AN EVALUATION OF THE FORENSIC DETECTION OF MICRORNAS IN DNA EXTRACTIONS FOR SOURCE IDENTIFICATION

C Lewis, T Layne, S-Seashols Williams, Department of Forensic Science, Virginia Commonwealth University

Molecular markers for forensic body fluid identification, such as microRNAs (miRNAs) or messenger RNAs, have been focused on in the recent forensic literature as an alternative method to the current presumptive serological techniques. MicroRNAs are small non-coding RNAs that regulate gene expression by binding to messenger RNA in the cytosol to prevent further translation. Their short length of 18-22 nucleotides, cellular function, and resistance to degradation allow for easy detection in highly degraded samples, as is often the case in forensic casework samples. The work in microRNAs naturally utilizes RNA extractions, which would add an additional step in the forensic DNA analysis workflow. A previous report has shown that microRNAs can potentially be detected in silica-column DNA extracts. Further exploration of microRNA detection within other types of DNA extraction methods is essential for implementation of microRNAs into forensic casework.

Liquid donations of blood, semen, and saliva were collected from three individuals, aliquoted onto swabs, and allowed to dry. Four of the most common DNA extractions methods used by forensic laboratories were performed on all samples, with a total RNA isolation method for each sample as a control. A commercially available co-extraction method was also evaluated for miRNA detection. A portion of all DNA and RNA extracts were DNase-treated to ensure that the data reflected true miRNA detection rather than genomic DNA contamination. MicroRNA presence was evaluated using RT-qPCR analysis of miRNAs let-7g and let-7i; microRNAs that have been shown to have relatively similar expression levels across multiple body fluids. An RT-qPCR panel of 750 human miRNAs was evaluated to compare global miRNA expression between RNA and DNA extracts. Preliminary data showed that DNase treatment of extracts has little effect on miRNA expression levels between untreated samples and DNase-treated samples. Subsequent work confirmed that miRNAs were best detected in silica-column DNA extracts; however, non-silica column-based DNA isolation methods also yielded comparable miRNA levels. Based on this data, miRNAs can be detected within a variety of DNA extracts, and they can provide more information about a forensic sample without an additional step in the DNA analysis process.