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A genetic analysis of arodermatitis enteropathica

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A Genetic Analysis of Acrodermatitis Enteropathica

by

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THESIS

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Introduction

Zinc is found in every tissue of the body and plays an important role in many biological processes. It is an essential cofactor for more than 300 enzymes and can affect catalytic activity and structural stability (Prasad, 1996). Zinc also plays an important role in gene transcription by binding to zinc finger-proteins. Due to the widespread need for zinc in multiple biochemical processes, zinc deficiency can have very serious effects on an animal's well-being. Zinc deficiency can result from many causes including poor nutrition, malabsorption syndromes (such as Crohn's disease), liver or renal disease, and parenteral alimentation. Zinc deficiency can also result from the inherited disease acrodermatitis enteropathica (AE).

AE is a very rare autosomal recessive disease that presents during early infancy. Patients initially develop a characteristic periorificial and acral dermatitis. If left untreated, other symptoms may develop. These include alopecia, diarrhea, growth retardation, neuropsychiatric disorders, and impaired immune response. All of these symptoms are characteristic of systemic zinc deficiency. Treatment consists of lifelong daily oral zinc supplementation, which ameliorates all symptoms. Studies using intestinal biopsies have shown that AE patients have a decreased ability to absorb zinc (Atherton *et al.*, 1979), however the molecular defect responsible for AE is unknown.

The goal of this project was to genetically map the AE disease locus. In order to accomplish this, blood samples from 21 families with cases of AE were collected (Figure 1 and Table 1). The families were from many different countries including Egypt, the Netherlands, France, the United States, and Jordan. The initial samples collected were from several inbred Egyptian families, enabling us to utilize homozygosity mapping in the initial search for the AE locus.

Homozygosity mapping is a technique used for mapping autosomal recessive diseases in the offspring of consanguineous marriages (Lander and Botstein, 1987). It

is based on the principle that a fraction of the genome in these inbred offspring will be homozygous because of identity by descent. If one looks at affected offspring from several different consanguineous matings, the regions of homozygosity will vary except at the disease locus. Thus, the common region of homozygosity will contain the disease locus.

Based on the phenotype of AE patients, it seems most likely that the gene responsible for the disease will be involved in the transport of zinc. At the time this study was initiated, very little was known about zinc homeostasis in mammals, but several genes have subsequently been identified. Researchers have cloned four genes in rodents that appear to play a role in the management of zinc at the cellular level: ZnT1 (Palmiter and Findley, 1995), ZnT2 (Palmiter *et al.*, 1996), ZnT3 (Palmiter *et al.*, 1996a), and ZnT4 (Huang and Gitschier, 1997). ZnT1 is ubiquitously expressed and is localized on plasma membranes. It appears to be involved in zinc efflux, and its orientation in enterocytes of the small intestine supports a role in zinc absorption/retention (McMahon and Cousins, 1998)). ZnT2 is expressed in intestine, kidney, seminal vesicles, and testis and is localized on vesicular membranes. ZnT3 is expressed in brain and testis and is associated with synaptic vesicles. Both ZnT2 and ZnT3 are presumably involved in the accumulation of zinc in vesicles. ZnT4 is highly expressed in the mammary gland and brain and appears to function as a zinc exporter. Mutations in ZnT4 have been identified as the cause of *lethal milk*, a mouse mutant with defects in zinc metabolism (Huang and Gitschier, 1997). The human homologues of ZnT1 and ZnT4 were identified during the course of this study (by Bing Zhou and Liping Huang respectively, unpublished results), so an evaluation of these as potential candidate genes for AE was also included in this work.

Materials and Methods

Sample Collection

Blood samples from patients and their families were collected in collaboration with Dr. Wedad Mostafa, Dr. Jacobus Van Wouwe, Dr. Kathleen Bohanon, Dr. Irene Buño, Dr. Jackson Smith, and Dr. Hatem El-Shanti. Samples were obtained using informed consent. 2-10 ml of blood (depending on the age of the patient and the doctor's discretion) was drawn into a yellow top Vacutainer Blood Collection Tube (Becton-Dickinson). The blood was then shipped to us at room temperature. For long term storage, blood was kept at -70° C.

Cell Lines

In cases where a sufficient quantity of fresh blood was received, lymphoblasts were isolated and transformed using Epstein-Barr virus. Transformations were performed by Hernando Consengco. These cells were grown in RPMI-1640 media + 20% fetal bovine serum + 100 units/ml penicillin/streptomycin + 2mM glutamine at 37°C, 7.5% CO_2.

Preparation of DNA

DNA was isolated from fresh or frozen blood or transformed lymphoblast cell lines using Easy-DNA Kit (Invitrogen). The protocol was modified to increase recovered yield by using Phase Lock Gel-Light (5 Prime→3 Prime) in the chloroform extraction steps. In the preparation of DNA from blood, the protocol was also modified to increase yield by adding the chloroform before Solution B.

Preparation of RNA

mRNA was isolated from transformed cell lines using QuickPrep Micro mRNA Purification Kit (Pharmacia).

RT-PCR

cDNA was made using Superscript cDNA Kit (Gibco BRL). Approximately 450 ng of mRNA was reverse transcribed using 150 ng of random primers.

Candidate genes were amplified by polymerase chain reaction (PCR) using gene specific primers (Table 2 and Figure 2). Primary PCR was performed using 2 ul of cDNA, 1X PCR buffer (10X = 100 mM Tris-HCl pH 8.3, 500 mM KCl), 2.5 mM $MgCl_2$, 200 uM dNTPs, 25 pmol of 5' and 3' primer, 1.5 units of AmpliTaq DNA Polymerase (Perkin-Elmer), and 0.1 units of Pfu Polymerase (Stratagene). The cycling conditions used for these reactions are listed in Table 3. Reactions were performed in a DNA Thermal Cycler 480 (Perkin-Elmer).

The PCR amplification of the C-terminus of hZnT1 in unaffected samples 401, 402, 403, 801, 802, and 1501 was performed under the same conditions, but using 100 ng of genomic DNA as template.

In some instances it was necessary to subject the primary PCR product to a second round of hemi-nested PCR to increase the yield and/or specificity. This secondary PCR was performed as in the primary PCR but using 1 ul of primary PCR product as template. Sample 803 did not require a second round of PCR amplification with either fragment of hZnT4, so the primary product was used in sequencing.

Sequencing

PCR products were purified for sequencing either by Qiaquick PCR Purification Kit (Qiagen) or by running the reaction out on an agarose gel and purifying the band according to the protocol for Geneclean Kit (Bio 101).

All sequencing was performed by Martha Gunthorpe and Keith Beall of the HHMI DNA Facility. Sequencing reactions were conducted as outlined in the ABI Prism Dye Terminator Cycle Sequencing Ready Reaction Kit (Perkin-Elmer) using 50 ng of

purified PCR product and AmpliTaq DNA Polymerase FS (Perkin-Elmer). The reactions were carried out in a GeneAmp PCR System 9600 (Perkin-Elmer) and then run on an ABI Prism 377 DNA Sequencer (Perkin-Elmer).

In most cases, the PCR products were sequenced using the same primer that was used in PCR amplification (Table 2 and Figure 2). The N-terminal portion of hZnT4 was sequenced using primers ZNT 4E and ZNT 4D. The C-terminal portion of hZnT4 was sequenced using primers ZNT 4G and ZNT 4I (samples 404 and 1502) or primers ZNT 4A and ZNT 4B (sample 803). The N-terminal portion of hZnT1 was sequenced using primers ZNT 1B and ZNT 1C. For sample 1502, primer ZN-9 was also used to sequence a small portion of the N-terminus. The C-terminal portion of hZnT1 was sequenced using primers ZNT 1A and ZN-10.

Genome Screen

Samples were genotyped using the ABI Prism Linkage Mapping Set (version 1, Perkin-Elmer). This work was done in collaboration with Dr. Kirk Wilhelmsen.

PCR

Each PCR reaction contained 40 ng DNA, 1X buffer (10X = 100 mM Tris-HCl pH 8.3, 500 mM KCl), 2.5 mM dNTPs, 2.5 mM $MgCl_2$, 250 nM 5' and 3' primers, and 0.4 units of AmpliTaq Gold DNA Polymerase (Perkin-Elmer). Each of the 358 primer pairs was aliquoted individually in 96-well reaction plates, and the plates were incubated at 55° C until dry (approximately 15 min). The remaining reaction components were then aliquoted as a cocktail to the plates containing the dried primers. The reactions were performed in an MJ Thermalcycler (PTC 200) using the following program: 95° C x 12 min, then 1 cycle of 95° C x 1 min, 65° C x 2 min, 72° C 2.5 min, followed by 9 cycles of the same but decreasing the annealing temperature by 1° C each cycle so that the ninth

cycle annealing temperature is 56° C, followed by 20 cycles of 95° C x 1 min, 55° C x 1.5 min, 72° C x 1.5 min, and then a final extension of 72° C x 10 min.

Gel Analysis of Fragments

The Linkage Mapping Set is organized into 28 different panels based on size range and fluorescent dye color so that all the markers within a panel can be loaded into a single lane during gel analysis. Equal amounts of each PCR reaction in the same panel were pooled together to make 28 different pooled samples. A loading cocktail was prepared which consisted of 200 ul formamide, 40 ul blue dextran (50mM EDTA, 50 mg/ml blue dextran), and 40 ul GS-500 TAMRA size standard (Perkin-Elmer). 3.5 ul of this loading cocktail was added to 1.5 ul of each pooled panel and the mixture was denatured at 95° C for 5 min. 1.5 ul of this denatured sample was run on a 4% acrylamide gel for 2 hours using an ABI Prism 373 DNA Sequencer (Perkin-Elmer).

The gel results were analyzed using ABI Prism Genescan software (Perkin-Elmer). Allele sizes for each marker were assigned by importing the Genescan results into ABI Prism Genotyper software (Perkin-Elmer) and assigning peak size based on the GS-500 TAMRA size standard in each lane.

Follow-up Genotyping

Samples were genotyped for various highly polymorphic microsatellite markers. DNA from CEPH individual 1347-02 (NIGMS) was used as a control.

Radiolabeling primers

One marker-specific primer was radiolabeled by kinase reaction. Each kinase reaction consisted of 7 pmoles/ul primer, 1X kinase buffer (10X = 700 mM Tris-HCl pH 7.6, 100 mM $MgCl_2$, 50 mM DTT), 0.1 unit/ul polynucleotide kinase (Promega),

and 1 uCi/ul ^{32}P gamma-ATP (3000 Ci/mmol, Amersham). The reactions were incubated at 37° C for 20-30 minutes.

PCR

Each PCR reaction consisted of 48 ng DNA, 1X PCR buffer (10X = 100 mM Tris-HCl pH 8.3, 500 mM KCl), 1 mM $MgCl_2$, 200 mM dNTPs, 320 nM unlabelled primer, 336 nM labeled primer, and 0.25 units AmpliTaq DNA Polymerase (Perkin-Elmer). Reactions were performed in a GeneAmp PCR System 9600 (Perkin-Elmer), using a program of 94° C x 3 min, followed by 9 cycles of 94° C x 30 sec, 55° C x 1 min 20 sec, 72° C 30 sec, followed by 15 cycles of 90° C x 15 sec, 55° C x 1 min 20 sec, 72° C 30 sec, with a final extension of 72° C x 10 min. Reactions were removed at the end of the program and either run on a gel immediately or stored at -20° C until they could be analyzed. Note: The annealing temperature and concentrations of $MgCl_2$ used were modified in some cases in order to optimize the intensity and/or specificity of the resulting product.

Gel Analysis of Fragments

The results of the PCR were analyzed on gels consisting of 5% Long Ranger Gel Solution (FMC), 0.6X TBE, and 7 M urea. An equal volume of denaturing dye (1X TBE, 80% formamide, 0.02% xylene cyanol, 0.02% bromophenol blue) was added to each PCR reaction and they were incubated at 94° C for 3 min and then on ice 3-5 min. The gel was loaded with 6 ul of denatured sample and run at 50 Watts for 1.5 - 4 hours depending on the size of the fragments. At the end of the run, the gel was dried and exposed to film at room temperature overnight.

Allele sizes for each marker were assigned by using the results with the CEPH 1347-02 DNA as a size standard.

Results

Genome Screen

Initial Genome Screen

The locus for the AE gene was initially sought by homozygosity mapping using several inbred Egyptian families. A genome wide screen was conducted using the ABI Prism Linkage Mapping Set. This set includes 358 highly polymorphic microsatellite markers spaced at average intervals of 10 cM. The screen was performed on inbred Egyptian patient samples III1, 2C, 905, 1102, and 1303. Samples IV1 and 303 were not included due to insufficient quantities of DNA. A general summary of the results of this screen is shown in Table 4.

