		1	1
Moi	[maoli]	0	1
	'ele'ele	1	1
	kea / ke'oke'o	1	1
	'ula	1	1
Mokihana		1	1
Mōlina		1	1
Na-kalo-aola-o-kalalau*		0	1
Nahiolea		1	1
Naio		1	1
Naioea	[maoli]	0	1
	'ele'ele	0	1
	ke'oke'o	0	1
	lehua / 'ula'ula	0	0
Nā-kalo-i-kū'ē*		0	0
Nāiliko'i*		1	1
Nana	[maoli]	1	1
	piko	1	1
Nana-i-puhene-na-kalo		0	1
Nao		1	1
Nāpili		1	1
Naua		1	1
Nāwao		1	1
Ne`ine ikekanaka*		0	1
Ne'ene'e		1	1
Nihopu'u		1	1
Nina		1	1
Ni'o		0	1
Nohiapele*		0	0
Nohu	[maoli]	0	1
	'ele'ele	0	1
Nūkea	dark [sic]	1	1
	light [sic]	1	1
Nuku 'e'ehu		1	1
Nuku kau		1	1
Nuku-manu		1	1

O-ka-he'e-kō-kai		0	1
ʻO(w)ā'o(w)ā		1	1
Oalu / Oolu		1	1
Oaulu nui		1	1
ʻOhe	[maoli]	0	1
	ʻeleʻele	1	1
	kea / keʻokeʻo	1	1
	ʻulaʻula / ʻula	1	1
ʻŌhiʻa		1	1
ʻŌhuehue		1	1
ʻŌlena		1	1
ʻŌnihinihi		1	1
ʻOʻopu		1	1
ʻOʻopukai / ʻOpukai	[maoli]	0	1
	keʻokeʻo	1	1
	ʻulaʻula	1	1
ʻOpae-ʻula		1	1
ʻŌpelu		0	1
ʻŌpelu haoee*		1	1
ʻŌpule		1	1
Owale*		1	1
ʻOwau		1	1
ʻO(w)ene	[maoli]	0	1
	ʻeleʻele	1	1
	keʻokeʻo	1	1
	lenalena / melemele	1	1
	māna	0	1
	ʻulaʻula	1	1
Paʻakai		1	1
Paʻakai mikomiko		0	0
Paʻakea		0	0
Paʻapaʻaʻina		1	1
Pae		1	1
Pa`ea			
Paʻele-hili-mānoanoa		0	1
Paʻiaha		1	1
Paipu lana		1	1
Pākē		0	0
Pākea		1	1
Pala	[maoli]	0	1
	hāokea / kea	0	0

	mahiki	1	1
Palapalaha / Pālaha*		1	1
Palaʻiʻi	[maoli]	0	1
	ʻeleʻele	1	1
	kea / keʻokeʻo	1	1
	pohā	1	1
	poni	1	1
	ʻulaʻula	1	1
Palakea	ʻeleʻele	1	1
	keʻokeʻo	1	1
Pālau		1	1
Palili	[maoli]	1	1
	ʻulaʻula	1	1
Pana		1	1
Pani-kohe		0	1
Papakole(a) kāwaʻa		1	1
Papakole(a) koaʻe		1	1
Papala kea*		0	0
Papamu		0	1
Papapueo		1	1
Pāʻū-o-hiʻiaka		1	1
Paua		1	1
Pauʻiole		1	1
Pehua		1	1
Peke		0	1
Pelu hāʻele		1	1
Pelu haole		0	0
Peu	[maoli]	0	1
	ʻeleʻele	1	1
	kea / keʻokeʻo	1	1
	lena	1	1
	ʻulaʻula	1	1
Peue		1	1
Pia / Piapia		1	1
Pihalole / Pihalale		1	1
Piʻi hālāwai		1	1
Piʻialiʻi	[maoli]	0	1
	ʻeleʻele	1	1
	keʻokeʻo	1	1
	melemele	1	1
	ʻulaʻula	1	1

