MO LOCI, MO PROBLEMS: AN EVALUATION OF OUTSIDE MARKER RANGE ALLELES IN MEGAPLEX KITS

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The forensic science community has recently transitioned to use of the newer megaplex amplification kits. These kits contain an expanded set of loci to facilitate increased confidence in database matches. Unfortunately, with the addition of more loci in a limited number of dye channels and smaller amplicons comes new concerns. Specifically, during the evaluation and implementation of PowerPlex Fusion® 6C, the Wisconsin DNA Databank observed a number of alleles that are designated as outside marker range or appear to fall into the marker range of adjacent loci. These alleles are often denoted as tri-alleles in the adjacent locus, but can also appear as a heterozygote with poor peak height balance. This problem does not appear to be limited to a single megaplex amplification system.

In a preliminary study using Fusion® 6C and Fusion® amplification kits, the Wisconsin DNA Databank observed instances of alleles designated as outside marker range or appearing to fall into the marker range of adjacent loci in approximately 0.1% of profiles. Further analysis revealed that D1S1656, D2S441, D10S1248, and D12S391 were most frequently impacted using these amplification kits. Additionally, although the order of many loci are unchanged between Fusion® 6C and Fusion®, differences in the initial allele designation were observed, which can likely be attributed to variation in spacing between loci.

Alleles inadvertently designated into the adjacent, but incorrect locus may increase the complexity of profile comparisons with different amplification kits, have an impact on statistical analysis and CODIS eligibility, or create the potential for missed matches in CODIS. Recent presentations demonstrate that this problem appears to be present in all available megaplex amplification kits. Based on this evaluation, the entry of offender DNA profiles in CODIS has been modified to incorporate both possible iterations of the DNA profile. Further, when evaluating matches and performing statistics for samples processed with the newer megaplex amplification kits greater caution is taken to ensure accuracy.