The genotyping data was examined to identify markers for which all five patients or at least four out of five patients were homozygous. Eleven such loci were identified: D1S413, D3S1293, D4S402, D5S433, D10S189, D11S934, D11S1345, D13S158, D15S131, D19S418, and D21S1256. One locus where four patients were homozygous and the reaction for the fifth patient failed (D2S319) was also included, making a total of twelve initial candidate loci.

Analysis of Initial Candidate Loci

In order to determine which of the markers at the twelve loci were actually homozygous by descent rather than homozygous by state, additional markers adjacent to the candidate loci were selected and genotyped using primers from Research Genetics (RG). The samples included were patients III1, IV1, 2C, 303, 905, 1102, and 1303, their parents (where available), and a small number of unaffected siblings.

During the course of this follow-up, markers for ten of the loci identified in the initial screen were genotyped again with the RG markers, and some discrepancies between the results were discovered (Table 5). Some of the patient samples that

appeared to be homozygous using the ABI markers were heterozygous for the same loci using RG markers. Sample mix-up prior to the second genotyping seems an unlikely explanation because some of the markers did give identical results in both instances. The discrepancy appears to be due to random weak amplification of alleles in the ABI genotyping. This created a false indication of homozygosity in several instances where patients were actually heterozygous. (See the re-evaluation section below for more discussion about the reliability of the ABI data).

The results with the RG markers showed that most of the original loci identified did not actually have that many patients who were homozygous and ruled these out as good candidates. Loci D10S189 and D19S418 could not be successfully genotyped using RG markers, but data for adjacent markers did not reveal substantial homozygosity in the region. Four out of seven patients were homozygous for D1S413 and D11S1345, but coverage of more markers near these loci showed no substantial homozygosity in the region.

Repeating Miscellaneous Markers That Did Not Work Well In the Screen

The data from the original screen was reviewed again with a focus on markers that had failed with some samples, leaving data for four or fewer patients at these loci. Any marker with no more than one sample heterozygous was selected for follow-up genotyping on the basis that if data for more patients were available, the location could turn out to be a good candidate. RG markers for these loci were genotyped in the same seven patients listed previously. In instances where the RG marker did not work well, another marker very close by was chosen (Table 6). Markers D4S1546 and D13S1275 appeared to be potentially interesting with 3 to 6 out of 7 and 6 out of 7 patients homozygous respectively. However, when additional markers near these loci were genotyped, no substantial homozygosity was found.

Filling In Gaps In the Screen

The marker spacing in the original screen was only an average of every 10 cM, and therefore it contained many intervals of much larger distances. The expected average length of regions homozygous by descent (HBD) was calculated for each of the seven inbred patients (Table 7). Based on this result, a marker interval of 15 cM should allow detection of regions that are HBD in these patients. Gaps of more than 15 cM in the original screen were identified and markers were chosen to fill in these regions (Table 8). The data from these markers produced five potentially interesting loci: D1S2785 (4/7 homozygous), D9S1826 (5/7 homozygous), D2S111 (3/6 homozygous), D4S1579 (4/7 homozygous), and D13S1315 (5/7 homozygous). Locus D19S884, with a somewhat borderline result of 3/6 homozygous, was also included as a possibility because 3/5 patients were homozygous for the adjacent marker D19S221 in the initial screen. Additional markers adjacent to these potential loci were genotyped, but no shared region of homozygosity could be detected.

Analysis of More Potential Loci

In an attempt to reduce the risk that something important was being missed in the original screen, loci for which three out of five patients were homozygous were considered. We were reluctant to pursue all of these loci because of the large number involved (twenty-seven) and because we knew many of them may actually be less than three due to the occurrence of false homozygosity in the screen results. Therefore, the loci were prioritized by considering the possibility of linkage disequilibrium.

Linkage disequilibrium is the nonrandom association of a specific marker allele and a disease allele in a population. When a disease mutation is introduced into a population, it exists on a background of specific marker alleles. Affected individuals who are descendants of this founder will have these alleles more often than expected based on the allele frequency in the population as a whole. There is no evidence that the six

inbred Egyptian families used for mapping in this study have descended from a common ancestor. However, a founder effect could explain the relatively high incidence of such a rare disease within the population from this region.

Loci where 3 out of 5 patients were homozygous and at least 6 out of 10 of the patient chromosomes had the same allele were chosen for follow-up (Table 9). The genotyping of adjacent markers identified D10S1652 (4/6 homozygous) and D20S886 (4/7 homozygous) as potentially interesting loci. Marker D2S142 was also highlighted because while it only had 3/7 patients homozygous, 9/14 chromosomes shared the same allele. Follow-up of these regions with additional markers did not identify any shared region of homozygosity.

Re-Evaluation of Original Screen Data

The discrepancies observed between the ABI results and the RG results had been very disconcerting. After consulting with several people experienced in using the ABI system, the data from the genome screen was re-evaluated. Previously, genotyping results for markers had been reviewed for each sample individually. This time the results for a particular marker were reviewed with all samples simultaneously in order to evaluate the general characteristics of each marker. This change in the reviewing process made it easier to score the precise alleles with more confidence and to spot aberrant results that might be questionable. Upon more careful review of the data, it was also possible to assign alleles for markers with complicated results that had not seemed scorable before. The criteria for what kind of results would actually be scored in the genotyping was also changed. During the initial analysis, many markers were scored but were noted as questionable. In the re-analysis, any marker that gave ambiguous results in a sample was not scored at all, and the minimum acceptable signal strength required to consider a peak real was increased. These higher standards in scoring meant

a lot of data previously included was now thrown out as potentially unreliable. A general summary of the screen results after re-analysis is shown in Table 10.

The re-analyzed data was reviewed for potential candidate regions. Many of the same regions identified from the initial analysis showed up again. One new potential locus was D6S271 with 5 out of 5 patients homozygous. However, this region had already been covered by other markers during the follow-up of loci with 3/5 patients homozygous, and no significant homozygosity had been observed.

Twenty-three markers failed completely with all five samples, creating gaps in the screen. Four of these gaps had already been covered during follow-up genotyping, and in the case of eight of these markers, the gap created by the failure was less than 15 cM. Two intervals larger than 15 cM in the ABI marker coverage which had been missed in the follow-up were also discovered. That makes a total of thirteen gaps of more than 15 cM in the screen that have not been covered.

Sixty-seven markers that did not work very well (data for four or fewer samples) and where no more than one patient was heterozygous were identified. Fifteen of these loci had already been covered in follow-up genotyping, and for six of the markers the gap in the screen without them was still less than 15 cM. That leaves forty-six loci which could have substantial homozygosity if the genotyping data for more samples were available.

A complete summary of all genotyping data, including the re-analyzed ABI results and the results with RG markers, is shown in Table 11.

Candidate Gene Evaluation

The zinc transporters ZnT1 and ZnT4 were evaluated as potential candidate genes. mRNA for patient samples 404, 803, and 1502 was reverse transcribed and then hZnT1 and hZnT4 were amplified by PCR with gene-specific primers. The resulting PCR products were sequenced and analyzed for any mutations. In hZnT4, the same two

differences were detected in all three patients: a change in nucleotide 844 from a C to a T, and a change in nucleotide 915 from a C to a T. Neither of these changes affect the amino acid sequence of hZnT4 and the mouse ZnT4 has the same nucleotide sequence for the corresponding amino acids. In hZnT1, one difference was detected in all three patients: an A to C substitution that changes amino acid 450 from an isoleucine to leucine. When this region was sequenced in six unaffected family members (samples 401, 402, 403, 801, 802, and 1501), the same A to C substitution was found, therefore, the change appears to be only a polymorphism. This amino acid is also not conserved in mouse where the corresponding residue is an arginine.

Based on these results, hZnT1 and hZnt4 are essentially ruled out as potential candidate genes for AE.

Discussion

Blood samples were collected from 21 families with cases of AE. A genome screen was conducted using 358 polymorphic microsatellite markers and patient samples from inbred Egyptian families. An additional 80 markers were also genotyped in following-up the results of this screen. No obvious candidate region for the AE disease locus could be identified from these results. In addition, the hZnT1 and hZnT4 genes were analyzed as possible candidate genes for AE, but no mutations were detected.

There are several possible reasons that the screen did not identify any candidate region. The most likely explanation is that the marker coverage was not dense enough to detect the chromosomal region that is homozygous by descent. The re-analysis of the original screen data reduced the number of markers that could be reliably scored, and sufficient coverage of these problem loci might reveal the location of the disease locus. However, even if the coverage were at 15 cM intervals, it still might not be possible to detect the candidate region. This screen density was chosen based on the predicted lengths of regions HBD in the patients, but this is only an estimate based on averages. The actual length of the region surrounding the disease locus could be smaller in some or all of the patients, and even denser marker coverage could be required to detect it.

One possible complication could be a mix-up in the patient samples. This would have a very negative effect on our ability to identify the disease locus from the genotyping data. We of course made every attempt to insure this did not happen, but human error can occur. All of the genotyping that included additional family members showed Mendelian patterns of inheritance for everyone involved which helps to rule out the possibility of any major mix-ups. However, this does not rule out the possibility of a patient sample being mistakenly swapped with that of an unaffected sibling. Even if such an unlikely event had occurred, a shared region of homozygosity would presumably still be detectable in six out of the seven patients, but no such region was observed.

Another possible complication could be misdiagnosis of a patient. Zinc deficiency and its related symptoms can result from causes other than AE. Marginal zinc deficiency is actually quite widespread, particularly among poor children of developing countries who consume very little red meat but do consume large amounts of cereal grains containing phytates which can decrease zinc absorption (Ploysangam *et al.*, 1997). Marginal zinc deficiency is typically associated with growth retardation and general failure to thrive. It seems unlikely that any of the Egyptian patients collected for this study are suffering from zinc deficiency solely due to poor nutrition. First, all of the children in these families are presumably consuming the same diet, but only a small number are developing symptoms of zinc deficiency. Second, if they were suffering from nutritional zinc deficiency, then presumably the symptoms would disappear as the children got older and their biological zinc requirements decrease. However, most of these patients have been taken off zinc supplementation at least once and sometimes multiple times, and in all cases their symptoms returned almost immediately.

A third possible complication could be genetic heterogeneity. While there has been no reason to suspect heterogeneity in AE, it cannot be ruled out, particularly if denser coverage with polymorphic markers reveals no shared region that is HBD among all of the patients. An attempt was made to limit the effects of any locus heterogeneity that may exist by only including patients from a particular ethnic background. The Egyptian population is highly inbred (approximately 29% of marriages are consanguineous) and has had very little migration until recently (Teebi, 1997). However, locus heterogeneity within this population is still a possibility. A better way to avoid this potential problem would be to utilize the samples from Pedigree 21 for analysis. This large kindred from Jordan was identified just recently and has five individuals affected with AE. Because these patients are all from the same family, genetic heterogeneity of the AE locus among them would be highly unlikely.

In terms of addressing the AE question with a candidate gene approach, it is possible that as more genes involved in zinc homeostasis are identified, another good candidate gene will be among them. One must also consider that the gene involved in AE might transport other metals in addition to zinc or might have a different function altogether which cannot be predicted based on our limited knowledge. One metal-ion transporter that could be evaluated in AE patients is the human NRAMP2 gene. This predicted transmembrane protein is expressed in many adult tissues, including the small intestine (Vidal *et al.*, 1995). Experiments with DCT1, a rat homologue of hNRAMP2, have demonstrated active transport of a wide range of metals, including zinc, copper, and iron (Gunshin *et al.*, 1997). Expression of DCT1 appears to be upregulated by dietary iron deficiency, suggesting a role in intestinal iron absorption. Perhaps a mutation in hNRAMP2 that affects only its specificity for zinc could be the cause of AE.

