Promega

PowerPlex® 35GY System

Optimized for Next Generation CE STR Analysis

The Power to Solve...from Sample to Analysis

PowerPlex® 35GY System: First Commercially Available 8-Dye STR Multiplex

Capillary electrophoresis (CE) technology remains the standard in forensic DNA laboratories for producing reliable data in a cost-effective and timely manner. The new Spectrum CE System builds on this CE tradition of 4-, 5- and 6-color separation and offers an expanded spectral capacity to allow for the separation of eight color channels.

The PowerPlex® 35GY System is our first 8-dye PowerPlex® STR System and offers the following features:

- Rapid cycling conditions for extracted DNA and database samples
- Inclusion of two Quality Indicators
- Consists of Amelogenin, 23 autosomal STR loci and 11 Y-STR loci in a single multiplex
- 15 autosomal STR loci are less than 250bp

The Power of 8-Dye Chemistry

The expansion of the number of Combined DNA Index System (CODIS) core loci in the United States from 13 to 20 short tandem repeat (STR) loci was designed to reduce the potential for adventitious matches occurring within the database, to increase international compatibility for data sharing and to increase discrimination power in missing persons cases.

Although manufacturers introduced 6-dye multiplexes to accommodate the additional loci, it became clear that the multiplexes had limited space for these loci and any future loci additions would not be ideal due to space limitations in each channel. Shifting larger markers into two additional dye channels allows for improved analyses (Figure 1). For instance, the addition of more loci in a kit provides more information from the sample. The narrow range of amplicon lengths enables more consistent results and allows for more mini-STRs, which can provide better results with degraded samples.

The PowerPlex® 35GY System (Figure 2) allows co-amplification and fluorescent detection of the 20 CODIS core loci plus Amelogenin and DYS391 for gender determination. Penta D, Penta E and SE33 also are included to increase discrimination and allow searching of databases that contain profiles with these loci. With the two additional colors for detection, there are a total of 15 autosomal loci smaller than 250bp, aiding the analysis of difficult samples. Additionally, we've designed the layout to allow smaller amplicon sizes for these particularly informative loci: SE33, Penta E and D2S1338.

The inclusion of Y-STR loci increases the amount of genetic information obtained in a single amplification and can provide valuable data for familial searching or can assist in the analysis of sexual assault evidence by helping to better determine the number of male contributors in mixture samples.

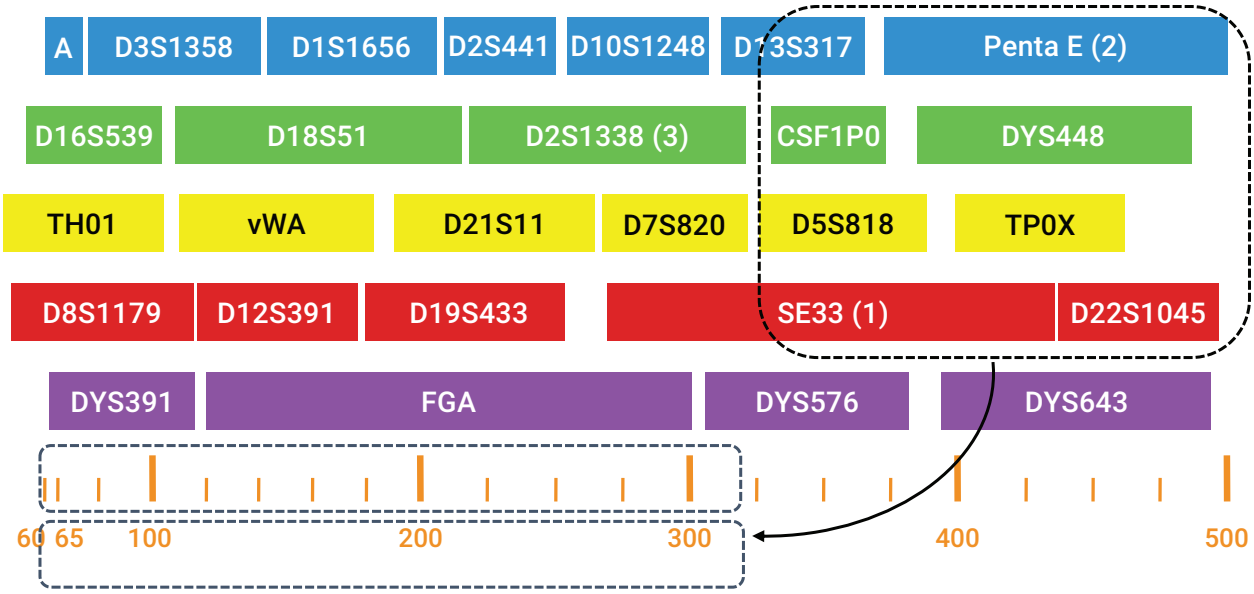

Figure 1. Highlighting the Power of 8-Dye Channels. In this figure, we've shifted loci from the PowerPlex® Fusion 6C System into the 2 additional dye channels. This frees up space for more loci to be included, providing more information from a sample and improving analyses.

