Guadian, Laura. Genetic Diversity of Easter Island (Rapanui) Population from Identifiler® Plus autosomal, Y-filer®, and Y-Plex™ 6 Y-STR Loci. Master of Science (Biomedical Sciences, Forensic Genetics). May 2015. Pages, tables, figures, references, titles.

This study investigated the genetic diversity of the Easter Island (Rapanui) population using data on 15 autosomal Short Tandem Repeats (STRs) typed with the commercial STR kits Identifiler® Plus and 23 Y-chromosome STRs typed using Y-filer (17 loci) and Y- PLEX™ 6 (6 loci). The analysis was conducted using genotype and haplotype data of 122 presumably unrelated individuals that included 48 males and 74 females. This study: (i) examined if Easter Island population had reduced genetic diversity in comparison with cosmopolitan populations such as Mainland Chilean, Polynesian, European, and African; (ii) compared genetic affinity of the Easter Island population with historically related cosmopolitan populations; and (iii) investigated the forensic utility of autosomal STRs and Y-STRs in the Easter Island population.

GENETIC DIVERSITY OF EASTER ISLAND (RAPANUI) POPULATION FROM IDENTIFILER® PLUS AUTOSOMAL, Y-FILER®, AND Y- PLEX™ 6 Y-STR LOCI

Laura Guadian, B.S.

APPROVED:

_____

Major Professor

_____

Committee Member

_____

Committee Member

_____

University Member

_____

Chair, Department of Molecular and Medical Genetics

_____

Dean, Graduate School of Biomedical Sciences

GENETIC DIVERSITY OF EASTER ISLAND (RAPANUI) POPULATION FROM IDENTIFILER® PLUS AUTOSOMAL, Y-FILER®, AND Y- PLEX™ 6 Y-STR LOCI

THESIS

Presented to the Graduate Council of the

Graduate School of Biomedical Sciences

University of North Texas

Health Science Center at Fort Worth

in Partial Fulfillment of the Requirements

For the Degree of

MASTER OF SCIENCE

By

Laura Guadian, B.S.

Fort Worth, TX

May 2015

ACKNOWLEDGEMENTS

I would like to express my appreciation to my major professor, Dr. Ranajit Chakraborty, for all his support, assistance, and guidance over the course of my graduate education at the University of North Texas Health Science Center. My gratitude also goes to my advisory committee: Dr. Bruce Budowle, Dr. Lisa Hodge, and Dr. Michael Forster for their time, patience, and understanding. I gratefully acknowledge Dr. Carlos Campusano from the University of Valparaiso, Chile for his help in collecting samples. Also, thanks to my classmates in the Forensic Genetics program for their support and help throughout the two years of my educational career. Thanks go to Michael Nolan for all his help and time offered. I offer my enduring gratitude to the faculty who have inspired me to continue my work in the Forensic Science field. I want to thank and express my love to my mother Dora and my brother Carlos, for always supporting me with all my decisions and my dreams. The most special thanks goes to my father, Jose. He is the main reason why I got this far in my career, I am sorry that he will not be with me to see me graduating, but his memory will be always present.

TABLE OF CONTENTS

LIST OF TABLES

LIST OF FIGURES

CHAPTER I

INTRODUCTION

1.1 Background

Easter Island, or Rapanui, is a volcanic island located east to Polynesia. Rapanui Island is triangular in shape, surrounded by South America, Tahiti, Hawaii, and north Antarctica (1). Easter Island is considered one most of secluded islands in the world; as of 2012, the total size of the Rapanui population was of only 5,761 inhabitants (2, 3). Archaeological evidence suggests an early establishment on the island by Polynesian individuals who earlier populated Fiji, Tonga, and Samoa (4). Tests performed on skeletal remains from Easter Island have discovered mitochondrial DNA (mtDNA), DNA that is inherited maternally, inferred to be from Polynesian origin (3). However, there is also indication of an early interaction with South American individuals, suggested from the presence of sweet potato and the bottle gourd, vegetables believed to have an American origin (3). It has been proposed that the existence of the sweet potato may have been the consequence of Polynesian trips back to South America. However, the possibility of South American individuals (Ameridians) arriving to Easter Island, as well as to other Polynesian islands in ancient times cannot be ruled out (5).

Furthermore, historical architecture present in Easter Island indicates a date of settlement about 400-500 AD (4). Data suggest that Rapanui first pioneers were inhabitants coming from the Marquesas or Mangavera Islands (4). Y-Chromosome STRs are uni-parental markers that are

inherited through the paternal lineage. Y-STR studies have found Native American Y-chromosomes present in Easter Island male (4). Studies propose that before the 20th century, Native American Y-chromosomes were introduced in the male Rapanui population (4). Moreover, Alu insertion polymorphism studies found suggestions of European and Amerindian admixture in Easter Island (3). Amerindian contribution is believed to be due to five crew members of a slave ship coming from Peru, which reached Easter Island before the island population crashed on the 19th century (4). On the other hand, European contribution in Easter Island occurred in 1888 after the annex of the island to the Republic of Chile (3). As previously mentioned, Easter Island population collapsed undergoing a severe decrease in number of inhabitants (1). The 19th century collapse arose after the introduction of epidemics, such as: dysentery, tuberculosis, and smallpox (1, 4). Rapanui population initial size before collapsed was approximately of 2,000 inhabitants, after the Easter Island population crashed, only 111 residents remained (4). Providing that Peruvians from the slave ship possessed better resistance to epidemics, Amerindian genetic contribution was amplified in following generations (1, 4).

1.2 Significance

Variety of theories about the biological origins of the native inhabitants of Easter Island, as well as, uncertainty regarding historic admixture with European and South American populations make the genetic history of Easter Island somewhat obscure (6, 7). Previously mentioned speculations limits the use of Easter Island population genetic data for research purposes. Mitochondrial DNA, HLA complex, and Y-chromosome genetic studies of the Rapanui population corroborate the idea of a Polynesian origin (8). These same studies, imply

that an interaction of Polynesians and Native Americans occurred previous to any contact with Europeans (8).

In this study 15 autosomal Short Tandem Repeats (STRs), or microsatellites, were analyzed. STRs are short sequences of DNA, 2-5 base pairs long, which due to the different repeats that vary among individuals make them effective for human identification and paternity testing purposes. Microsatellites are widely used genetic markers, employed for the purpose of population characterization to individual identification (9). Autosomal STRs contribute to a better understanding of the Easter Island population structure and its gene diversity because of inherent hypervariability of these loci (9). In addition, STRs provide an insight of the history related to past migrations, affinity, and association between genotypes of Mainland Chile, Polynesia and other cosmopolitan populations (9).

In order to study local population structure and recent demographic history the markers chosen were Y-chromosome STRs. Y-STRs are able to determine male genealogies and are generally employed to construct extended multilocus haplotypes (4). Because the Rapanui population has not been deeply studied and open questions still remain in regards to its history, affinity, and association between genotypes of other populations further study is required (9). Autosomal STRs and Y-STRs were chosen for their capability in providing a large understanding of gene diversity and genetic affinity (ancestry relationship) among populations. Moreover, comparison of Easter Island with other populations aided in determining if autosomal and Y-chromosome STR loci used in cosmopolitan populations are enough to use as a forensic tool for the Easter Island population.

Furthermore, because the study focus is on the genetic analysis at an individual level, the minimum degree of substructure present in the Easter Island population could be determined.

This later subject is important because it has become a major issue for research performed on complex genetic disorders (10). Consequently, knowledge about the existence of population substructure in Rapanui population can be used in future disease gene association studies considering the existence of some biomedical phenotypes that are prevalent in the Easter Island population. According to Lagos et al. (2011) children from Rapanui are at higher risk of experiencing attention- deficit/hyperactivity disorder (ADHD) in comparison to Chilean resident children from the Arica-Parinacota Region (11). Children suffering ADHD undergo hyperactivity, impulsivity, and periods of inattention (12). Despite the fact that genome wide association studies are based on single nucleotide polymorphisms (SNPs), it is possible to account for the minimum amount of substructure present in the Easter Island population, because STRs mutate faster than SNPs (13). Hence, information generated will be potentially helpful in future disease gene association studies in order for researchers to establish the minimum substructure present in the population of Easter Island.

1.3 Aims and hypothesis

Aim 1: To examine the extent of gene diversity in the Easter Island (Rapanui) population with data on 15 autosomal Short Tandem Repeats (STRs) typed with the commercial STR kits Identifiler® Plus and 23 Y-chromosome STRs typed using Y-filer (17 loci) and Y- PLEX™ 6 (6 loci). Hypothesis 1: Because of the evolutionary isolation of the Easter Island population both autosomal and Y-chromosome gene pool diversity in this population is expected to be reduced in comparison with those of the cosmopolitan populations.

Aim 2: To investigate the genetic affinity of the Easter Island population in relation to other historically related populations, such as Mainland Chile, Polynesia and other populations.

Hypothesis 2: Easter Island population consists of a gene pool mainly made up of Polynesian populations combined with some other cosmopolitan populations as a result of migration.

Aim 3: To investigate forensic utility of autosomal and Y-chromosome Short Tandem Repeats.

Hypothesis 3: In spite of reduced genetic diversity, Identifiler® Plus autosomal STRs, Y-filer® and Y- PLEX™ 6 Y-STR loci have considerable utility for human identification and paternity testing in the Easter Island population.

