

## **VALIDATION AND IMPLEMENTATION OF PowerPlex® Fusion 6C: A 27-LOCUS MULTIPLEX AND EXPANDED CODIS CORE LOCI AMPLIFICATION**

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The CODIS Core Loci will be expanded from the original 13 to 20 loci in January of 2017. A requirement of all CODIS participating laboratories will be to validate one of the new expanded core multiplex amplification kits before this transition. At the Columbus Police Forensic Services Center, three expanded core multiplex amplification kits were evaluated and Promega's PowerPlex® Fusion 6C amplification kit was chosen. PowerPlex® Fusion 6C is a six-dye autosomal and Y-STR amplification kit that amplifies 27 loci in a single reaction. The kit contains all 20 autosomal loci in the new expanded CODIS core loci. Promega's PowerPlex® Fusion 6C was validated alongside Promega's SwabSolution™ kit for direct amplification of reference buccal swabs and FTA blood cards. The FBI QAS standards and SWGDAM guidelines were followed for the validation design and included the following studies: analytical threshold, sensitivity, base pair precision, mock casework samples, mixture samples, contamination, concordance, reproducibility, and artifacts. Differential, touch, blood, hair, and buccal sample extractions were performed to encompass samples routinely encountered in DNA casework. All non-direct amplification samples were quantified with Promega's Plexor® HY quantification system and amplified targeting 1ng of total DNA. Direct amplification was performed following Promega's Technical Manual for the SwabSolution™ kit with a few modifications such as sample amount, volume, and cycle number. All samples were run on Applied Biosystems® 3500 Series Genetic Analyzer using a seven second default injection time. The results indicated that PowerPlex® Fusion 6C was sensitive down to 15pg and reliably generated full profiles at or above 250pg. PowerPlex® Fusion 6C was robust in amplifying various samples routinely encountered in casework. Two, three, and four person mixtures were interpretable at or above 250pg with the major component preferentially amplified in the majority of mixtures. All standards were concordant. The PowerPlex® Fusion 6C amplification kit was reproducible across analysts, sample types, and instruments. One concern that arose during the mixture study was the inability of the GeneMapper™ ID-X v1.4 software to consistently resolve alleles that were one base pair apart when one allele was significantly higher than the other. Overall the PowerPlex® Fusion 6C kit was robust, reproducible, and reliable. The PowerPlex® Fusion 6C kit was implemented into our casework flow at the Columbus Police Forensic Services Center.