Technical Information

Investigator® Quantiplex® Pro Kit

Highest sensitivity and quality assessment for your forensic samples

The Investigator Quantiplex Pro Kit quantifies human and male DNA in a sample using quantitative real-time PCR, based on TaqMan® technology and QIAGEN's fast reaction chemistry. An internal PCR control (IPC) is incorporated into the kit to reliably and consistently identify PCR inhibitors that may interfere with downstream processes, such as STR PCR. In addition, the effect of DNA degradation on downstream STR analysis can be accurately assessed. The kit works with any human ID workflow, regardless of the upstream sample prep or downstream STR kit used, and is designed to complement the Quality Sensor included in several of our Investigator STR PCR assays (Investigator ESSplex SE QS, Investigator 24plex QS and Investigator 24plex GO! kits), enabling an unparalleled level of workflow optimization.

- Highly accurate results for male DNA, even with a high background of female DNA (up to 400,000:1)
- Precise assessment of DNA degradation
- High dynamic range from 0.5 pg/µl – 200 ng/µl
- Reliable information about inhibitors that correlates well with STR results
- Fast-cycling technology and rapid quantification in 1 hour

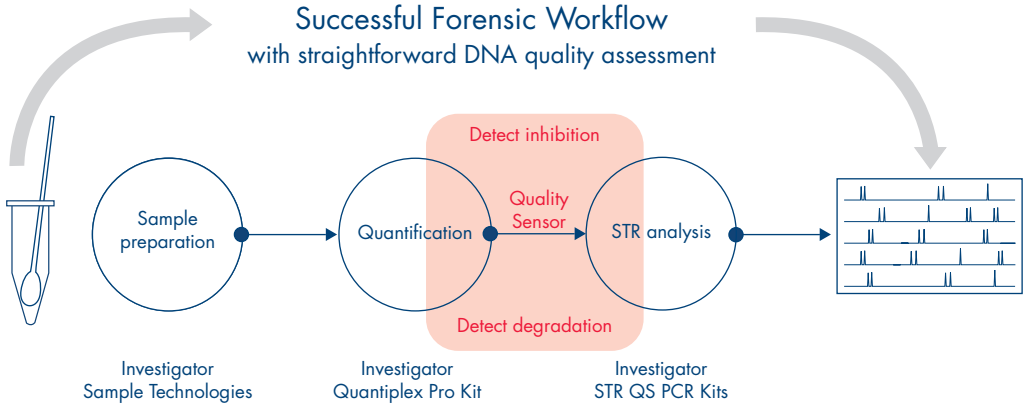

Figure 1. The Investigator Quantiplex Pro Kit provides information on inhibition and degradation, as well as accurate human and male DNA quantification for downstream STR PCR. When STR kits containing the Quality Sensor are used in combination with the Investigator Quantiplex Pro Kit, workflow synergy is maximized since quantification and the STR PCRs work together to predict and confirm PCR performance.

Partners against crime

Quantification is only one part of a complex workflow, where all the steps need to be optimized and aligned to get the best possible results from your samples. This is why we have developed the Investigator Quantiplex Pro Kit so you can achieve the most meaningful data in the context of downstream STR PCR. The alignment of quantification and the STR PCRs is achieved using the IPC in our Investigator Quantiplex Pro Kit and the Quality Sensor included in our Investigator STR PCR Kits. The IPC is designed to behave similarly to the Quality Sensor in the presence of inhibitors. Therefore, the two different PCR assays work together to predict and confirm the performance of the STR PCR, allowing you to interpret your results with confidence.

Sensitivity and dynamic range

Forensic and human identity samples are notoriously challenging to work with because the amount of DNA can vary from extremely high (e.g., visible blood stains) to trace levels (e.g., single cells or cell-free DNA). Furthermore, in many sexual assault samples, the quantity of male DNA within a much larger background of female DNA needs to be accurately determined. For these reasons, the Investigator Quantiplex Pro Kit has been developed with reliable, multi-copy male and human DNA targets to provide unparalleled sensitivity and dynamic range. These targets enable accurate and reproducible quantification of both male and human DNA, regardless of the input DNA amount used (Figure 2).

"The Quantiplex Pro DNA quantification kit from QIAGEN is very sensitive and does an exceptional job of detecting low levels of male DNA in mixtures with an overwhelming majority of female DNA." **– DNA Technical Lead of a North American Beta Testing Forensic Laboratory**

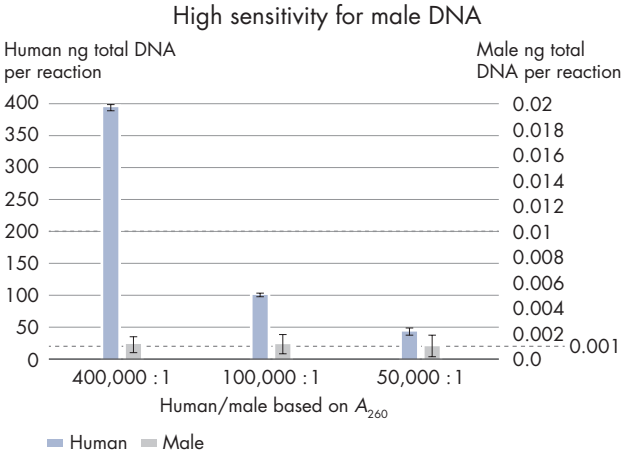

Figure 2. Highly accurate and sensitive results for male DNA. Three different mixture sets were tested in quadruplicate. Each set contains a total of 1 pg of male DNA, while the amount of female DNA was increased from 50–400 ng. The Investigator Quantiplex Pro Kit accurately quantified the increasing amount of female DNA, whereas the amount of male DNA was consistently quantified at approximately 1 pg in total.

