DIRECT AMPLIFICATION FROM BUCCAL SWABS AND NON-FTA CARDS

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The PowerPlex® 18D System allows fast and efficient amplification directly from 1.2mm diameter punches of unwashed FTA® cards containing either a blood or buccal sample. FTA® cards allow for efficient lysis of cells, denaturation of proteins, and long term storage. Most direct amplification STR kits are designed to facilitate amplification from FTA® cards. However, many other substrates are used for collection of single source samples. These include buccal swabs of numerous varieties (e.g. OmniSwabs™, cotton swabs, Dacron swabs) and non-FTA cards (e.g. Bode DNA Collector). While non-FTA cards may be punched directly into wells of a PCR amplification plate, this is not always feasible for buccal swabs. An additional issue is that the cellular material deposited onto these surfaces is not subjected to the same lytic/denaturing agents present on FTA® cards with the consequence that the genomic DNA is not as readily accessible to the amplification reagents. Some loci may also be more accessible than others when working with chromatin as opposed to "naked DNA," introducing issues with balance not observed with an FTA® card. Any method that allows direct amplification from both buccal swabs and non-FTA® cards should therefore be able to work with both sample types and eliminate issues associated with accessibility to the genomic DNA.

We describe a method combining a proprietary formulation called SwabSolution™ in conjunction with a protease that allows simple pre-processing of both buccal swabs and punches from non-FTA® cards prior to cycling with a direct amplification compatible STR system. In the case of whole buccal swabs, the entire swab is incubated with 1mL of a low protease concentration in SwabSolution™ at 70 °C for 1 hour. At the end of this time, a fixed volume of the crude extract is removed and amplified in a standard 25µL amplification reaction. In contrast, for non-FTA® cards, 10µL of protease in SwabSolution™ is added to one 1.2mm diameter punch in a well of a PCR plate and incubated, without sealing, at 70 °C for 30 minutes until dry. The complete 25µL amplification mixture is then added to this well and cycling performed. Due to the formulation and incubation conditions used there is no carryover of active protease into the amplification reaction, eliminating any risk of proteolysis of the amplification components, even with prolonged incubation at ambient temperature prior to cycling. No protease inhibitors are required.