B71 The Development of Epigenetic Methylation Markers for Skin/Sweat

Nicole Fernandez Tejero, BS, Sun City Center, FL 33573; Quentin T. Gauthier, MSFS, Miami, FL 33174; Bruce R. McCord, PhD, Florida International University, Miami, FL 33199*

Learning Overview: After attending this presentation, attendees will have gained valuable insight on the research efforts being made to use DNA methylation patterns at different genome locations as a tool to identify skin cells.

Impact on the Forensic Science Community: This presentation will impact the forensic science community by determining skin/sweat from other body fluids.

The determination of tissue type is important when reconstructing a crime scene. However, at present there is no specific methylation-based marker to distinguish touch DNA from other body fluids. Because epigenetic markers capable of identifying sweat and/or skin have not yet been developed, the epigenome is being explored to discover loci of interest. Typically, the DNA in such samples results from free DNA secreted in sweat and cellular material and may result from skin sweat or other catabolic processes. Work in the lab has successfully identified DNA methylation in various body fluids, but not in skin or sweat. Moreover, the development of a body fluid identification multiplex in the lab would greatly benefit from the inclusion of a skin/sweat marker. The ultimate goal is to develop multiplexed methylation-based epigenetic loci for use in forensic analysis.

This study compared DNA obtained from skin with semen, saliva, blood, and vaginal epithelia. An Illumina® MethylationEPIC Beadchip Array was utilized to perform an Epigenome Wide Association Study (EWAS) to determine suitable methylation markers for touch DNA. The results indicated that CpGs from five different genes were potentially of use: WDR11, PON2, NAA16, MRPS15, and NHSL1. A set of Polymerase Chain Reaction (PCR) primers were designed to encompass these loci. Next, DNA was extracted, quantified, and bisulfite converted using the QIAGEN® EpiTect™ Bisulfite Kit to convert all the unmethylated cytosine to uracil on the DNA template. Bisulfite-converted DNA was amplified using the Rotor-Gene® SYBR® Green PCR Kit on a Rotor-Gene® Q, then analyzed for melting temperature.

This high-resolution melt analysis was then used to examine various epigenetic loci with potential for the discrimination of touch DNA. In these studies, several loci showed bisulfite-modified skin amplicons that melted at lower temperatures when compared to blood, saliva, semen, and vaginal epithelia. These results demonstrate that High Resolution Melt (HRM) analysis may be a promising technology to identify skin/sweat from other body fluids of a DNA sample.

DNA Methylation, Skin/Sweat, High Resolution Melt

*Presenting Author