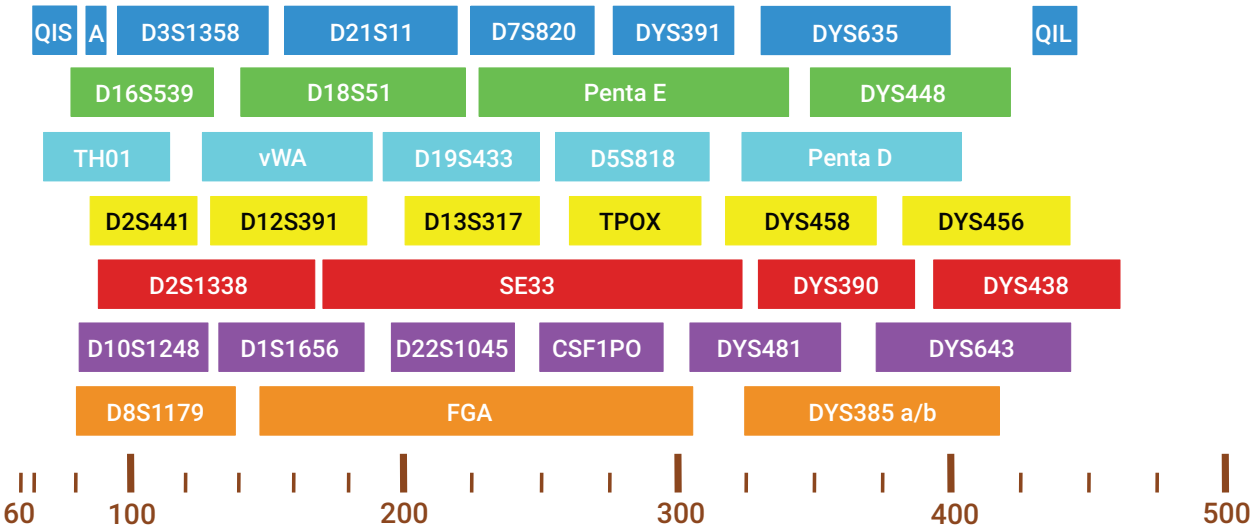

Figure 2. Configuration of the PowerPlex® 35GY System. The PowerPlex® 35GY System allows co-amplification and detection of 35 loci: Amelogenin; 23 autosomal STR (D3S1358, D21S11, D7S820, D16S539, D18S51, Penta E, TH01, vWA, D19S433, D5S818, Penta D, D2S441, D12S391, D13S317, TPOX, D2S1338, SE33, D10S1248, D1S1656, D22S1045, CSF1PO, D8S1179, and FGA); and 11 Y-STR loci (DYS391, DYS635, DYS448, DYS458, DYS456, DYS390, DYS438, DYS481, DYS643, and DYS385a/b). Two Quality Indicators (QIS and QIL) are also included to assist with data interpretation.

Get The Most Information from Your Sample

With current caseloads increasing, no one wants to have to repeat their STR analyses on DNA samples that failed to yield a full profile due to the presence of inhibitors or degradation. The PowerPlex® 35GY System is the first PowerPlex® STR system to include two Quality Indicators, QIS and QIL. These two amplification products provide additional valuable information about your samples to help you interpret your STR results.

The Quality Indicators can take the guesswork out of determining whether the amplification was successful by confirming if there is DNA present or not. Additionally, they can indicate inhibition that was not observed during the quantification step. Lastly, knowing whether the sample was inhibited or degraded enables you to make an informed decision to rework the samples. If inhibited, laboratories can choose to dilute samples for PCR or re-purify them to reduce inhibitor load. If the sample is degraded, a laboratory may choose to add more template DNA (if available) to push the high molecular weight markers above threshold.

