

DEVELOPMENT AND VALIDATION OF A RAPID qPCR ASSAY FOR ASSESSING THE QUALITY OF HUMAN DNA IN FORENSIC SAMPLES

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Short tandem repeat (STR) analysis is the most commonly used technique in human identification and forensic testing. STR assays are complex multiplex systems that require a defined range of input template to perform accurately. This requirement makes the quantification of human DNA in a sample a necessary step prior to the STR analysis.

DNA quantification is the only pre-STR step, so it is crucial to extract as much information as possible from this reaction to support both the setup of the STR reaction and the interpretation of unexpected results. For example, the presence of amplifiable DNA in the sample and the absence of inhibitors can be verified during this step. Moreover, highly sensitive and accurate DNA quantification are critical factors that ensure a high correlation between quantification and STR results.

This is the first presentation of validation data for a new qPCR assay for forensic testing of human DNA, the Investigator Quantiplex Kit, which utilizes novel PCR fast-cycling technology and Scorpion primers. It provides rapid and accurate quantification of human DNA with a high sensitivity (down to 0.3 pg/ μ l) and high accuracy thanks to the newly identified multi-copy target 4NS1C. Moreover, inhibitor detection is ensured by a balanced internal amplification control. This quality sensor is designed to be less stable than the 4NS1C target, showing a shift (mostly not a complete failure) of the CT value in the presence of inhibitors.

Data from both the developmental validation and the external study on the confirmation of the 4NS1C target copy-number will be shown, along with the field test results on inhibitor detection sensitivity and accuracy.