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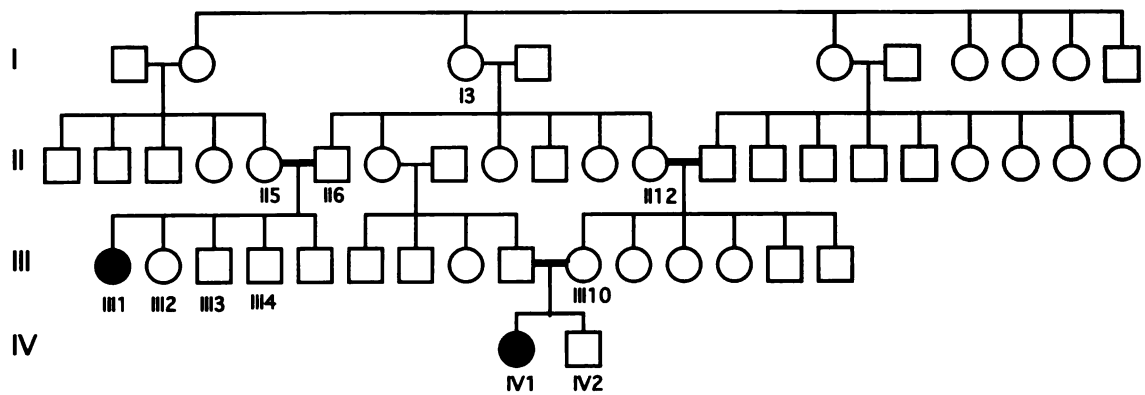
Figure 1 - Pedigrees

Pedigree #1



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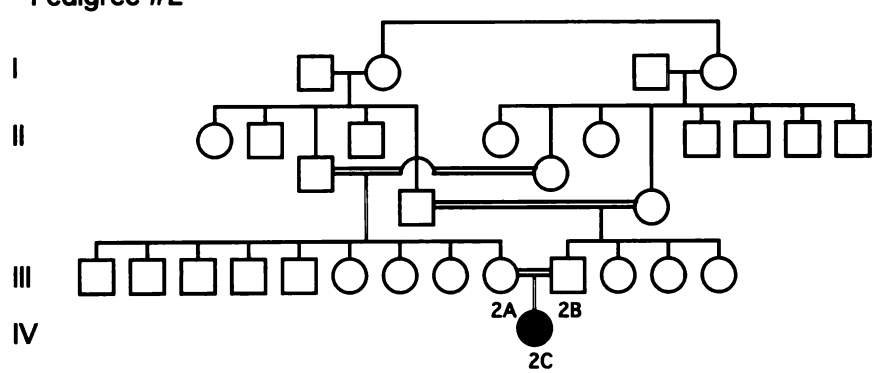


Pedigree #3

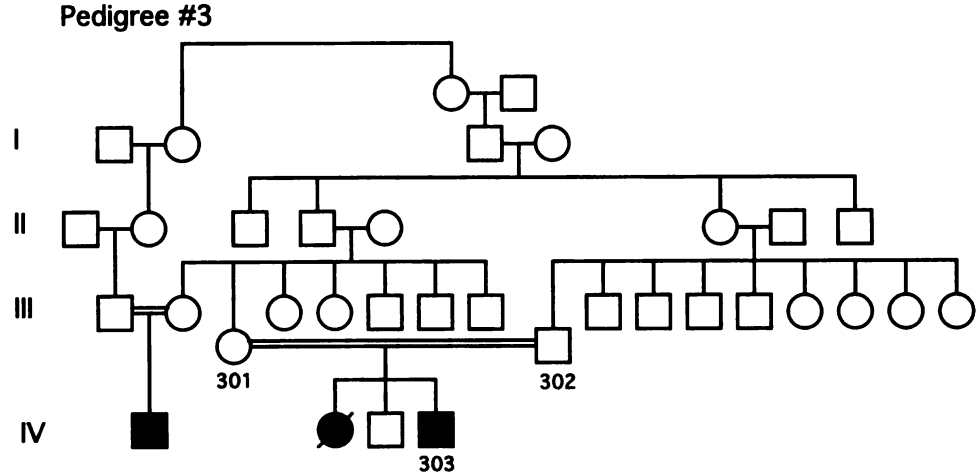
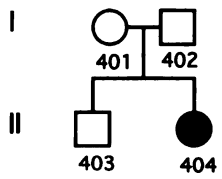


Figure 1 - Pedigrees (Continued)

Pedigree #4



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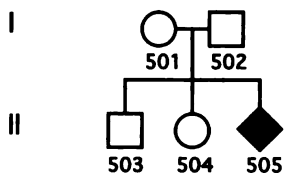
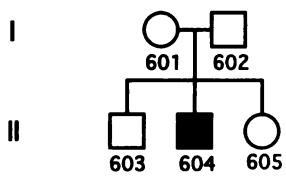
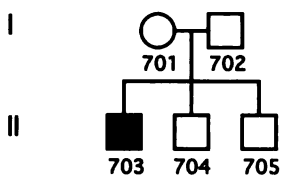


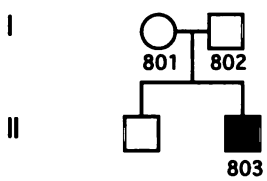
Pedigree #6



Pedigree #7



Pedigree #8



Pedigree #9

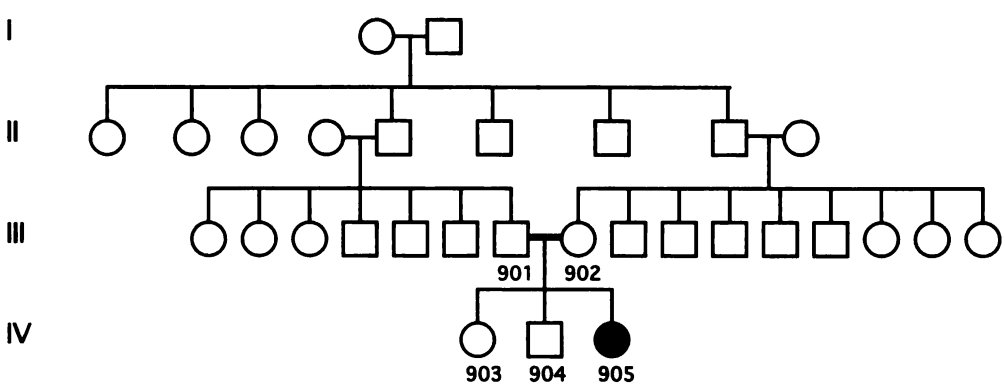
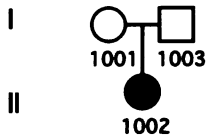
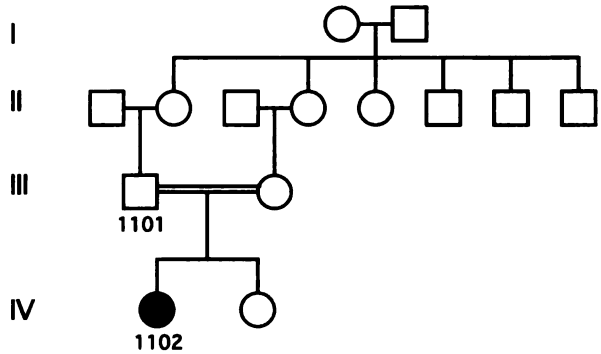


Figure 1 - Pedigrees (Continued)

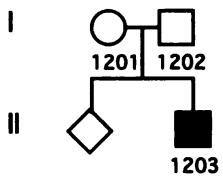
Pedigree #10



Pedigree #11



Pedigree #12



Pedigree #13

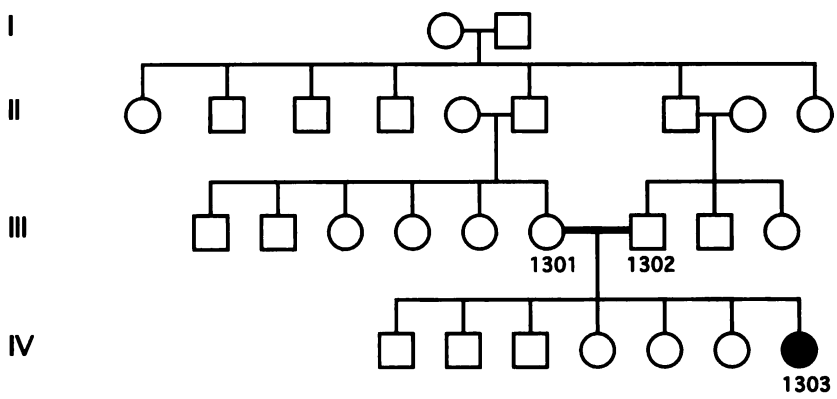
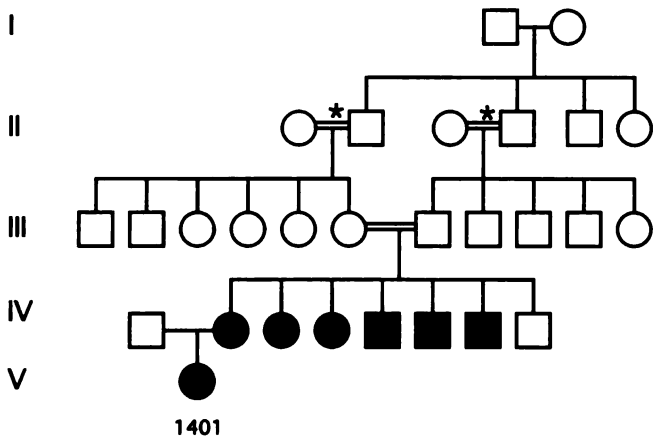


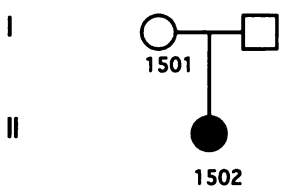
Figure 1 - Pedigrees (Continued)

Pedigree #14 - Questionable AE

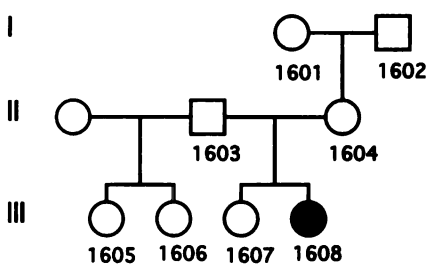


*Distant Relative

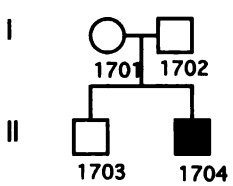
Pedigree #15



Pedigree #16



Pedigree #17



Pedigree #18

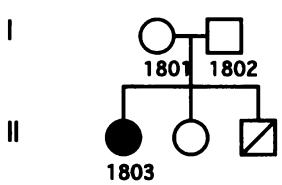
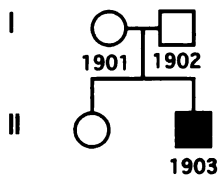
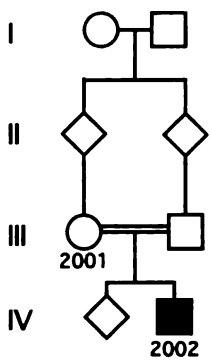


Figure 1 - Pedigrees (Continued)

Pedigree #19



Pedigree #20



NOTE: not certain if parents are first or second cousins

Pedigree #21

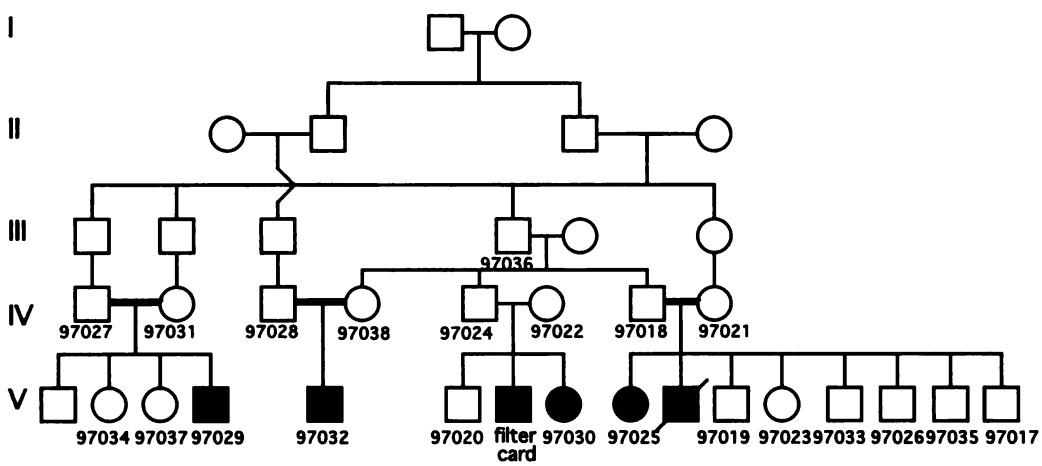


Table 1 - AE Patients

Ped#	ID#	Consang?	Country	Cell line	Comments
1	III1	Yes	Egypt		Affected relative
1	IV1	Yes	Egypt		Affected relative
2	2C	Yes	Egypt		
3	303	Yes	Egypt		Affected relative
4	404	No	Netherlands	96-103	
5	505	No	Netherlands	96-104	
6	604	No	Netherlands	96-105	
7	703	No	Netherlands		
8	803	No	USA	96-111	
9	905	Yes	Egypt		
10	1002	No	Egypt		
11	1102	Yes	Egypt		
12	1203	No	Egypt		
13	1303	Yes	Egypt		
14	1401	Yes	Egypt		Questionable AE
15	1502	No	USA	97-104	
16	1608	No	France	97-111	
17	1704	No	USA		Mom-N.Africa
18	1803	No	USA	97-118	Atypical AE
19	1903	No	USA	97-126	
20	2002	Yes	Egypt		
21	97029	Yes	Jordan		Affected relatives
21	97032	Yes	Jordan		Affected relatives
21	Filter card	Yes	Jordan		Affected relatives and sib
21	97030	No	Jordan		Affected relatives and sib
21	97025	Yes	Jordan		Affected relatives and sib