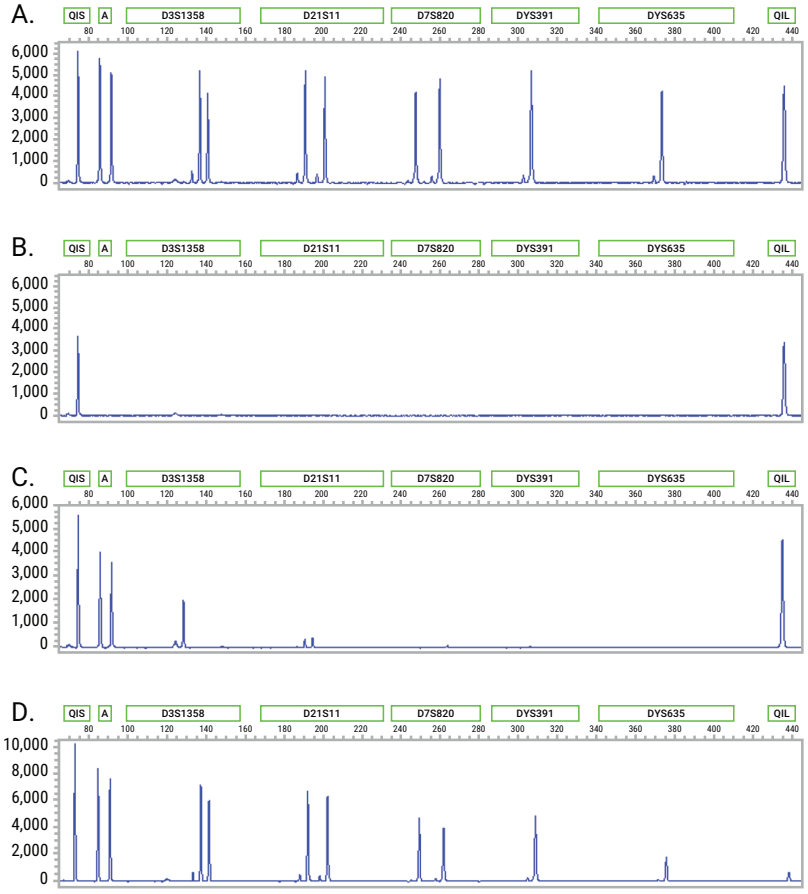

Figure 3. Quality Indicators Aid Data Interpretation.

Panel A shows a successful, balanced STR profile with balanced QIS and QIL peaks, indicating successful amplification from high quality template DNA.

Panel B shows no STR peaks with balanced QIS and QIL peaks, indicating that no template DNA was amplified in the sample.

Panel C is an example of a sloping STR profile with balanced QIS and QIL peaks, indicating that the sample is degraded.

Panel D is a sloping STR profile but with a low or missing QIL peak on the right. This suggests that the sample is inhibited.

In cases where the electropherogram shows that neither QI peak is present, then amplification has failed.

The PowerPlex® 35GY System is designed for amplification of extracted DNA and direct-amplification samples. Slight protocol variations are recommended for optimal performance with each template source. PowerPlex® 35GY System is optimized to work with 1ng of extracted DNA in a 25µl reaction volume (see Figure 4).