CHAPTER II

MATERIALS AND METHODS

2.1 Population samples

Samples were collected by Dr. Carlos Campusano from the University of Valparaiso, Chile with an IRB approval from the same university and the University of North Texas Health Science Center. Samples were collected as blood spots on FTA cards and were sent to UNTHSC without personal identification information. Originally FTA cards were available for 52 males and 75 females.

2.2 DNA Analysis

DNA extraction, amplification, genotyping and haplotyping were performed at the UNTHSC Research and Development Laboratory of the Institute of Applied Genetics using partial clippings of these anonymized blood-containing FTA cards. Originally FTA cards were available for 52 males and 75 females, however, autosomal genotype data were obtained for only a total of 122 individuals of the Easter Island population, of which 48 were males and 74 were females. Individuals were typed for 15 autosomal Short Tandem Repeats (STRs) employing the commercial STR kit Identifiler® Plus (Applied Biosytems, Foster City, CA, USA). Specifically, the loci analyzed were: CSF1PO, D13S317, D16S539, D18S51, D19S433, D21S11, D2S1338, D3S1358, D5S818, D7S820, D8S1179, FGA, TH01, TPOX, vWA and the gender determining

locus amelogenin. In addition, Y-STR haplotype data were obtained for the original total of 52 Rapanui males and typed for 23 Y-STRs utilizing Y-filer (17 loci) (Applied Biosytems, Foster City, CA, USA) and Y- PLEX™ 6 (ReliaGene Technologies, New Orleans, LA, USA) commercial kits. Y-STR haplotype data obtained encompassed the loci: DYS389I, DYS389II, DYS390, DYS456, DYS19, DYS385a, DYS385b, DYS458, DYS437, DYS438, DYS448, GATA_H4, DYS391, DYS392, DYS393, DYS439, DYS635, DYS481, DYS576, DYS549, and DYS57.

2.3 Statistical Analyses

Autosomal and Y-STRs allele frequencies were computed by simple gene counting method. Observed and expected genotype frequencies, as well as each autosomal locus allele and genotype counts were calculated using a NIST Excel-based tool named STR Genotype (14). Next, autosomal loci were tested for Hardy-Weinberg equilibrium with Fisher's exact test with 3200 shuffling's using GDA version 1.1 Software. Hardy-Weinber equilibrium test allows correlation of allele frequencies with genotype frequencies (15). Pairs of loci were tested for linkage disequilibrium where genotypes were preserved to avoid any within-locus disequilibrium take an effect on the results. Linkage disequilibrium test enables the multiplication of genotype frequencies across all tested loci (15). PowerStatsV1.2 Excel spreadsheet was employed in order to calculate several parameters of population genetics interest including Power of Discrimination (*PD*), Polymorphism Information Content (*PIC*), typical Paternity Index (*PI*), Power of Exclusion (*PE*) Match Probability (*MP*), observed heterozygosity (H_{obs}) and expected heterozygosity (H_{exp}) (16). GDA also was employed to calculate the inbreeding coefficient (F_{IS}) of individuals per locus.

Y-STRs haplotype frequencies were calculated using Arlequin (17). Also, Arlequin Software was used to compare haplotypic content within the Easter Island population samples to search for shared haplotypes (17). Haplogroups were predicted employing the software tool Haplogroup Predictor provided by Whit Athey (18, 19). A median-joining network using Y-STRs was constructed by Network v 4.6 software (20). Network analysis by median joining (MJ) was utilized to construct haplogroup networks from recombination-free population data; MJ combines features of Kruskal's algorithm for finding minimum spanning trees by favoring short connections, and Farris's maximum-parsimony (MP) heuristic algorithm, which sequentially adds new vertices called "median vectors", except that our MJ method does not resolve ties (19).

Moreover, the STRUCTURE software was used to estimate individual-level gene admixture, and to identify overall genetic affinity of 48 male individuals from Easter Island (21). Next, a comparison of STRUCTURE software results of autosomal and Y-STRs was performed in order to obtain information about patrilineal versus bi-parental ancestry of Easter Island individuals. Subsequently, comparisons based on autosomal allele frequency distribution were performed from the Easter Island (RPN) population and other populations, such as Chile (CHI), Samoa (SAM), Tonga (TON), Croatia (CRO), Spain (ESP), Angola (ANG), and Equatorial Guinea (EGU). These populations were chosen based on historic events that related them to Rapanui. The ancestry coefficient F_{ST} and Slatkin's genetic distances R_{ST} (or Φ_{ST}) were estimated from autosomal allele counts using the Arlequin software.

Furthermore, to determine phylogenetic relationships the MEGA 5.2 software was used to draw phylogenetic trees from F_{ST} and R_{ST} genetic distances employing the neighbor-joining method (22). Finally, descriptive statistics of forensic relevance (such as Probability of Discrimination, Match Probability, and Probability of Exclusion) from Easter Island, European,

South American, and African populations were compared in order to ensure the utility of autosomal STR loci for human identification and paternity testing (23-28).

CHAPTER III

RESULTS

3.1 Allele frequencies, and deviation from Hardy-Weinberg and Linkage Equilibrium at the autosomal STR loci in Easter Island

Allele frequencies of 122 individuals of the Rapanui populations were calculated and are displayed in Table A1. Loci were tested for Hardy-Weinberg equilibrium and it was found that four loci (D3S1358, D5S818, TPOX, and vWA**)** presented detectable deviation from a significance probability expectation of $\alpha = 0.05$, but only the D5S818 locus continued to be significant after applying a Bonferroni correction of multiple testing ($\alpha' = 0.0034 = 0.05/ 15$). A linkage disequilibrium test was performed assuming Hardy-Weinberg Equilibrium, where thirty six pairs of loci exhibiting significant disequilibrium at $p=0.05$ level. Only two of these remained significant after a Bonferroni correction of multiple testing ($\alpha' = 0.00048 = 0.05/ 105$). Loci presenting deviations are shown in Table 1, along with their respective *p*-values. Using 15 autosomal STRs, several statistical parameters of the Rapanui population were calculated. Due to missing data, the total number of individuals for each locus range between 121 and 122. Number of alleles observed at each locus and descriptive statistics are summarized in Table 2. When comparing heterozygosity proportions across loci, the locus presenting highest heterozygosity was vWA (H_{obs}=0.861). Loci presenting a lower heterozygosity than expected were: TPOX (H_{obs}=0.607), D3S1358 (H_{obs}=0.689), and D5S818 (H_{obs}=0.762). As well, the inbreeding coefficient (F_{IS}) present in the Rapanui population was calculated resulting in average F_{IS} of 0.008. In other words, even if the significant deviations of genotype frequencies from their

expectations of Hardy-Weinberg equilibrium are to be considered as real the effect of isolation and genetic drift would be adjustable in this population with the substructure parameter estimates as 0.008.

3.2 Reduced genetic diversity of autosomal STR loci in Easter Island

Reduction of genetic diversity was seen when comparing Easter Island to seven cosmopolitan populations based on number of alleles observed at each locus and observed heterozygosity. Table 3a presents the comparisons of the populations locus by locus. Due to the substantially larger number of individuals sampled in the Chilean population, a sample size adjustment was required in order to properly compare its genetic diversity with the Easter Island population. As explained by Chakraborty et. al (1988), the expected number of alleles in a smaller sample size can be calculated if allele frequencies in the population are known from a larger sample (29). Expected number of alleles for a sample size of 122 individuals from the Chilean population is summarized on Table 3b. Adjustment to observed heterozygosity (H_{obs}) was not required because, as opposed to number of alleles (k), it is not sensitive of sample size (29). Taken together, data shown in Table 3a and 3b suggest a modest degree of reduced genetic diversity in Easter Island compared with the chosen cosmopolitan populations of this study. On an average, Easter Islanders exhibited 1 to 2% reduction in average heterozygosity at these 15 autosomal STR loci; the reduction in the observed number of alleles also was not so pronounced (1 to 2 alleles per locus).

3.3 Genetic affinity of Rapanui with other populations studied with the autosomal STR loci

The estimated F_{ST} from autosomal STR loci obtained from the comparison of Rapanui with the seven populations showed a close relationship of Easter Island to Chile and Samoa, although Tonga and Croatia populations were also not far distant from the Easter Island population. While in these calculations the repeat size variability is not accounted for, genetic distances based on allele size, the R_{ST} calculations, also reached the same conclusion, establishing a close relationship of Easter Islanders with Chileans and Samoans. A summary of the genetic distances obtained employing both of these distance coefficients is presented in Table 4. In addition, employing Wright's and Slatkin's genetic distances, neighbor-joining trees were constructed as shown in Figure 1. A neighbor-joining tree utilizing F_{ST} values (Figure 1a) indicates that Samoa and Tonga share common ancestry, and at the same time, share the closest ancestry with Rapanui. These three populations, formed a clade with Angola and Equatorial Guinea. On the other hand, Chile, in spite of being genetically close to Easter Island, formed a clade with the two European populations, Croatia and Spain, before clustering with the clade of Easter Island, Tonga, Samoa, and the two African populations of Angola and Equatorial Guinea. Overall, the phylogenetic tree based on the R_{ST} values (Figure 1b) produced the same results except for the fact that Croatia and Chile appeared to be more closely related than Spain. Table 5 presents the comparisons of the descriptive statistics relevant for DNA forensics of 15 STR loci from Easter Island and other cosmopolitan populations. At a locus-by-locus level, it was found that despite the reduced genetic diversity that is present in the Rapanui population, autosomal STRs are almost as useful in Rapanui for paternity testing, and human identification purposes, as is the case for the other cosmopolitan populations examined here.

