## EVALUATION OF COLLECTION METHODS FOR DIRECT PCR AMPLIFICATION OF TOUCH DNA SAMPLES

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Improved methods for generating high-quality DNA profiles from touch DNA samples are of considerable interest to forensic DNA laboratories. Direct polymerase chain reaction (PCR) amplification, a sample processing method in which an evidence swab or substrate punch is added directly to an amplification reaction without prior extraction or quantification, has been identified as a method that may improve DNA profiles from low-yield touch DNA evidence samples. Direct PCR maximizes the amount of DNA template in a reaction by eliminating the DNA loss that occurs during DNA extraction, quantification, and concentration; however, the amount of DNA available for direct PCR can be affected by the method used for DNA sample collection. This study aimed to identify collection methods that maximize DNA recovery from common touch DNA substrates and to determine whether standard or direct PCR processing methods result in higher quality DNA profiles.

Nine direct PCR-compatible collection methods were used to collect touch DNA from cotton denim fabric, wool fabric, polyester fabric, plastic microscope slides, metal tools, handgun grips, vinyl shutter samples, brass cartridge casings, foam cups, concrete bricks, and unfinished wooden tool handles. Collection was performed with cotton swabs, Copan microFLOQ<sup>®</sup> direct swabs, and FTA paper that were moistened with sterile water, moistened with 0.1% Triton X-100, or left dry. For each collection method, processing method, and substrate type, eight replicates were prepared from three donors. Samples were processed with two methods: (1) standard processing with DNA extraction and quantification and (2) direct PCR. The samples that underwent standard processing were extracted with Qiagen's Investigator<sup>®</sup> STAR<sup>™</sup> Lyse&Prep Kit, concentrated with Microcon<sup>®</sup> DNA FastFlow concentration devices, and quantified with Quantifiler<sup>™</sup> Trio. For direct PCR, the collected samples were placed directly in a 96-well plate for amplification. Amplification for both the extracted and direct PCR samples was performed with GlobalFiler<sup>™</sup>.

Direct PCR was successful for samples collected from plastic slides, polyester fabric, metal tools, handgun grips, vinyl shutters, foam cups, and unfinished wooden tool handles. For these substrates, the highest quality direct PCR results were obtained using microFLOQ swabs for collection. MicroFLOQ swabs & water or Triton X-100 produced the highest quality direct PCR profiles for plastic slides, handgun grips, and vinyl shutters; dry microFLOQ swabs produced the highest guality direct PCR profiles for metal tools, foam cups, and unfinished wooden tool handles. Although standard and direct PCR processing produced generally comparable DNA profile results, direct PCR of samples collected from metal tools and wooden tool handles with dry microFLOQ swabs and handgun grips with microFLOQ swab & water resulted in significantly higher quality DNA profiles than standard processing. However, direct PCR was unsuccessful for samples collected from concrete bricks, cartridge casings, denim fabric, wool blend fabric, and 100% wool, regardless of the method used for collection. Therefore, the success of direct PCR is highly dependent on the substrate from which samples are collected, whereas profile quality is affected by the efficacy of the collection method. These results indicate that direct PCR is an effective method for processing evidentiary touch DNA samples collected from certain substrates, and microFLOQ swabs were the most effective collection method for direct PCR of those tested.