A173 Enhanced Quantitation Data Analysis Using Next Generation Real - Time PCR Analysis Software

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After attending this presentation, attendees will learn about the next generation real-time PCR analysis software which streamlines quantitation set-up and data analysis as well as provides sample dilution and STR reaction set-up tools.

This presentation will impact the forensic community by explaining how the next generation real-time PCR analysis software will provide efficient analysis of quantitation data, assist in obtaining optimal results from downstream STR reactions, and enable continued high quality laboratory throughput.

As forensic laboratories continue to seek ways to increase throughput, the need for advanced software has become paramount. The use of real-time PCR systems for DNA quantification and quality evaluation has reduced analysis time and provided highly informative results. The ability to quantify the DNA from large amounts of samples requires software that can aid the forensic scientist in the critical analysis of data from a wide range of sample types. The next generation real-time PCR analysis software has been developed to meet this need by establishing a Quality Control Flag system to ease data analysis by quickly and accurately identifying sample or quantitation assay anomalies. Furthermore, simplified background, ROI, optical, and pure dye spectral calibrations complete with wizard-based instructions and automated analysis have been incorporated into the next generation real- time PCR analysis software.

The next generation real-time PCR analysis software employs a Quality Control Flag system to assist the analyst with streamlined data analysis and evaluation of critical information obtained from quantitation assays. Such information includes the detection of PCR inhibition, reagent contamination, and mixtures of male and female DNA. Quality flags evaluate the slope, R^2, and Y-intercept standard curve metrics, Internal PCR Control (IPC) C_T value, high or low quantity samples, and instrument performance. In addition, a Male to Female ratio flag specific to the multiplexed quantitation assay indicates the presence of samples containing a mixture of male DNA combined with excess female DNA prior to STR analysis.

Each of these quality flags not only eases data analysis but provides guidance for downstream STR analysis. By automatically assessing sample quality and quantity, the software helps to facilitate the selection of the appropriate STR amplification kit and DNA input amount, or whether further processing is required. For example, when the IPC C_T flag is activated, the analyst can evaluate sample specific amplification plots to determine if dilution and requantification is required or if the sample should be amplified using a mini-STR kit due to sample inhibition. Also, if the user defined Male to Female ratio flag is triggered in the multiplexed quantitation assay, the analyst can determine if autosomal or Y chromosome amplification should be performed. The software also contains workflow enhancements to assist the analyst in preparing sample dilutions for STR reactions of choice as well as provide STR reaction set-up parameters. The next generation real-time PCR analysis software has been designed to provide efficient analysis of quantitation data, assist in obtaining optimal results from downstream STR reactions, and enable continued high quality laboratory throughput.

Next Generation Real-Time PCR Analysis Software, Quality Control Flags, Multiplexed Quantitation

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