A58 Comparative Evaluation of Methods to Remove Exogenous DNA From Tooth Samples

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After attending this presentation, attendees will have an understanding of the development of a decontamination method that is effective for the removal of nanogram quantities of exogenous DNA from tooth samples.

This presentation will impact the forensic science community by demonstrating the effectiveness of a decontamination method that will remove amounts of exogenous DNA from a tooth expected from normal handling by examiners not protecting the samples from contamination. Forensic laboratories will be able to adopt this method to prevent contamination of these types of samples.

This presentation will summarize the evaluation of several methods for the removal of exogenous DNA deposited during previous examination, handling, or comingling of forensic tooth samples. All treatments for tooth and bone decontamination, including the original Harris County Institute of Forensic Sciences (HCIFS) method, were tested by adding and attempting to remove increasing amounts of DNA from teeth. An optimal method for DNA removal was identified and subsequently validated for use on forensic casework tooth and bone samples submitted to the HCIFS laboratory.

DNA extracted from tooth samples can provide powerful information in both forensic casework and missing person identification. The presence of surface DNA contamination from previous examinations or comingling can provide misleading or confusing results, however. This study approached systematically the development of a method to effectively remove surfacecontaminating DNA by assessing a variety of removal methods in various combinations. These techniques included prolonged soaking in bleach and/or deionized water, rinsing with deionized water and/or ethanol, and exposure to UV irradiation.

Sixteen combinations of the techniques were tested for their ability to remove five levels of exogenous DNA, at 5ng, 10ng, 25ng, 50ng, and 100ng. One combination, treatment “M,” comprised of 10 minutes of exposure to 5% bleach, to deionized water, and to UV irradiation at 120,000 µJoules/cm^2 followed by a physical scrub and an ethanol rinse was the most effective method. Treatment M removed all but 0.4% of the highest level of exogenous DNA, a 36-fold improvement from the original HCIFS method.

Using the improved decontamination method M, results were reproducible, with no exogenous DNA detected for eight of the ten animal tooth-human saliva samples tested. Two samples, dosed with 25ng and 50ng amounts of exogenous DNA, yielded DNA residual amounts of 0.04ng and 0.16ng, respectively. For those two samples, 4.0% and 6.0% of the donor profiles were observed, on average, with standard deviations of 0.14 and 0.04, respectively. All other samples, with donor DNA amounts ranging from 0.1 to 22.4ng of DNA, yielded negative amplification results (that is, no alleles detected). Six of the ten samples decontaminated using the original HCIFS method yielded sufficient exogenous DNA to produce full STR profiles.

All non-probative human tooth samples cleaned using treatment M produced single-source profiles from the tooth donors only; no exogenous DNA was detected. Using the original HCIFS method, one tooth, a molar with 1µL of human saliva containing 22.4ng of DNA added, yielded a mixture of DNA from the exogenous DNA source and the tooth donor. These results indicate that treatment M is a more effective method for decontamination of a tooth than the original HCIFS procedure, removing substantially more exogenous DNA than all other methods tested. The procedure known as treatment M, a multi-step process removing exogenous DNA, may be of general utility of cleaning teeth prior to analysis.

Tooth, Bone, Decontamination

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