THE ANALYSIS OF CHALLENGING HUMAN REMAINS SAMPLES USING NOVEL RETROTRANSPOSABLE ELEMENT-BASED TECHNOLOGIES

J. Carlos Alvarez, Ph.D.[1], Jose Lorente, Ph.D.[1], Hiromi Brown, Ph.D.[2], Gina Murphy, M.S.[2], Jonathan Tabak, B.A.[2], Anne Montgomery, M.S.[2], and Sudhir K. Sinha, Ph.D.[2],
[1]Laboratorio Identificación Genética, Dpto. de Medicina Legal, Toxicología y Antropología Física FACULTAD DE MEDICINA
[2]InnoGenomics Technologies USA

Alternative nuclear DNA typing systems that can successfully type highly compromised samples with limited DNA quality and/or quantity are needed. The InnoTyper® 21 kit is a small amplicon DNA typing system for challenging forensic samples compatible with commonly used PCR/CE instrument platforms. The system uses a novel primer design to multiplex 20 ALU retrotransposon element bi-allelic markers, plus Amelogenin, with amplicon sizes ranging from 60-124 bp, thus making the assay highly sensitive for extremely degraded and/or low-level forensic samples, and enabling recovery of statistically discriminatory results from samples that would typically require mtDNA sequencing.

In conjunction with InnoTyper® 21, a next-generation DNA quantitation kit, InnoQuant® HY, can be used to more effectively target the amplification and to assess the presence of PCR inhibitors in the sample. This quantitation kit employs targets with large copy numbers (>1000 copies/genome), which provides high sensitivity while minimizing the effect of variation between individuals, enabling high reproducibility for low level samples. The results indicate the observed InnoQuant® HY DI values from the skeletal remains correlate well with the samples' condition and known environmental stresses, and allow more informative decisions to be made prior to PCR amplification.

The application of the InnoTyper® 21 and InnoQuant® HY systems to heavily degraded skeletal remain samples will be presented. A total of 4 exhumed specimens were analyzed with these new systems producing highly useful results in samples that previously yielded no results with autosomal STR and gender informative markers. The samples had different ages and preservation conditions. For example, sample 1 was from the exhumation of a corpse from a family of an historical person who died in the year 1887 (130 years). These analyses will be crucial to enable the use of the obtained nuclear DNA profiles as a reference in the identification process, as it had not previously been possible to obtain sufficient nuclear DNA results for comparison. In this case, mitochondrial DNA was not analyzed because the biological relationship in question was paternal. Another sample was a bone from a mass grave exhumed as part of an extensive project to identify victims of the Spanish Civil War (1936-1939). In this case, the sample was 80-years-old and, because of the specific circumstances of its location, was quite a challenge. The sample was buried near a stream in which successive floods subjected the sample to an increase in moisture, and the effects of this continuous flushing of the sample were evident in the badly damaged appearance with which the sample presented itself. Both the InnoQuant® HY and InnoTyper® 21 testing employed with these samples resulted in highly informative information, yielding interpretable nuclear DNA profiles from highly degraded remains for relationship testing.