

## **MICROFLOQ® DIRECT CONVEYS AN ALTERNATIVE FORENSIC WORKFLOW FOR PATERNITY, KINSHIP AND REFERENCE SAMPLE TESTING**

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Forensic DNA analysis should become less time consuming, labor intensive and costly for paternity, kinship and reference sample testing. The preservation of collected samples for Court debates and further analyses is also crucial. The microFLOQ® (MF), recently introduced to the market by Copan, allows to use a tiny portion of buccal sample collected with 4N6FLOQSwabs® (4N6FS) Genetics and to directly profile human DNA (hDNA) in less than 2 hours with standard instruments, eliminating the extraction and quantification steps.

The objective of the study was to validate an alternative direct workflow for MF processing of buccal cell samples collected with 4N6FS, in comparison with the standard procedures of the same buccal swabs.

A set of buccal 4N6FS was collected from 10 donors following Copan collection procedure. One swab per donor was processed with the standard workflow: DNA extraction with PrepFiler® Express kit, using NAO® baskets (Copan) to optimize DNA recovery; quantification by real-time PCR with Quantifiler® Trio Kit on AB 7500; profiling with the GlobalFiler™ (GF) kit on Veriti™ thermal cycler and fragment run on the AB 3500 Genetic Analyzer (Thermo Fisher Scientific). In parallel, another swab per donor was used to set up the MF workflow, through direct amplification with GF Express (GFE) kit (Thermo Fisher Scientific), testing: MF collection on buccal swabs fresh vs dried; the number of MF taps on the 4N6FS; PCR cycle number for direct amplification; different pre-wetting methods of collection devices.

The optimized direct workflow consists of pre-wet (pipetting 1 µl of PCR grade water) MF collection with a single tap on buccal 4N6FS dried; direct amplification set at 26 cycles with GFE kit.

Concordant and full DNA profiles were obtained with both direct MF workflow and with standard protocols from the 4N6FS of the same donors. The average peak height and quality of the DNA profiles are within the parameters suggested by the profiling kit manufacturer, with higher variability for direct workflow, as expected.

Ongoing experiments aim to extend the optimized direct workflow validation to a wider number of donors and to compare the performance of the 4N6FS vs traditional cotton swabs.

The microFLOQ® allows direct and rapid DNA profiling from buccal 4N6FLOQSwabs® collected for paternity or kinship testing and reference sample comparison in forensic investigations; an effective alternative to the standard swab processing; and conserving the original buccal swab for Court cases retesting.