# Development of the PowerPlex® Fusion System

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#### **Abstract**

The PowerPlex® Fusion System includes all the loci in Section A of the proposed expanded CODIS core loci (D.R. Hares, Addendum to expanding the CODIS core loci in the United States, Forensic Sci. Int. Genet. 6 (2012) e135.). It also includes two loci from CODIS Section B (TPOX and D22S1045) and two highly polymorphic pentanucleotide loci, Penta E and Penta D. These Penta loci have been used worldwide for more than 10 years, and are included in greater than one million profiles in the CODIS database. The system is based upon the same proven five color chemistry used in several of the systems Promega introduced in the last two years. This allows for use of the kits on the Applied Biosystems 31xx and 3500 series Genetic Analyzers. The combination of these 23 STR loci and Amelogenin make this multiplex an effective tool for human identification. The PowerPlex® Fusion System is optimized for extracted DNA, but allows for direct amplification from samples on FTA® paper (GE Healthcare/Whatman). It can be used in combination with Promega's PunchSolution™ Kit to amplify samples on other paper substrates or with Promega's SwabSolution™ Kit for buccal swabs. The system employs rapid cycling (one hour thirty minutes) providing significant time savings.

## **System Configuration**



Layout of PowerPlex® Fusion Loci

#### Includes:

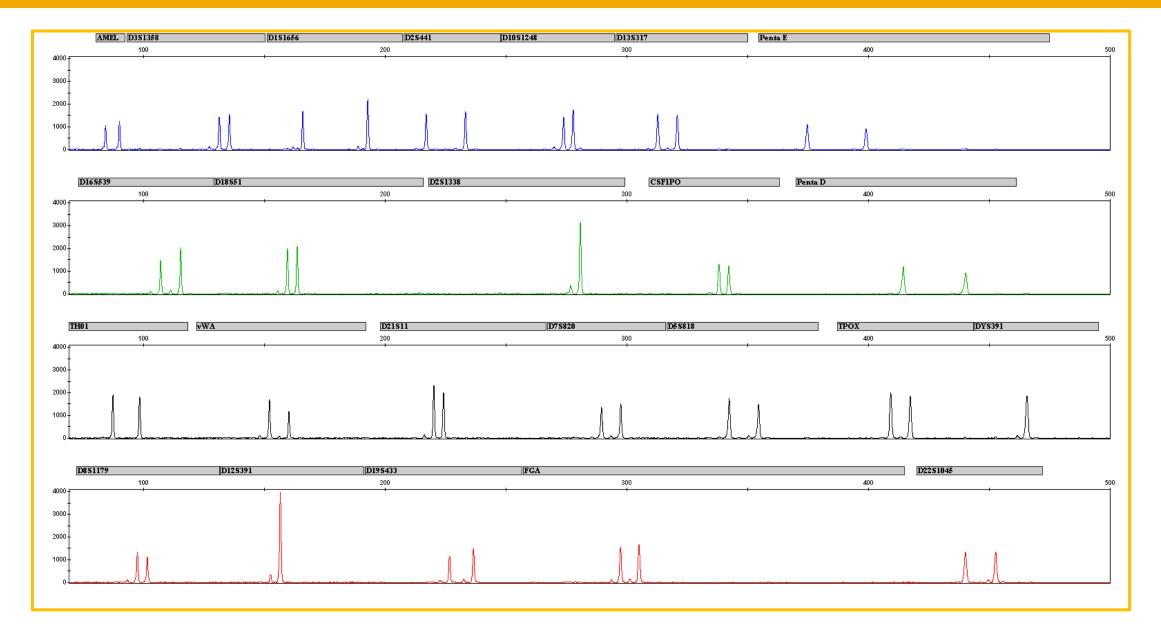
- All required "new CODIS loci" from "Panel A"
- Two out of three "recommended" loci, TPOX and D22S1045 from "Panel B"
  Does not include SE33
- Penta E and Penta D
  - Used routinely in Asia, Europe, North and South America
  - >1M samples in US NDIS contain Pentanucleotide profiles
- Easy mixture interpretation due to low stutter
- Increased discrimination by the addition of these two highly polymorphic loci