Table 2 - Gene Specific Primers

Primer name	Primer sequence
ZNT 4A	gctgtgcaaagaactatccatat
ZNT 4B	gctctcaagtctgtgactgca
ZNT 4C	ataaataaagtgaattcagcaatgc
ZNT 4D	ctgcaactccaacagctgcg
ZNT 4E	cggccgctcagcctctg
ZNT 4G	atgaactatgaaataaatggag
ZNT 4I	ggcttttacaacatttcgaatc
ZNT 1A	gtgtgaatatgtttccctgacc
ZNT 1B	tgcaccagtctccgggc
ZNT 1C	ccagcctcataaactgatgcatgag
ZNT 7	tatagcacccagcaaggaccagc
ZN-9	atgctctccgactccttccac
ZN-10	ctggctttccaaaacagtcac

Figure 2 - Map of Gene Specific Primers

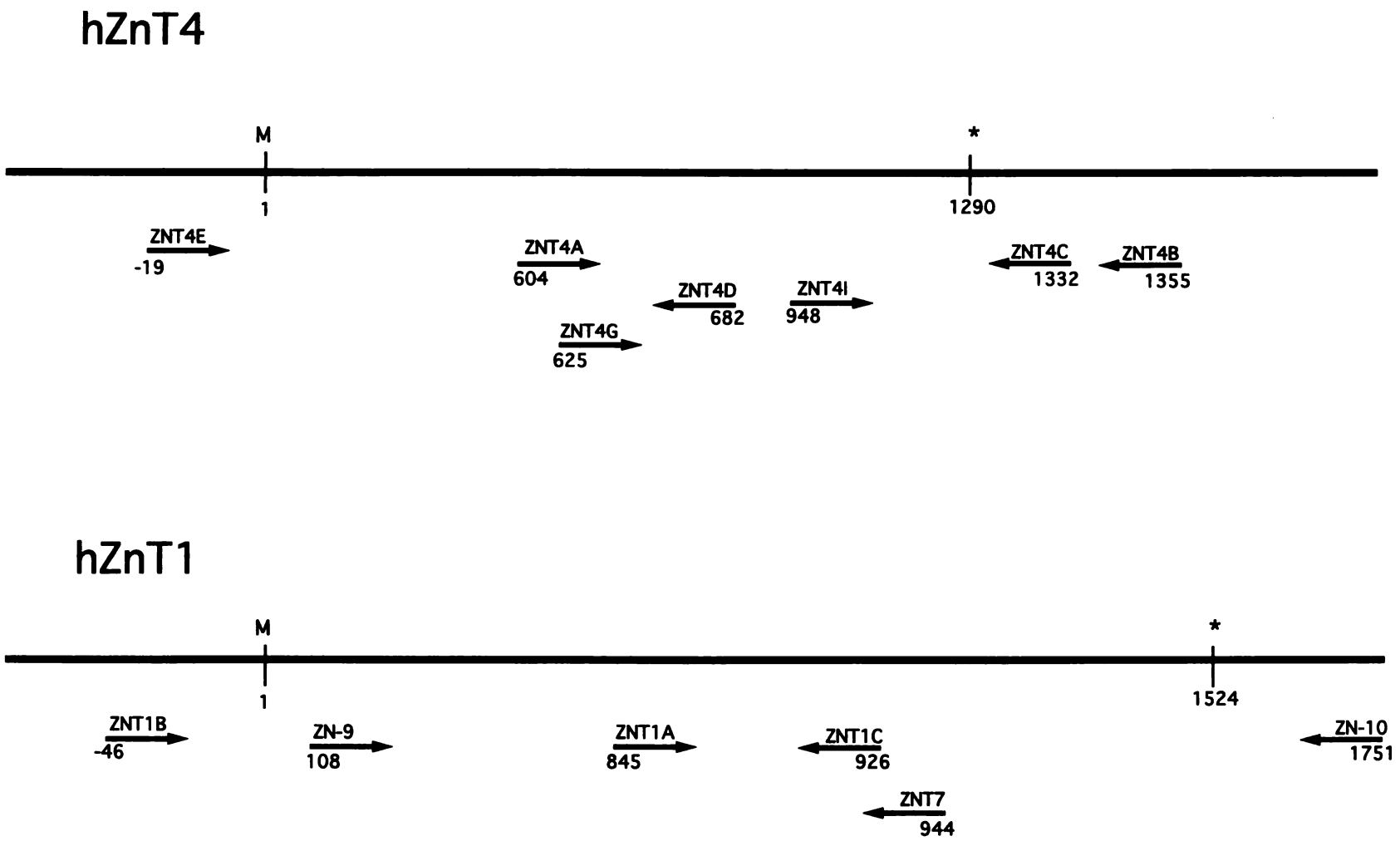


Table 3 – RT-PCR Conditions

Gene	Fragment	PCR type	Primers		Annealing temp	Extension time	Number of cycles
			5'	3'			
hZnT1	N-terminus	Primary	ZNT 1B	ZNT 7	58° C	3 min	36
		Secondary	ZNT 1B	ZNT 1C	58° C	1 min 30 sec	25
hZnT1	C-terminus	Primary	ZNT 1A	ZN-10	55° C	1 min 20 sec	30
hZnT4	N-terminus	Primary	ZNT 4D	ZNT 4E	68° C	1 min	30
		Secondary	ZNT 4D	ZNT 4E	70° C	1 min	25
hZnT4	C-terminus	Primary	ZNT 4A	ZNT 4B	59° C	1 min	30
		Secondary	ZNT 4A	ZNT 4C	61° C	1 min	25

Table 4 - General Summary of ABI Screen Results

Sample	Number of failed markers	Marker success rate	Number of markers homozygous	Percent of markers homozygous
1111	34	90%	82	27%
2C	43	87%	114	38%
905	30	91%	91	29%
1102	26	92%	94	30%
1303	17	95%	82	25%

Note: Only autosomal markers are included in these numbers (343 total)

Table 5 - Summary of Initial Candidate Loci Results

Locus	Homozygosity Status (ABI)	Homozygosity Status (Research Genetics)	
	5 patients	5 patients	All 7 patients
D1S413	4/5	4/5	4/7
D2S319	4/4	3/4	3/7
D3S1293	4/5	1/5	1-2/7
D4S402	4/5	1/5	1/7
D5S433	4/5	1/5	2/7
D10S189	5/5	N/A	N/A
D11S934	4/5	1/5	2/7
D11S1345	4/5	4/5	4/7
D13S158	4/5	2/5	2/7
D15S131	4/5	2/5	2-3/7
D19S418	4/5	N/A	N/A
D21S1256	4/5	3/5	2/7

Table 6 - Summary of Miscellaneous Marker Results

ABI Marker	Homozygosity status	Gap without marker (cM)	RG marker	Distance from ABI marker	Homozygosity status
D1S235	2/3	24.5	D1S2800	3 cM	3/7
D1S255	2/3	17.8	Same	N/A	2-3/7
D2S281	3/4	15.6	D2S2211	1 cM	2/6
D2S338	3/4	17.7	Same	N/A	3/7
D2S368	3/4	20.2	D2S112	2 cM	0/5
D2S396	3/3	19.6	D2S2158	5 cM	2/7
D4S1535	3/3	26.6	Same	N/A	3/7
D4S419	2/3	19.6	D4S1546	2 cM	3-6/7
D5S422	3/4	20.5	Same	N/A	2/5
D7S507	3/4	18.1	Same	N/A	2/5
D8S272	0/1	12.4+	D8S534	1 cM	3/7
D8S556	3/3	27.1	D8S1830	1 cM	3/6
D12S310	1/1	24.4	D12S1669	2 cM	1/7
D12S83	3/4	21.9	D12S1585	1 cM	2/7
D13S175	3/4	6.8+	D13S1275	1 cM	6/7
D15S165	3/4	20	D15S1031	?	1-2/6
D17S945	1/2	17.8	D17S1879	1 cM	2/7
D18S52	3/4	16.7	Same	N/A	1/6
D20S173	2/2	16.3	Same	N/A	2-3/5

? Indicates unresolved map distance

Table 7 - Expected Lengths of Regions HBD

Patient	# Generations btwn patient and common ancestor	Number of meioses	Number of crossovers[1]	Average length of regions (cM)
III1	3	6	180	16.7
IV1	4	8	240	12.5
2C	4	8	240	12.5
303	5	10	300	10
905	3	6	180	16.7
1102	3	6	180	16.7
1303	3	6	180	16.7

[1] Using recombination frequency of 1/100 cM and genome size of 3000 cM

Table 8 - Summary of Gap Marker Results

ABI flanking markers	Gap length (cM)	RG marker to fill	Homozygosity Status
D1S214 - D1S228	15.5	D1S450	1/7
D1S228 - D1S199	17.6	D1S436	1/6
D1S235 - D1S423	23	D1S2785	4/7
D1S249 - D1S213 *	21	D1S471	1/7
		D1S490	1/7
D1S484 - D1S196	15.7	D1S2844	0/7
D2S142 - D2S326	16.3	D2S111	3/6
D3S1566 - D3S1271	20.8	D3S3681	0/7
D3S1580 - D3S1311	15.6	D3S3562	2/7
D4S1575 -D4S424	19.6	D4S1579	4/7
D4S1597 - D4S415	15.1	D4S2992	2/7
D4S392 - D4S1534	15.3	D4S2963	3/7
D4S412 - D4S403	22	D4S394	1/7
D5S424 - D5S428	15.9	D5S641	1/7
D6S257 - D6S462	20.2	D6S460	2/7
D6S264 - D6S281	22	D6S446	3/7
D7S519 - D7S669	22	D7S2483	3/7
D7S636 - D7S550	16.1	D7S2462	2-3/7
D7S669 - D7S657	16.2	D7S2540	2-4/7
D7S684 - D7S636	15.2	D7S661	1-3/6
D8S258 - D8S283	20	D8S1839	2-3/7
D8S550 - D8S258	20	D8S1731	1-2/7
D9S164 - D9S158	15.2	D9S1826	5/7
D9S279 - D9S290	24	D9S1872	0/5
D9S286 - D9S157	16.9	D9S269	0/7
D9S287 - D9S279	20.4	D9S1784	2/7
D12S310 - D12S345	19	D12S1591	2-3/7
D13S171 - D13S263	16.2	D13S218	3/7
D13S173 - D13S285	18.4	D13S1315	5/7
D15S165 - D15S126	27	D15S1040	3/7
		D15S146	2/7
D16S405 - D16S420	16.7	D16S3036	1/7
D16S511 - D16S520	19.5	D16S3037	0/7
D17S799 - D17S798 *	22	D17S1843	3/6
D19S216 - D19S221	18.4	D19S884	3/6
D19S420 - D19S418	26	D19S866	1/7

* Gaps created by failed markers

Table 9 - Summary of Potential Loci Results

ABI marker	Chromosomes sharing allele	RG marker	Distance from ABI marker	Homozygosity Status
D2S142	6/10	D2S284	1 cM	3/7
D6S271	8/10	D6S282	?	0/7
D9S171	6/10	D9S1679	?	1/7
D10S597	7/10	D10S543	1 cM	0/6
D10S561	7/10	D10S1652	?	4/6
D14S288	8/10	D14S1013	?	0/7
D20S178	approx 6/10	D20S886	?	4/7

? Indicates unresolved map distance

Table 10 –
General Summary of Re-Analyzed ABI Screen Results

Sample	Number of failed markers	Marker success rate	Number of markers homozygous	Percent of markers homozygous
1111	69	80%	74	27%
2C	124	64%	79	36%
905	95	72%	76	31%
1102	84	76%	68	26%
1303	78	77%	67	25%

Note: Only autosomal markers are included in these numbers (343 total)

