B1 Different Effects of PCR Inhibitors on Multiplex STR Assays

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After attending this presentation, attendees will learn the impact of six common forensic PCR inhibitors on the performance of multiplex STR assays.

This presentation will impact the forensic community by showing the forensic community that PCR inhibitors can have different effects on multiplex STR assays. It will also be demonstrate that multiplex STR assays have varying degrees of tolerances to the presence of PCR inhibitors in the PCR reaction.

DNA samples recovered from crime scenes are often commingled with contaminants that present a significant challenge for PCR amplification. Outdoor crimes may leave body fluids such as blood and semen on soil, sand, wood or leaf litter that contain substances which can co-extract with the perpetrator's DNA and prevent PCR amplification. Textile dyes, leather and wood from interior crime scenes can also contain inhibitors that interfere with the DNA polymerase's activity. The impact of these contaminants on the multiplex STR assays can vary from attenuation to complete inhibition of the amplification process, resulting in partial STR profiles or profiles with unusual peak morphology.

In the present study, a systematic approach was utilized to evaluate the effect of six PCR inhibitors commonly found in forensic samples on different multiplex PCR assays. Each multiplex PCR assay has unique primer sequences and buffer formulation. The six PCR inhibitors used in the study were hematin, indigo, melanin, humic acid, collagen, and calcium. For each multiplex PCR reaction, a range of inhibitor concentrations was included during PCR amplification. The amplification results were evaluated based on: 1) the ability of each multiplex PCR assay to generate full STR profiles, and 2) the quality of the STR profiles obtained.

The results revealed that the STR profiles obtained from multiplex PCR assays can be severely compromised by various PCR inhibitors. Within the same multiplex PCR assay, the degree of inhibition varies greatly with different types of PCR inhibitors. Between different multiplex PCR assays, the tolerance to PCR inhibitor also differed considerably. The results clearly indicated that with optimal primer sequences and buffer formulation, PCR inhibition can be kept to a minimal. Furthermore, the results also demonstrated that PCR cycling conditions can influence the peak morphology of the PCR-inhibited samples.

STR Typing, MiniSTR, PCR Inhibition

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