Table 1. Loci and pairs of loci showing deviations from Hardy-Weinberg Equilibrium (*HWE*) and Linkage Equilibrium (*LE*) with *p*-values.

Locus	Deviation from *HWE*
D3S1358	0.0150
D5S818	**0.0025**
TPOX	0.0328
vWA	0.0263

Significant *p*-values after a Bonferroni correction (α'=0.0034) are shown in gray.

Loci	Deviation from *LE*
CSF1PO/D13S317	0.0294
CSF1PO/D19S433	0.0356
CSF1PO/**D5S818**	0.0197
CSF1PO/D7S820	0.0428
CSF1PO/D8S1179	0.0022
CSF1PO/**TPOX**	0.0159
CSF1PO/**vWA**	0.0159
D13S317/D21S11	0.0184
D13S317/**D5S818**	0.0009
D13S317/D8S1179	0.0319
D13S317/FGA	0.0313
D13S317/**TPOX**	0.0228
D16S539/D18S51	0.0081
D16S539/**D5S818**	0.0156
D16S539/D8S1179	0.0172
D18S51/D19S433	**$<10^{-4}$**
D18S51/D21S11	0.0078
D18S51/D2S1338	0.0228
D18S51/**D3S1358**	0.0253
D18S51/**D5S818**	0.0022
D18S51/D8S1179	0.0059
D18S51/**vWA**	0.0163
D19S433/D2S1338	0.0106
D19S433/D8S1179	0.0022
D19S433/FGA	0.0163
D21S11/**D5S818**	0.0009
D2S1338/**D3S1358**	0.0353
D2S1338/**D5S818**	0.0481
D2S1338/**TPOX**	0.0338
D3S1358/D5S818	0.0216
D5S818/D7S820	0.0244
D5S818/D8S1179	**$<10^{-4}$**
D5S818/TPOX	0.0063
D5S818/vWA	0.0441
D8S1179/FGA	0.0388
D8S1179/**TPOX**	0.0094

Significant *p*-values after a Bonferroni correction (α'=0.00048) are shown in gray. Loci presenting deviations from *HWE* are shown in bold.

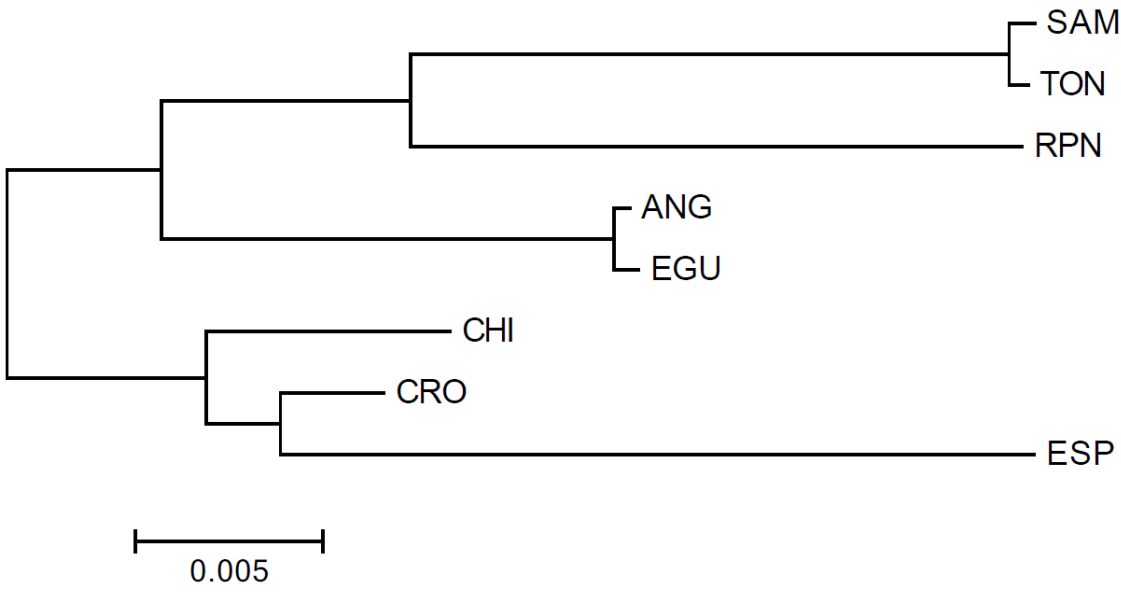
Table 2. Statistical parameters of 15 autosomal STR loci in the Easter Island population.

	# of individuals	**# of alleles observed**	H_{obs}	H_{exp}	***PIC***	***PD***	***MP***	***PE***	***PI***	F_{IS}
CSF1PO	*n*=121	*k*=6	0.653	0.650	0.583	0.797	0.203	0.359	1.44	-0.005
D13S317	*n*=121	*k*=7	0.820	0.791	0.762	0.907	0.093	0.636	2.77	-0.036
D16S539	*n*=122	*k*=6	0.721	0.726	0.684	0.874	0.126	0.462	1.79	0.006
D18S51	*n*=122	*k*=12	0.787	0.836	0.790	0.952	0.048	0.575	2.35	0.059
D19S433	*n*=122	*k*=9	0.828	0.815	0.790	0.930	0.070	0.652	2.90	-0.016
D21S11	*n*=122	*k*=11	0.738	0.780	0.750	0.918	0.082	0.489	1.91	0.054
D2S1338	*n*=122	*k*=10	0.844	0.865	0.851	0.961	0.039	0.684	3.21	0.023
D3S1358	*n*=122	*k*=5	**0.689**	**0.734**	0.688	0.874	0.126	0.411	1.61	0.062
D5S818	*n*=122	*k*=7	**0.762**	**0.806**	0.778	0.924	0.076	0.531	2.10	0.054
D7S820	*n*=121	*k*=7	0.703	0.685	0.633	0.847	0.153	0.432	1.68	-0.026
D8S1179	*n*=122	*k*=8	0.721	0.726	0.693	0.878	0.122	0.462	1.79	0.007
FGA	*n*=122	*k*=10	0.836	0.819	0.797	0.938	0.062	0.668	3.05	-0.021
TH01	*n*=122	*k*=6	0.787	0.731	0.688	0.877	0.123	0.575	2.35	-0.076
TPOX	*n*=122	*k*=5	**0.607**	**0.808**	0.780	0.818	0.182	0.299	1.27	0.249
vWA	*n*=122	*k*=7	**0.861**	**0.808**	0.780	0.916	0.084	0.716	3.59	-0.066
MEAN ± SD	*n*=121.8±0.41	*k*=7.73±2.19	0.76± 0.08	0.77± 0.06						0.008

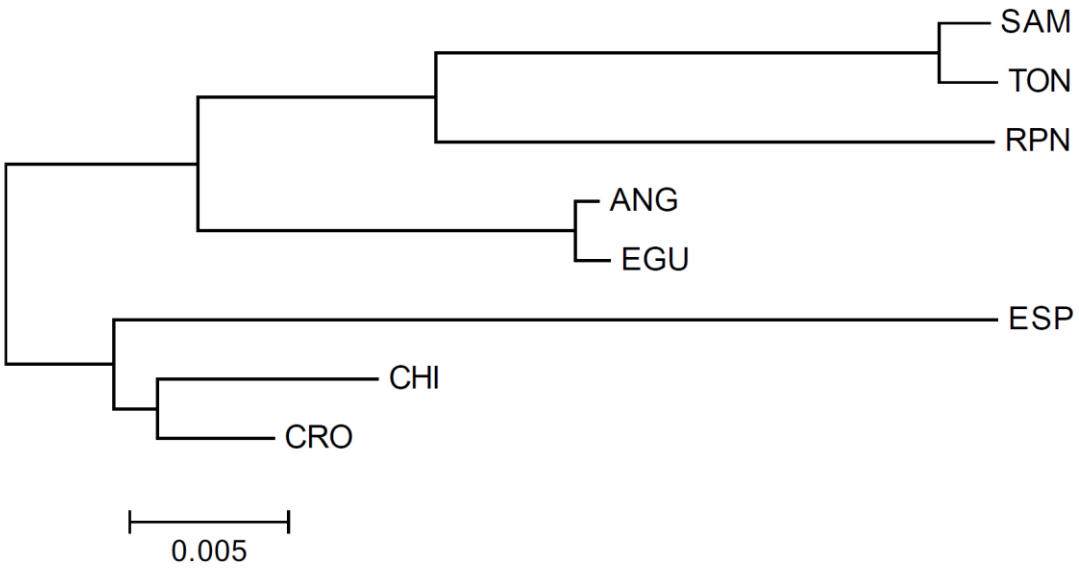
H_{obs}*:* Observed heterozygosity; H_{exp}: Expected heterozygosity; *PIC:* Polymorphic Information Content; *PD:* Power of Discrimination; *MP*: Match Probability; *PE:* Power of Exclusion; *PI:* Paternity Index; F_{IS}*:* Inbreeding coefficient

Figure 1. Neighbor-joining trees showing genetic affinities of Easter Island and other cosmopolitan populations using genetic distances based on autosomal STRs.

a) Wright's F_{ST}.



b) Slatkin's R_{ST}.



