

Identity



NEWS FOR THE GENETIC IDENTITY COMMUNITY • WINTER 2009

Product Updates

PowerPlex® 16 HS System

Use a single kit for multiple applications. Both databasing and casework samples can be efficiently processed with the PowerPlex 16 HS System. The kit includes hot-start *Taq* DNA polymerase, which makes it more convenient to order, and an improved buffer and PCR protocol, which is 9 minutes shorter and makes it easier to use.

The current PowerPlex 16 System, which satisfies the needs of several major standardization bodies throughout the world, including INTERPOL, ENFSI, GITAD and FBI, will be soon available with a hot-start *Taq* DNA polymerase. All you need for PCR – PowerPlex 16 HS 10X Primer Pair Mix, PowerPlex HS 5X Master Mix (including hot-start *Taq* DNA polymerase), PowerPlex 16 HS Allelic Ladder Mix, Internal Lane Standard 600 and 9947A DNA for 100 or 400 reactions of 25µl each – is now in one kit.

• Power of Robustness

The improved buffer system helps you get a profile with difficult casework samples, which are often inhibited. With the PowerPlex 16 HS System, 0.5ng of DNA will give you a full profile in the presence of 200µM hematin or 50ng/µl humic acid.

• Power of Sensitivity

The PowerPlex 16 HS System provides the solution for degraded and low-level DNA samples by offering maximum sensitivity. It is optimized for use with 0.5ng of DNA; however, each lot is performance tested to provide reproducible results with as little as 0.1ng of DNA. Additional studies show interpretable results can be obtained with less than 0.1ng of DNA. Superior sensitivity means you can obtain full profiles with low-level samples.

• Power of Discrimination

The PowerPlex 16 HS System also contains two low-stutter, highly polymorphic pentanucleotide repeat loci, Penta E and Penta D. These loci add significantly to the discrimination power of the system. The low

stutter makes them ideal loci to evaluate DNA mixtures often encountered in forensic casework. Finally, the amelogenin locus is included to allow gender identification of each sample.

• Power of Flexibility

The PowerPlex 16 HS System is designed specifically for use with variety of genetic analyzers and includes both Internal Lane Standard 600 and allelic ladders. If you need bulk components, we can provide a solution tailored just for you.

We also provide panel and bin files for use with GeneMapper®ID software or the PowerTyper™ Macro for use with Genotyper® software. These tools may be downloaded at: www.promega.com/geneticidtools/panels_bins www.promega.com/geneticidtools/powertyper

When you are ready to validate a new system, Promega offers the Validation Reference Guide to simplify the validation process in your lab. If you need help, experienced technical support is ready to help you.

New ILS 600 eliminates artifacts at vWA locus

In recent years we have received reports of split or n-1 peaks at the vWA locus from PowerPlex®16 and PowerPlex® ES users. In addition, a vWA artifact has been observed that runs at approximately the n-10 position (10 bases smaller than the main peak) on the multicapillary ABI PRISM® 3100 and 3100-Avant and Applied Biosystems 3130 and 3130xl Genetic Analyzers and at approximately the n-18 position on the single-capillary ABI PRISM® 310 Genetic Analyzer (1).

The vWA split peak appears as two forms in electropherograms. In the most extreme cases it manifests as two distinct peaks, with the second peak migrating at the n-1 position relative to the main peak. In other cases the split peak presents as a shoulder on the left (i.e., smaller) side of the main peak (Figure 1). From the results of several experiments and observations that we recently published (2), we determined that the split peak is due to free

Spotlight

Laura Bimbashi



Laura Bimbashi joined the European Genetic Identity team in November.

She is based in London and will be a Forensic Regional Account covering UK, Scandinavia and other parts of Europe. Laura has an extensive experience in a forensic lab and will handle any of your sales or technical questions. You can contact her at:

laura.bimbashi@promega.com

Meeting



The 10th Meeting of the European STR Working Group

was held in Mannheim on November 25-27, 2009. Fifty attendees from 13 European countries were present. The key note speaker, Prof. Dr. Walther Parson, held an educational session on "Forensic mtDNA analysis – new developments". Additional presentations on automation, including high-throughput solutions for DNA extraction and differential extraction, miniSTRs, real-time PCR quantitation – advantages of new technologies, interesting and difficult cases, and PowerPlex® user experiences.

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unlabeled vWA primer annealing to its complementary sequence at the 3' end of the denatured TMR-labeled strand after injection into the capillary array. This happens despite the fact that the POP-4™ polymer should maintain DNA in a denatured state. To prevent this annealing we devised a simple competitor oligonucleotide, or sacrificial hybridization sequence (SHS), which is complementary to the unlabeled vWA primer. This SHS

oligonucleotide is present in the new Internal Lane Standard 600 (ILS 600) and, thus, is added to the amplified product post-amplification. Following electrokinetic injection, the SHS oligonucleotide anneals to the unlabeled vWA primer, thereby preventing formation of the partially double-stranded species and consequently the split peak.