Pi'ikea		1	1
Piko	[maoli]	0	1
	'ele'ele	1	1
	hao*	1	1
	kea	1	1
	ke'oke'o	1	1
	lehua-'āpi'i	0	1
	uaua	1	1
	uliuli / uli	1	1
Piko-a-Wākea		1	1
Piko'ele		1	1
Piko-nui		1	1
Pilimai		1	1
Pipiko		1	1
Pōhina		1	1
Poi pulana* / P. pulaua*		1	1
Pololū		1	1
Poni	[maoli]	0	1
	'ele'ele / uliuli	1	1
	kea	1	1
	mana	1	1
	'ula'ula	0	1
Po'o hunue* [honua]		0	1
Pōpolo		0	1
Pua kawaihae		1	1
Puakai mikomiko		1	1
Pueo	[maoli]	0	1
	hālenalena	1	1
	ke'oke'o	1	1
Puhi		1	1
Pulani*		1	1
Pu'u		1	1
Pu'u kōnane		1	1
Pu'u nānā		1	1
Puwalu/Pualu		1	1
U(w)ahi-a-pele	[maoli]	1	1
	'ula'ula	1	1
Ua lehua		1	1
Uahi 'oki*		1	1
Ualehu / Ūlehu		0	1
Uaua	[maoli]	0	0

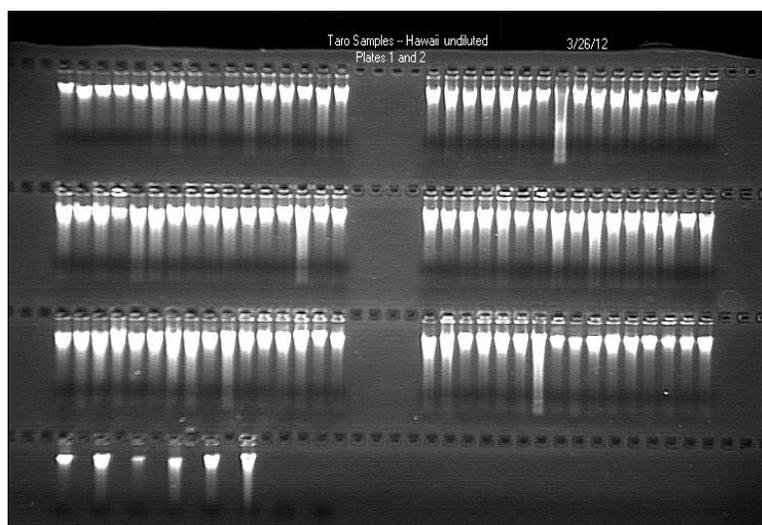
	'ele'ele	0	0
	ke'oke'o	0	0
	mōlina	1	1
	piko	0	0
Uhai*		1	1
Uhu		1	1
Uia		1	1
Ula	[maoli]	0	1
	li'i	1	1
	nui / mani / mau / mau	1	1
	i'a*	1	1
'Ula'ula	[maoli]	0	0
	'āhiu	1	1
	'āniholoa	1	1
	kūmū	0	0
	moano	1	0
	poni	0	0
	uahi-a-pele	0	1
'Ūlei		1	1
Uli / Uliuli		0	1
'Ume / 'Umi / 'Umi'umi		1	1
Uwauwahi		0	1
Wa`e		0	0
Waiākea		1	1
Wai'anae		1	1
Waiānuenuē		1	1
Wailana		1	1
Wehiwa / Wehewa / Wewehiwa	[maoli]	0	1
	'ōni'oni'o	1	1
	uliuli	1	1
Welehu		1	1
Welowelolā		0	1
Weo	[maoli]	0	1
	black [sic]	1	1
	red [sic]	1	1
Wewemana		1	1
Wia		1	1
Wolu		1	1
<b>TOTALS:</b>		<b>368</b>	<b>482</b>

## Appendix 2. Lab notes for taro AFLP study

National Clonal Germplasm Repository (University of California, Davis)  
March – May 2012

### 03/22-23/2012

All DNA samples (except #023) extracted from dried leaf tissue by the CTAB method (Appendix 1).