3.4 Y-STR allele frequencies in haplogroup network in Easter Island

Y-STRs allele frequencies of 52 male individuals also were computed and are shown in Table A2. Subsequently, when comparing the haplotypic content of male Easter Islanders thirty seven haplotypes were found, where only six haplotypes were shared. The composition of the different haplotypes, as long with its frequencies, are shown in Table A3. Prediction of haplogroups yielded a result of five different groups: E1b1a, Q, R1b, Q2a, and J2a4b present in Easter Island. Figure 2a displays a pie chart representing the haplogroup distribution. The E1b1a haplogroup (generally believed to be of African male ancestry) is found in 38% males and the Q haplogroup (considered to be of Native American male ancestry) has a frequency of 35% in Easter Island males. In other words, among the males of Easter Island, the most prevalent Y-STR haplogroups were either of African or Native American ancestry. Construction of a median-joining network (Figure 2b) using Y-STRs, confirmed that the haplogroup of African origin E1b1a, possesses the greatest number of shared haplotypes among the males of Easter Island.

3.5 STRUCTURE analyses of Easter Island males based on autosomal and Y-STR data

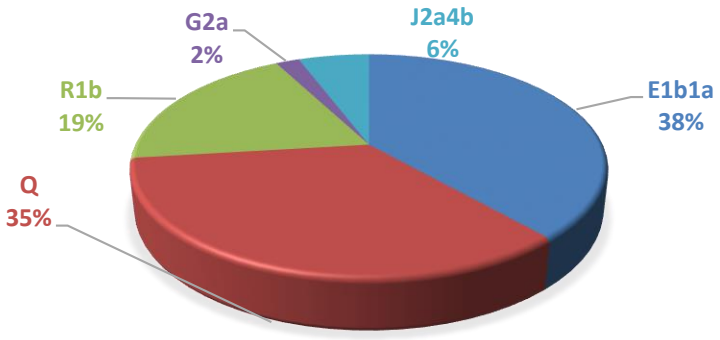
Genetic structure patterns obtained from autosomal and Y-STRs indicated that male individuals from the Rapanui population are admixed from three different populations. In order to estimate the number of populations present (*K*), three replicates for *K*=1, *K*=2, *K*=3, *K*=4, and *K*=5 were performed. A posterior probability was obtained for every run, this probability stabilizes when *K* values increase (30). Therefore, it can be concluded that when numerous *K* values have similar posterior probabilities, the lowest *K* value is the most suitable for the data (30).

As it can be seen in Figure 3a, a bar chart illustrates the various degrees of admixture present in individuals, as reflected with the autosomal STRs. Figure 4a, serves the purpose of a better visualization, and proximity to any triangle vertex relates individuals to a high membership value for a specific population. Obviously, representation of the 48 males of Easter Island within the triangle (equivalent of the assumption of three ancestral population) signified that these individuals do not correspond entirely to a single population with reference to their bi-parental ancestry. In other words, at the autosomal level, the gene pool of Easter Island males appear to have ancestral contributions of three populations (that may be inferred to be of Native American, African, and Polynesian ancestry)

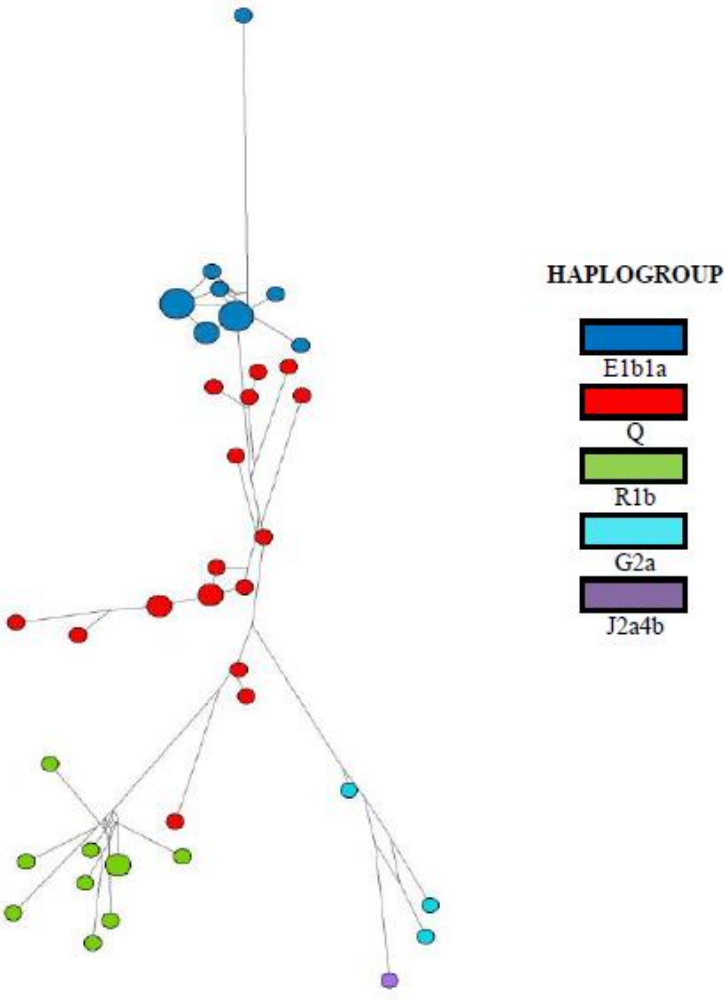
On the other hand, male lineage STRs showed an individual-level genetic structure composed of mainly a single population. In Figure 3b shows that individuals are primarily members of a single population. A triangle representation of the Easter Island population based on Y-STRs (Figure 4b), shows that individuals belong entirely to a single population. Since concentration of the data points near all three vertices of the triangle was observed, at the population level, genetic origin of Easter Island males can again be traced to the same three ancestral gene pools, though at individual level their Y-chromosomes appeared to be of a single population ancestry.

Figure 2. Y-STR haplogroups present in the population of Easter Island

a) Percentage distribution of haplogroups in the Easter Island population (*n*=52).



b) Network analysis of 37 different haplotypes in the Easter Island male population (*n*=52).



Nodes are proportional to haplotype frequency. E1b1a: Africa; Q: Native American; R1b: Worldwide; G2a: Middle Eastern; J2a4b: East Europe.

Table 3.

a) Genetic diversity of Easter Island compared with seven cosmopolitan populations.

Pop		CSF1PO	D13S317	D16S539	D18S51	D19S433	D21S11	D2S1338	D3S1358	D5S818	D7S820	D8S1179	FGA	TH01	TPOX	vWA	MEAN ± SD
RPN	***n***	121	121	122	122	122	122	122	122	122	121	122	122	122	122	122	---
	H_{obs}	0.653	0.820	0.721	0.787	0.828	0.738	0.844	0.689	0.762	0.703	0.721	0.836	0.787	0.607	0.861	0.76±0.08
	k	6	7	6	12	9	11	10	5	7	7	8	10	6	5	7	7.73±2.19
CHI	***n***	986	986	986	986	986	986	986	986	986	986	986	986	986	986	986	---
	H_{obs}	0.741	0.859	0.783	0.879	0.802	0.838	0.851	0.743	0.707	0.777	0.804	0.872	0.754	0.668	0.765	0.79±0.06
	k	12	8	8	19	17	20	13	10	8	10	10	17	6	10	10	11.87±4.37
SAM	***n***	95	95	95	95	95	95	95	95	95	95	95	95	95	95	95	---
	H_{obs}	0.737	0.758	0.916	0.821	0.747	0.768	0.821	0.684	0.747	0.853	0.800	0.821	0.684	0.611	0.863	0.78±0.08
	k	5	9	6	12	8	10	10	6	6	8	9	11	6	4	7	7.80±2.34
TON	***n***	51	51	51	51	51	51	51	51	51	51	51	51	51	51	51	---
	H_{obs}	0.745	0.784	0.804	0.843	0.902	0.745	0.902	0.686	0.824	0.784	0.765	0.804	0.745	0.608	0.765	0.78±0.08
	k	6	7	7	13	8	7	10	6	6	8	11	10	7	4	6	7.73±2.34
CRO	***n***	195	195	195	195	195	195	195	195	195	195	195	195	195	195	195	---
	H_{obs}	0.723	0.774	0.800	0.897	0.805	0.841	0.821	0.708	0.677	0.826	0.739	0.872	0.805	0.687	0.805	0.79±0.07
	k	8	7	7	13	13	11	12	7	7	8	9	15	6	6	9	9.20±2.88
ESP	***n***	114	114	114	114	114	114	114	114	114	114	114	114	114	114	114	---
	H_{obs}	0.763	0.789	0.746	0.886	0.702	0.851	0.807	0.798	0.684	0.789	0.833	0.825	0.737	0.605	0.825	0.78±0.07
	k	7	8	8	12	10	12	11	7	7	8	9	11	6	6	7	8.60±2.10
ANG	***n***	110	110	110	110	110	110	110	110	110	110	110	110	110	110	110	---
	H_{obs}	0.755	0.618	0.818	0.855	0.891	0.864	0.864	0.682	0.682	0.809	0.782	0.836	0.727	0.8364	0.8364	0.79±0.08
	k	9	9	9	15	13	17	12	5	8	8	6	15	6	7	10	9.93±3.67
EGU	***n***	134	134	134	134	134	134	134	134	134	134	134	134	134	134	134	---
	H_{obs}	0.761	0.612	0.776	0.873	0.858	0.836	0.895	0.761	0.798	0.724	0.679	0.918	0.709	0.791	0.724	0.78±0.08
	k	9	8	8	13	14	16	12	8	7	7	10	17	6	8	10	10.20±3.43

H_{obs}*:* Observed heterozygosity; *n:* Number of individuals; *k:* Number of alleles observed; RPN: Rapanui; CHI: Chile; SAM: Samoa; TON: Tonga; CRO: Croatia; ESP: Spain; ANG: Angola; EGU: Equatorial Guinea.

b) Expected number of alleles in Chile population when sample size is reduced to 122 individuals.