Reliable detection of PCR inhibition

Forensic samples can be delivered on any matrix depending on the case circumstances, and therefore, they frequently contain substances that can negatively impact DNA profiling. Even with the most robust sample prep method, some of these substances are still present in the purified DNA and will inhibit the downstream STR PCR, unless identified and corrected for. The Investigator Quantiplex Pro Kit includes an IPC that is both sensitive to PCR inhibitors and responds to inhibition in a highly linear way (Figure 3).

Therefore, the IPC not only identifies inhibition, at a sensitivity closely matched to that of STR assays, but also provides accurate quantitative feedback on inhibitor levels that can be used as a basis for choosing the most appropriate downstream analysis for your samples.

"In the presence of inhibitors, it outperformed the DNA quantification kit currently in use at our laboratory." **– DNA Technical Lead of a North American Beta Testing Forensic Laboratory**

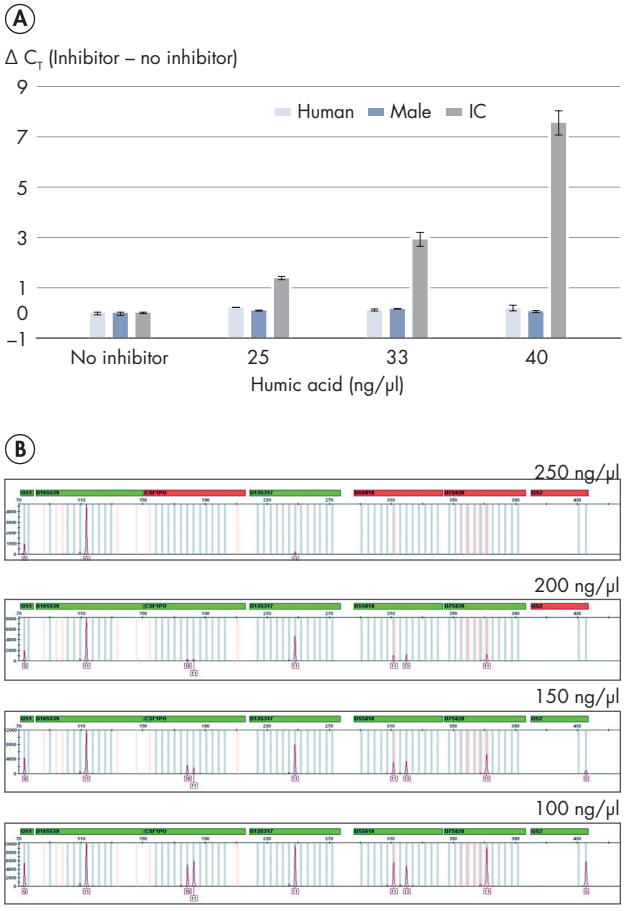

Figure 3. Identification of PCR inhibition. A The Investigator Quantiplex Pro Kit was run in the presence of 0, 25, 33 and 40 ng/µl humic acid on the Applied Biosystems® 7500 Real-Time PCR System. The internal control (in gray) acts as quality sensor and reports the presence of the inhibitor with a C_T shift, while quantification (in light blue and dark blue for human and male DNA) remains reliable up to a final humic acid concentration of 40 ng/µl. **B** The Investigator 24plex QS Kit shows resistance to humic acid up to 200 ng/µl (based on the final concentration in the STR reaction), while the Quality Sensor in the Investigator 24plex Kit (the far right homozygous locus of each line) reports the presence of inhibitors with the same level of sensitivity as the Investigator Quantiplex Pro Kit.

Degradation

Even with a high quantity of DNA in your sample, your STR PCR is unlikely to be successful if the DNA is severely degraded. In order to save time, reagents and costs, and to enable you to choose the right downstream analysis for your samples, the Investigator Quantiplex Pro Kit includes Degradation and Human DNA targets to provide an accurate assessment of DNA degradation. The Degradation target is over 350 base pairs in size, enabling the maximum possible insight into the quality of high-molecular-weight loci in the STR profile. Moreover, because the Human and Degradation targets are so different in size, the effect of degradation on their respective amplification is significantly different. The relative amplification of these two fragments is a powerful tool for predicting DNA degradation in your samples and this can be directly correlated to the likely performance of your STR PCR.