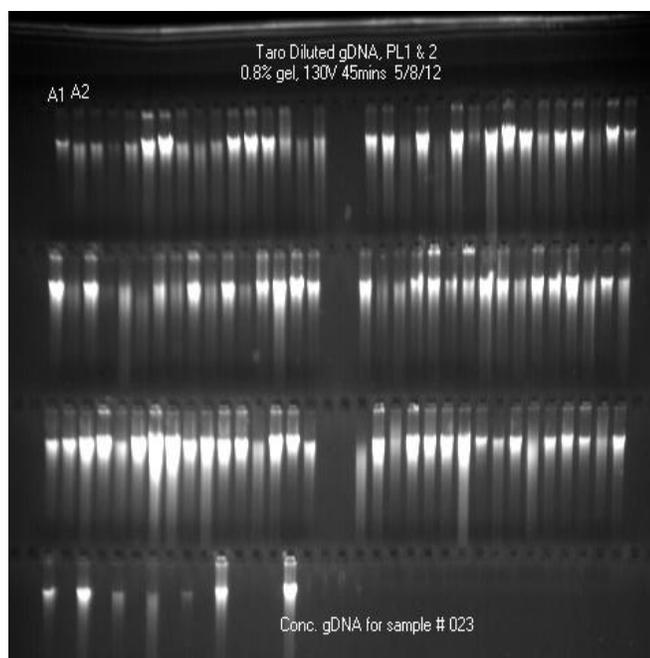


### 05/7-8/2012

Sample #023 DNA extracted by CTAB method (in single tube) (Appendix 2A).

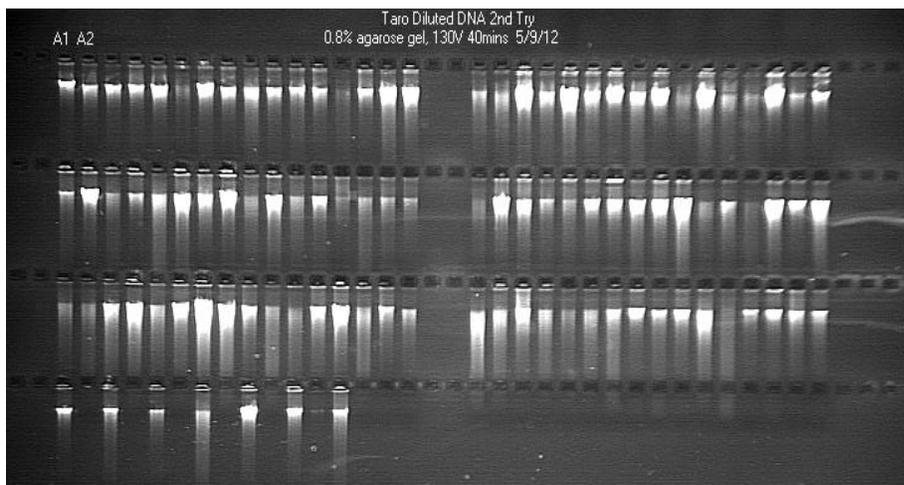
**05/08/2012**

Both DNA plates initial dilution were done. All but C7 was diluted by 90% (10ul/90ul dH<sub>2</sub>O). The results are seen below.



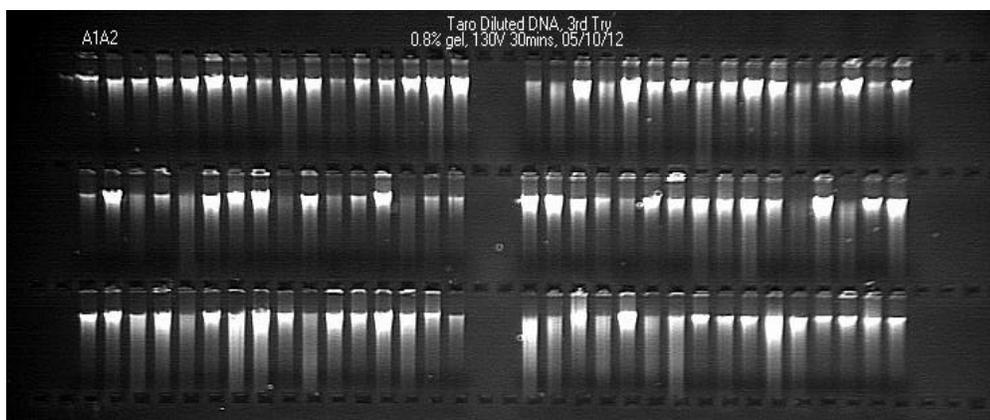
**05/09/2012**

The first concentration adjustments were done by removing 25ul of solution and adding 25ul of dH2O to reduce by 50% to samples that were too concentrated. In the samples that were not of sufficient concentration, 25ul of genomic DNA was added. The results are seen below.



**05/10/2012**

The final concentration adjustments were done. Dilutions and adding was done in the same manner as above except 20ul was used instead of 25ul. The results are seen below.