Table 11 - Genotyping Data

Chromosome 1																	
Marker	**cM**	**III1**	**III1**	**IV1**	**IV1**	**2C**	**2C**	**303**	**303**	**905**	**905**	**1102**	**1102**	**1303**	**1303**	**Status**	
D1S468	9.4	200	202	xx	xx	197		xx	xx	201				197	202	2/4	
D1S214	15.5	120	135	xx	xx	119	136	xx	xx	119	137	135		119	135	1/5	
D1S450#		249	251	251	257	249	261	251	257	247	259	249	253	253		1/7	
D1S228	17.6	128	132	xx	xx			xx	xx	129	131	129		127	129	1/4	
D1S436#		202	228	196	228	202		202	238	n/a	n/a	202	230	220	238	1/6	
D1S199	9.5	95	107	xx	xx	101	111	xx	xx	97	99	101	105	95	101	0/5	
D1S234	10.3	265	273	xx	xx	264	273	xx	xx	265	273			265	269	0/4	
D1S255#	7.5	74		74	80	78	?	74	80	82	92	80	84	80		2-3/7	(ABI 1/2)
D1S197	12	122	130	xx	xx	119	121	xx	xx	117		123	125	115	123	1/5	
D1S220	5.7	246	250	xx	xx	235	251	xx	xx	237		242	248	235	247	1/5	
D1S209	10.1			xx	xx	165		xx	xx							1/1	
D1S216	11.6	245	247	xx	xx			xx	xx	249	251			246	250	0/3	
D1S207	14.3	148	156	xx	xx	148		xx	xx	148	156			148	158	1/4	
D1S424	9.3	210	228	xx	xx			xx	xx	224	226	210	228	210	212	0/4	
D1S206	11.3	206		xx	xx	206	210	xx	xx	206	210	206	210	206	216	1/5	
D1S502	4.4			xx	xx			xx	xx							failed	
D1S252	5.6	99	110	xx	xx			xx	xx	108		98	102	108	110	1/4	
D1S498	13.1	195	197	xx	xx	193	199	xx	xx	199		191	193	195	197	1/5	
D1S484	15.7	275		xx	xx	273	277	xx	xx	271	275	271	277	270		2/5	
D1S2844#		175	189	179	183	179	181	177	179	179	183	181	183	177	183	0/7	
D1S196	8.9	315	319	xx	xx	319		xx	xx	309		309	321	309		3/5	
D1S218	10.9	266	278	xx	xx			xx	xx	272	278	268		274	280	1/4	
D1S238	8.5	303	314	xx	xx	301	307	xx	xx	301	307	299	309	303	307	0/5	
D1S2840#*		3		1	3	1	3	3	4	2	3	3	4	3		2/7	
D1S413#*	9.8	252		254	256	246		246	254	250	258	254		246		4/7	(ABI 4/5)
D1S249	11.4	163	175	xx	xx	163		xx	xx	163		163	174	165	175	2/5	
D1S471#*		191	195	185	189	189	191	189	193	187	191	189		187	189	1/7	
D1S425	10.2			xx	xx			xx	xx							failed	
D1S490#*		197		197	199	199	213	197	211	203	207	197?	209?	197	213	1/7	
D1S213	13.5	109		xx	xx	101	107	xx	xx	107	109	103	113	103	109	1/5	
D1S2800#		178	180	186	188	178	184	178	182	178		192		178		3/7	
D1S235	21.2			xx	xx			xx	xx			208				1/1	
D1S2670#		3	7	3	8	1	5	5	8	1	5	7		4		2/7	
D1S2785#*		166	172	172	176	178		174	178	170		168		166		4/7	
D1S423		164	166	xx	xx	164	166	xx	xx	164	166	164	166	164		1/5	

Results shown are with RG marker

* Results for unaffected relatives not shown

Table 11 - Genotyping Data (Continued)

Chromosome 2																	
Marker	**cM**	**III1**	**III1**	**IV1**	**IV1**	**2C**	**2C**	**303**	**303**	**905**	**905**	**1102**	**1102**	**1303**	**1303**	**Status**	
D2S319#*	7.8	128		128	132	128		130	132	132		130	132	124	130	3/7	(ABI failed)
D2S281	8.1	252		xx	xx	248	250	xx	xx			250				2/3	
D2S2211#		7		5	7	n/a	n/a	5		1	9	6	7	8	9	2/6	
D2S162	5.8	119	137	xx	xx	130	132	xx	xx	125	129	130		119	137	1/5	
D2S168	13.4	167	169	xx	xx	167		xx	xx	159	175	157	169	163	171	1/5	
D2S305	7.1	323		xx	xx	318	324	xx	xx	322	326			326		2/4	
D2S165	7.8	158		xx	xx	151	153	xx	xx	158	160	154	160			1/4	
D2S367	14.9	307	313	xx	xx	313	323	xx	xx	305	323	323	325	315		1/5	
D2S391	11.4	146	151	xx	xx	152		xx	xx	146	149	146	148	142		2/5	
D2S337	5.5	287	306	xx	xx			xx	xx	296						1/2	
D2S285	7.5	91	93	xx	xx	95	104	xx	xx	89	93	93	106	91		1/5	
D2S286	8.8	83	87	xx	xx			xx	xx			91	98			0/2	
D2S139	8.7	115	124	xx	xx	126		xx	xx	108	114	108	128	115	128	1/5	
D2S113	12.5	214	223	xx	xx	224		xx	xx	223	225			208	220	1/4	
D2S160	10.7	210		xx	xx	212		xx	xx	212		208	210	210	212	3/5	
D2S347	12.8			xx	xx	288	291	xx	xx	267						1/2	
D2S112#		n/a	n/a	140	142	n/a	n/a	140	148	140	146	146	148	140	148	0/5	
D2S368	7.4			xx	xx			xx	xx							failed	
D2S151	11.6			xx	xx			xx	xx					227	233	0/1	
D2S142	16.3	238	240	xx	xx	239		xx	xx	238		240		238	240	3/5	
D2S284#		234	238	234	238	238	244	234	244	234		234		234		3/7	
D2S111#		126	142	126		126	142	130		n/a	n/a	126	136	138		3/6	
D2S326	9.2			xx	xx			xx	xx							failed	
D2S364	8.7	236	240	xx	xx			xx	xx	232	240	226	248			0/3	
D2S117	10.2			xx	xx			xx	xx					198		1/1	
D2S325	8.4	156	173	xx	xx	161	163	xx	xx			165	174	155	163	0/4	
D2S164	9.1			xx	xx			xx	xx							failed	
D2S126	13.4	298	313	xx	xx	308	310	xx	xx	298	308	298		298	310	1/5	
D2S2158#		235	243	243	249	235	241	237	241	235		235	239	231		2/7	
D2S396	6.2			xx	xx			xx	xx							failed	
D2S206	10.6	150		xx	xx			xx	xx	141		147	149	141	145	2/4	
D2S338#	7.1	279	291	285	291	273	277	279		279		289		277	279	3/7	(ABI 1/2)
D2S125		96		xx	xx			xx	xx			99	103	85	87	1/3	

Results shown are with RG marker

* Results for unaffected relatives not shown

Table 11 - Genotyping Data (Continued)

Chromosome 3																	
Marker	**cM**	**III1**	**III1**	**IV1**	**IV1**	**2C**	**2C**	**303**	**303**	**905**	**905**	**1102**	**1102**	**1303**	**1303**	**Status**	
D3S1297	13.3	346	354	xx	xx	354		xx	xx	348	352	348		354	356	2/5	
D3S1304	13.8			xx	xx			xx	xx							failed	
D3S1263	7.2	180	190	xx	xx	192	197	xx	xx	188	192	190	207	196	200	0/5	
D3S1293#*	7.6	122	138	124	132	124?	134	122	124	132		124	132	124	130	1-2/7	(ABI failed)
D3S1266	11	289	291	xx	xx	293	299	xx	xx	289	291	289	299	289	293	0/5	
D3S1298	11.6			xx	xx			xx	xx							failed	
D3S1289	8.4	207	209	xx	xx	208	214	xx	xx	210	212	198	206	210	214	0/5	
D3S1300	11.8	243	249	xx	xx	231		xx	xx	231	253	247		243	253	2/5	
D3S1285	7.1	234	238	xx	xx			xx	xx	238		236	242			1/3	
D3S1566	20.8	151	155	xx	xx	155	160	xx	xx	155	168	155	162	155	157	0/5	
D3S3681#		226	228	226	242	210	238	236	242	210	244	236	240	226	238	0/7	
D3S1271	11	92		xx	xx	96	98	xx	xx	95	97	94	98	92		2/5	
D3S1278	8.2	233	237	xx	xx	238		xx	xx			230	246	244	250	1/4	
D3S1267	9	108	112	xx	xx			xx	xx	98	116	114	116	110	118	0/4	
D3S1292	12.3	117	141	xx	xx	116		xx	xx	117	127	130	135	139	141	1/5	
D3S1569	9.6	164	172	xx	xx			xx	xx	156	158	164	166	154		1/4	
D3S1279	9.9	263		xx	xx	268		xx	xx	270	274	268	279	268	270	2/5	
D3S1614	12.3	117	121	xx	xx	109	119	xx	xx	110	116	116		109	117	1/5	
D3S1565	13.5	180	184	xx	xx			xx	xx			180	182			0/2	
D3S1262	9	121	124	xx	xx	116	122	xx	xx	115		111	113	114	116	1/5	
D3S1580	15.6	228	232	xx	xx			xx	xx	234		228	232	224		2/4	
D3S3562#		222	228	220	224	222		220	228	220		222	226	220	222	2/7	
D3S1311		139	148	xx	xx	139	148	xx	xx	139	150	144	146	146	148	0/5	

Results shown are with RG marker

* Results for unaffected relatives not shown

Table 11 - Genotyping Data (Continued)

Chromosome 4																	
Marker	**cM**	**III1**	**III1**	**IV1**	**IV1**	**2C**	**2C**	**303**	**303**	**905**	**905**	**1102**	**1102**	**1303**	**1303**	**Status**	
D4S412	21.3	157		xx	xx	162	164	xx	xx	157		157	164	157	164	2/5	
D4S394#*		205		201	203	203	207	197	207	201	215	205	207	197	205	1/7	
D4S403	9.3	177	181	xx	xx	169	179	xx	xx	169	177	177	181	177	179	0/5	
D4S419	10.3	223		xx	xx			xx	xx							1/1	
D4S1546#		148		148		148?	152	152	154	148?	152	152?		152		3-6/7	
D4S3013#		152		148		148		152	160	144	152	148	156	144	156	3/7	
D4S391	14.3			xx	xx			xx	xx					215	227	0/1	
D4S405	8.3			xx	xx			xx	xx					280	295	0/1	
D4S428	7.6	193	197	xx	xx	193		xx	xx	191	193	187	191	187	197	1/5	
D4S398	7.7			xx	xx			xx	xx					134		1/1	
D4S392	15.3	90	92	xx	xx	88	100	xx	xx	94		98	100	96	98	1/5	
D4S2963#		252		246		246	252	252		252	256	248	250	252	254	3/7	
D4S1534	5.2	157		xx	xx	153		xx	xx	149	157	158	162	155	158	2/5	
D4S414	7.1	241		xx	xx	237		xx	xx			231	235	234	239	2/4	
D4S1572	10.4	199		xx	xx	205		xx	xx	203		191	197	193	201	3/5	
D4S406	6.7			xx	xx			xx	xx			247	249			0/1	
D4S402#	7.1	293	307	287	293	287		309	317	287	315	287	293	305	309	1/7	(ABI 4/5)
D4S1575	19.6	290	292	xx	xx	295		xx	xx	293	296	291	293	291		2/5	
D4S1579#		146	154	146		150		140	146	146	154	146		146		4/7	
D4S2939#		153?	157	153		157		153?	157	143	149	143	157	143	157	2-4/7	
D4S424	10.8			xx	xx			xx	xx	193						1/1	
D4S413	12.1	282	309	xx	xx	293	319	xx	xx	307		292	297	284	317	1/5	
D4S1597	15.1			xx	xx			xx	xx			289	291			0/1	
D4S2992#		240	242	240		246	254	250		242	254	250	252	248	250	2/7	
D4S415	12.4	263		xx	xx			xx	xx	283	287	284	290	264	267	1/4	
D4S1535#	14.2	179	191	191		183		191	199	191	193	185	191	179		3/7	(ABI failed)
D4S426		161		xx	xx	161	166	xx	xx	161		157	166	159	170	2/5	

Results shown are with RG marker

* Results for unaffected relatives not shown

Table 11 - Genotyping Data (Continued)

Chromosome 5																	
Marker	**cM**	**III1**	**III1**	**IV1**	**IV1**	**2C**	**2C**	**303**	**303**	**905**	**905**	**1102**	**1102**	**1303**	**1303**	**Status**	
D5S406	7.6	168	180	xx	xx	178	182	xx	xx	168	172	168		168	176	1/5	
D5S630	9			xx	xx	346		xx	xx	323	340	327	344	330	352	1/4	
D5S416	12.7	283		xx	xx			xx	xx			283				2/2	
D5S419	12.9	268	270	xx	xx			xx	xx			264	270	264	270	0/3	
D5S426	7.2			xx	xx	286	290	xx	xx			272	280	282	294	0/3	
D5S418	5.9	215		xx	xx			xx	xx	207	213	213	215	207	209	1/4	
D5S407	10.2	95	101	xx	xx	95	101	xx	xx	87	95	89	99	83	97	0/5	
D5S647	6.2	333	341	xx	xx			xx	xx	339		337	339			2/3	
D5S424	15.9	209	211	xx	xx	211	215	xx	xx	215		205	209	209	217	1/5	
D5S641#		271	275	279		265	269	253	267	271	275	267	269	263	271	1/7	
D5S428	10.4	246	248	xx	xx			xx	xx	246	252	242	248	242	246	0/4	
D5S644	8.8	94	96	xx	xx			xx	xx							0/1	
D5S433#*	8.2	216		216		206	220	212	216	190	208	206	218	204	216	2/7	(ABI 2/3)
D5S421	8.6	176	181	xx	xx	168	181	xx	xx	166		175	179	166	179	1/5	
D5S471	11.8	238		xx	xx	248		xx	xx	248		246	248			3/4	
D5S393	7	149		xx	xx			xx	xx							1/1	
D5S436	6.7	237	247	xx	xx	239	241	xx	xx	247	253	241	245	237	243	0/5	
D5S673	7.4	297	301	xx	xx			xx	xx	299	301	299	301	293	299	0/4	
D5S422#	13.1	125$		5	6	n/a	n/a	3	4	3		3		1	2	3/6	(ABI 3/4)
D5S400	4.9	227	231	xx	xx	223	227	xx	xx	227		217	221	226	228	1/5	
D5S429	13.9	255	279	xx	xx	268	279	xx	xx			264	268	268		1/4	
D5S408		258	260	xx	xx	250	260	xx	xx	260		250	260	256	262	1/5	

