# DIRECT PCR AMPLIFICIATION USING POWERPLEX® FUSION KIT, POWERPLEX® Y23 AND WASHING REAGENTS

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The goal of this research was to generate DNA profiles from body fluids deposited on different swab substrates. Each substrate containing one body fluid sample was treated with a purification reagent prior to amplification with the PowerPlex® Fusion kit from Promega Corporation. Male body fluids were also pretreated prior to amplification for Y-STR analysis.

The following two substrates were used for this research project: Pur-Wraps® sterile cotton tipped applicator, and Pur-Wraps® Foam Swab. 1.2mm punch of each substrate was prepared for deposition of body fluids. Saliva and blood samples from male and female donors were deposited on each punch. 0.5 µL to 5 µL of saliva or 0.1 µL to 2 µL of blood was added to each punch. These specimens containing body fluids were kept at room temperature for varying lengths of time ranging from immediate extraction to processing after several weeks. Each punch containing only one body fluid was then washed with either the SwabSolution™ or the PunchSolution™ from Promega Corporation.

Each punch was incubated for the appropriate amount of time recommended or as deemed necessary. Each sample was amplified using either the PowerPlex® Fusion, or the PowerPlex® Y23 amplification kits from Promega Corporation.

Substrate types, amount of reagents needed, and, if necessary, amplification parameters were varied in this study to detect complete autosomal and Y-STR profiles. Recommended reaction volumes were used first to determine if complete profiles can be generated from each substrate using the amplification reagents. Once it was successful, half reaction volumes were used for this research. Y-STR profiles were generated from male blood and saliva samples. Analysis of the amplified product was performed by capillary electrophoresis injection on the Applied Biosystems 3130xl Genetic Analyzer. The generated DNA profiles were analyzed using GeneMarker® HID Software Version 2.2.0 from SoftGenetics®.

Autosomal STR profiles were generated successfully from both of the swab substrates containing the two body fluids. The research indicated that 0.1µL blood and 1.0 µL saliva were the optimal volumes necessary to obtain complete profiles. Half reaction volumes produced complete autosomal and Y-STR profiles. Concordant profiles were obtained within and between both substrates and the two reagents.

In the forensic science community, direct PCR amplification could have an important impact. Swabs are frequently used to collect reference samples, and having the ability to process different types of swabs and amplify the body fluids more quickly using different types of amplification kits is beneficial to the crime laboratories, paternity testing laboratories and to data banking institution. This research project indicates that the samples can be processed faster, and it also allows for decreased reagent consumption and, consequently, reduces cost.