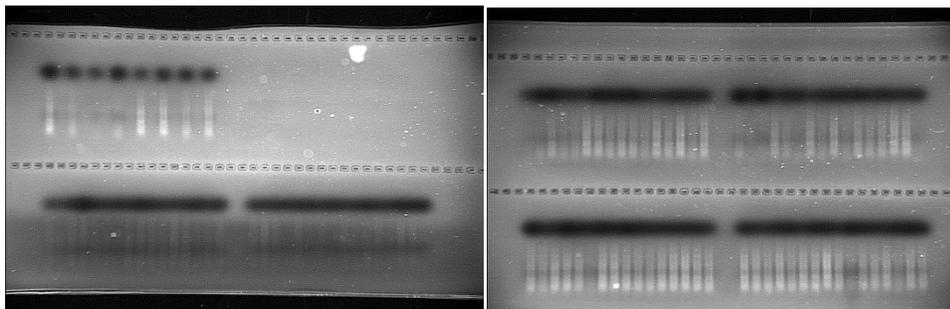
**5/11/2012**

Restriction-Ligation (Appendix B) for both plates done, incubated from about 5pm to next morning, when Malli diluted each sample with TE buffer, pH=8.

**05/14/2012**

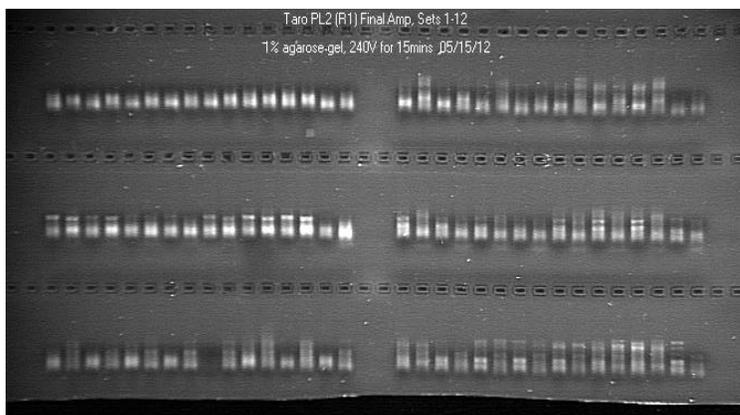
Pre-Selective Amplification on both plates done. Most samples diluted 20-fold (10ul product in 180ul TE), but some diluted less (10ul product in 100ul TE).

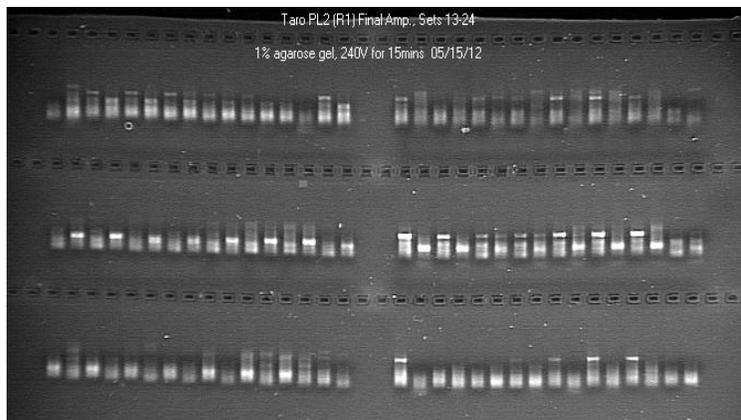
1.5% agarose gel done to check Pre-Amp results - results were good.

**05/15/2012**

Final Selective Amplification for PL2 (which is actually only the first row of 8 samples) done on all 24 different primer combinations (24 combos of 3 different MseI-EcoRI primer pairs, i.e. 72 primer pairs total attempted).

Ran 1% agarose gel on PL2 final amp products to check if they worked - results were good for all 24 primer combinations.





