B187 An Evaluation of the Differential Stability of Nucleic Acids in Biological Fluids Compromised by Environmental Exposure

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After attending this presentation, attendees will better understand how much of an effect the environment has on evidence and biological material through a discussion of relative degradation rates of DNA, messenger RNA (mRNA), and microRNAs (miRNAs). This presentation will also increase awareness of RNA use in the forensic community and promote research in novel body fluid identification methods.

This presentation will impact the forensic science community by clarifying relative stabilities between RNAs and DNA within the same sample and the stability of RNA for body fluid identification purposes.

The visualization, presumptive, and confirmatory tests used for body fluid identifications in forensic casework have remained static for many years. This is problematic because many of these tests are known to yield false positive results with other biological fluids, foods, or chemicals. Recent work in the forensic science field has explored RNAs for a molecular-based, forensic body fluid identification method. Many researchers have assessed mRNA to identify body fluids. While these research studies provide good evidence for mRNA as being useful in body fluid identification, some research has provided unsupportive evidence and thus mRNA analysis has been slow to catch on for casework. The miRNAs are small RNAs that have the ability to suppress translation of mRNA into proteins and are shorter in length than mRNAs. Because of these advantages and the remarkable stability observed by the species, they are also being considered as markers for body fluid identification. This study evaluated the relative stability of DNA, RNA, and miRNAs in the same samples under conditions mimicking an outdoor crime scene.

Samples of blood, urine, semen, and saliva were placed in an environmental chamber for defined periods of time corresponding from 24 hours to 14 days during a Virginia summer. The environmental chamber manages the irradiance, air temperature, and humidity for a more controlled assessment of sample degradation. DNA and RNA were isolated from each stain, and quantitative Polymerase Chain Reaction (qPCR) for DNA and Real-Time (RT) qPCR for RNA and miRNAs was performed. Expression levels were calculated relative to the positive untreated control samples. DNA stability was evaluated using both standard TPOX and "mini" length primers for the Short Tandem Repeat (STR locus) TPOX. Both mRNA and miRNA expression were evaluated using GAPDH and/or ACTB, and Let-7g, respectively.

The miRNA expression was not significantly impacted by treatment in the environmental chamber, unlike the impacted levels of DNA and mRNA. When measuring miRNA stability, Let-7g levels were not significantly different from the untreated control levels for all four body fluid samples. The DNA and mRNA data showed how environmental effects can greatly degrade biological material after exposure. The data from this project drives home the environmental effects on biological material, clarifies differential stability of the nucleic acids, and, consequently, can provide the practitioner with options for analysis workflows in compromised samples.

RNA, Degradation, Body Fluid Identification

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