#### **Probability of Identity**

STR Typing Kits (Locus Combinations)	Total (N=1036)	African American (N=342)	U.S. Caucasian (N=359)	U.S. Hispanic (N=238)	U.S. Asian (N=97)
CODIS 13	5.02 x 10 <sup>-16</sup>	1.13 x 10 <sup>-15</sup>	3.01 x 10 <sup>-15</sup>	1.39 x 10 <sup>-15</sup>	1.71 x 10 <sup>-14</sup>
PowerPlex <sup>®</sup> 16	2.82 x 10 <sup>-19</sup>	6.08 x 10 <sup>-19</sup>	4.22 x 10 <sup>-18</sup>	1.29 x 10 <sup>-18</sup>	2.54 x 10 <sup>-17</sup>
PowerPlex <sup>®</sup> 18D	3.47 x 10 <sup>-22</sup>	5.55 x 10 <sup>-22</sup>	9.76 x 10 <sup>-21</sup>	2.58 x 10 <sup>-21</sup>	7.87 x 10 <sup>-20</sup>
PowerPlex <sup>®</sup> 21	6.71 x 10 <sup>-27</sup>	2.08 x 10 <sup>-26</sup>	2.51 x 10 <sup>-25</sup>	7.96 x 10 <sup>-26</sup>	5.80 x 10 <sup>-24</sup>
CODIS Panel A (-DYS391)	1.95 x 10 <sup>-23</sup>	1.08 x 10 <sup>-22</sup>	1.66 x 10 <sup>-22</sup>	1.93 x 10 <sup>-22</sup>	6.01 x 10 <sup>-21</sup>
CODIS Panels A&B (-DYS391)	1.60 x 10 <sup>-27</sup>	5.63 x 10 <sup>-27</sup>	2.95 x 10- <sup>26</sup>	5.10 x 10 <sup>-26</sup>	2.57 x 10 <sup>-24</sup>
PowerPlex® Fusion (-DYS391)	1.36 x 10 <sup>-28</sup>	2.83 x 10 <sup>-28</sup>	5.25 x 10 <sup>-27</sup>	4.81 x 10 <sup>-27</sup>	2.01 x 10 <sup>-25</sup>

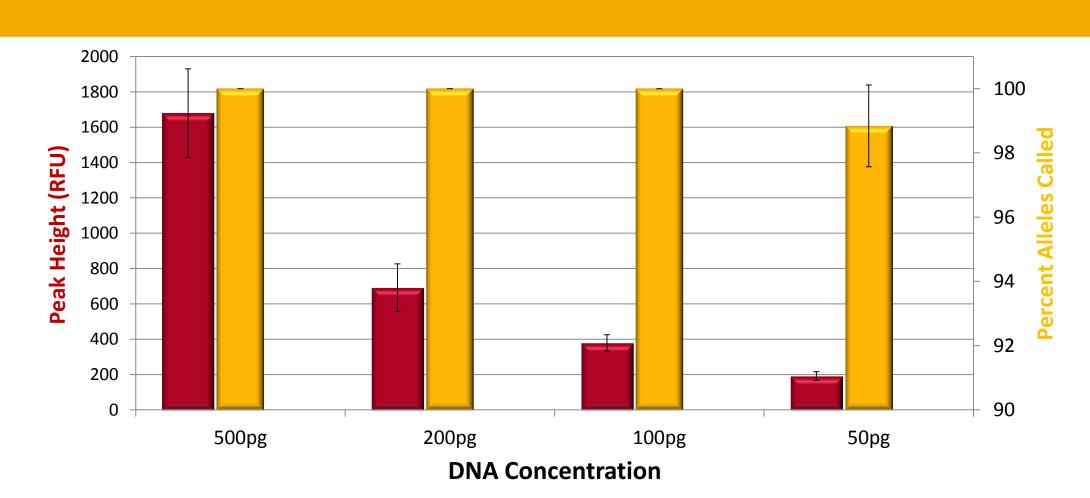
Data kindly provided by John Butler (NIST) using data collected by Becky Hill & analysis tools developed by Dave Duewer