Results shown are with RG marker

* Results for unaffected relatives not shown

$ Genotype with RG failed. Showing ABI result

Table 11 - Genotyping Data (Continued)

Chromosome 6																
Marker	**cM**	**III1**	**III1**	**IV1**	**IV1**	**2C**	**2C**	**303**	**303**	**905**	**905**	**1102**	**1102**	**1303**	**1303**	**Status**
D6S344	12.9			xx	xx			xx	xx							failed
D6S309	5	306	312	xx	xx	312		xx	xx	308	314	302	308	312	316	1/5
D6S470	12.5	120	130	xx	xx	120	127	xx	xx	120	130	121	128			0/4
D6S289	6.2	165	171	xx	xx	169		xx	xx	161	171	169		163	169	2/5
D6S422	7.9	307	313	xx	xx	295	301	xx	xx	303	305	295	303	291	299	0/5
D6S276	13.3	205	223	xx	xx	197	215	xx	xx	197	205	221		221	223	1/5
D6S426	6.4	211	217	xx	xx	203		xx	xx	207	217	203	215	211	217	1/5
D6S271	11.8	129		xx	xx	128		xx	xx	129		128		129		5/5
D6S282#		114	122	114	120	112	120	110	116	116	118	108	120	108	120	0/7
D6S257	20.2	174	176	xx	xx	172	176	xx	xx	168	170	176	178	166	178	0/5
D6S460#		148	160	146	152	144		156	158	144	150	144		154	158	2/7
D6S462	9.3	108	110	xx	xx	107		xx	xx			105		108	110	2/4
D6S434	11.7	210	220	xx	xx	206		xx	xx	202	206	204	206	206	210	1/5
D6S287	9.7	128	130	xx	xx	129		xx	xx	127	129	121	127	127	129	1/5
D6S262	5.2			xx	xx			xx	xx							failed
D6S292	9.1	151	154	xx	xx			xx	xx					153	164	0/2
D6S308	8.6	337		xx	xx	331	337	xx	xx	335	339	333	337	337		2/5
D6S441	12.3			xx	xx	180		xx	xx	168						2/2
D6S305	10	265	271	xx	xx	250	273	xx	xx	250	275	261	269	261	263	0/5
D6S264	22.6	111	113	xx	xx	113		xx	xx	107	111	113		112	118	2/5
D6S446#*		201		201		201	209	201	203	201		203	209	203	209	3/7
D6S281				xx	xx			xx	xx					133	137	0/1

Results shown are with RG marker

* Results for unaffected relatives are not shown

Table 11 - Genotyping Data (Continued)

Chromosome 7																	
Marker	**cM**	**III1**	**III1**	**IV1**	**IV1**	**2C**	**2C**	**303**	**303**	**905**	**905**	**1102**	**1102**	**1303**	**1303**	**Status**	
D7S531	3.9	283		xx	xx	278	285	xx	xx	287	289	284		278	283	2/5	
D7S517	8.7	244	254	xx	xx	248	258	xx	xx	252	256	248	250	248	251	0/5	
D7S513	11.8			xx	xx	178	187	xx	xx	187	189			175	187	0/3	
D7S507#	6.3	158		n/a	n/a	101$		150	158	158	166	158		150	162	3/6	(ABI 3/4)
D7S493	7.8	203	208	xx	xx			xx	xx			222	226	222	224	0/3	
D7S516	13.2			xx	xx			xx	xx	315	319			307	313	0/2	
D7S484	5.1	104	112	xx	xx	104		xx	xx	104	108	104		106	112	2/5	
D7S510	12.4	87	91	xx	xx	92		xx	xx	89	91	87	94	87	89	1/5	
D7S519	25.2	261	265	xx	xx	257	268	xx	xx	263	266	261	262	262	268	0/5	
D7S2483#		121	125	121	125	121		119	129	125	129	125		125		3/7	
D7S669	16.2			xx	xx			xx	xx	179		185		179		3/3	
D7S2540#		197	203	209		197	211	209		197?	199	207?	209	197	209	2-4/7	
D7S657	9.8	243		xx	xx	256	258	xx	xx	251	258	260		248	260	2/5	
D7S515	13.2			xx	xx			xx	xx							failed	
D7S486	10.2	224	233	xx	xx	217	228	xx	xx	230	232	226	229	227	230	0/5	
D7S530	3.5			xx	xx			xx	xx			107	110			0/1	
D7S640	11.2	119	121	xx	xx	119		xx	xx			117	125	115	127	1/4	
D7S684	15.2	335		xx	xx			xx	xx	341	347			338	347	1/3	
D7S661#		262	270	270?	272	270		270?	272	n/a	n/a	270	280	264	272	1-3/6	
D7S636	16.1	137	154	xx	xx	165	169	xx	xx			146	150	133	148	0/4	
D7S2462#		129	139	133?	135	129		137		135	139	129	139	131	135	2-3/7	
D7S550		311	313	xx	xx	321	323	xx	xx	313	317	317	321	319	323	0/5	

Results shown are with RG marker

$ Genotype with RG failed. Showing ABI result

Table 11 - Genotyping Data (Continued)

Chromosome 8																
Marker	**cM**	**III1**	**III1**	**IV1**	**IV1**	**2C**	**2C**	**303**	**303**	**905**	**905**	**1102**	**1102**	**1303**	**1303**	**Status**
D8S504	8.5	140	142	xx	xx	139	148	xx	xx	137	140	139		131	139	1/5
D8S277	8	159		xx	xx			xx	xx	149	159					1/2
D8S550	19.1	190	206	xx	xx	188	192	xx	xx	192	206	184	188	190	212	0/5
D8S1731#*		219	221	225		231?		221	233	225	235	217	229	215	235	1-2/7
D8S258	24	144	148	xx	xx	148		xx	xx	152		144	148			2/4
D8S1839#		177	179	183?		179		179		177?	181	177	179	179	187	2-3/7
D8S283	10.5			xx	xx			xx	xx					103	108	0/1
D8S285	8.3	313	316	xx	xx	309	324	xx	xx	313	318	320		308		2/5
D8S260	10.1	199	201	xx	xx	203		xx	xx	193	209	199	207	199	209	1/5
D8S279	14	236	240	xx	xx	229		xx	xx	239	241	229	237	235	249	1/5
D8S270	13	109	111	xx	xx	102	113	xx	xx	100		103		104	105	2/5
D8S556	14.1			xx	xx			xx	xx							failed
D8S1830#		163	173	163	173	n/a	n/a	167	175	175		149		149		3/6
D8S514	12.5			xx	xx	222	224	xx	xx	218	220	216	218	216	222	0/4
D8S284	12.4			xx	xx			xx	xx	269		294		281	292	2/3
D8S272				xx	xx			xx	xx							failed
D8S534#		174	204	204		196		198	208	194	206	192		198	204	3/7
# Results shown are with RG marker																
* Results for unaffected relatives not shown																

Table 11 - Genotyping Data (Continued)

Chromosome 9																	
Marker	**cM**	**III1**	**III1**	**IV1**	**IV11**	**2C**	**2C**	**303**	**303**	**905**	**905**	**1102**	**1102**	**1303**	**1303**	**Status**	
D9S288	9.3	141		xx	xx	132	136	xx	xx	132	143	145	147	140	142	1/5	
D9S286	16.9	139	141	xx	xx	143	153	xx	xx	139		139	155	139	161	1/5	
D9S269#		173	175	179	181	173	181	173	177	169	185	165	171	173	181	0/7	
D9S157	9.1	223	225	xx	xx	223	227	xx	xx	227	235	227	235	227	235	0/5	
D9S1679#		139	141	139		135	139	135	139	137	139	135	137	135	141	1/7	
D9S171	7.1	173	177	xx	xx	167		xx	xx	167		160	173	167		3/5	
D9S161	14.8	120		xx	xx	119	130	xx	xx	132		120	130	120	134	2/5	
D9S273	4.5	207	211	xx	xx	209	213	xx	xx	208	211	208	211	205	215	0/5	
D9S175	13.9			xx	xx	258	276	xx	xx	259	276	258	260	266	274	0/4	
D9S167	11.1	321		xx	xx			xx	xx	310				318		3/3	
D9S283	10	90	102	xx	xx	90	100	xx	xx	92	100	95	106	89	95	0/5	
D9S287	20.4	297	299	xx	xx	295		xx	xx	297	299	295	299	295	297	1/5	
D9S1784#		180	182	182?	184?	184		184		184	190	178	184	178	184	2/7	
D9S279	21.3	224	226	xx	xx	229	230	xx	xx	226	230	226	230	231	233	0/5	
D9S1872#*		2	4	n/a	n/a	n/a	n/a	2	3	2	3	2	3	1	4	0/5	
D9S290	7.2	244	252	xx	xx	242	244	xx	xx	242	244	244	248	246	248	0/5	
D9S164	15.2	90	94	xx	xx	94	96	xx	xx	93	95	94	96	92	96	0/5	
D9S66#		4	6	6		6		7		2	12	7	11	6	7	3/7	
D9S1818#		151	153	151	153	151		157		155	157	157		151	157	3/7	
D9S67#		2	4	2	4	2	4	2	4	3	5	n/a	n/a	2	4	0/6	
D9S1826#*		135		135		139	151	139		135		135		139	145	5/7	
D9S158#		219	227	227	229	221	233	219	223	219	221	229		223	225	1/7	(ABI 2/3)

Results shown are with RG marker

* Results for unaffected relatives not shown

Table 11 - Genotyping Data (Continued)

Chromosome 10																
Marker	**cM**	**III1**	**III1**	**IV1**	**IV1**	**2C**	**2C**	**303**	**303**	**905**	**905**	**1102**	**1102**	**1303**	**1303**	**Status**
D10S249	11.3	118	122	xx	xx	117	129	xx	xx	113	121	115	129			0/4
D10S591	5.2	325		xx	xx	330	331	xx	xx	324	327	327		326		3/5
D10S1713#*		249		243	249	247	251	249		249		251	255	247	249	3/7
D10S189	11.9	186		xx	xx	183		xx	xx	182		180		188		5/5
D10S1751#*		251		247		251		247	251	247	251	247		247	251	4/7
D10S547	9.2	239		xx	xx			xx	xx	237	239	237	239			1/3
D10S191	8.7	126		xx	xx			xx	xx	133	137			137	139	1/3
D10S548	6	187		xx	xx	185		xx	xx	183	187	183		183	187	3/5
D10S197	9.7	167		xx	xx	171		xx	xx	169	171	169		168	170	3/5
D10S208	9.3	180	188	xx	xx			xx	xx	184				178	182	1/3
D10S220	10.6	113		xx	xx			xx	xx					113		2/2
D10S1790#		187	191	189	191	185	187	193	195	n/a	n/a	187	195	185	189	0/6
D10S1652#		172		170	172	170		n/a	n/a	174		170		174?	178?	4/6
D10S561	11.4	286		xx	xx	286		xx	xx			285				3/3
D10S581#		139	145	139	145	135	139	137	141	n/a	n/a	133	139	137	143	0/6
D10S537	9.7	144	156	xx	xx	150	156	xx	xx	152		144		144		3/5
D10S201	13.5	297	304	xx	xx	285	291	xx	xx	293	297	285	306	287	291	0/5
D10S583	8.4			xx	xx			xx	xx			201		203	205	1/2
D10S192	3	254	256	xx	xx			xx	xx							0/1
D10S543#		127	141	137	139	137	141	141?	145?	n/a	n/a	133	141	135	137	0/6
D10S597	10.5	286		xx	xx	286		xx	xx	276	288	286		284	286	3/5
D10S190	9.9	208		xx	xx			xx	xx							1/1
D10S587	8.4	100		xx	xx	100	101	xx	xx	100	102	95	102	102		2/5
D10S217	14.7	98	102	xx	xx	98	102	xx	xx	96	102	102		100	104	1/5
D10S212		191		xx	xx	195		xx	xx					191	197	2/3