Pop		CSF1PO	D13S317	D16S539	D18S51	D19S433	D21S11	D2S1338	D3S1358	D5S818	D7S820	D8S1179	FGA	TH01	TPOX	vWA	MEAN ± SD
RPN	*n*	121	121	122	122	122	122	122	122	122	121	122	122	122	122	122	---
	k	6.0	7.0	6.0	12.0	9.0	11.0	10.0	5.0	7.0	7.0	8.0	10.0	6.0	5.0	7.0	7.73±2.19
CHI	*n*	122	122	122	122	122	122	122	122	122	122	122	122	122	122	122	---
	k	6.6	7.4	7.2	12.8	10.2	11.3	11.1	6.9	7.2	7.5	8.9	5.7	5.8	5.7	6.9	8.09±2.25

Table 4. Genetic distance of Easter Island with other cosmopolitan populations.

	RPN	CHI	SAM	TON	CRO	ESP	ANG	EGU
RPN	---	0.0321	0.0318	0.0386	0.0358	0.0675	0.0448	0.0501
CHI	0.0307	---	0.0428	0.0417	0.0106	0.0373	0.0342	0.0340
SAM	0.0303	0.0403	---	0.0034	0.0440	0.0691	0.0287	0.0293
TON	0.0355	0.0389	0.0012	---	0.0430	0.0682	0.0270	0.0286
CRO	0.0341	0.0104	0.0416	0.0416	---	0.0318	0.0307	0.0283
ESP	0.0573	0.0295	0.0600	0.0574	0.0229	---	0.0455	0.0435
ANG	0.0419	0.0326	0.0277	0.0261	0.0290	0.0401	---	0.0018
EGU	0.0464	0.0324	0.0282	0.0275	0.0269	0.0382	0.0011	---

R_{ST}

F_{ST}

Genetic distances: Wright's F_{ST} bottom left gray boxes (white font) and Slatkin's R_{ST} upper right gray boxes (black font).

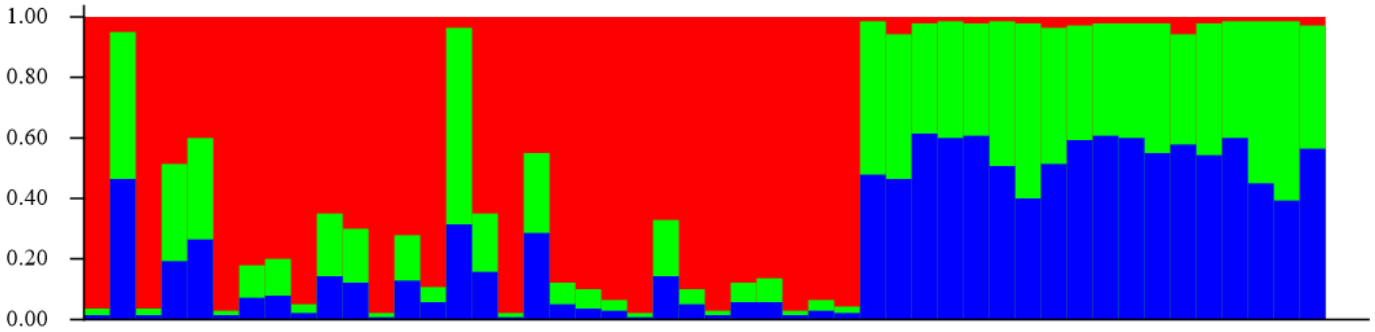
Table 5. Comparison of statistical parameters from 15 autosomal STR loci in Easter Island with 7 cosmopolitan populations.

Pop		CSF1PO	D13S317	D16S539	D18S51	D19S433	D21S11	D2S1338	D3S1358	D5S818	D7S820	D8S1179	FGA	TH01	TPOX	vWA
RPN	***PD***	0.810	0.927	0.883	0.954	0.941	0.922	0.968	0.884	0.934	0.849	0.892	0.945	0.885	0.936	0.936
	PE	0.359	0.636	0.462	0.575	0.652	0.489	0.684	0.411	0.531	0.432	0.462	0.668	0.575	0.299	0.716
CHI	***PD***	0.875	0.950	0.924	0.972	0.936	0.952	0.965	0.896	0.876	0.907	0.934	0.971	0.913	0.832	0.909
	PE	0.311	0.501	0.408	0.599	0.449	0.505	0.562	0.345	0.308	0.372	0.440	0.594	0.376	0.250	0.377
SAM	***PD***	0.832	0.929	0.926	0.937	0.930	0.933	0.945	0.855	0.900	0.946	0.927	0.889	0.883	0.840	0.913
	PE	0.425	0.599	0.636	0.694	0.629	0.629	0.679	0.440	0.544	0.678	0.606	0.867	0.506	0.402	0.589
TON	***PD***	0.837	0.905	0.917	0.941	0.927	0.913	0.926	0.866	0.890	0.929	0.947	0.931	0.913	0.809	0.877
	PE	0.501	0.570	0.606	0.681	0.799	0.501	0.799	0.407	0.643	0.570	0.535	0.606	0.501	0.300	0.535
CRO	***PD***	0.862	0.919	0.913	0.964	0.935	0.954	0.967	0.930	0.855	0.924	0.927	0.963	0.915	0.782	0.942
	PE	0.465	0.552	0.599	0.790	0.609	0.677	0.638	0.457	0.393	0.647	0.490	0.738	0.599	0.416	0.599
ESP	***PD***	0.870	0.925	0.906	0.960	0.896	0.951	0.961	0.933	0.856	0.921	0.935	0.951	0.925	0.820	0.910
	PE	0.533	0.580	0.502	0.767	0.431	0.697	0.612	0.596	0.404	0.580	0.662	0.645	0.488	0.297	0.645
ANG	***PD***	0.918	0.858	0.909	0.967	0.970	0.965	0.977	0.885	0.897	0.923	0.909	0.975	0.869	0.918	0.954
	PE	0.555	0.436	0.529	0.713	0.725	0.705	0.763	0.477	0.503	0.565	0.528	0.753	0.452	0.552	0.659
EGU	***PD***	0.925	0.860	0.932	0.952	0.953	0.952	0.968	0.892	0.976	0.914	0.900	0.968	0.870	0.912	0.938
	PE	0.512	0.305	0.555	0.741	0.711	0.667	0.786	0.529	0.741	0.466	0.397	0.832	0.442	0.583	0.466

PD: Power of Discrimination; *PE:* Power of Exclusion.

Figure 3. STRUCTURE bar charts showing patterns of genetic structure in the Easter Island population.

a) Genetic structure of 48 male individuals using 15 autosomal STRs (*K*=3)



b) Genetic structure of 48 individuals using 23 Y-STRs (*K*=3).

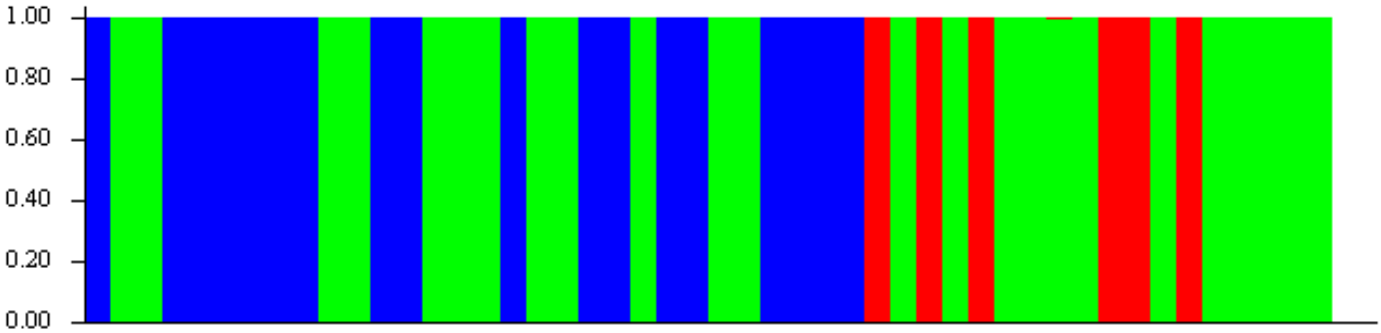
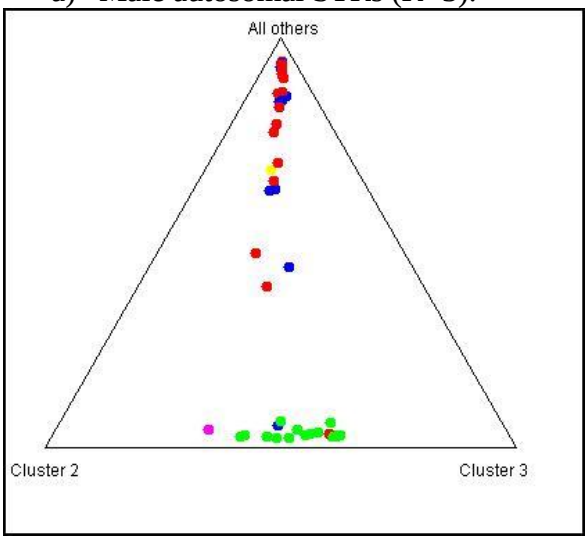
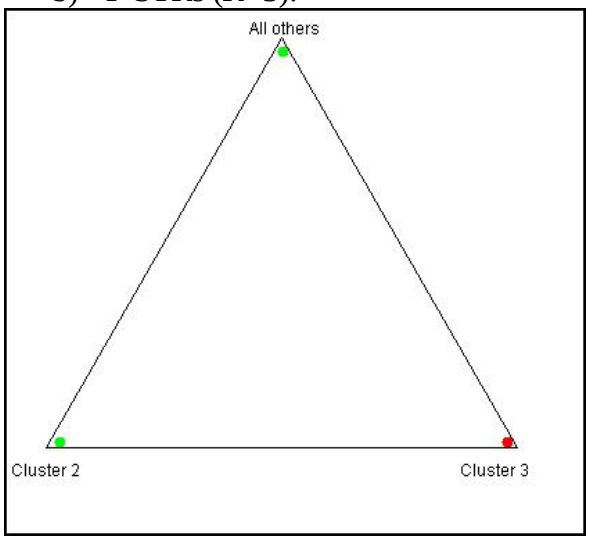