## **Amplification of 500pg Extracted DNA**



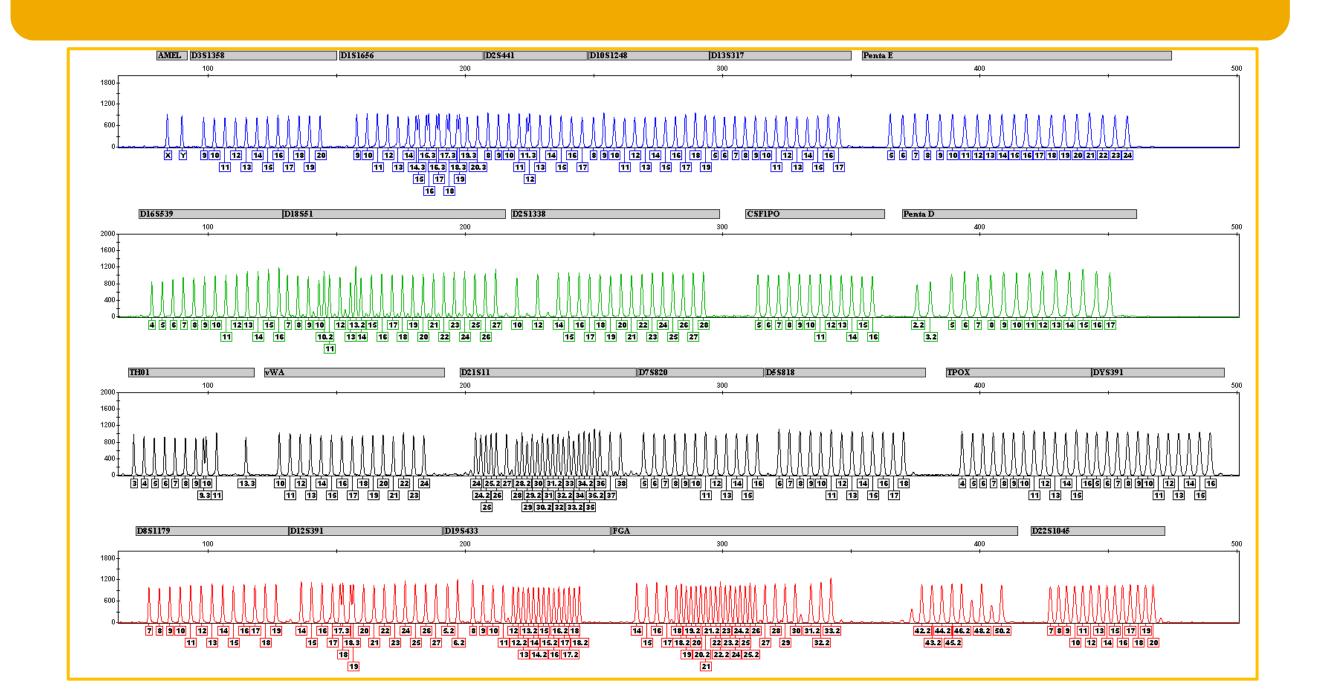
Applied Biosystems 3130xl Genetic Analyzer using a 3kV, 5 second injection

#### **Sensitivity: Titration of Extracted DNA**



Genomic DNA samples from two individuals were amplified in triplicate using the PowerPlex® Fusion System with 500pg, 200pg, 100pg or 50pg. The average overall peak height at each concentration was determined and is shown in red. Error bars represent the standard deviation between the average of both DNA samples at the stated concentration. The percentage of alleles (using a 50rfu threshold) called is shown in orange. Error bars represent the standard deviation between replicate reactions of both DNA samples.