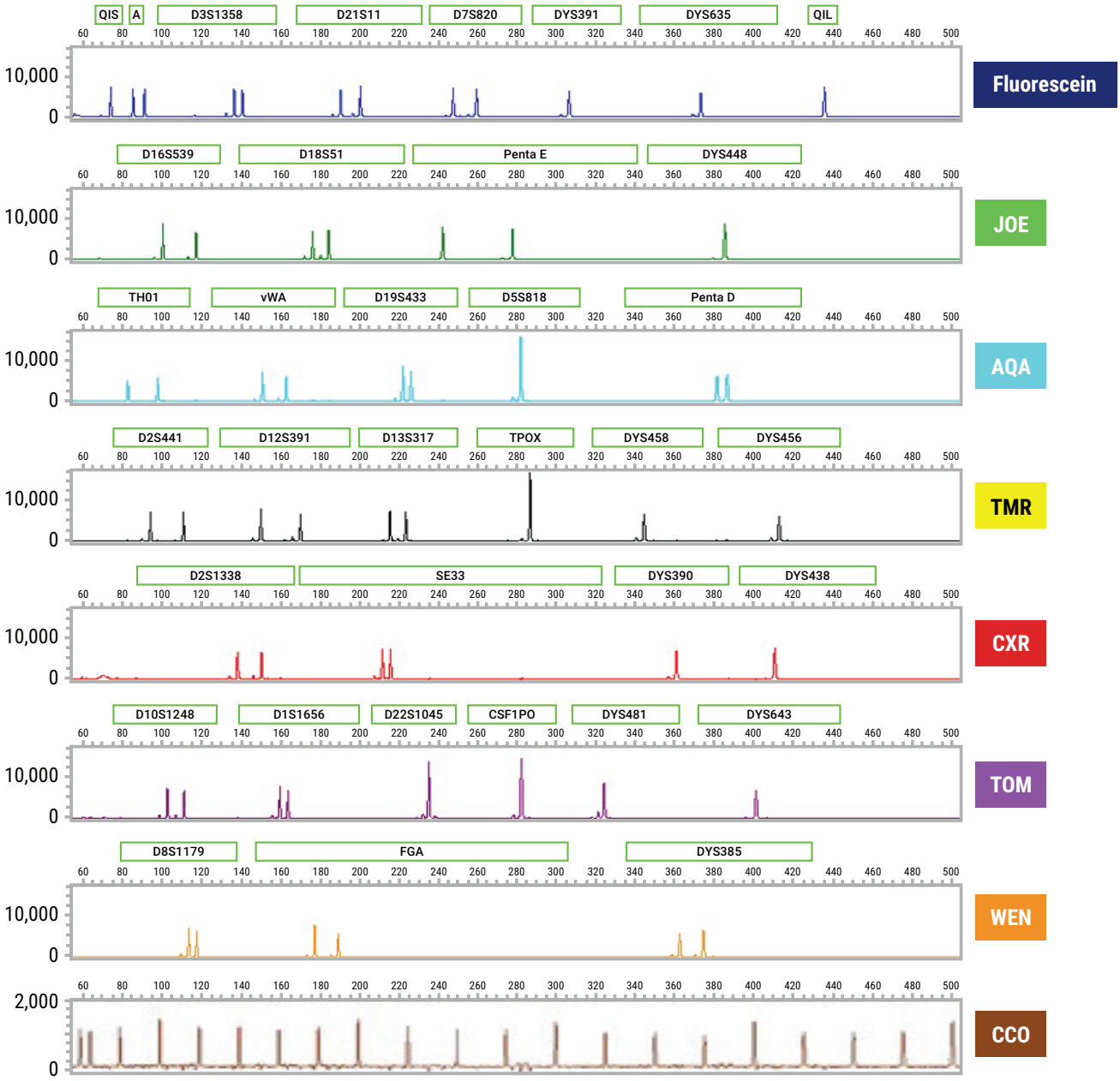

Figure 4. Amplifying Extracted DNA with the PowerPlex® 35GY System. The 2800M Control DNA (1ng) was amplified using the PowerPlex® 35GY System. Amplification products were mixed with CCO Internal Lane Standard 500 and separated on the Spectrum CE System. Electropherogram was generated using GeneMarker® HID Software for Spectrum CE Systems. Dye names are shown on the right.

As with all our PowerPlex® Systems, the PowerPlex® 35GY System enables half volume direct amplification reactions with buccal swabs and blood and buccal samples on lytic and nonlytic storage card punches. In the run below, DNA from a buccal FTA® punch was amplified for 25 cycles in a 12.5µl reaction volume. Amplified samples were separated on the Spectrum CE System.

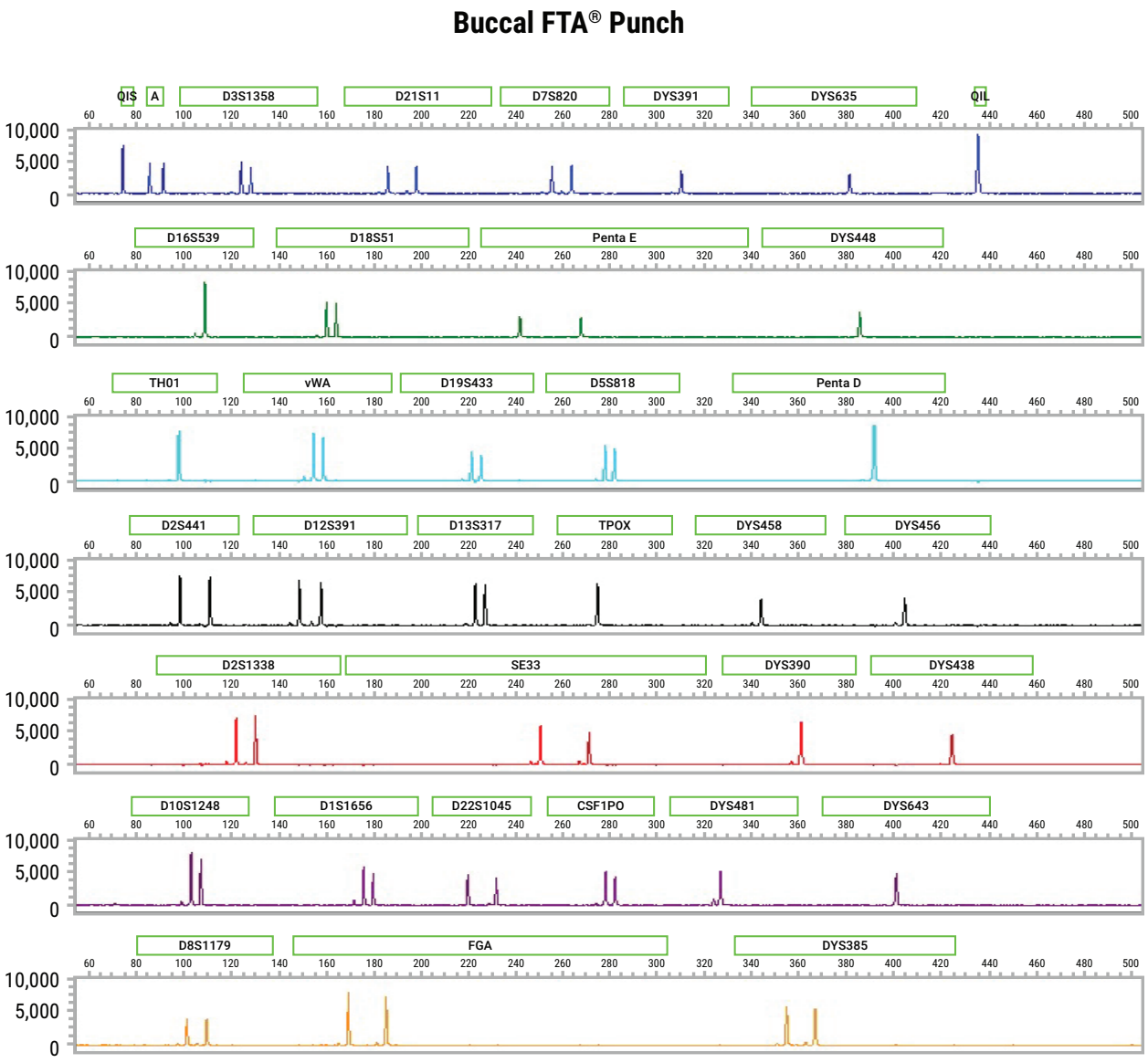

Figure 5. Direct Amplification with the PowerPlex® 35GY System. An FTA® punch of buccal cells was direct-amplified using the PowerPlex® 35GY System. Amplification products were separated on the Spectrum CE System and analyzed using GeneMarker® HID Software for Spectrum CE Systems.

