

## **An Alternate Workflow for DNA Analysis with Increased Sensitivity of Detection and Reduced Consumption of Evidence: Casework and Legal Implications**

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A number of issues continue to be raised in court proceedings regarding potential low quantity and low quality (LQLQ) samples. The arguments focus on sample splitting and whether it is appropriate. LQLQ DNA samples are at the crux of the decision process for deciding whether a swab in its entirety should be subjected to DNA extraction or instead be split in half and each half sequentially subjected to DNA extraction. The difference between the opposing positions is 1) the government tends to advocate that it is better to consume an entire swab containing LQLQ biological evidence so the most DNA possible can be recovered to increase the chances of generating a DNA profile; and 2) the defense tends to argue that a swab should be split in half, and only half of the swab should be extracted and quantitated; if the amount of DNA obtained is insufficient for analysis then the second half of the swab should be extracted and combined with the first DNA extract. This sample splitting approach requires more manipulations, results in loss of precious sample, increases chances of contamination, and dilutes the sample if the two portions subsequently are pooled. The decision to consume an entire swab or to split a swab should be based on reasonable expectations of the amount of LQLQ DNA that may reside in a swab.

A better way to address these opposing positions is to minimize sample consumption. A novel methodology/workflow has been developed that subsamples a minute portion of low quantity and/or touch samples. The subsampling consumes such a small portion of the stain that essentially the entire sample is preserved for additional testing or re-analysis. After collection the sample is amplified directly. Under the amplification conditions there appears to be an enhanced sensitivity of detection likely due to a localized PCR effect. Indeed, controlled studies of 1:99 dilutions of blood and saliva consistently yield higher STR peak heights than standard procedures with 1 ng input DNA from the same samples. Touch samples from common items yielded results consistent with the types of items. With these features it may be worthwhile to consider an alternate workflow in which subsampling is performed first on all stains, and if the results are acceptable, no additional testing is performed. This approach would preserve precious sample for additional forensic analyses, if desired. If the subsampling results are limited or inconclusive, then the entire stain can be extracted using traditional methods. The results of this study potentially may have important implications for analysis of low quantity and/or degraded samples that plague forensic casework. This presentation will describe the methodology, strengths of the alternate workflow (cost/benefit analysis and labor reduction), rationale for why quantitation of a subsample is unnecessary, problems encountered with sample splitting, benefits of consuming less evidentiary material, and the legal issues that can be overcome.