#### PowerPlex® Fusion Allelic Ladder



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### Direct Amplification with the PowerPlex® Fusion System

The PowerPlex® Fusion System is compatible with direct amplification from a variety of substrates including:

- FTA® Paper including the Indicating FTA® Cards (GE Healthcare/Whatman)
- S&S 903 Paper (GE Healthcare/Whatman) and Bode Buccal DNA Collector™ (Bode Technologies) pretreated with Promega's PunchSolution™
- Cotton swabs and OmniSwabs<sup>™</sup> (GE Healthcare/Whatman) pretreated with Promega's SwabSolution<sup>™</sup>

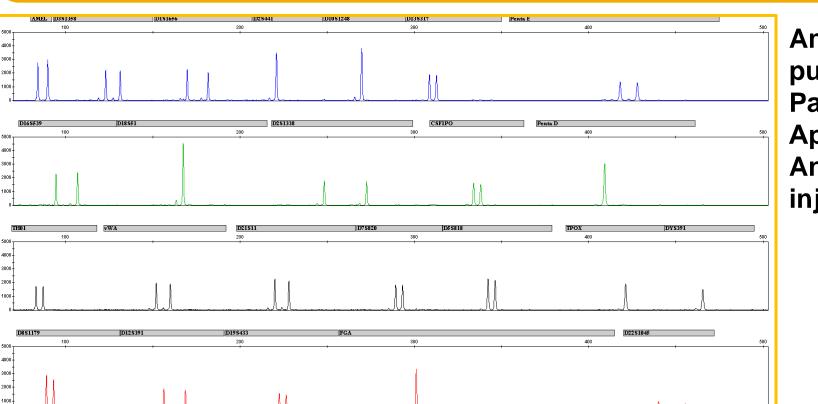




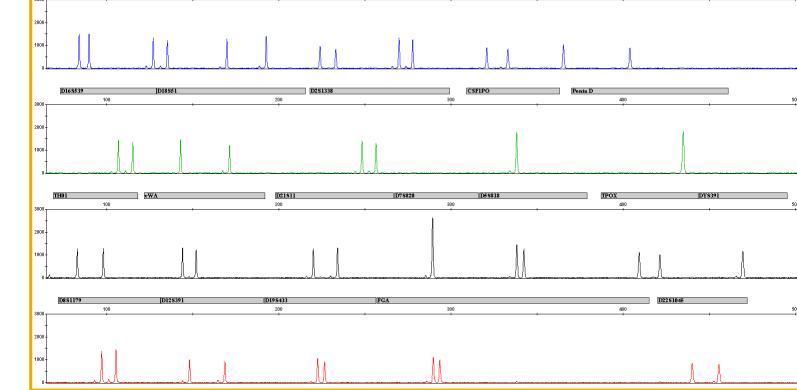




Direct Amplification with the PowerPlex® Fusion System



Amplification (27 cycles) of a 1.2mm punch of a buccal sample on FTA® Paper followed by electrophoresis on an Applied Biosystems' 3130xl Genetic Analyzer using a 3kV, 5 second injection.



Amplification (27 cycles) of 2µl of a swab extract pretreated with SwabSolution™ followed by electrophoresis on an Applied Biosystems' 3130xl using a 3kV, 5 second injection.

## Summary

- The PowerPlex® Fusion System is a rapid (1.5 hour thermal cycling), robust and sensitive system that meets the requirements of the new CODIS Standard.
- The combination of 23 STR loci, including Penta E and Penta D along with Amelogenin make this multiplex an effective tool for human identification.
- The PowerPlex® Fusion Allelic Ladder has been expanded from the PowerPlex® 16 and 18D with more alleles for several of the STR loci.
- The system is based upon the same proven five color chemistry used in several of the systems Promega introduced in the last two years. This allows for use of the kits on existing configurations of the Applied Biosystems 3100/3130 and 3500 series Genetic Analyzers.
- The PowerPlex® Fusion System is a one kit solution for both extracted DNA and direct amplification. The PowerPlex® Fusion System is optimized for extracted DNA, but allows for direct amplification from a variety of substrates.