Results shown are with RG marker

* Results for unaffected relatives not shown

Table 11 - Genotyping Data (Continued)

Chromosome 11																	
Marker	**cM**	**III1**	**III1**	**IV1**	**IV1**	**2C**	**2C**	**303**	**303**	**905**	**905**	**1102**	**1102**	**1303**	**1303**	**Status**	
D11S922	10.6			xx	xx			xx	xx			133	139	130	155	0/2	
D11S1338	7.6	257	263	xx	xx			xx	xx	257	263	263	265	257	265	0/4	
D11S902	13.3			xx	xx			xx	xx					150		1/1	
D11S904	11.5	192	200	xx	xx	188	190	xx	xx	196		200		202		3/5	
D11S935	8.9	196	200	xx	xx	198	206	xx	xx	196	204	206		196	206	1/5	
D11S905	5.9	267	271	xx	xx			xx	xx							0/1	
D11S1313	6.5			xx	xx			xx	xx	189						1/1	
D11S987	7.4	117	121	xx	xx	120	122	xx	xx	119		114	126	115	119	1/5	
D11S1314	6.9			xx	xx			xx	xx					99		1/1	
D11S937	4.6	158	160	xx	xx			xx	xx			158				1/2	
D11S901	7.3	313		xx	xx	311		xx	xx	306	321	305	313	314	321	2/5	
D11S1358	6.3	288		xx	xx			xx	xx	293		291	293			2/3	
D11S898	9.5	153		xx	xx	151		xx	xx	145	149	145	149	145	151	2/5	
D11S908	13.6	178		xx	xx	174	178	xx	xx	174	176	174	178	178		2/5	
D11S1345#*	5.7	236		234	236	236		232	234	232	236	236		236		4/7	(ABI 4/5)
D11S1353#*		2		5	6	3	4	4	5	3	4	4	5	2	4	1/7	
D11S4158#*		1	5	1	3	1	4	3	4	3		3	5	3	4	1/7	
D11S934#*	13.1	186	190	182	184	180	186	180		182	184	180	182	180		2/7	(ABI 4/5)
D11S1320	8	261	265	xx	xx	259	261	xx	xx	259	261	259		261	263	1/5	
D11S968		141	152	xx	xx	143	152	xx	xx	148	150	143	147	148	152	0/5	
# Results shown are with RG marker																	
* Results for unaffected relatives not shown																	

Table 11 - Genotyping Data (Continued)

Chromosome 12																
Marker	**cM**	**III1**	**III1**	**IV1**	**IV1**	**2C**	**2C**	**303**	**303**	**905**	**905**	**1102**	**1102**	**1303**	**1303**	**Status**
D12S352	13.8	158	160	xx	xx			xx	xx	155	157	158	160	155	158	0/4
D12S99	8.3	265	275	xx	xx	273	281	xx	xx	267	269	273	288	271		1/5
D12S336	9.8	120		xx	xx	112	119	xx	xx	112	120	112	120	112		2/5
D12S364	5.4			xx	xx			xx	xx					311	313	0/1
D12S310	19			xx	xx			xx	xx					245		1/1
D12S1669#		105	109	105	109	105	109	105	113	103		101	105	99	115	1/7
D12S1591#		268	274	268?	272	268	274	268		268		264	272	268	276	2-3/7
D12S345	8.3	214	220	xx	xx	235	239	xx	xx	211	219	215	227	217	227	0/5
D12S85	4.2	116	118	xx	xx	104		xx	xx	118	128	104	122	120	122	1/5
D12S368	8.2	200	206	xx	xx	200	208	xx	xx	200		200		200	210	2/5
D12S83	13.7			xx	xx	106		xx	xx					101	111	1/2
D12S1585#		2	4	1	4	2		2	4	2	4	1	4	2		2/7
D12S326	9.3	205	223	xx	xx	221	227	xx	xx	217	224	204	220	205		1/5
D12S351	10	153	155	xx	xx			xx	xx	157	161	153		157	161	1/4
D12S346	7.5	193	203	xx	xx	188	203	xx	xx	189	201	185	195	181	183	0/5
D12S78	12.8	177	185	xx	xx	185	189	xx	xx			171	200	173	189	0/4
D12S79	9	158	159	xx	xx	156	157	xx	xx	157	163	157	169	163	171	0/5
D12S86	13.5			xx	xx			xx	xx					135	146	0/1
D12S324	14	240	244	xx	xx	240	244	xx	xx	240		244		244		3/5
D12S367		321	325	xx	xx	327		xx	xx	323	333	321	325	321	327	1/5
# Results shown are with RG marker																

Table 11 - Genotyping Data (Continued)

Chromosome 13																	
Marker	**cM**	**III1**	**III1**	**IV1**	**IV1**	**2C**	**2C**	**303**	**303**	**905**	**905**	**1102**	**1102**	**1303**	**1303**	**Status**	
D13S1236#		128		126		122	130	126	128	126	130	122		128	130	3/7	
D13S175#	6.8	105	113	113?		99	103	105	111	105	107	103	105	101		1-2/7	(ABI 1/3)
D13S1275#		202		202		206	214	202		206		208		206		6/7	
D13S1243#		246	252	246	252	250		250	252	252	256	248	250	246	250	1/7	
D13S217	8.3	248	251	xx	xx	249	254	xx	xx	249	250	252	256	245	247	0/5	
D13S171	16.2	179	183	xx	xx	179	183	xx	xx	179	189	179	183	190	192	0/5	
D13S218#		191		191		191	195	187	193	191	195	189	195	191		3/7	
D13S263	9.5	163	167	xx	xx	163		xx	xx	156	162	163	165	151	153	1/5	
D13S153	9.3	92	96	xx	xx			xx	xx	101		95	98	95	98	1/4	
D13S156	8.6	275	283	xx	xx	278	282	xx	xx	280	284	278	282	280	282	0/5	
D13S170	7.1			xx	xx	143	157	xx	xx			157		139	159	1/3	
D13S265	9.2	103	107	xx	xx	107		xx	xx	106	110	106		101	106	2/5	
D13S159	7.9	152	164	xx	xx	151	164	xx	xx	161		151	181	155	161	1/5	
D13S158#*	6.5	107		101	107	105	109	103	107	107	113	99	111	107		2/7	(ABI 4/5)
D13S174#*		177	193	177	191	177	183	183	187	177	?	177	193	183	191	0-1/7	
D13S173#	18.4	172		170		170	174	168	174	172	174	150		172	174	3/7	(ABI 1/4)
D13S1265#		132		146		134	150	128		128	142	152		150		5/7	
D13S895#		6		4	5	3		3	4	3	4	3		2	3	3/7	
D13S1315#		160		172	174	172		172		172	176	178		162		5/7	
D13S285		96		xx	xx	99		xx	xx	94	101	101	112	101		3/5	
D13S1295#		116		108	116	116		108	116	108	112	108		102	108	3/7	

Results shown are with RG marker

* Results for unaffected relatives not shown

Table 11 - Genotyping Data (Continued)

Chromosome 14																
Marker	**cM**	**III1**	**III1**	**IV1**	**IV1**	**2C**	**2C**	**303**	**303**	**905**	**905**	**1102**	**1102**	**1303**	**1303**	**Status**
D14S261	7.1	299		xx	xx			xx	xx	291	293	270				2/3
D14S283	12.8	125	148	xx	xx			xx	xx			124				1/2
D14S80	13.3	144		xx	xx	156		xx	xx			151	152			2/3
D14S70	7	101	106	xx	xx			xx	xx	105	106			103	107	0/3
D14S288	7.8	187	199	xx	xx	199	201	xx	xx	199		199	203	182	200	1/5
D14S1013#		225	233	219	239	237	239	233	239	235	237	229	233	219	237	0/7
D14S276	14.3	245		xx	xx			xx	xx	239	243			242		2/3
D14S63	4.9	179	183	xx	xx	179	187	xx	xx	181	185	179	189	183		1/5
D14S258	13			xx	xx	193	195	xx	xx	197	199	199		196		2/4
D14S74	8.9			xx	xx	300		xx	xx							1/1
D14S68	7.8	321	325	xx	xx			xx	xx	325	328	321	328			0/3
D14S280	11.3	242	244	xx	xx	242		xx	xx	242	244	242	246	242		2/5
D14S65	11.5	142	146	xx	xx	144		xx	xx	125	146	139	148	148	150	1/5
D14S78	10.7	225	233	xx	xx	229	231	xx	xx	219	225	229		225		2/5
D14S292		85	87	xx	xx	92		xx	xx	85	87	94		85	87	2/5
# Results shown are with RG marker																

Table 11 - Genotyping Data (Continued)

Chromosome 15																	
Marker	**cM**	**III1**	**III1**	**IV1**	**IV1**	**2C**	**2C**	**303**	**303**	**905**	**905**	**1102**	**1102**	**1303**	**1303**	**Status**	
D15S128	14.7	201	207	xx	xx	201	203	xx	xx	205	207	203	205	197	201	0/5	
D15S165	30.8			xx	xx			xx	xx							failed	
D15S1031#		299?	301?	301	313	303	307	291	301	303	307	n/a	n/a	303		1-2/6	
D15S1007#*		185	187	163	175	175	187	179	185	169	173	173		179	181	1/7	
D15S1040#*		199		201	205	199		199	205	201	209	199		197	207	3/7	
D15S118#*		228		220	230	220	228	226		220	222	226		228		4/7	
D15S146#*		217	223	221		221	223	221	223	219	227	221		219	223	2/7	
D15S126	5.7	188	200	xx	xx			xx	xx	210		202		188	200	2/4	
D15S117	10.6			xx	xx			xx	xx	317	323	332				1/2	
D15S153	6.9	252	257	xx	xx	262		xx	xx	261		238	261	252	262	2/5	
D15S216#*		233		225	233	231	233	231	233	231	235	231		233		3/7	
D15S131#*	9.4	1	2	1	?	1		5	11	1	9	8	12	11		2-3/7	(ABI 2/2)
D15S980#*		185	197	185	189	185	209	189	193	181	187	179	193	189	191	0/7	
D15S205	7.9			xx	xx	144	150	xx	xx			127	154	142	150	0/3	
D15S127	12.7			xx	xx			xx	xx			134	145	113	145	0/2	
D15S130	11.6	284	294	xx	xx			xx	xx	284	288	282	294	286	292	0/4	
D15S120		170	174	xx	xx	162	170	xx	xx	170		160	174	168		2/5	
Chromosome 16																	
Marker	**cM**	**III1**	**III1**	**IV1**	**IV1**	**2C**	**2C**	**303**	**303**	**905**	**905**	**1102**	**1102**	**1303**	**1303**	**Status**	
D16S423	7.6			xx	xx	137		xx	xx	137		137	141			2/3	
D16S407	10.2	262	277	xx	xx			xx	xx			271	279			0/2	
D16S405	16.7			xx	xx	130	138	xx	xx	134		134	136			1/3	
D16S3036#		154	170	162	166	164	170	160	166	154		154	168	154	166	1/7	
D16S420	4.8	262		xx	xx	255	272	xx	xx	266		257	266	267		3/5	
D16S401	12.8	166	174	xx	xx	172	176	xx	xx	172	176	170	172	166	176	0/5	
D16S411	11.5	216		xx	xx	212	220	xx	xx	216	220	202		202	224	2/5	
D16S415	8	211	222	xx	xx	221	227	xx	xx	218	222	216	220	209		1/5	
D16S503	11.2	294	300	xx	xx			xx	xx			298	300	300		1/3	
D16S515	10.4	328	332	xx	xx	327	331	xx	xx			326	348			0/3	
D16S516	7.3			xx	xx			xx	xx			245	253	246	253	0/2	
D16S511	19.5			xx	xx			xx	xx							failed	
D16S3037#		205	211	205	209	203	211	205	209	205	209	n/a	n/a	205	209	0/7	
D16S520				xx	xx			xx	xx	151						1/1	

Results shown are with RG marker

* Results for unaffected relatives not shown

Table 11 - Genotyping Data (Continued)

