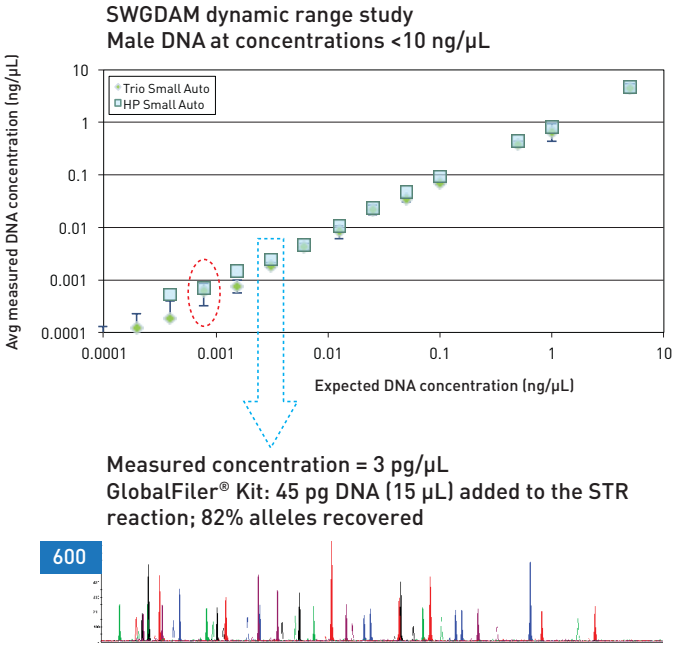
Quantifiler® HP and Quantifiler® Trio DNA Quantification Kits

Offering accurate results in less time for your most challenging casework samples

Quantifiler® DNA Quantification Kits, including the Quantifiler® HP and Trio kits, are used by human identification (HID) laboratories to make informed decisions on the downstream processing of casework samples. The Quantifiler® HP and Trio kits enable efficient and accurate quantification of human DNA and are the first kits to provide a Quality Index to detect the presence of degraded DNA along with PCR inhibitors. In addition, the Quantifiler® Trio kit determines the quantity of male DNA present in your samples. All of these results guide the selection of the most appropriate Applied Biosystems® STR kits in order to help maximize the chances of casework sample analysis success. For high sample processing efficiency, the kits run on the Applied Biosystems® 7500 Real-Time PCR System and utilize HID Real-Time PCR Analysis Software v1.2. Together, this system provides a streamlined HID-specific quantitation workflow with enhanced data review functionality and the ability to improve STR analysis efficiency.



Quantification range: 5 pg/µL–100 ng/µL

DNA was detected for all 3 replicates: 0.8 pg/µL–120 ng/µL

Figure 1. Dynamic range and sensitivity study. The performance of the small autosomal target was evaluated in the Quantifiler® Trio and HP kits. Quantification and Globalfiler® amplification was performed in triplicate. A representative electropherogram is shown.

Quantify with confidence

To accommodate the most challenging casework samples, the Quantifiler® HP and Trio kits use a more robust reaction mix along with multiple-copy target loci to provide 6x greater detection sensitivity than single-copy assay targets. The targets in the Quantifiler® HP and Trio kits have been selected to minimize copy number variation and maximize quantification sensitivity for challenging, low-level samples, down to sub-picogram DNA concentrations (Figure 1). The Quantifiler® Trio kit also utilizes an improved male target to enable more accurate results for samples containing low concentrations of

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male DNA in the presence of high amounts of female DNA (Figure 2). The improved sensitivity at sub-picogram levels of DNA, combined with greater reproducibility, enables casework laboratories to more accurately identify the samples that may contain sufficient DNA to proceed with STR analysis and deliver the most informative results.

Evaluate sample quality and quantity in a single reaction

The quantification step of the casework workflow is the key to determining how a sample will be processed downstream. There are many factors determining the success that will be seen post-amplification in the STR analysis. The Quantifiler® HP and Trio kits are the first kits to provide both a quantity and quality assessment of casework samples in a single reaction. Both kits utilize primers for small and large autosomal amplicons that together enable a more comprehensive view of sample degradation (Figure 3). The kits also contain an enhanced internal positive control (IPC) that enables rapid identification of samples that may also contain high levels of PCR inhibitors. The IPC together with the degradation evaluation provides a Quality Index that enables casework laboratories to accurately predict amplification performance and downstream sample success.

