

DIRECT AMPLIFICATION APPROACHES TO STREAMLINING DATABASING SAMPLE PROCESSING

Chang Zhong, Dennis Wang, Mary Ma, Andrea Carbonaro, Lisa Calandro, Lori Hennessy
Life Technologies, 850 Lincoln Centre Drive, Foster City, CA 94404

Laboratories involved in convicted offender and arrestee sample processing are continuously seeking opportunities to increase throughput and minimize the potential for sample backlogs. Increasing the turnaround time from sample submission to genetic profile upload provides earlier access to law enforcement enabling more expeditious resolution of no suspect cases and providing investigative leads in a timely fashion. Direct amplification approaches allow laboratories to eliminate time-consuming sample processing steps which may be prone to sample switching errors or cross contamination. The AmpFISTR® Identifiler® Direct PCR Amplification Kit was initially developed to take advantage of the lysis capabilities inherent in FTA® paper. The Identifiler® Direct Kit contains a robust master mix allowing users to directly punch discs from blood or buccal samples deposited on FTA® paper into the amplification master mix, eliminating the need for DNA extraction and purification. The increased efficiency is accompanied by high data quality with first pass success rates (full, interpretable profiles) at a 150 RFU analysis threshold of 94.7% for buccal samples (N=703) and 99.8% for blood samples (N=437). Recently, we have developed a protocol which extends direct amplification capabilities to non-FTA® substrates. The Prep-n-Go™ Buffer effectively lyses buccal samples collected on punchable non-FTA® substrates such as the Bode Buccal DNA Collector™ and is compatible with the Identifiler® Direct kit. The protocol is similar to direct amplification from FTA® paper: a 1.2 mm disc is punched into 2 ul of Prep-n-Go™ Buffer prior to addition of the amplification reagents. This protocol is easily automated and does not require an extended incubation or heating step to facilitate lysis. Success was demonstrated by the generation of interpretable, well-balanced, complete DNA profiles without introduction of artifact peaks. The first pass success rate of 95.9% (N=270) for buccal samples collected with the Bode Buccal DNA Collector™ was similar to that achieved for buccal samples on FTA® cards at an analysis threshold of 150 RFU. Evaluations of non-punchable substrates such as cotton swabs indicate that this streamlined protocol can be extended to a wide range of substrates. Side-by-side comparative studies showed that the Prep-n-Go™ Buffer helps improve the workflow efficiency and the STR profile quality of samples collected from buccal swabs and when amplified with the AmpFISTR® NGM™ and NGM SElect™ PCR Amplification kits. The Prep-n-Go™ Buffer used in combination with AmpFISTR® kits such as the Identifiler® Direct PCR Amplification kit will provide direct benefits to laboratories seeking to increase their efficiency for processing databasing samples.