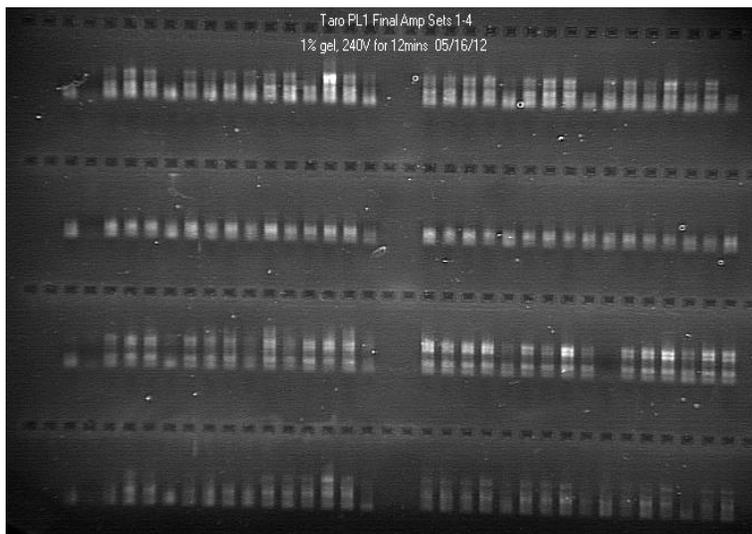
Final Selective Amplification for PL1 done on primer combinations #1-4 (also called "Sets 1-4").

Setup plates for PL2 Sets 1-24 for the Applied Biosystems 3130xl Genetic Analyzer capillary electrophoresis system (Hi-Di Formamide + GS500 ROX size standard master mix, then add final amp product and denature).

Ran PL2 Sets 1-24 on 3130 machine.

### **05/16/2012**

Ran 1% agarose gel on PL1 Sets 1-4 final amp product on - results were good for all 4 sets.

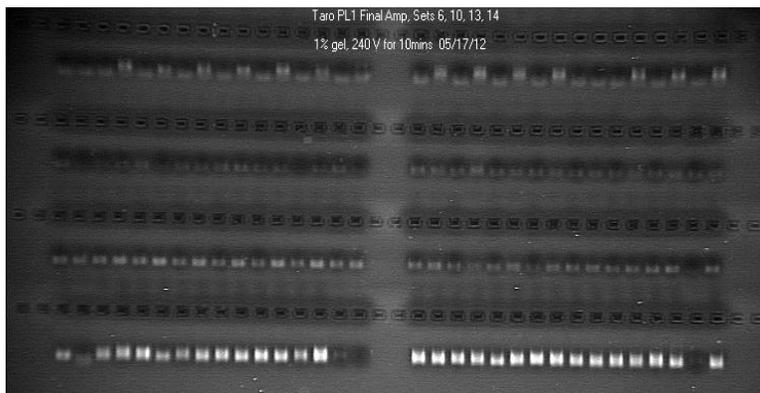


Setup and ran PL1 Sets 1-4 on 3130 overnight 05/16-17/2012.

**05/17/2012**

Final Selective Amplification for PL1 done on primer combinations #6, 10, 13, and 14 (also called "Sets 6, 10, 13, 14").

Ran 1% agarose gel on PL1 Sets 6, 10, 13, 14 final amp product on 05/17/2012 - results showed sets 6 and 14 good, but 10 and 13 failed.

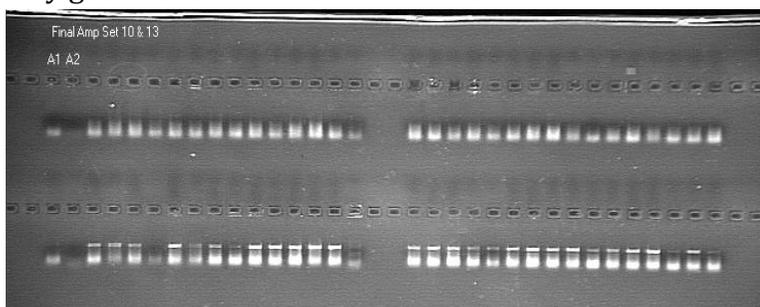


A "redo" final amp reaction for PL1 Sets 10 and 13 was done.

Setup and ran PL1 Sets 6 and 14 on 3130, but first injection showed that Set 6 actually failed (i.e. we interpreted a "false positive" of the agarose gel results for Set 6), so the run for Set 6 was stopped. Set 14 results were good, however.

**05/18/2012**

Ran 1% agarose gel on "redo" final amp reaction for PL1 Sets 10 and 13 on - results were inconclusive for Set 10 (i.e. bands visible, but wary of another "false positive" result), but clearly good for Set 13.



Setup and ran PL1 Set 10 test run of only first injection (first two rows), and full plate of Set 13, on the 3130.