Figure 4. STRUCTURE triangle representing 48 male individuals.

a) Male autosomal STRs (*K*=3).



b) Y-STRs (*K*=3).



CHAPTER IV

DISCUSSION

Joint analyses of autosomal and Y-STRs provide a deeper understanding of patterns of human genetic diversity and population structure of the Easter Island population. However, in order to properly interpret these results of genetic structure of Rapanui it is important to take into account its demographic history.

It is believed that Easter Island population underwent a collapse, this occurred as a consequence of epidemics (31). Population bottlenecks occur when a population's size is decreased for one generation or more (32). Genetic drift acts faster in reducing genetic variation within smaller populations (32). As a consequence, reduced genetic variation can be seen within Easter Island individuals. The Hardy-Weinberg assumptions appeared to be violated at several loci in the Rapanui population (Table 1), which can be ascribed to possible bottleneck effect of the past population collapse in Easter Island or substructure present in the Rapanui population. The presence of linkage disequilibrium between several pairs of loci (Table 1) could also be an effect of genetic drift due to the past population collapse and persistent small population size present in the island. The same factors would also reflect a reduced level of genetic diversity in the Easter Island population. Examined by the two most commonly used measure of genetic diversity (Heterozygosity and observed number of segregating alleles), this study demonstrated a somewhat reduced level of genetic diversity among the Easter Islanders (Table 3). On an average, Easter Islanders have 1 to 2 alleles less per locus, and a 1 to 2% reduced heterozygosity

per locus, in comparison to the cosmopolitan populations studied here. Reduction of genetic diversity is probably lesser than that observed in other Native American populations of continental USA (33). This is likely to be due to the fact that the loci studied here are the ones generally used in DNA forensics, chosen for their hypervariability. It is well-known that this hypervariability is due to the high mutation rate inherent to these STR loci, which facilitates recovery from bottleneck effect and consequently has a decreased impact of population substructure (33). The estimates of F_{IS} (presented in Table 2) exhibits this effect; namely, average F_{IS} value for these 15 loci for Easter Island is 0.008, much smaller than the one anticipated for isolated populations at such loci (34).

Studies of Rapanui Y-STRs have suggested the presence of predominantly African and Amerindian admixture in this population inferred from high prevalence of haplogroups E1R1a, and Q, respectively). Haplogroups frequency distribution (Figure 2a) and haplotype network resulst (Figure 2b) also demonstrates evidence of European admixture. These results can be explained by an expansion out from Africa into the Pacific that occurred around 3,500 AD (35). In this migration the people went east to Samoa and Tonga, who's inhabitants later reached Easter Island. Rapanui autosomal STRs were found to be admixed of three different populations, even at individual level (Figures 3a and 4a), while their Y-STR structure, was predominantly constituted of a single population ancestry (Figure 3b and 4b). Y-STR results point toward that male lineages are probably a consequence of conquerors who reproduced with Rapanui women.

Selection of seven cosmopolitan populations for comparisons with Easter Island, were based on its history with the island. Tonga and Samoa, were chosen because these islands populations were previously colonized by Polynesians, which would represent the initial settlers of the island. Chile was selected because it is the most affine population from the political view-

point, and would be a good surrogate of Native American gene pool that may explain the genetic origin of Easter Islanders. Angola and Equatorial Guinea were selected because they would represent contribution of gene pools from the African continent, also due to migration events that occurred between these countries and South America. It has been documented that individuals from Europe have migrated to Chile. For this reason, Spain and Croatia were selected for comparison with Easter Islanders. At the end, the relationship of Rapanui with Samoa and Tonga populations was clear (Figure 1a and 1b), suggesting a colonization from Polynesians rather than South Americans.

The utilization of autosomal allele frequencies based on 15 markers from these previously mentioned cosmopolitan populations, helped validate the usefulness of these markers under paternity and forensic testing. Regardless of the decrease of genetic diversity present in Easter Island, application of these loci for human identification and paternity testing purposes are still expected to be efficient for this population (Table 5). Lastly, an important outcome was the computation of the inbreeding coefficient (F_{IS}) present in the Rapanui population. In this regard, the observation of a low F_{IS} value (average value of 0.008, mentioned earlier, see Table 2) is reassuring not only for DNA forensic work in Easter Island, but also for probable research of disease-gene association studies in Easter Island. This is so, because the estimate reflects a minimum indicator of population substructure effect in Easter Island that would give guidance of design of disease-gene association studies of complex disorders in Ester Island.

CHAPTER IV

CONCLUSION

Rapanui, the most isolated island in the world, remains one of the most intriguing historic places on earth. Evidence of moderate levels of reduced genetic diversity has been identified due to small population size, genetic drift and founder effects. Since the study was based only on autosomal and Y-chromosome STRs; results are limited in several respects. First, generalizations of reduction of genetic diversity may not be extendable to other loci (such as SNPs and Alu markers) which have much lower mutation rates. Second, absence of markers of maternal lineage (X-chromosome markers and mtDNA markers) did not allow an indication of female gene flow in Easter Island. Next, individuals were presumably unrelated but the possibility of the presence of relatives in the sample cannot be excluded, and if it was the case, reduced diversity could have been present due to allele sharing. Also, sample size was a limitation, particularly for Y-STR haplotypes, this was due to the fact that only 52 individuals were males. Nonetheless, significant information about Easter Island was obtained. Rapanui was found to be closely related to Samoa and Tonga, pointing to a predominantly Polynesian origin of Easter Islanders. However, the possibility of multiple recent admixtures cannot be excluded, since suggestions of contributions from African and Native American gene pools also were found in this study.

The investigation also concluded that, autosomal and Y-chromosome STRs are robust tools for human identification and paternity testing in this island population. However, further studies involving autosomal, mitochondrial, and Y-chromosome markers are needed for a deeper

understanding of demographic genetics of Easter Island and genetic structure of the contemporary population of this island.

APPENDIX

Table A1. Allele frequencies for 15 autosomal loci in Easter Island with *p*-values for Hardy-Weinberg Equilibrium test for each locus: *n* denotes the number of individuals for whom genotype data is available for the locus.

CSF1PO			**D13S317**			**D16S539**		
Allele	Count	Frequency	Allele	Count	Frequency	Allele	Count	Frequency
9	4	0.017	8	7	0.029	9	16	0.066
10	92	0.380	9	39	0.161	10	47	0.193
11	34	0.140	10	19	0.079	11	100	0.410
12	105	0.434	11	75	0.310	12	59	0.242
13	7	0.029	12	61	0.252	13	19	0.078
14	1	0.004	13	29	0.120	14	3	0.012
			14	13	0.054			
p= 0.0838 (*n*=121)			*p*= 0.0519 (*n*=121)			*p*= 0.2866 (*n*=122)		

D18S51			**D19S433**			**D21S11**		
Allele	Count	Frequency	Allele	Count	Frequency	Allele	Count	Frequency
11	2	0.008	12	9	0.037	25.2	1	0.008
11.2	0	---	13	20	0.082	27	3	0.012
12	17	0.070	13.2	3	0.012	28	7	0.029
13	12	0.049	14	57	0.230	29	74	0.303
14	34	0.139	14.2	32	0.131	30	78	0.320
15	62	0.254	15	16	0.066	30.2	14	0.057
16	17	0.070	15.2	39	0.160	31	24	0.094
17	58	0.238	16	1	0.004	31.2	22	0.090
18	17	0.070	16.2	67	0.279	32	2	0.008
19	19	0.078				32.2	15	0.061
20	2	0.008				33	0	---
21	3	0.012				33.2	4	0.016
22	0	---						
23	1	0.004						
p= 0.1275 (*n*=122)			*p*= 0.0581 (*n*=122)			*p*= 0.1194 (*n*=122)		