Chromosome 17																	
Marker	**cM**	**III1**	**III1**	**IV1**	**IV1**	**2C**	**2C**	**303**	**303**	**905**	**905**	**1102**	**1102**	**1303**	**1303**	**Status**	
D17S849	17.1	253	256	xx	xx	255	259	xx	xx	256		253	254	253	255	1/5	
D17S938	7.4			xx	xx			xx	xx							failed	
D17S945	10.4			xx	xx			xx	xx					304	306	0/1	
D17S1879#		153	167	151	153	153?	155?	155		151	155	151		153	167	2/7	
D17S799	17.6	190	192	xx	xx	192		xx	xx	186	192	184	188	186		2/5	
D17S1843#*		199 $		199		n/a	n/a	185	205	189	203	183	203	189		3/6	
D17S925	5.9			xx	xx			xx	xx							failed	
D17S798	10.9	310		xx	xx			xx	xx	294	302			308	310	1/3	
D17S791	11.1			xx	xx			xx	xx							failed	
D17S787	7.7	137	141	xx	xx	139	141	xx	xx	139	158	137	141	139	141	0/5	
D17S808	9.3	340	344	xx	xx			xx	xx	344		332	344			1/3	
D17S949	14.7	220	226	xx	xx	215	223	xx	xx	215		214	220	215		2/5	
D17S802	10.8	178	188	xx	xx	194		xx	xx			184	188	189		2/4	
D17S784	11.6	235		xx	xx			xx	xx			233	235	228		2/3	
D17S928		84	88	xx	xx			xx	xx	84		91		84		3/4	
Chromosome 18																	
Marker	**cM**	**III1**	**III1**	**IV1**	**IV1**	**2C**	**2C**	**303**	**303**	**905**	**905**	**1102**	**1102**	**1303**	**1303**	**Status**	
D18S59	7.8	151	159	xx	xx	151	164	xx	xx	155	159	151	158	151	160	0/5	
D18S52#	9.1	120	124	120	124	n/a	n/a	122	124	120	122	124		120	128	1/6	(ABI 1/1)
D18S452	13.9	121	133	xx	xx	126	138	xx	xx	127	131	138	141	126	134	0/5	
D18S464	9	306	309	xx	xx	302	308	xx	xx	306	308	306		300	304	1/5	
D18S53	13.1	162	171	xx	xx	162	168	xx	xx	160	162	162	164	158	166	0/5	
D18S478	9.1	241	251	xx	xx	241	251	xx	xx	241	245	241	259	245	247	0/5	
D18S57	7.9	95	99	xx	xx	101		xx	xx	93	105	105	106	99	103	1/5	
D18S474	13.8	122	134	xx	xx	122	136	xx	xx	122	124	122	124	138		1/5	
D18S64	12	320	332	xx	xx			xx	xx			324	332	320		1/3	
D18S68	11	272	283	xx	xx			xx	xx	272	280	274	283			0/3	
D18S61	9.2	224	226	xx	xx			xx	xx	222	228			208	226	0/3	
D18S469	4			xx	xx			xx	xx			234		216	218	1/2	
D18S462	6.9	303	309	xx	xx			xx	xx	301		299		299	303	2/4	
D18S70		110	113	xx	xx	109	118	xx	xx	113		122		114	116	2/5	

Results shown are with RG marker

* Results for unaffected relatives not shown

$ Reaction failed. Genotype constructed from parents

Table 11 - Genotyping Data (Continued)

Chromosome 19																	
Marker	**cM**	**III1**	**III1**	**IV1**	**IV1**	**2C**	**2C**	**303**	**303**	**905**	**905**	**1102**	**1102**	**1303**	**1303**	**Status**	
D19S209	5.9	241	246	xx	xx	239		xx	xx	239	249	239	241			1/4	
D19S216	18.4	262	266	xx	xx	254		xx	xx	262		260	262	254	260	2/5	
D19S884#		222		222	224	230	234	234		n/a	n/a	234		224	234	3/6	
D19S916#		236	256	232	244	234	238	244	248	240	246	248	250	238	254	0/7	
D19S221	7.3	103		xx	xx			xx	xx							1/1	
D19S226	12.6	238		xx	xx	236	238	xx	xx	234	236	246	251	242	246	1/5	
D19S414	7.9	164		xx	xx	179	183	xx	xx	183	185	179	183	183		2/5	
D19S220	5.5	268	272	xx	xx	268	276	xx	xx	274	280	268		264	274	1/5	
D19S420	27	82	96	xx	xx	98		xx	xx	87	95	97	98	97	100	1/5	
D19S866#		183	199	183	199	183	185	183		183	201	189	203	191	211	1/7	
D19S924#*		197		207		201	203	205		201	203	201	207	197	203	3/7	
D19S418	9.8	85#		85#		94		xx	xx	89		92	95	90		5/6	(ABI 4/5)
D19S926#*		101	105	105?		99	107	95	113	103	107	95	107	109		1-2/7	
D19S210		174	176	xx	xx	174	176	xx	xx	182		174	182	176		2/5	
Chromsome 20																	
Marker	**cM**	**III1**	**III1**	**IV1**	**IV1**	**2C**	**2C**	**303**	**303**	**905**	**905**	**1102**	**1102**	**1303**	**1303**	**Status**	
D20S117	12.9	155	174	xx	xx			xx	xx					155	176	0/2	
D20S95	4.9			xx	xx	103	112	xx	xx	105	114	103	105			0/3	
D20S115	11	234	235	xx	xx	234	236	xx	xx	234	237	236		234	236	1/5	
D20S189	0.9	304	306	xx	xx	306	308	xx	xx	294	306	302		306	308	1/5	
D20S186	7.1	118	120	xx	xx	122	124	xx	xx	124		114		124		3/5	
D20S118	12.6			xx	xx	207	212	xx	xx	206	215	216		215	219	1/4	
D20S195	5.9	131	146	xx	xx	139	146	xx	xx	133	139			142		1/4	
D20S107	5.6	208	212	xx	xx	212		xx	xx	206	210	202	208	206		2/5	
D20S119	5.7	107	111	xx	xx	111	116	xx	xx	111	117	109	110	109		1/5	
D20S178	7.4	184		xx	xx	184	190	xx	xx	178		180	182	184		3/5	
D20S886#		145		151?	153?	141	147	145		145		143	153	145		4/7	
D20S196	8.7			xx	xx	280	288	xx	xx	282	286	277	284	260	284	0/4	
D20S100	13.1	217	221	xx	xx			xx	xx					215	219	0/2	
D20S173#	3.2	183	187	185		n/a	n/a	n/a	n/a	179	183	187?		185		2-3/5	(ABI failed)
D20S171				xx	xx			xx	xx			134		140		2/2	
# Results shown are with RG marker																	

Table 11 - Genotyping Data (Continued)

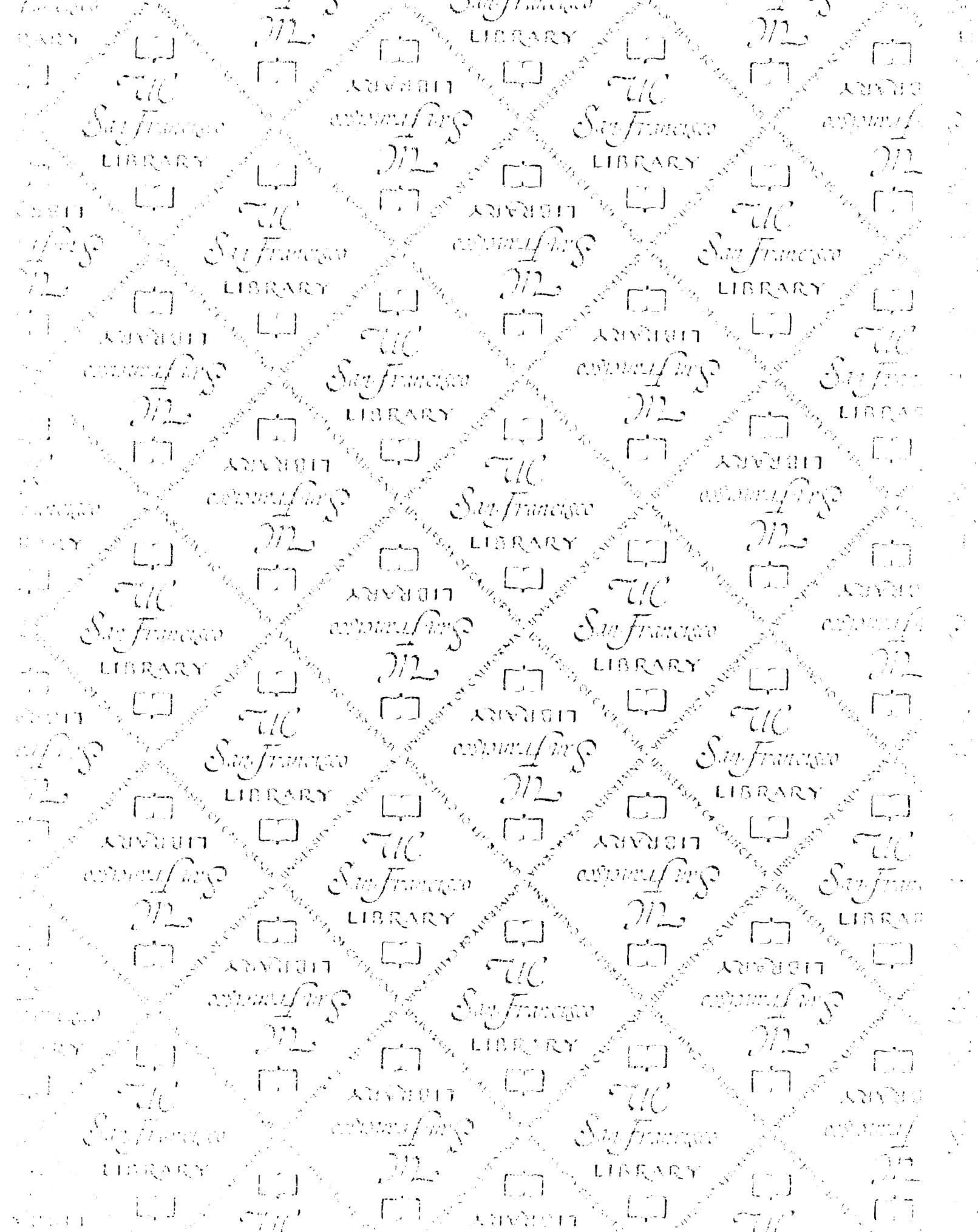
Chromosome 21																	
Marker	**cM**	**III1**	**III1**	**IV1**	**IV1**	**2C**	**2C**	**303**	**303**	**905**	**905**	**1102**	**1102**	**1303**	**1303**	**Status**	
D21S1256*#	13.8	184	186	182	190	182	184	176	182	182		184		182	188	2/7	(ABI 4/5)
D21S1253	7.6	271	279	xx	xx	279		xx	xx	273	279			275	279	1/4	
D21S263	6.7			xx	xx			xx	xx							failed	
D21S1252	12.6	139	159	xx	xx	157	161	xx	xx	144	169	143	156	143	156	0/5	
D21S266		154	174	xx	xx	157	171	xx	xx	157	163	157		153	157	1/5	
Chromosome 22																	
Marker	**cM**	**III1**	**III1**	**IV1**	**IV1**	**2C**	**2C**	**303**	**303**	**905**	**905**	**1102**	**1102**	**1303**	**1303**	**Status**	
D22S420	15.3	153		xx	xx			xx	xx	152						2/2	
D22S315	10.7	192	194	xx	xx	194		xx	xx	190	196					1/3	
D22S280	7.3	210	216	xx	xx	216	218	xx	xx	210	216	210	214	208	210	0/5	
D22S283	6.9	141	152	xx	xx	145	147	xx	xx	133	150	130	154	145	149	0/5	
D22S423	7.8			xx	xx	290		xx	xx	299				290	291	2/3	
D22S274		277	281	xx	xx	281	287	xx	xx	281	287	281	285	281		1/5	

* Results for unaffected relatives not shown

Results shown are with RG marker

Table 11 - Genotyping Data (Continued)

Chromosome X																
Marker	**cM**	**III1**	**III1**	**IV1**	**IV1**	**2C**	**2C**	**303**	**303**	**905**	**905**	**1102**	**1102**	**1303**	**1303**	**Status**
DXS1060	15	240		xx	xx	250		xx	xx	248	254	242	250	245	249	2/5
DXS987	11	215		xx	xx	215	218	xx	xx	206	211	206	212	216		2/5
DXS1226	5	356		xx	xx	359		xx	xx			346	353			2/3
DXS1202	9	217		xx	xx	218		xx	xx	206	219	218		202	216	3/5
DXS1214	9	284		xx	xx	281		xx	xx	282	286	282		283	286	3/5
DXS1068	9	253		xx	xx			xx	xx			246		250	254	2/3
DXS993	11	286		xx	xx	266		xx	xx	284		282	284	267	274	3/5
DXS1055	12	84		xx	xx	88		xx	xx	84		88	90	83	85	3/5
DXS991	9			xx	xx	322		xx	xx	323	327	322				2/3
DXS986	7	175		xx	xx	161		xx	xx	161	163	163		161	171	3/5
DXS990	12	123		xx	xx	123	127	xx	xx	121	125	127		124	127	2/5
DXS1106	10			xx	xx			xx	xx					113	117	0/1
DXS1001	13			xx	xx			xx	xx							failed
DXS1047	11	161		xx	xx			xx	xx			157		155	157	2/3
DXS1227		81		xx	xx	87	92	xx	xx	81		90		87		4/5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