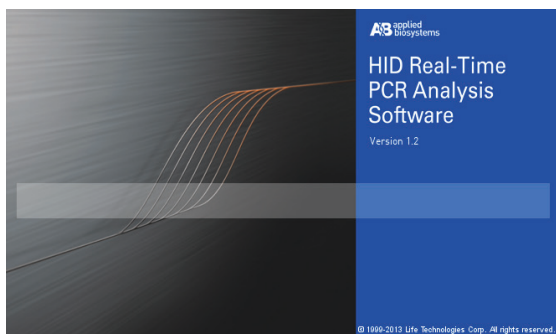# HID Real-Time PCR Analysis Software v1.2

Enhanced Quantifiler® kit data analysis on Windows® 7 and Windows® XP platforms

Quantifiler® DNA Quantification Kits are used by human identification (HID) laboratories to aid forensic analysts in the processing of casework samples. The quantification results help guide selection of the most appropriate Applied Biosystems® STR kits in order to maximize the chances of obtaining optimal STR results on the first attempt.

To take full advantage of the data and capabilities offered by the Quantifiler® kits and to maximize sample-processing efficiency, software designed specifically for the HID application is a must.

HID Real-Time PCR Analysis Software v1.2 has been developed to provide an integrated solution for DNA quantification. The software has quantification and STR setup features and data quality review functionality that integrate and streamline the quantification and STR analysis steps. By providing predefined templates for Quantifiler® and Applied Biosystems® STR kit runs, sample setup time is minimized. Incorporating HID-specific data quality assessment and STR setup tools into a single software package enables faster and better decisions.

Accuracy
- Designed and validated specifically for human identification applications on the Applied Biosystems® 7500 Real-Time PCR System (Microsoft® Windows® 7 and Microsoft® Windows® XP platforms)

- Automatically provides sample quantity and quality information to help maximize data recovery

- Integrated STR reaction setup tools automatically calculate the sample normalization and STR reaction setup volumes based on the quantification data, minimizing the potential for errors

Efficiency
- Intuitive, easy-to-navigate user interface streamlines run setup with predefined Quantifiler® assay run templates

- Simple, faster sample processing that integrates the quantification and STR analysis setup steps with automated instrument calibration, data quality assessment, and sample normalization/STR reaction setup tools

- Faster run setup through optimized and validated robotic integration scripts for commonly used robotic platforms

Flexibility
- Optimized and validated for use with Quantifiler® kits, or perform custom assays by running in the Custom Assay Mode

- Configurable run templates, experimental and data quality parameters, data display, and reporting features to fit lab-specific protocols

- Flexible for use with the instrument or as a stand-alone analysis software on a user's laptop

life technologies

Streamlined HID-specific workflow

HID Real-Time PCR Analysis Software v1.2 provides HID-specific templates including Quantifiler® Trio, HP, Duo, Human, Male, and Human and Male (hybrid plate). Customize and set up a run by clicking one of the Quantifiler® assay template icons on the home screen (Figure 1).

In each template, predefined experimental properties, targets, sample types, and standard concentrations simplify the quantification run setup process. To set up a run, simply enter or import the sample names and well positions of any unknown samples.

The intuitive, easy-to-navigate user interface further streamlines the workflow (Figure 2). Plate Setup, Run, and Analysis functions are accessed in the Experiment Menu on the left navigation pane. Detailed experimental parameters and results are displayed conveniently on the right pane. For advanced data analysis there are multiple ways to view and filter data, including plate map and table views, amplification plots, standard curve plots, multi-component plots, and raw data views that support fast QC and sample data review and troubleshooting.

Rapid sample quality assessment

The software employs a Quantifiler® assay–specific quality control flag to help facilitate data analysis and more informed decision-making for STR analysis by identifying samples that require further analysis. Quality flags alert you to standard curve performance or dilution preparation issues, as well as samples that contain potential PCR inhibition (IPC C_t flag), high or low quantities of DNA, or male-and-female mixtures (Figure 3).