Small amplicons for challenging samples

The Quantifiler® HP and Trio DNA Quantification Kits utilize multiple-copy target loci for improved detection sensitivity. The human- and male-specific target loci each consist of multiple copies, dispersed on various autosomal chromosomes or multiple copies on the Y-chromosome. To maximize the consistency of quantification results, genomic targets were selected with conserved primer and probe binding sites within individual genomes, and with minimal copy number variability between different individuals and population groups. The primary quantification targets (small autosomal and Y-chromosome

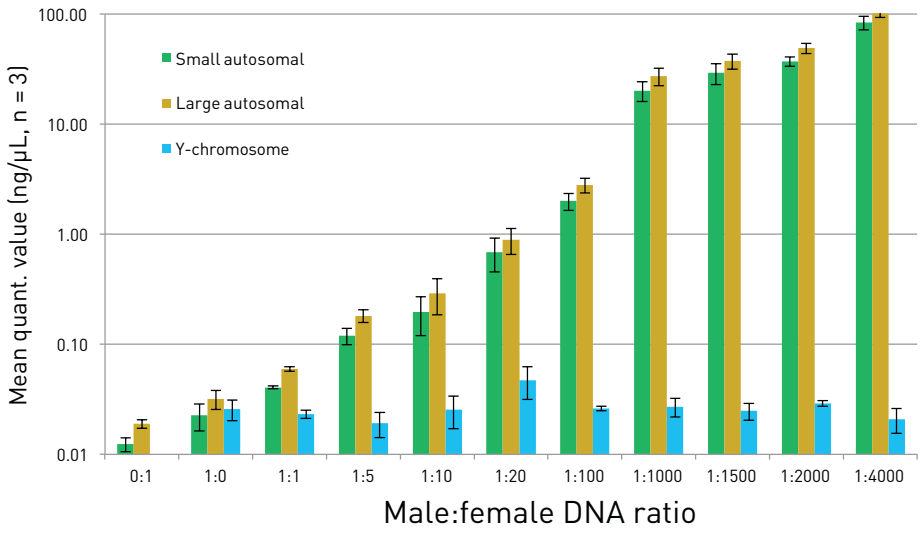
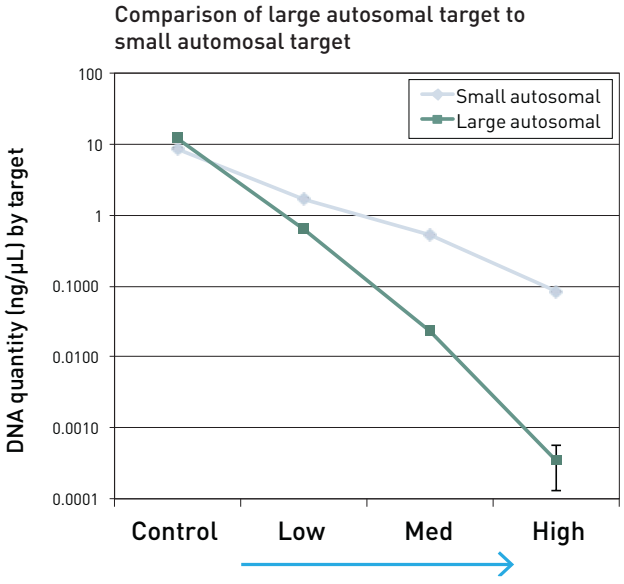


Figure 2. Balanced and consistent Y chromosome quantification in the presence of high concentrations of female DNA. Mixture samples containing 20 pg/µL of human male DNA were prepared with varying amounts of female DNA. Samples were processed for quantification in triplicate using the Quantifiler® Trio assay.



Sample quality determined by evaluating Degradation Index (DI) and IPCCT flag to identify the level of DNA degradation

Figure 3. Predictive sample quality assessment. Degradation Index (DI) is calculated using quantification results from small and large amplicons. An increase DI means that there is a decrease in the measured amount of large DNA fragments compared to small fragments. Large DNA fragments may perform more poorly relative to small DNA fragment in STR reactions.

targets) consist of short amplicons that range from 75 to 80 bases, which improve the detection of degraded DNA samples (Figure 4). Utilizing multicopy targets and smaller amplicons, the Quantifiler® HP and Trio kits enable improved detection sensitivity when compared to previous Quantifiler® kits.

Increase throughput and drive workflow efficiency

The Quantifiler® HP and Trio kits have been optimized and validated as part of a complete solution when used with HID Real-Time PCR Analysis Software v1.2 and the AmpFℓSTR® Identifiler® Plus*, AmpFℓSTR® NGM SElect™*, or GlobalFiler™ PCR Amplification Kits*. Validation studies have shown that the latest generation of quantification kits function as critical components of an improved and integrated workflow for more effective processing of forensic casework samples.