## Appendix 2A: CTAB Protocol

NCGR-Davis CTAB protocol for 96-well extraction

CTAB buffer (25 mL for 50 samples x 2 for one plate)

Tris HCl, pH 8.0	100 mM	
NaCl	1.4 M	
EDTA	20 mM	
CTAB	2% w/v	
PVP-40	2% w/v	
PVPP	4% w/v	Not soluble in buffer; mix regularly (4% = 1 g)
$\beta$ -mercaptoethanol	1%	Add just before use (1% = 250 $\mu$ L)

**DANGER: Beta-mercaptoethanol and chloroform are hazardous chemicals! Alcohols, e.g. isoamyl alcohol, are highly flammable liquids!** Use personal protective equipment at all times during the DNA extraction protocol and use particular caution when working with these chemicals. Use these chemicals in the fume hood whenever possible. Never work with an open flame near any alcohol or other flammable substance.

1. Turn on water bath and preheat to 65C.
2. Place CTAB buffer and PVPP in 50 mL Falcon tube and place in 65C water bath.
3. Sample Preparation: Mark one end of strip tubes with column number. Using bead dispenser place one 4mm stainless steel ball in each tube of 96-well plate. Load tissue (~100 mg fresh tissue) into each well and record on 96-well record sheet. (Depending on the tissue, dispense a second ball into each tube of 96-well plate and cover tubes with strip caps).
4. For fresh tissue proceed to step 5, for desiccated/lyophilized tissue proceed to step 6.
5. Grinding Fresh Tissue: Place rack with tubes in liquid nitrogen for one minute to freeze tissue (for better contact between tubes and liquid nitrogen remove bottom of rack). Repeat between grinding sessions. **DANGER: Liquid nitrogen is an extremely cold, liquefied gas!**
6. **Replace bottom and cover on rack.** Place rack (with bottom and cover) into GenoGrinder and secure with clamp.
7. Disrupt tissue at 1200 cycles per minute (200 on dial, 1X on toggle switch) for one minute.
8. Repeat step 7 as necessary to grind tissue to fine powder. (Between grinding sessions it is often helpful to knock rack against a bench, upright or inverted,

- to move samples from top to bottom and check if steel balls are able to move freely.)
9. After final grinding session, hold rack upright and knock against countertop again to loosen powdered tissue from caps. (For fresh tissue using liquid nitrogen, place rack in hood and allow caps to thaw slightly for removal.)
  10. Add  $\beta$ -mercaptoethanol to CTAB buffer and mix.
  11. Remove strip caps from tubes and discard.
  12. Add 500  $\mu$ L buffer to each well.
  13. Place new strip caps on tubes and place rack on GenoGrinder.
  14. Mix samples on GenoGrinder by running at 1200 cycles per minute for one minute.
  15. Remove rack from GenoGrinder, cover and place in 65C water with weight on cover. Check that the CTAB buffer has penetrated the powdered samples before proceeding to step 16. If the buffer has not penetrated the samples, run for one more cycle on the GenoGrinder.
  16. Incubate at 65C for 10 minutes with weight holding down the cover of the rack, remove, drain excess water from the plate, manually shake, and incubate at 65C for an additional 10 minutes. Caution: caps can become loose, ensure they are fastened before shaking plate. Note: if the water level is higher than where the cover and rack meet, remove some of the water.
  17. Centrifuge rack(s) briefly to move material away from caps. (Allow to reach 2000-3000 rpm and stop centrifuge.)
  18. In the hood remove strip caps and save.
  19. Add 300  $\mu$ L of 24:1 chloroform-isoamyl alcohol to each sample and recap. Cover plate and place into clamp on GenoGrinder.
  20. Mix samples on GenoGrinder at 500 cycles per minute (Left-000 on dial, 0.5X on toggle switch) for 10-20 seconds.
  21. Leave samples on GenoGrinder to incubate at room temperature for 10 minutes.
  22. Repeat steps 20 and 21 once.
  23. Remove rack from GenoGrinder and centrifuge at 6000 rpm for 15 minutes. Caution: ensure base is reinstalled prior to centrifuging. Centrifuging without the base can cause tubes to break releasing Chloroform-Isoamyl alcohol and may damage centrifuge components.
  24. While samples are centrifuging, prepare a new 96-well plate by labeling plate and marking ends of strip tubes with column numbers, then add 300  $\mu$ L of isopropanol to each tube.
  25. Remove samples from centrifuge and transfer supernatant to new tubes containing isopropanol. Seal tubes with new strip caps and mix by inverting, then write column number on cap ends.
  26. Incubate samples in 4C refrigerator for a minimum of 15 minutes (may be left longer, even overnight) to precipitate DNA.
  27. Remove samples from refrigerator and centrifuge at 6000 rpm for 10 minutes.
  28. Remove from centrifuge, remove caps, retain for reuse.

