A147 Internal Validation of Y-STRs for Casework at the Kentucky State Police Crime Laboratory

Sally Edwards, BS, Marshall University Forensic Science Center, 1401 Forensic Science Drive, Huntington, WV 25701*

The goal of this presentation is to make the forensic science community aware of the benefits of Y-STRs for casework at the Kentucky State Police Crime Laboratory.

This presentation will impact the forensic science community by showing the importance and effectiveness of Y-STRs.

Y-STRs allow amplification of human male DNA in a single reaction. Crime laboratories are interested in amplification kits for situations such as sexual assaults. In cases of sexual assault, often times a complete profile for a male contributor is difficult to obtain due to high levels of female DNA and low levels of male DNA. These kits are also effective for blood-blood or saliva-blood mixtures, samples from azospermatic males, and other samples containing male DNA not optimally amplified with autosomal STR kits. One Y-STR kit targets 17 Y-STR markers including the European minimal haplotype markers (DYS19, DYS385a/b, DYS389I, DYS389II, DYS390, DYS391,
DYS392, and DYS393), two markers recommended by SWGDAM (DYS438 and DYS439), and six polymorphic haplotype markers (DYS437, DYS448, DYS456, DYS458, DYS635, and Y GATA H4)
while another kit targets the European minimal haplotype markers (DYS19, DYS385a/b, DYS389I, DYS389II, DYS390, DYS391, DYS392, and DYS393), two markers recommended by SWGDAM (DYS438 and DYS439), and one polymorphic haplotype marker (DYS437). Some markers, such as DYS385a/b, are multi-copy markers. At this specific marker, the a/b indicates two peaks at this locus. In actuality, the locus is amplified with a single set of primers, which may result in the peaks being the same height (seen as one peak) or different heights (seen as two peaks).

An internal validation for Y-STRs was performed at the Kentucky State Police Central Laboratory in order to determine which commercial amplification kit meets the needs for forensic casework at the KSP. The validation studies included: sensitivity, female/male mixtures, male specificity, allelic dropout, distinguishing profiles from both contributors in male/male mixtures, adjusting stutter filters, documentation of common artifacts, reproducibility of results, accuracy of results using NIST Standard Reference Material® (SRM) # 2395 samples, contamination, precision, and Y-STR database statistics. One Y-STR amplification kit was chosen for validation purposes because more markers were amplified as well as an established relationship with the manufacturer. In the validation studies performed, this kit proved to be sensitive and able to produce complete profiles from samples containing template DNA as low as 125 pg. Samples with as little as 50 pg had 88.2% alleles called. Female DNA is not amplified at strengths of 200 times that of male DNA (e.g., 100 ng:0.5 ng mixtures) because the primers bind specifically to the 17 Y-STR markers. This means that at concentrations as strong as 100 ng, female DNA will not interfere. Male/male mixture samples were created to mimic casework-type samples. These mixtures could be discerned at a ratio of 70:30 in samples with 1.2 ng, 0.6 ng, and 0.3 ng total DNA and most 60:40 samples. For 90:10 mixtures, samples with 1.2 ng total DNA had 95.70% of alleles called and for samples with 0.3 ng total DNA, 77.42% of alleles were called. Stutter peaks were documented and adjustments to stutter filters were based on the highest stutter value observed. During this validation, it was found that the following loci needed higher stutter filters than those recommended by the manufacturer: DYS456, DYS390, DYS389II, DYS385 a/b, DYS393, and DYS392. Based on the results, this internal validation shows that reproducible results from a Y-STR amplification kit will be beneficial for forensic casework purposes.

A future study using a Y-STR amplification kit may be performed to determine whether or not there will be any interference with male DNA amplification if female DNA is present at stronger concentrations (e.g., >100 ng). Other studies of interest include increasing the PCR cycle number during amplification, analyzing male/male mixtures with additional genotypes, and performing post-amplification purification to remove primer peaks.

Y-STR, Validation, Y-filer

 ** Presenting Author*