D2S1338

Allele	Count	Frequency
16	12	0.049
17	14	0.057
18	22	0.090
19	49	0.201
20	16	0.066
21	21	0.086
22	32	0.131
23	19	0.078
24	52	0.213
25	7	0.029

p= 0.3381 (*n*=122)

D3S1358

Allele	Count	Frequency
14	22	0.090
15	88	0.361
16	58	0.238
17	64	0.262
18	12	0.049

p= 0.0150 (*n*=122)

D5S818

Allele	Count	Frequency
7	8	0.033
8	0	---
9	5	0.020
10	40	0.164
11	45	0.184
12	67	0.275
13	50	0.205
14	29	0.119

p= 0.0025 (*n*=122)

D7S820

Allele	Count	Frequency
7	1	0.004
8	8	0.033
9	12	0.050
10	45	0.186
11	109	0.450
12	66	0.273
13	1	0.004

p= 0.8888 (*n*=121)

D8S1179

Allele	Count	Frequency
9	1	0.004
10	55	0.225
11	11	0.045
12	17	0.070
13	108	0.443
14	29	0.119
15	18	0.074
16	5	0.020

p= 0.0859 (*n*=122)

FGA

Allele	Count	Frequency
19	30	0.123
20	9	0.037
21	18	0.074
22	17	0.070
23	68	0.279
24	63	0.258
25	11	0.045
26	21	0.086
27	6	0.025
29	1	0.004098

p= 0.3441 (*n*=122)

TH01

Allele	Count	Frequency
6	49	0.201
7	95	0.389
8	23	0.094
9	14	0.057
9.3	62	0.254
10	1	0.004

p= 0.6191 (*n*=122)

TPOX

Allele	Count	Frequency
8	133	0.545
9	8	0.033
10	28	0.115
11	56	0.230
12	19	0.078

p= 0.0328 (*n*=122)

vWA

Allele	Count	Frequency
14	45	0.184
15	69	0.283
16	36	0.148
17	48	0.197
18	28	0.115
19	17	0.070
20	1	0.004

p= 0.0263 (*n*=122)

Table A2. Allele frequencies of 23 Y-STR loci in Easter Island and PD locus by locus (*n*=52)

DYS389I			**DYS389II**			**DYS390**		
Allele	Count	Frequency	Allele	Count	Frequency	Allele	Count	Frequency
12	5	0.096	28	4	0.077	20	19	0.365
13	43	0.827	29	11	0.212	21	1	0.019
14	4	0.077	30	30	0.577	22	3	0.058
			31	5	0.096	23	7	0.135
			32	2	0.038	24	18	0.346
						25	4	0.077
PD:	0.3009		*PD:*	0.6055		*PD:*	0.7192	
DYS456			**DYS19**			**DYS385a**		
Allele	Count	Frequency	Allele	Count	Frequency	Allele	Count	Frequency
14	3	0.058	13	8	0.154	10	2	0.038
15	36	0.692	14	13	0.25	11	8	0.154
16	12	0.231	15	10	0.192	12	3	0.058
18	1	0.019	16	21	0.404	13	22	0.423
						14	14	0.269
						15	2	0.038
						16	1	0.019
PD:	0.4641		*PD*:	0.7137		*PD:*	0.7184	
DYS385b			**DYS458**			**DYS437**		
Allele	Count	Frequency	Allele	Count	Frequency	Allele	Count	Frequency
12	1	0.019	15	3	0.058	14	40	0.769
13	1	0.019	16	16	0.308	15	9	0.173
14	9	0.173	17	11	0.212	16	3	0.058
15	7	0.135	18	22	0.423			
16	19	0.365						
17	6	0.115						
18	3	0.058						
19	1	0.019						
20	5	0.096						
PD:	0.7917		*PD*:	0.6779		*PD*:	0.3753	
DYS438			**DYS448**			**GATA_H4**		
Allele	Count	Frequency	Allele	Count	Frequency	Allele	Count	Frequency
9	3	0.058	17	1	0.019	10	1	0.019
10	1	0.019	18	3	0.058	11	4	0.077
11	36	0.692	19	15	0.288	12	45	0.865
12	11	0.212	20	12	0.231	13	2	0.038
13	1	0.019	21	21	0.404			
PD:	0.4629		*PD*:	0.6968		*PD*:	0.2440	

DYS391

Allele	Count	Frequency
9	3	0.058
10	30	0.577
11	19	0.365

PD: 0.5305

DYS392

Allele	Count	Frequency
11	5	0.096
12	19	0.365
13	10	0.192
14	13	0.25
15	3	0.058
16	2	0.038

PD: 0.7534

DYS393

Allele	Count	Frequency
12	3	0.058
13	29	0.558
14	20	0.385

PD: 0.5370

DYS439

Allele	Count	Frequency
10	3	0.058
11	13	0.25
12	25	0.481
13	10	0.192
14	1	0.019

PD: 0.6689

DYS635

Allele	Count	Frequency
20	1	0.019
21	4	0.077
22	14	0.269
23	31	0.596
24	1	0.019
25	1	0.019

PD: 0.5654

DYS481

Allele	Count	Frequency
20	1	0.019
21	4	0.077
22	5	0.096
23	5	0.096
24	9	0.173
25	25	0.481
26	2	0.038
27	1	0.019

PD: 0.7122

DYS576

Allele	Count	Frequency
15	2	0.038
16	2	0.038
17	6	0.115
18	20	0.385
19	20	0.385
20	2	0.038

PD: 0.6860

DYS549

Allele	Count	Frequency
11	3	0.058
12	24	0.462
13	19	0.365
14	6	0.115

PD: 0.6367

DYS643

Allele	Count	Frequency
8	1	0.019
10	43	0.827
11	7	0.135
14	1	0.019

PD: 0.2971

DYS533

Allele	Count	Frequency
9	1	0.019
10	1	0.019
11	20	0.385
12	29	0.558
13	1	0.019

PD: 0.5393

DYS570

Allele	Count	Frequency
15	2	0.038
16	9	0.173
17	26	0.5
18	8	0.154
19	6	0.115
22	1	0.019

PD: 0.6813

PD: Power of discrimination.

Table A3. Y-STR haplotypes in Easter Island (n=52).

Haplotype	*#*	*389I*	*389II*	*390*	*456*	*19*	*385a*	*385b*	*458*	*437*	*438*	*448*	*GATA*	*391*	*392*	*393*	*439*	*635*	*481*	*576*	*549*	*643*	*533*	*570*	*Freq.*
H1	6	13	30	20	15	16	13	16	18	14	11	21	12	11	12	14	12	23	25	19	12	10	12	17	0.1150
H2	1	13	30	22	16	15	11	12	17	16	12	17	12	11	15	13	11	23	22	18	12	10	11	17	0.0192
H3	1	12	28	24	15	14	11	14	16	15	12	19	12	11	13	13	11	23	23	18	14	11	11	18	0.0192
H4	7	13	30	20	15	16	13	16	18	14	11	21	12	10	12	14	12	23	25	18	12	10	12	17	0.1350
H5	2	13	30	24	16	16	11	14	16	15	12	19	12	11	13	13	11	23	21	18	13	10	12	16	0.0385
H6	1	12	28	22	15	15	14	14	16	16	10	20	12	10	11	13	12	20	21	15	12	11	9	17	0.0192
H7	1	13	29	25	15	14	10	14	17	15	12	18	12	11	13	13	11	25	21	20	13	10	12	17	0.0192
H8	1	13	30	22	15	14	13	15	16	15	9	20	11	10	11	12	11	21	23	18	13	8	11	16	0.0192
H9	1	13	29	24	16	14	10	14	17	15	12	19	12	11	13	13	11	23	22	19	13	11	12	18	0.0192
H10	1	14	30	24	16	13	12	14	17	14	12	18	11	11	13	13	13	23	22	18	13	10	11	17	0.0192
H11	1	13	29	24	18	14	11	13	16	14	12	18	11	11	13	13	11	23	22	19	14	10	12	16	0.0192
H12	1	13	30	20	15	16	13	15	18	14	11	21	12	11	12	14	12	23	25	19	12	11	12	16	0.0192
H13	1	12	29	20	15	16	13	15	18	14	11	21	13	11	12	14	14	24	25	18	12	10	11	17	0.0192
H14	1	12	29	24	16	14	11	14	18	15	12	19	12	10	13	13	12	23	22	19	12	10	12	15	0.0192
H15	1	13	30	20	15	16	13	16	18	14	11	21	12	10	12	14	12	23	25	17	12	10	12	17	0.0192
H16	1	13	30	20	15	16	13	16	17	14	11	21	12	10	12	14	12	23	25	18	12	10	12	17	0.0192
H17	2	13	30	20	15	16	13	16	18	14	11	21	12	10	12	14	12	23	25	19	12	10	12	17	0.0385
H18	1	13	30	24	15	15	14	20	16	14	11	19	12	10	14	13	13	22	25	19	13	10	11	18	0.0192