Raising the bar in quality

In addition to sample quantity and flagging samples that do not meet data quality requirements, the software automatically calculates the level of DNA degradation (Degradation Index)* and PCR inhibition (IPC) as well as male:female mixture ratios,* providing a more comprehensive view of sample quality to help determine the optimal approach to maximize data recovery during STR analysis (Figure 4). More informed, up-front decisions can be made, such as whether to amplify using an STR kit with a high number of miniSTRs or Y-STRs, to add more DNA to the reaction to improve allele recovery, or to perform additional cleanup or sample dilution prior to STR analysis to remove or minimize potential inhibitors.

Validated for the Windows® 7 platform

HID Real-Time PCR Analysis Software v1.2 is designed and validated for use on both Windows® 7 (32- and 64-bit) and Windows® XP operating systems.

*For use with Quantifiler® Duo and HP kits.

Figure 1. The home screen provides direct access to HID-specific templates for all Quantifiler® DNA Quantification Kits.
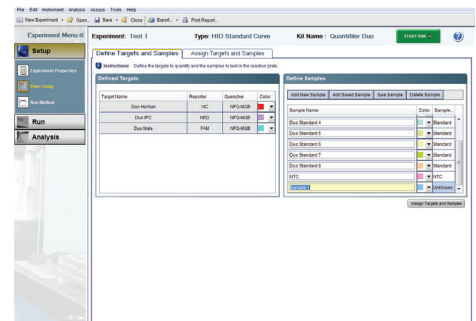

Figure 2. The easy-to-navigate user interface streamlines the workflow with easy access to Plate Setup, Run, and Analysis menus in the left navigation pane.
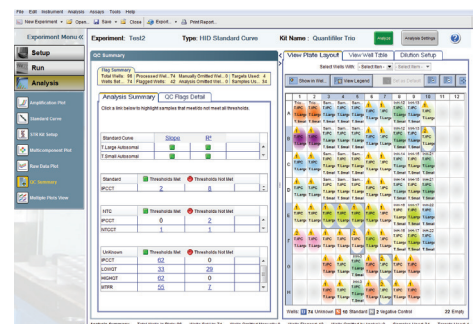

Figure 3. The Analysis Summary provides a quick overview of samples that meet or do not meet thresholds, with hyperlinks to view more detailed information on flagged samples in the well table and plate layout screens.
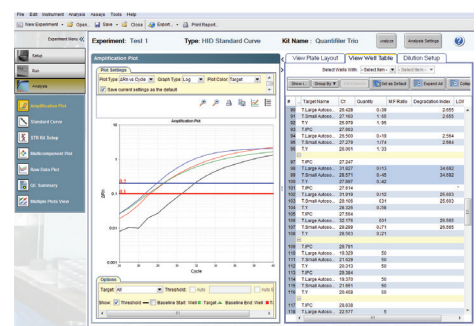

Figure 4. Sample quality information, such as the level of DNA degradation and PCR inhibition, as well as the ratio of male to female DNA in mixed samples are automatically provided.

Dilution Calculation Tool
This feature provides instructions based on user preferences to normalize samples prior to STR amplification. Define the dilution scheme parameters, including pipetting overage (%) and minimum pipetting and maximum sample volumes; also choose either the one-step or two-step dilution method as well as a desired dilution factor to meet the laboratory's protocols (Figure 5).

STR Kit Reaction Setup Tool
This tool utilizes customizable STR kit and sample dilution information to generate sample dilution and STR reaction setup worksheets (Figure 6). Customizable parameters include a list of PCR master mix components, component and sample volumes, target DNA concentration, and pipetting overage allowances. Select the STR kit(s) for downstream analysis in the well table, and the sample dilution and STR kit parameters are automatically applied. Dilution preferences or DNA target concentration can be edited in real time for each sample. This tool makes STR reaction setup quick and easy and minimizes the potential for errors.

Configurable reporting features
A report in PDF format can be exported for inclusion in a case file. A report can also be customized and exported in Microsoft® Excel® or text format to perform additional analysis, if needed. To customize an Excel® or text file, select the desired information and how it should be organized in the report, such as plate setup, results, amplification data, standard curve, STR dilution and reaction setup, and the file format (Figure 7).

