

ENHANCED QUANTIFILER[®] DATA ANALYSIS USING THE 7500 REAL TIME PCR SYSTEM AND 7500 HID SOFTWARE V1.0

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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 7500 HID Software v1.0 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 Quantifiler[®] assay anomalies. Furthermore, simplified background, ROI, optical, and pure dye spectral calibrations complete with wizard-based instructions designed for Quantifiler[®] assays and automated analysis have been incorporated into the 7500 HID Software v1.0.

Much like GeneMapper[®] ID-X software, the 7500 HID Software v1.0 employs a Quality Control Flag system to assist the analyst with streamlined data analysis and evaluation of critical information obtained from Quantifiler[®] assays. Such information includes the detection of PCR inhibition, reagent contamination, and mixtures of male and female DNA. Two quality flags evaluate the slope and R2 standard curve metrics providing evaluation of quantification standard preparation and assay set-up. Another quality flag focuses on the Internal PCR Control (IPC) amplification alerting the analyst to the specific wells in which the IPC CT value does not meet user defined thresholds indicating potential inhibition or non-optimal assay performance. Other quality flags notify the analyst of high or low quantity samples as well as many instrument and data collection issues. In addition, a Male to Female ratio flag specific to the Quantifiler[®] Duo 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 AmpF λ STR[®] kit and DNA input amount, or whether further processing is required. For example, when the IPC CT 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 with the AmpF λ STR[®] MiniFiler kit due to sample inhibition. Also, if the user defined Male to Female ratio flag is triggered in the Quantifiler[®] Duo 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 7500 HID Software v1.0 has been designed to provide efficient analysis of Quantifiler[®] data, assist in obtaining optimal results from downstream STR reactions, and enable continued high quality laboratory throughput.