More Data with Low DNA Input

The kit is optimized to work with 1ng of extracted DNA, but the kit performs well with lower amounts of input DNA. Because the total number of loci is higher in the PowerPlex® 35GY System than in other commercially available kits, the total number of alleles will be higher as well, providing more useful results to casework laboratories. Figure 6 shows that 69% of alleles are called at 31.25pg input DNA, and almost 100% of alleles are called at 62.5pg input.

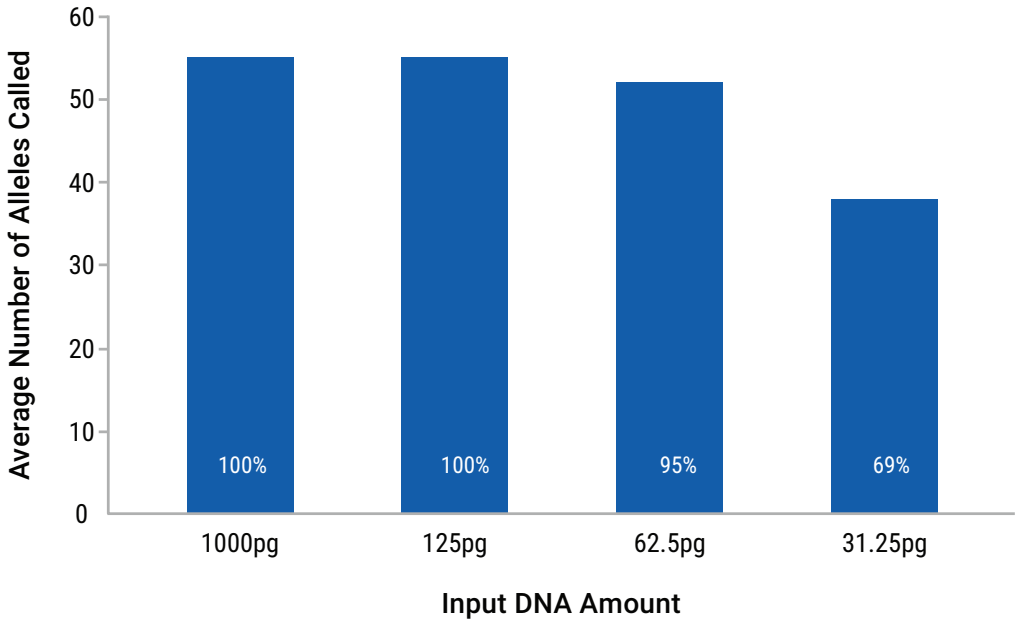

Figure 6. Sensitivity of the PowerPlex® 35GY System. Varying amounts of extracted DNA were amplified using the PowerPlex® 35GY System. Amplification products were separated on the Spectrum CE System and analyzed using GeneMarker® HID Software for Spectrum CE Systems. The number of and percent alleles called are an average of four amplifications with each input DNA amount.

PowerPlex® 35GY has more loci than any other commercially available kits, meaning more alleles and more useful results.

Robust Performance in the Presence of Strong Inhibitors

Forensic DNA samples may contain inhibitors that cannot be effectively removed after the extraction and purification procedures. These inhibitors can interfere with PCR amplification and reduce PCR efficiency or even cause complete amplification failure. The superior buffer system included in the PowerPlex® 35GY System ensures strong performance even with the most inhibited samples. The kit minimizes the need to re-amplify samples generally thought to be too challenging to run, saving your lab countless hours of repeat analyses.

Figure 7 below highlights the average number of alleles called for all loci in the kit as well as only the autosomal loci included in the kit. Complete STR profiles were generated in the presence of 500µM hematin as well as in samples containing 100ng/µl of humic acid. A slight drop in the average number of alleles called did occur when samples were exposed to 750µM hematin, and a larger drop was seen when samples were subjected to 1000µM of hematin and to higher amounts of humic acid.