Table A3. *(continued)*

Haplotype	*#*	*389I*	*389II*	*390*	*456*	*19*	*385a*	*385b*	*458*	*437*	*438*	*448*	*GATA*	*391*	*392*	*393*	*439*	*635*	*481*	*576*	*549*	*643*	*533*	*570*	*Freq.*
H19	1	13	30	23	15	13	14	17	18	14	11	19	12	10	13	13	10	22	24	19	14	10	12	19	0.0192
H20	1	13	30	24	15	15	14	19	16	14	11	19	12	10	14	13	13	22	24	19	13	10	11	18	0.0192
H21	1	14	31	24	15	13	14	15	18	14	13	20	12	9	14	13	13	22	26	20	14	10	11	16	0.0192
H22	2	13	30	24	15	15	14	20	16	14	11	19	12	10	14	13	13	22	24	19	13	10	11	18	0.0385
H23	1	14	32	24	15	15	15	17	16	15	11	19	12	10	14	13	10	22	24	18	14	10	12	19	0.0192
H24	1	13	29	23	15	14	13	15	15	14	9	20	11	10	11	12	10	21	24	17	11	10	11	22	0.0192
H25	1	12	28	23	16	14	14	18	15	16	9	20	10	10	11	12	11	21	23	16	11	10	10	18	0.0192
H26	1	12	28	23	16	14	14	18	15	16	9	20	10	10	11	12	11	21	23	156	11	10	10	18	0.0192
H27	1	13	32	25	16	14	14	17	16	14	11	19	12	11	14	13	13	22	25	19	12	11	12	17	0.0192
H28	1	13	31	21	15	15	16	17	16	14	11	21	12	11	11	13	11	21	27	15	11	14	11	19	0.0192
H29	2	13	30	24	15	15	14	20	16	14	11	19	12	10	14	13	13	22	24	18	13	10	11	19	0.0385
H30	1	13	29	24	14	14	14	16	17	14	11	20	12	10	15	13	12	23	23	17	13	10	11	17	0.0192
H31	1	13	29	25	16	14	11	14	15	15	12	19	12	11	13	13	12	23	24	17	12	10	12	19	0.0192
H32	1	13	31	23	16	13	12	15	17	14	11	20	12	9	14	14	11	22	25	18	13	10	12	16	0.0192
H33	1	13	28	24	15	13	13	17	17	14	11	20	12	10	14	13	13	22	25	19	13	10	11	18	0.0192
H34	1	14	31	23	15	14	15	18	18	14	12	20	12	10	16	13	12	23	25	19	13	11	11	15	0.0192
H35	1	13	29	24	14	14	14	17	17	14	11	20	12	10	16	13	12	23	20	17	13	10	11	17	0.0192
H36	1	13	29	23	16	13	11	18	16	14	11	21	13	10	14	13	12	22	26	16	13	11	13	16	0.0192
H37	1	13	31	23	16	13	12	15	17	14	11	20	12	9	14	13	11	22	25	18	14	10	11	16	0.0190

REFERENCES

1. González-Pérez E, Esteban E, Via M, García-Moro C, Hernández M, Moral P. Genetic change in the polynesian population of easter island: Evidence from alu Insertion polymorphisms. Annals of Human Genetics. 2006;70(6):829-840.

2. http://www.citypopulation.de/Chile-IsladePascua.html

3. Lie BA, Dupuy BM, Spurkland A, Fernández-Viña MA, Hagelberg E, Thorsby E. Molecular genetic studies of natives on easter island: Evidence of an early european and amerindian contribution to the polynesian gene pool. Tissue Antigens. 2007;69(1):10-18.

4. Ghiani ME, Moral P, Mitchell RJ, Hernandez M. Y-chromosome-specific STR haplotype data on the rapanui population (easter island). Human Biology. 2006;78(5):Article 3.

5. Thorsby E, Flåm ST, Woldseth B, Dupuy BM, Sanchez-Mazas A, Fernandez-Vina MA. Further evidence of an amerindian contribution to the polynesian gene pool on easter island. . Tissue Antigens. 2009;73(6):582-585.

6. Hunt TL, Lipo CP. Late colonization of easter island. Science. 2006;311:1603-1606.

7. Chapman PM. A biological review of the prehistoric rapanui. 1997;106(2):161-174.

8. Moreno-Mayar J , Rasmussen S, Seguin-Orlando A, Rasmussen M, Liang M, Flåm S, et al. Genome-wide ancestry patterns in rapanui suggest pre-european admixture with native americans. Current Biology. 2014.

9. Silva NM, Pereira L, Poloni ES, Currat M. Human neutral genetic variation and forensic STR data. PLos ONE. 2012:p. e49666.

10. Tian C, Gregersen PK, Seldin MF. Accounting for ancestry: Population substructure and genome-wide association studies. Human Molecular Genetics. 2008;17(143-150).

11. Paz Lagos L, Silva C, Rothhammer P, Carrasco X, Llop E, Aboitiz F, et al. Risk of attention deficit/hyperactivity disorder in aymara and rapa-nui school children. Association with dopaminergic system polymorphisms. Revista Medica de Chile. 2011;139 (5):600-605.

12. Comas D. Mapping genes involved in psychiatric disorders by admixture linkage disequilibrium in chilean populations. Report Summary. CORDIS: European Commission; 2012. Report No.: 236836.

13. Chakraborty R, Jin L. Heterozygote deficiency, population substructure and their implications in DNA fingerprinting. 1992. Hum. Genet. 88:267-272

14. Hill, C.R., Duewer, D.L., Kline, M.C., Sprecher, C.J., McLaren, R.S., Rabbach, D.R., Krenke, B.E., Ensenberger, M.G., Fulmer, P.M., Stort, D.R., Butler, J.M. (2011) Concordance and population studies along with stutter and peak height ratio analysis for the PowerPlex® ESX 17 and ESI 17 Systems. Forensic Sci. Int. Genet. 5(4): 269-275. Available from: http://www.cstl.nist.gov/strbase/software.htm.

15. Butler JM. Statistics and population genetics. NJSP Training Workshop. 2006:1-18. Available from: http://www.cstl.nist.gov/biotech/strbase/training.htm

16. Powerstats version 1.2, Promega corporation website, 2009. Available from: http://www.promega.com/geneticidtools/powerstats/.

17. Excoffier L, Laval LG, Schneider S. Arlequin ver. 3.0: An integrated software package for population genetics data analysis. Evol Bioinform Online. 2005;1:47–50.

18. Athey TW. Haplogroup prediction from Y-STR values using an allele frequency approach. Journal of Genetic Genealogy. 2005;1:1-7. Available from: http://www.hprg.com/hapest5/

19. Athey TW. Haplogroup prediction from Y-STR values using a Bayesian-allele frequency approach. Journal of Genetic Genealogy. 2006;2:34-9

20. Bandelt HJ, Forster P, Röhl A. Median-joining networks for inferring intraspecific phylogenies. Molecular Biology and Evolution. 1999 January 01;16(1):37-48. Available from: http://www.fluxus-technology.com

21. STRUCTURE version 2.3.4, Jul 2012. Available from: http://pritch.bsd.uchicago.edu/software.html.

22. Tamura K, Peterson D, Peterson N, Stecher G, Nei M, Kumar S. MEGA5: Molecular evolutionary genetics analysis using maximum likelihood, evolutionary distance, and maximum parsimony methods. Molecular Biology and Evolution. 2011 October 01;28(10):2731-9.

23. E. M. Shepard. and R. A. Chow. and Epifania Suafo'a. and David Addison. and A. M. Perez-Miranda. and R. L. Garcia-Bertrand. and Rene J. Herrera. "Autosomal STR Variation in Five Austronesian Populations." Human Biology 77.6 (2005): 825-851.

24. Projić P, Škaro V, Šamija I, et al. Allele Frequencies for 15 Short Tandem Repeat Loci in Representative Sample of Croatian Population. Croatian medical journal. 2007;48(4):473-477.

25. Coudray C, et al. Allele frequencies of 15 tetrameric short tandem repeats (STRs) in andalusians from huelva (spain). Forensic Science International. 2007 online; Volume 168(2):e21-e24.

26. Regueiro M, Mirabal S, Caeiro JL, Garcia-Bertrand RL, Herrera RJ. Austronesian genetic signature in east african madagascar and Polynesia. J Hum Genet. 2007/12/14 online; 53(2):106.

27. Beleza S, Alves C, Reis F, Amorim A, Carracedo A, Gusmao L. 17 STR data (AmpF/STR identifiler and powerplex 16 system) from cabinda (angola). . 2004;141(2):193-196.

28. Alves C, et al. STR allelic frequencies for an african population sample (equatorial guinea) using AmpFlSTR identifiler and powerplex 16 kits. Forensic Science International. 2004;148(2):239-242.

29. Chakraborty R, Smouse PE, Neel JV. Population amalgamation and genetic variation: observations on artificially agglomerated tribal populations of Central and South America. American Journal of Human Genetics. 1988;43(5):709-725.

30. Porras-Hurtado L, Ruiz Y, Santos C, Phillips C, Carracedo A, Lareu M. An overview of STRUCTURE: Applications, parameter settings and supporting software. Frontiers in Genetics. 2013;4:1-31.

31. Hunt TL. Rethinking easter Island's ecological catastrophe. Journal of Archaeological Science. 2007;34:485-502.

32. http://evolution.berkeley.edu/evosite/evo101/IIID3Bottlenecks.shtml

33. Chakraborty R, Stivers DN, Su B, Zhong Y, Budowle B. The utility of STR loci beyond human identification: Implications for development of new DNA typing systems. 1999. Electrophoresis 20:1682-16960.

34. National Research Council. NRC II: The evaluation of forensic DNA evidence. Washington, D.C.: National Academy Press, 1996. See Population Genetics Chapter 4. p.120

35. Denny M, Matisso-Smith L. Rethinking polynesian origins: Human settlement of the pacific. LEN Science. 2010:1-9.