29. Pour off supernatant from all samples being careful not to lose the DNA pellet.
30. Add 500 uL wash buffer to each sample, cap tubes, mix, and incubate for 15 minutes (may be left longer if necessary).
31. Pour off supernatant, being careful not to lose the DNA pellet. Optional: centrifuge samples briefly (1 minute) at 6000 rpm to move DNA pellet to bottom of the tube. Caution: Optional step does not always cause DNA pellet to stay at the bottom of the tube.
32. If DNA pellet appears dirty repeat steps 30 through 31.

Optional step: Let samples air dry for 10 minutes and prepare 'master mix' of 100 uL TE pH 8.0 and RNase A (4 uL of 10mg/mL or 0.4 uL of 100mg/mL) for each sample. When samples have dried for 10 minutes, add 100 uL of 'master mix' to each sample and mix to re-suspend sample. Incubate at room temperature for at least 15 minutes. Precipitate samples with 800 uL of 95% ethanol (re-cap and invert after adding) and incubate at room temperature for five minutes. Centrifuge for five minutes at 6000 rpm then pour off supernatant and continue with procedure. (This combines an RNase treatment and ethanol precipitation – it produces clean samples as part of the extraction procedure.)

33. Turn rack on side in hood to dry pellet.
34. After pellet is dry, resuspend sample in 300 uL of TE buffer.
35. Treat with RNase A (unless optional step between steps 32 and 33 was followed).
36. Clean up.
  - a. Dispose of discarded reagents in appropriately marked waste disposal container(s).
  - b. Allow tubes with plant material, chlorform-isoamyl alcohol, and steel beads to dry in hood, this may take multiple days. Once the tissue is completely dry it will have shrunk somewhat in size and is relatively easy to knock out of the tube along with the stainless steel beads. Throw used tubes (dry only) and caps in the trash.
  - c. Wash stainless steel beads in soapy water and rinse well. When dry, place in beaker and add approximately twice the volume of 0.4 M HCl needed to cover the beads and incubate for 5 minutes. Neutralize HCl with appropriate volume of NaOH (volume dependent on concentration), check solution with pH meter to verify it is between pH 6 and pH 8, preferably close to pH 7. Neutralized solution (Na<sup>+</sup>, Cl<sup>-</sup>, H<sub>2</sub>O) may be disposed down the drain. Rinse beads with DI water, dry and store in sealed container.

## Appendix 2B: AFLP Protocol for ABI Genetic Analyzer 3100

### Step I. Restriction-ligation reaction

<u>Reagent/Item</u>	<u>1X</u>	<u>100X</u>
ddH <sub>2</sub> O	0.100	10.000
10X T4 Ligase Buffer	0.550	55.000
0.5M NaCl	0.550	55.000
BSA (1 mg/mL)	0.275	27.500
EcoRI Adapter**	0.500	50.000
MseI Adapter**	0.500	50.000
EcoRI Enzyme	0.125	12.500
MseI Enzyme	0.125	12.500
T4 DNA Ligase*	0.025	2.500
DNA (0.5 ug)\$	2.750	
Total Volume	5.500	275.000

*Restriction-ligation is done at room temperature (24C) overnight*

\* 0.25 uL of T4 DNA Ligase in 4.75 uL of T4 Ligase Buffer

\*\*Adapters are first aliquoted into single thermal cycler tubes, and denatured on thermal cycler at 95C for 5 minutes. Then the tubes are placed in styrofoam container and let cool slowly before used in the master mix.