Validate with confidence

We performed developmental validation experiments for the Quantifiler® HP and Trio kits according to the Revised Guideline issued by the Scientific Working Group on DNA Analysis Methods (SWGDAM). Complete protocols and a summary of the experiments performed along with results obtained for developmental validation are available in the manuals [1, 2]. Each laboratory using the Quantifiler® HP and Trio kits should perform its own internal validation studies.

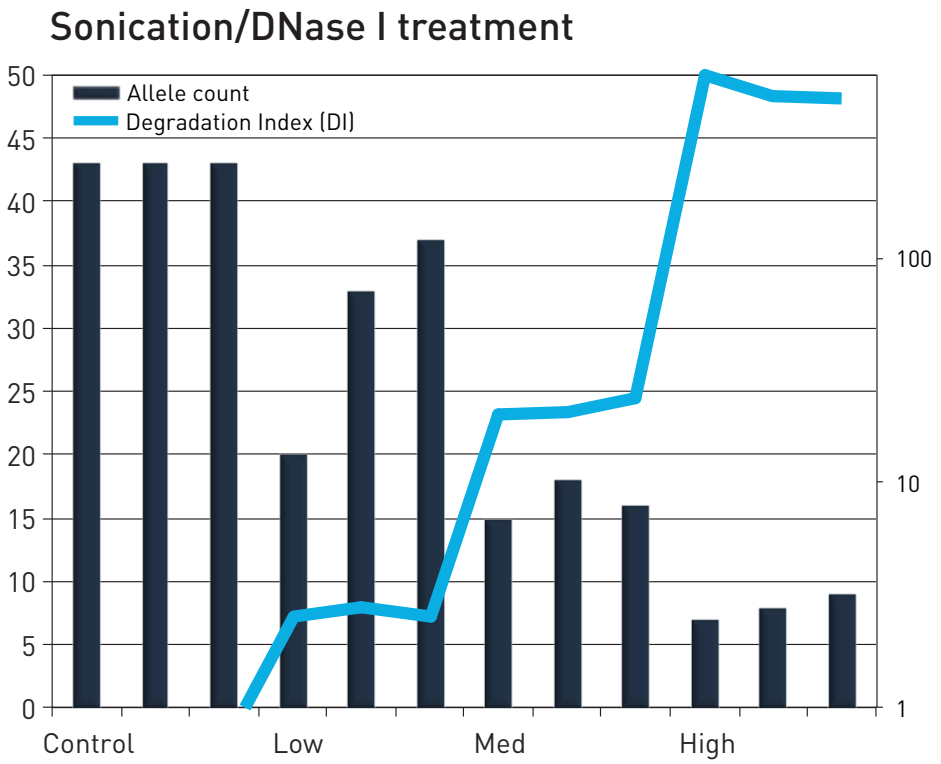


Figure 4. Degraded DNA study. Samples of purified human male genomic DNA were mechanically sheared with a sonicator to break up longer DNA strands, and then sheared DNA samples were digested with varying amounts of DNase I enzyme and different incubation times. STR allele recovery was inversely proportional to the Degradation Index.

Improved packaging minimizes the risk of reagent contamination and enhances ease of use

Each Quantifiler® HP and Trio kit includes all reagents necessary for the amplification, detection, and quantification of 400 reactions using the Applied Biosystems® 7500 Real-Time PCR System. The Quantifiler® HP and Trio DNA Quantification Kits have changed from a single box that contained one reagent bottle of 400 reactions to a two-box configuration, with one box containing a set of four screw-cap tubes of 100 reactions each for both the Primer Mix and PCR Reaction Mix, and a separate box for the DNA standard. This new configuration reduces the number of times analysts enter a bottle, helping to limit the possibility of contaminating the reagents.

Each set of PCR Reaction Mix and Primer Mix tubes in the kit contains enough reagents to set up one 96-well reaction plate. This minimizes the time it takes to plan and set up reaction plates. The DNA standard has been moved to a separate box, allowing for easy storage in the appropriate temperature conditions. Additionally, the kit box utilizes peel-and-stick reagent lot barcode labels, which allow customers to separate out and track the individual reagents that will be moved to separate storage conditions upon arrival at the laboratory.

Ordering information

Product	Size	Cat. No.
Quantifiler® HP DNA Quantification Kit	400 reactions	4482911
Quantifiler® Trio DNA Quantification Kit	400 reactions	4482910
HID 7500 Calibration Starter Kit	NA	A25019
HID Real-Time PCR Analysis Software v1.2	Single User License	A24664

References

1. Quantifiler® HP and Trio DNA Quantification Kits User Guide. Pub. No. 4485354, Rev B.
2. Quantifiler® HP and Trio DNA Quantification Kits Quick Reference Card. Pub. No. 4485356, Rev A.

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