B130 Optimized Extraction of Nuclear DNA From Hair Shafts

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The goals of this presentation are to describe a variation of an organic extraction used on samples obtained from the World Trade Center victims and reference samples from these victims to obtain nuclear DNA from hair shafts.

Extracting nuclear DNA from hair has mainly been performed if the root of the hair is present. Nuclear DNA typing on hair shaft material has not been very successful since it is known that the amount of nuclear DNA in hair shafts is low and that the keratinized cells present in hair as well as the hair pigments are inhibitors to the PCR reaction. mtDNA testing of hair shafts has been proven to be a more successful and very effective method but most laboratories are not equipped for mtDNA testing as it is time-consuming and of less discriminatory value. Attempts to extract nuclear DNA from hair shafts have recently been reported in the literature. However, results of these experiments indicate that hair shafts are poor sources of nuclear DNA and are generally not suitable for STR testing.

In response to the World Trade Center DNA identification project it was necessary to optimize a method for extracting DNA from hair shafts. Hair samples removed mainly from hairbrushes or combs had been submitted as personal reference samples. Toothbrushes and other personal effects were the preferred source of comparison of DNA but in several instances it was necessary to test the hair. Additionally, clumps of hair without adhering tissue had been recovered at the World Trade Center disaster site. In order to find a biological trace of as many victims as possible, these samples could not be left untested.

The hairs were cleaned when placed into 5% Tergazyme, sonicated for 15 minutes, rinsed in deionized water several times, and allowed to air dry. After the cleaning, the hair or hairs were examined and 5-10 hair shafts per extraction were cut into 2 mm lengths. The hair shafts were incubated in organic extraction buffer overnight at 56°C in thermal shakers at 14,000 RPM. If the hair had not dissolved, the mixture was transferred to a glass mortar and pestle and the sample was ground until it disappeared. The extraction method for the World Trade Center samples employed a phenol/chloroform/Microcon 100 combination procedure. DNA was quantified, amplified using Promega PowerPlex□16, and run on ABI 3100's.

This modified procedure for extraction of nuclear DNA from hair shafts has shown that the "last resort" of biological samples suitable for DNA extraction (hairs) may be used. Hair shafts are a viable source of nuclear DNA for human identification.

DNA Extraction, Nuclear DNA From Hair Shafts, Mass Disasters

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