Dilute Reaction mixture 20 folds with TE pH 8 (i.e. 5.5 uL of reaction mixture and 94.5 uL of TE)

Desired DNA concentration is 35-95 ng/uL (or 95-265 ng for half reaction or 200-500 ng for full reaction (11 uL reaction) - precheck for completion of digestion

### Step II. Preselective amplification reaction

<u>Reagent/Item</u>	<u>1X</u>	<u>100X</u>
ddH <sub>2</sub> O	10.08	1008.00
10X Taq Pol. Buffer	2.00	200.00
MgCl <sub>2</sub> (25 mM)	2.00	200.00
dNTPs (2.5 mM ea.)	1.60	160.00
EcoRI primer (100 pmol/uL)	0.06	6.00
MseI primer (100 pmol/uL)	0.06	6.00
Taq DNA polymerase (5U/uL)	0.20	20.00
diluted DNA from Step I	4.00	
Total Volume	20.00	1600.00

Dilute reaction mixture 20 fold with TE pH 7.4 (i.e. 10 uL of rxn mix and 180 ul TE)

**Step III. Selective multiplex amplification reaction**

<u>Reagent/Item</u>	<u>1X</u>	<u>100X</u>
ddH2O	2.650	265.000
10X Taq Pol. Buffer	1.500	150.000
MgCl <sub>2</sub> (25 mM)	1.500	150.000
dNTPs (2.5 mM ea.)	3.000	300.000
MseI primer (5 uM) @	1.500	150.000
EcoRI dye-labeled primer (1 uM) @	0.400	40.000
EcoRI dye-labeled primer (1 uM) @	0.400	40.000
EcoRI dye-labeled primer (1 uM) @	0.400	40.000
Taq DNA polymerase (5 U/uL)	0.150	15.000
diluted DNA from Step II	3.500	
Total volume	15.000	1150.000

@ Part #4303050

**Step IV. Preparation of samples for ABI 3100 Genetic Analyzer**

<u>Reagent/Item</u>	<u>1X</u>	<u>96X</u>
Hi-Di Formamide	10.0	960
Genescan 500ROX	0.4	39
Mixture Total	10.4	999

F-mide/500 ROX mixture	10.0
Selective Amp. Product	1.6 or 2.0
Total	11.6 or 12.0

Denature samples at 95C for 5 minutes then place immediately on ice for 5 minutes or until ready to use

**Selective Amplification Primer Combinations**

Yellow highlighted primer combos were used in this study.

The green highlighted one was attempted but failed.

<u>Set</u>	<u>MseI</u>	<u>EcoRI-6FAM</u>	<u>EcoRI-HEX</u>	<u>EcoRI-NED</u>
1	D1-CAA	DB09-ACT	DG11-AGG	DY13-AGC
2	D2-CTT	DB09-ACT	DG11-AGG	DY13-AGC
3	D1-CAA	DB10-ACA	DG12-ACG	DY14-ACC
4	D2-CTT	DB10-ACA	DG12-ACG	DY14-ACC
5	D3-CTC	DB09-ACT	DG11-AGG	DY13-AGC
6	D4-CAG	DB09-ACT	DG11-AGG	DY13-AGC
7	D3-CTC	DB10-ACA	DG12-ACG	DY14-ACC
8	D4-CAG	DB10-ACA	DG12-ACG	DY14-ACC
9	D5-CAT	DB09-ACT	DG11-AGG	DY13-AGC
10	D6-CTA	DB09-ACT	DG11-AGG	DY13-AGC

11	D5-CAT	DB10-ACA	DG12-ACG	DY14-ACC
12	D6-CTA	DB10-ACA	DG12-ACG	DY14-ACC
13	D7-CAC	DB09-ACT	DG11-AGG	DY13-AGC
14	D8-CTG	DB09-ACT	DG11-AGG	DY13-AGC
15	D7-CAC	DB10-ACA	DG12-ACG	DY14-ACC
16	D8-CTG	DB10-ACA	DG12-ACG	DY14-ACC
17	D1-CAA	DB15-AAC	DG16-ATC	DY17-AAG
18	D2-CTT	DB15-AAC	DG16-ATC	DY17-AAG
19	D3-CTC	DB15-AAC	DG16-ATC	DY17-AAG
20	D4-CAG	DB15-AAC	DG16-ATC	DY17-AAG
21	D5-CAT	DB15-AAC	DG16-ATC	DY17-AAG
22	D6-CTA	DB15-AAC	DG16-ATC	DY17-AAG
23	D7-CAC	DB15-AAC	DG16-ATC	DY17-AAG
24	D8-CTG	DB15-AAC	DG16-ATC	DY17-AAG