"The addition of the degradation index provides useful information about the quality of the DNA present in a sample, and would be particularly useful in deciding between amplification with a traditional autosomal STR kit versus a mini-STR kit for laboratories with the option of either kit."
– DNA Technical Lead of a North American Beta Testing Forensic Laboratory

Technical specifications

Parameter	Investigator Quantiplex Pro Kit
Human (length/channel)	91 bp / FAM™
Degradation (length/channel)	353 bp / ATTO® 550
Male (length/channel)	81 bp / ATTO 647N
IC (length/channel)	434 bp / JOE®
Volume per reaction	20 µl
PCR speed on the ABI 7500	60 min

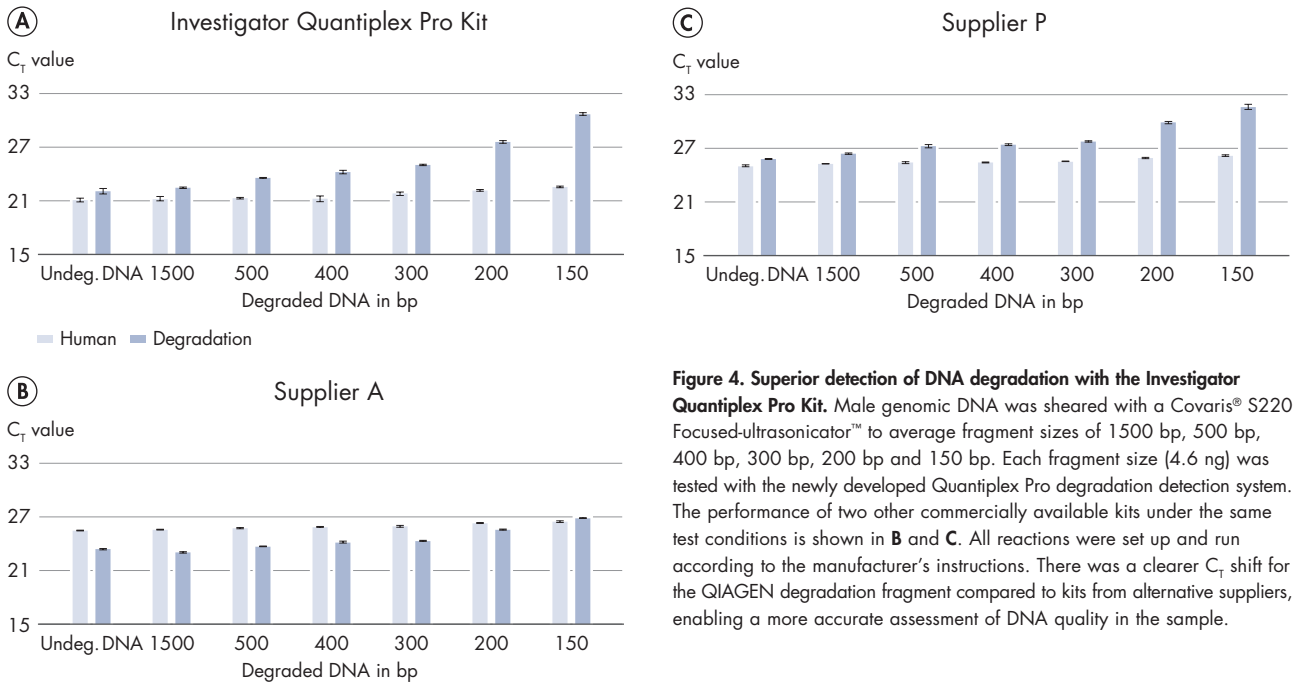

Figure 4. Superior detection of DNA degradation with the Investigator Quantiplex Pro Kit. Male genomic DNA was sheared with a Covaris® S220 Focused-ultrasonicator™ to average fragment sizes of 1500 bp, 500 bp, 400 bp, 300 bp, 200 bp and 150 bp. Each fragment size (4.6 ng) was tested with the newly developed Quantiplex Pro degradation detection system. The performance of two other commercially available kits under the same test conditions is shown in **B** and **C**. All reactions were set up and run according to the manufacturer's instructions. There was a clearer C_T shift for the QIAGEN degradation fragment compared to kits from alternative suppliers, enabling a more accurate assessment of DNA quality in the sample.

Ordering Information

Product	Contents	Cat. no.
Investigator Quantiplex Pro Kit (200)	Primer Mix, Reaction Mix, Control DNA M1, Nucleic Acid Dilution Buffer	387216
QIAgility® System HEPA/UV (incl. PC)	Robotic workstation for automated PCR setup (with UV light and HEPA filter); including notebook computer, QIAgility Software, installation and training, 1-year warranty on parts and labor	9001532
QIAsymphony® AS	QIAsymphony assay setup module, 1-year warranty on parts and labor	9001301

For up-to-date licensing information and product-specific disclaimers, see the respective QIAGEN kit handbook or user manual. QIAGEN kit handbooks and user manuals are available at **www.qiagen.com** or can be requested from QIAGEN Technical Services or your local distributor.

Learn more about our quantification solutions for human identity and forensic testing at **www.qiagen.com/quantiplexkits**.

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