B112 The Examination of DNA Extraction Lysis and Wash Step Modification for Low Template DNA Sample Processing

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Learning Overview: After attending this presentation, attendees will have a better understanding of alternative methods of DNA extraction that can be used to potentially increase overall DNA yields and improve short tandem repeat (STR) profiles of DNA extracted from archived latent fingerprints.

Impact on the Forensic Science Community: This presentation will impact the forensic science community by exploring methods that can be used in forensic laboratories to increase the success rate of obtaining a profile from low copy number fingerprint samples.

In recent years, as DNA analysis methods have become more sensitive, more attention has been paid to reevaluating closed and/or cold cases. In some of these cases, archived latent fingerprints may be the only source of biological evidence to revisit. Previous work on this project explored a variety of sampling techniques, extraction kits, and purification methods to establish an optimized "best practice" workflow for these challenging samples. Those studies found that directly extracting cuttings of the tape *and* paper sides of an archived print using QIAGEN QIAamp® DNA Investigator Kit, followed by quantification, pre-amplification purification via Centri-Sep™ columns, and concentration via vacuum centrifugation produced the best DNA yields and STR profiles. However, that study was completed using recently collected archived latent fingerprints; it is suspected that older samples may be more challenging. Thus, it is important to examine other potential procedural modifications that may minimize the DNA loss that occurs during laboratory handling of these samples. A preliminary study was completed to more closely examine where DNA loss occurs during the extraction process. On average, 83 ng of DNA (4.3% of the total DNA from each original buccal swab processed) did not bind to the column and was eluted with the initial lysate flow-through. This was enough DNA to obtain full STR profiles for 9 of 10 lysate flow-through samples tested. Consequently, a study was completed using 20 aged (2 years) archived latent fingerprint samples that were extracted using the QIAGEN QIAamp® DNA Investigator Kit, but the initial lysate filtrate was kept and extracted separately. The DNA extract and lysate filtrate samples from each individual sample tested were combined after quantification; combined samples were again quantified then concentrated and amplified (as a single sample) following the optimized methods previously described. While this modification increased DNA yields by 30% (from 0.183 ng to 0.263 ng), combining these samples reduced the number of expected STR alleles by 27% when compared to aged samples that were processed without the modification. Subsequently, a second study was conducted using a "double lysis" technique, with the aim of maximizing cell lysis and exposing more DNA for purification. For this technique, another 20-aged archived latent fingerprints (magnetic-powder treated and untreated) were processed using the same extraction method, but with one modification – after the initial lysis step, both the tape and paper sides of the sample were lysed twice and both lysates were combined for DNA extraction. Although this modification led to more samples producing detectable DNA, a more than 2-fold decrease in total DNA yields and a 22% decrease in the number of detected STR alleles were observed overall when compared to aged samples that were processed without the modification. Interestingly, though not improved with the method modification, magnetic-treated samples specifically were not affected, producing approximately the same number of expected STR alleles regardless of the lysis procedure used. Overall, our results suggest that while some DNA is lost during the laboratory extraction process, the amount recoverable may not be sufficient to improve the STR profile obtained from archived latent fingerprint samples using DNA extraction procedure modifications.

DNA Loss, Low Template DNA, Fingerprints