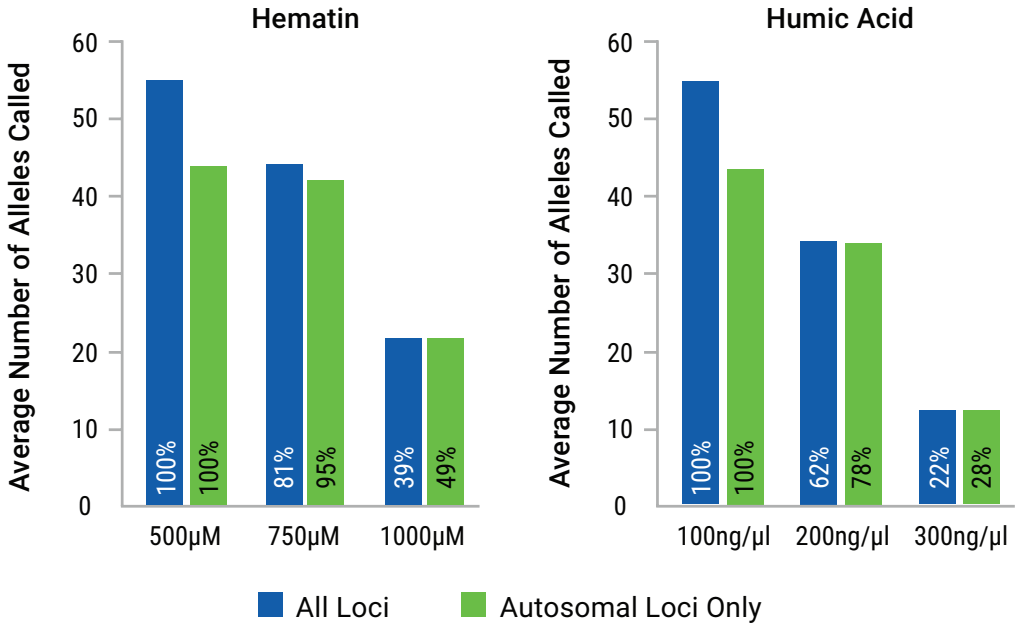

Figure 7. Robustness of the PowerPlex® 35GY System. Extracted DNA (1ng) in the presence of varying concentrations of PCR inhibitors was amplified using the PowerPlex® 35GY System. Amplification products were separated on the Spectrum CE System and analyzed using GeneMarker® HID Software for Spectrum CE Systems. The number of and percent alleles called are an average of four amplifications with each inhibitor concentration.

More Mini-STRs for Improved Success with Difficult Samples

Highly degraded samples are often seen in missing persons cases, forensic casework and mass disasters. Various environmental conditions can lead to severe degradation of human DNA samples, which often produce incomplete or no STR profiles due to larger loci that often "drop out". The inclusion of more loci as mini-STRs in the PowerPlex® 35GY System results in better and more complete profiles, making it an ideal option to process cold case samples as well as newer, more difficult samples. Figure 8 shows an electropherogram of degraded DNA from a femur where up to 22 loci are called. Also note the high QIL peaks, indicating that the amplification was not inhibited.

