MITOCHONDRIAL DNA SEQUENCE POLYMORPHISMS IN ARGENTINE POPULATION

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Introduction

Mitochondrial DNA sequencing, D-Loop Region HVI Segment, has been incorporated since 1993 in our laboratory in order to study maternal lineage. At the moment, we have studied 558 individuals to investigate maternal relationship. Most of the sequences have been obtained from blood samples, and others from bone, hairs, and soft tissue samples, for this reason mitochondrial DNA sequencing has become a tool of choice for forensic casework. The analysis of mitochondrial DNA sequence can be reported as a list of differences from the "Anderson Sequence " or the "Cambridge Reference Sequence " (CRS); most of them are base transitions; and in second term transversions; the presence of two bases at approximately equal intensities defined as "heteroplasmy", has also been observed. We studied the hypervariable region I of the D-Loop in order to analyze the DNA sequence polymorphism present in our population.

Materials and methods

We studied 279 unrelated Argentine individuals, most of them from Buenos Aires city. DNA from blood samples was extracted by Miller's method and total DNA from bone, hairs, and soft tissue samples by the Chelex® extraction method. PCR was performed for 34 cycles with 2µl of template, in a 9600 Perkin Elmer thermal cycler consisting of 1 min. at 94 C, 30 s. at 60 C and 30 s. at 72 C. PCR product was run in agarose gel and a plug was extracted to perform a single strand DNA PCR (Gyllenstein and Erlich). Sequencing reaction was done by Sanger's method with *Sequenase* 2 enzyme (USB) and radioactive ^{35}S. HVI region sequences analyzed were 390bp size, between 16023 and 16412 nucleotide positions.

Results

Mitochondrial sequence data show 127 polymorphic sites, respect to Cambridge Reference Sequence (CRS) and 200 different genotypes, for the HVI region studied. We calculated Genetic Identity: $P=\Sigma x^2$ (x=mitochondrial DNA sequence frequency) with a value of P=0.011 for HVI. We also determined Genetic Diversity $h=(1-\Sigma x^2).n/n-1$ (n=sample size) with a value of h= 0,9926. The Identity Probability is 1.1%.

Conclusions

These results confirm mitochondrial DNA sequencing as a useful tool for maternal lineage determination and the importance of this valuable information for forensic casework in human remains identification.