Several lines of evidence suggested that the n-10 artifact also was due to double-stranded DNA. We chose to add two 30mer oligonucleotides, which we named complementary oligonucleotide target 1 and 2 (COT1 and COT2; Figure 2). The presence of these three oligonucleotides (SHS, COT1 and COT2) in the new ILS 600 effectively eliminated both the split peak and n-10 artifacts. This also resolved the n-18 artifact seen on the ABI PRISM® 310 Genetic Analyzer (2). Testing of the new ILS 600 by laboratories outside of Promega has confirmed this result.

This article is intended as a brief overview. A more detailed report is available online at: www.promega.com/profiles/1102/ProfilesInDNA_1102_13.html The full report was published recently (2).

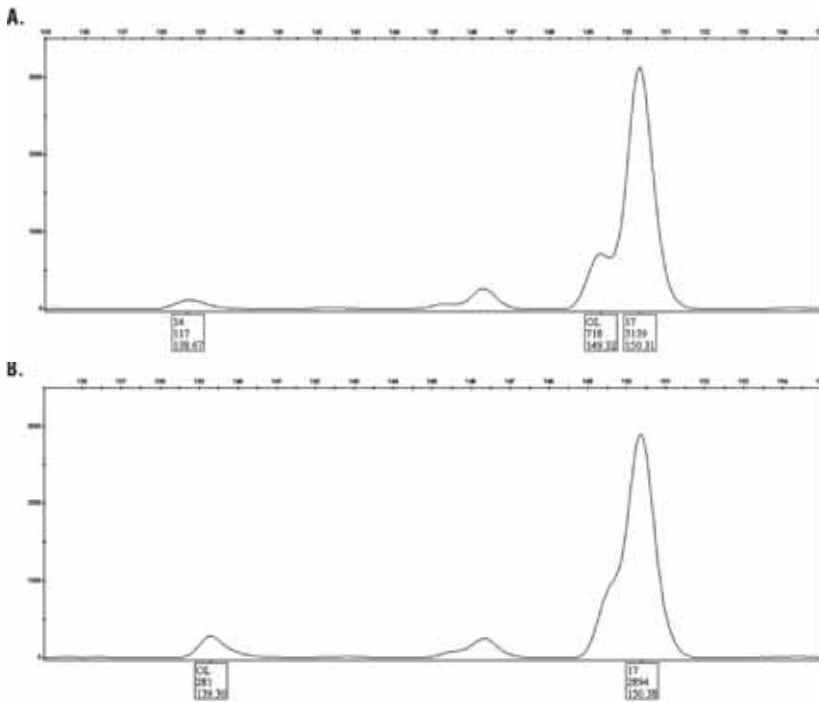


Figure 1: Artifacts at the vWA locus in the PowerPlex 16 System. We amplified 1ng of human genomic DNA using the PowerPlex 16 System and GeneAmp® PCR System 9700 for 32 cycles, then analyzed amplified products using an Applied Biosystems 3130 Genetic Analyzer.

Panel A: The vWA locus shows a defined split peak in roughly the n-1 position relative to the main peak and an n-10 peak, which migrates 11.64 bases shorter than the main peak at the 14 allele position.

Panel B: The split peak manifests as a shoulder. Note that the n-10 peak now migrates 11.08 bases shorter than the main peak and migrates "off-ladder". Peak labels are (from top to bottom) allele designation, peak height and size.



Figure 2: Schematic diagram of COT1, COT2 and SHS oligonucleotide hybridization to the vWA amplicon. The yellow line represents the TMR-labeled strand of the vWA amplicon. The black line represents the complementary unlabeled strand. The grey box denotes the location of short tandem repeats. The sites where the COT1, COT2 and SHS oligonucleotides anneal to the unlabeled strand are indicated.

References

1. *PowerPlex® 16 System Technical Manual #TMD012*, Promega Corporation.
2. McLaren, R. *et al.* (2008) Post-injection hybridization of complementary DNA strands on capillary electrophoresis platforms: A novel solution for dsDNA artifacts. *Forensic Sci. Int. Genetics* **2**, 257-73.

Meetings

Save the date

The original meeting on human identification is turning 20 years old and still going strong. **The 20th International Symposium on Human Identification** will be held October 12-15, 2009. The venue will be the JW Marriott Las Vegas Spa & Resort in Summerlin, Nevada. Pre-and post-show workshops will be held on system validation, statistics, mixture interpretation and ethics in forensics. Specialized trainings will include a DNA auditors workshop and Technical Leaders session.



Remember the date with a copy of our special commemorative poster. We hope to see you there!

Promega will attend the **29. Spurenworkshop der Deutschen Gesellschaft für Rechtsmedizin**, which will take place in Münster, February 27-28, 2009.



A user workshop will be organized in the morning of February 27, 2009. Watch for your invitation!