Easy switch to Custom Assay Mode
Users who need to perform custom assays can easily switch to the Custom Assay Mode by simply clicking "Assays" on the top menu bar and selecting "Custom Assays". This enables you to use the software for different applications or configurations.

Simplified instrument calibration
In addition to simplifying sample analysis, step-by-step wizard-based instructions guide you through the system calibration process (ROI, Background, Optical, Pure Dye, RNase P Verification Plate). At the end of each calibration, the software automatically analyzes the calibration data and displays the result as Pass or Fail. Time-consuming manual calibration analysis is no longer required. The software automatically records the date and time of each calibration and notifies you when a calibration is due.
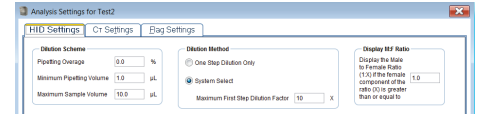

Figure 5. The Dilution Calculation Tool enables users to easily normalize samples.
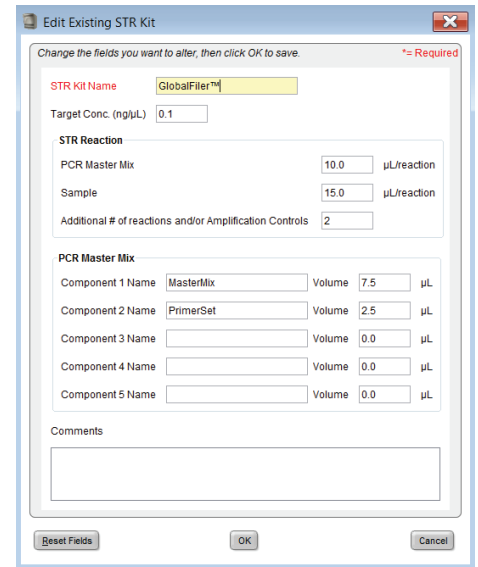

Figure 6. The STR Kit Reaction Setup Tool guides users to set up downstream STR reactions in a simple and easy manner.
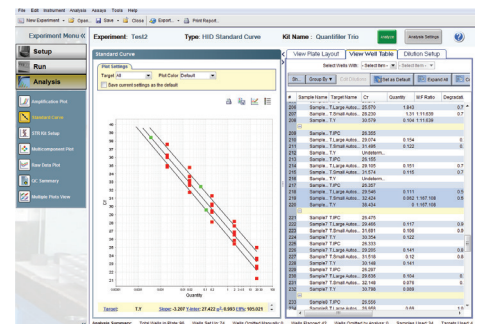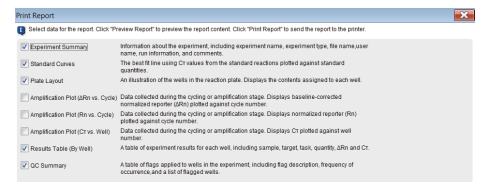

Figure 7. Customizable reporting features provide flexibility to select desired information for reporting in PDF format.

System requirements

Component	Recommended requirements	Minimum requirements
Computer	• Pentium® 4 or compatible processor, 2.0 GHz • 1 GB RAM • One hard drive with 10 GB available • 20/48X IDE CD-ROM drive • USB v2.0 • Ethernet network interface adapter (10BASE-T)	• Pentium® 4 or compatible processor, 1.2 GHz • 1 GB RAM • One hard drive with 10 GB available • 20/48X IDE CD-ROM drive • USB v1.1 • Ethernet network interface adapter (10BASE-T)
Monitor	• 1280 x 1024 pixel resolution for full-screen display • 19-inch • 32-bit color • UL listed	
Operating system	• Microsoft® Windows® 7, Service Pack 1 (32- and 64-bit) or • Microsoft® Windows® XP, Service Pack 3	
7500 instrument firmware	• G v2.10	

Ordering information

Description	Cat. No.
HID Real-Time PCR Analysis Software v1.2—Single User License	A24664
HID Real-Time PCR Analysis Software v1.2—5 User License	A24612
HID Real-Time PCR Analysis Software v1.2—Single Upgrade User License	A24665
HID Real-Time PCR Analysis Software v1.2—5 Upgrade User License	A24614

Find out more at **lifetechnologies.com/hid**

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