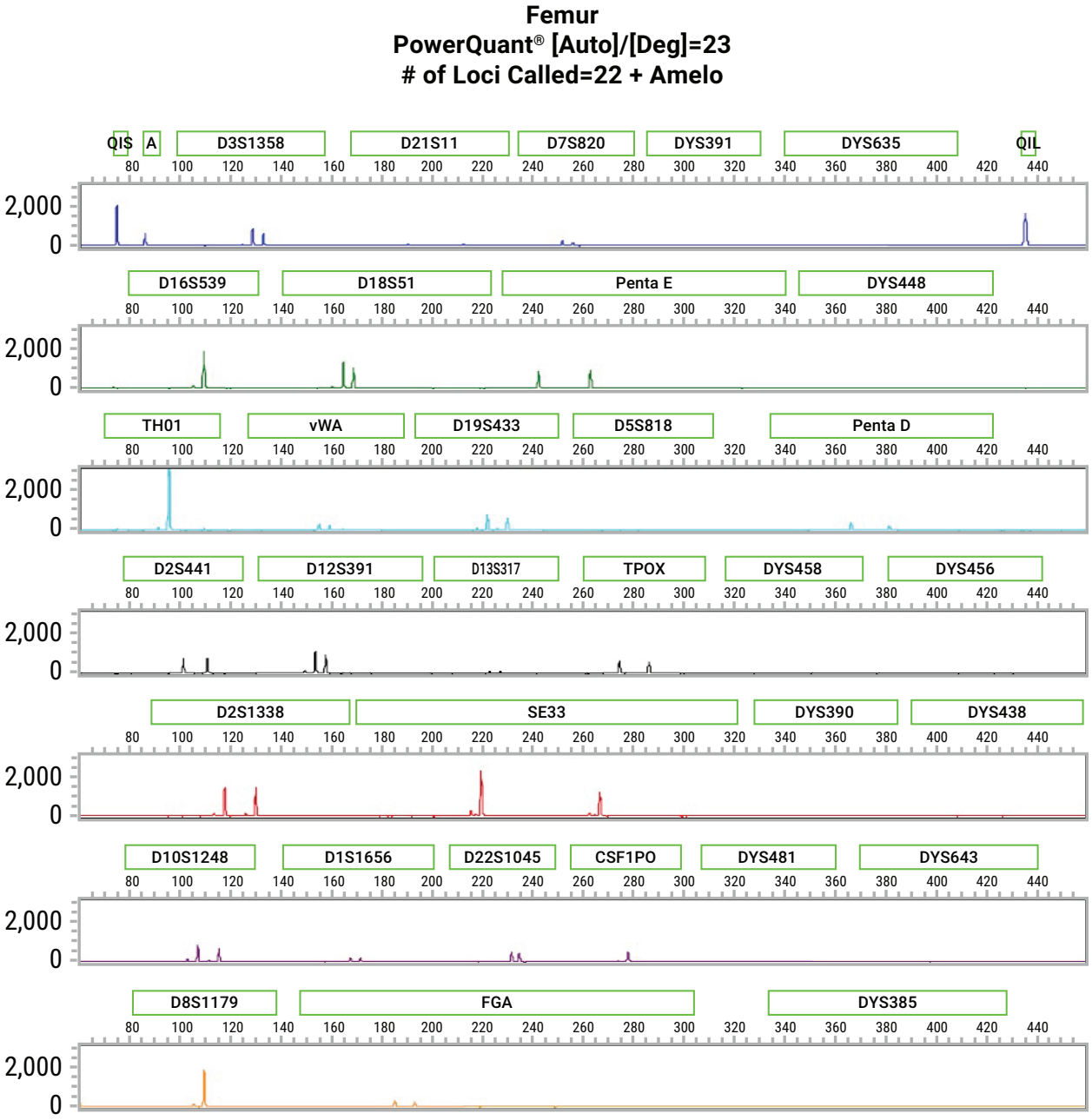

Figure 8. Better Results with More Mini-STRs in the PowerPlex® 35GY System. Extracted DNA (1ng) from a femur was amplified using the PowerPlex® 35GY System. Amplification products were separated on the Spectrum CE System and analyzed using GeneMarker® HID Software for Spectrum CE Systems.

More Successful Mixture Analysis

The enhanced sensitivity, superior capability to overcome inhibition and the inclusion of Y-STRs in the PowerPlex® 35GY System will enable laboratories to analyze and interpret mixtures more easily and reliably than was previously possible. The autosomal loci included in the PowerPlex® 35GY System help determine the total number of contributors in mixture samples, while the Y-STRs estimate the total male contributors, thereby enabling more genetic information to be obtained in a single amplification. This additional information can be invaluable in cases where familial searching is done or in the analysis of sexual assault kit evidence.

Figure 9 shows electropherograms for two 4-person mixtures. The Y-STR loci in the electropherogram in Figure 9A clearly indicates that the mixture contains at least 2 male. For the electropherogram shown in Figure 9B, DYS391 underestimates the number of male contributors in the mixture. However, multiple other Y-STRs correctly estimates the number of male contributors.

