COMPARATIVE EVALUATION OF THREE COMMERCIAL QUANTITATIVE PCR KITS WITH INHIBITED AND DEGRADED SAMPLES

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Forensic STR typing is currently the gold standard in the forensic DNA community. However, casework samples are often degraded, in low amounts and/or inhibited, which may complicate successful DNA typing. Therefore tools that better predict any DNA degradation or presence of PCR inhibitors in a sample prior to STR amplification could benefit laboratories by reducing the time for analysis and use of resources. Various commercial quantitative PCR kits that simultaneously provide the amount of human and male DNA whilst also predicting the level of DNA degradation (via a degradation index; DI) and presence of PCR inhibitors in a sample are available to forensic DNA laboratories. This study compares the performance of three commercial quantitative kits: 1) Quantifiler® Trio (Life Technologies), 2) PowerQuant® (Promega), and 3) InnoQuant™ HY (InnoGenomics Technologies) with degraded (N=15) and inhibited (N=100) DNA samples. We tested each kit's tolerance to a variety of inhibitors including hematin, calcium, humic acid, melanin, and salt in aqueous solution.
We compared the quantitation values and degradation indices for 3 sets of degraded samples (bone, formalin damaged, and decomposed tissues). Quantitation values and degradation indices were generally comparable between the kits; although the PowerQuant® kit generated notably higher DI values in some samples. We also assessed quantitation values and the level of PCR inhibition (ΔC_T) for standard DNA samples spiked with various inhibitors. Overall, Quantifiler® Trio was the most tolerant system to all inhibitors and concentrations tested in this study. PowerQuant® was tolerant to all inhibitors except for salt, and InnoQuant™ HY was most susceptible to melanin. InnoQuant™ HY also generally produced higher ΔC_T values and lower DNA concentrations than the other two kits, suggesting that this system may be less tolerant to common PCR inhibitors found in forensic samples.

Keywords
Quantitative PCR, degradation index, PCR inhibition, Quantifiler® Trio, PowerQuant®, InnoQuant™ HY