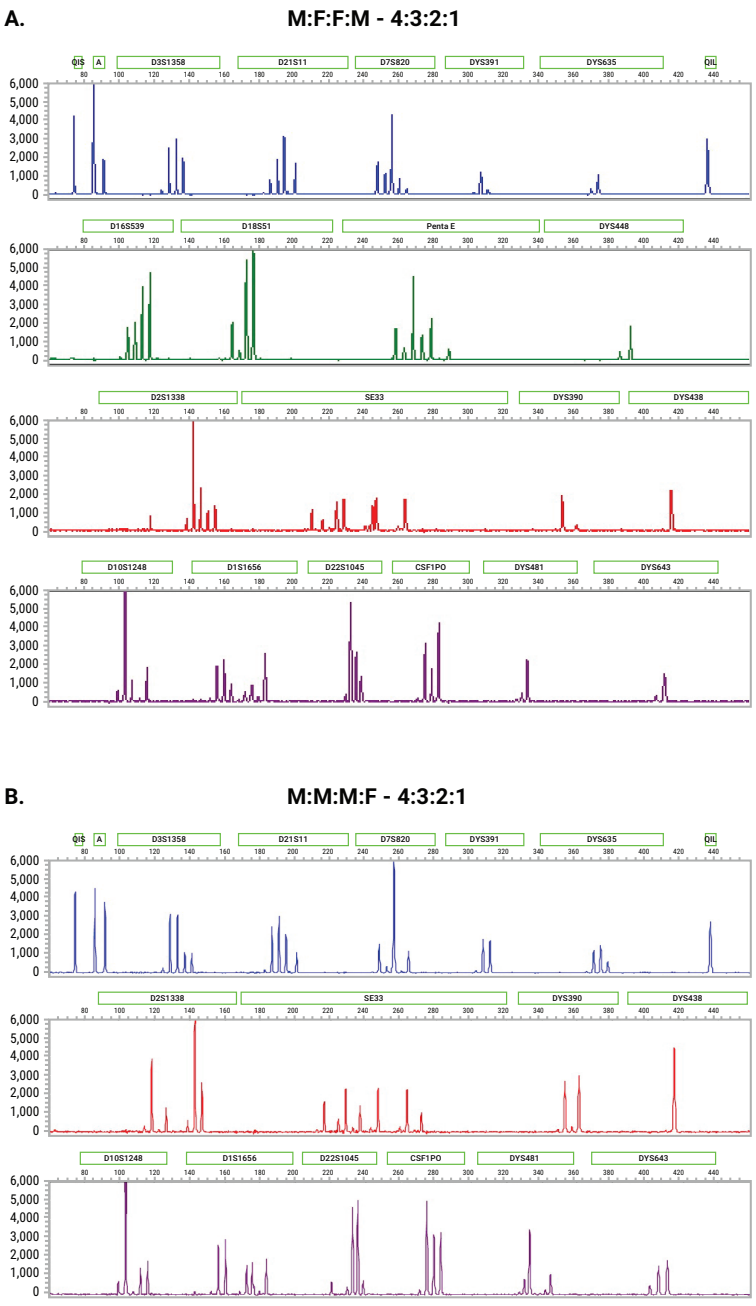

Figure 9. Better Mixture Analysis with More Y-STRs in the PowerPlex® 35GY System. Extracted DNA (1ng) from 4-person mixtures were amplified using the PowerPlex® 35GY System. Amplification products were separated on the Spectrum CE System and analyzed using GeneMarker® HID Software for Spectrum CE Systems.

Ordering Information

Product	Size	Cat.#
PowerPlex® 35GY System	200 (or 400 direct amp) reactions	**DC3520**
PowerPlex® 8C Matrix Standard	5 preps	**DG5010**
PowerPlex® 35GY QI Reagent	800µl	**DM1551**

Related Products	Size	Cat.#
Spectrum CE System, 8-Capillary	1 each	**CE1008**
GeneMarker®HID Software for Spectrum CE Systems	Local (1 seat)	**CE3001**
	Network (1 seat)	**CE3010**
	Client (1 seat)	**CE3011**
PunchSolution™ Kit	100 preps	**DC9271**
SwabSolution™ Kit	100 preps	**DC8271**
PowerQuant® System	200 reactions	**PQ5002**
	800 reactions	**PQ5008**

The enhanced sensitivity, superior capability to overcome inhibition and the inclusion of Y-STRs in the PowerPlex® 35GY System allows laboratories to analyze and interpret mixtures more easily and reliably than was previously possible.

The Power to Solve...*from Sample to Analysis*

DNA Isolation

Maxwell® FSC Instrument
A compact, plug-and-play instrument for automated DNA extraction

Maxwell® RSC 48 Instrument
Compact, automated nucleic acid purification platform that processes up to 48 samples simultaneously to yield high-quality nucleic acids

Maxprep™ Liquid Handler
A complete robotic nucleic acid purification system

Maxwell® FSC DNA IQ™ Casework Kit
Optimal extraction of DNA from forensic casework samples

Casework Extraction Kit
Preprocessing reagents to assist in DNA IQ™ chemistry extraction of DNA from challenging samples

Differex™ System
Easy separation of sperm and epithelial fractions

DNA IQ™ System
Manual and large platform automatable purification of DNA free of PCR inhibitors

Casework Direct System
Rapid screening of sexual assault evidence and processing of "Touch" DNA samples prior to quantification of human DNA using the PowerQuant® System and amplification using PowerPlex® Systems

Bone DNA Extraction Kit
Preprocessing reagents to assist the DNA IQ™ System in extracting DNA from skeletal remains

Quantification

Plexor® HY System
Quantitative PCR for both total human and male DNA in a single reaction

PowerQuant® System
A 5-dye, 4-target hydrolysis probe-based quantitative PCR assay for assessing total human and male DNA concentrations, degradation levels and the presence of inhibitors

STR Amplification

STR Amplification

PowerPlex® Fusion System (DA)
A rapid 24-plex suitable for casework, paternity and database testing and designed to meet the CODIS recommendations

PowerPlex® Fusion 6C System (DA)
A rapid 27-plex, including SE33, suitable for casework and database testing and designed to meet the CODIS recommendations

PowerPlex® Y23 System (DA)
Male-specific STR genotyping kit with 23 Y-STR loci; includes protocols for both casework and databasing

PowerPlex® ESX and ESI Fast Systems (DA)
Rapid human identification STR assays that meet ENFSI recommendations for use in casework, paternity and database testing

PowerPlex® 35GY System (DA)
An 8-color multiplex containing autosomal and Y-STR loci designed for use on the Spectrum CE System

Analysis

Spectrum CE System
8-capillary electrophoresis instrument that combines state-of-the-art STR analysis with increased run flexibility and 4-plate capacity

Spectrum Compact CE System
Benchtop, 4-capillary electrophoresis instrument capable of Sanger sequencing and fragment analysis

Massively Parallel Sequencing

Massively Parallel Sequencing

PowerSeq® 46GY System (DA)
Amplification of autosomal and Y-STR loci in a single multiplex for massively parallel sequencing on an Illumina MiSeq® platform

PowerSeq® CRM Nested System, Custom
Amplification and library preparation of mitochondrial control region for massively parallel sequencing on an Illumina MiSeq® platform

MPS Library Quantification

PowerSeq® Quant MS System
Quantification of prepared MPS libraries for Illumina platforms, enabling efficient pooling and flow cell representation

(DA) Direct Amplification Compatible

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