



### A19 Internal Validation of Yfiler® for Casework at the Saint Louis Metropolitan Police Department

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After attending this presentation, attendees will be able to perform an internal validation of Yfiler®. Attendees will have an idea of what tests should be conducted for a Yfiler® internal validation and the results that could be expected. They will also gain an understanding of why Yfiler® is important technology for DNA casework and its limitations.

This presentation will impact the forensic science community by informing attendees about the Yfiler® benefits and limitations.

AmpFISTR® Yfiler® amplification kit is a Short Tandem Repeat (STR) kit specific to the sex determining region of the Y-chromosome. Yfiler® kit can be used to detect male DNA from samples where a male profile cannot be generated from an autosomal kit. Yfiler® kits are valuable when a large presence of female DNA is in a sample as compared to the male DNA, when absence of sperm in an azoospermic male prevents a differential extraction, and in determining the number of individuals in a multiple assailant rape. Most of these instances prevent a full male profile from being generated. Single source Yfiler® profiles typically consist of haplotypes containing one allele at each locus. A single source Autosomal kit profile consists of diplotypes containing one or two alleles at each locus. Multiple alleles can be expected in situations of multiple assailant rape. Thus the Yfiler® kit may be useful in determining the number of assailants in a multiple assailant rape. Haplotypes only show one allele at most loci for each person involved. The manufacture conducts developmental validations that aids in the laboratory's internal validation when implementing the Yfiler® STR kit. The St. Louis Metropolitan Police Department hypothesized, based on SWGDAM guidelines, the developmental validation performed by Applied Biosystems and the work of Gross et al. (Journal of Forensic Science, 2008, Vol. 53, No. 1), the internal validation of Yfiler® kit for casework could be performed by completing sensitivity, precision, mixture, probative/non-probative, stutter, and specificity studies. DNA was extracted from male and female lab members and from probative and non-probative sources for use in the internal validation studies. The non-probative samples were collected in various common places in and around the lab. Examples include door handles and other commonly touched items including a parking pass button from a nearby parking lot. Probative sources were collected from cases previously adjudicated. The concentration of the DNA was determined using Quant Duo and diluted to appropriate concentrations depending on the study being conducted. DNA was amplified on an AB 9700 using the Yfiler® kit and electrophoresis was conducted using a 3130 genetic analyzer. Results of the experiments were analyzed using the AB software Genemapper v3.2. The results of the sensitivity study showed the target amount of DNA for an amplification using Yfiler® is 0.8 ng, which gave average RFU levels of 1012.25. A full profile was generated with 0.1 ng of DNA. Allele drop

out was seen with 0.08 ng template DNA in most of the loci. The peaks were still present below Genemapper's threshold for allele calls of 50 RFU. The results of the precision study suggested amplifications using Yfiler® were precise across multiple injections of samples. The mixture study showed Yfiler® to amplify only male DNA in the presence of a large amount of female DNA. Male mixtures could still generate male profiles, which could be deconvoluted in some cases or at least suggest how many people were in the mixture. The probative/non-probative study suggested the Yfiler® kit could work with casework consistent with the samples encountered by the lab. The stutter study generated an anomaly in the male B NIST standard that was reproducible in multiple extractions and in multiple injections. The specificity study showed the primers used in the Yfiler® kit were specific to human male DNA and did not cross react with dog, chicken, cow, orangutan, baboon, deer, or mouse DNA. The Yfiler® kit has limitations of not being as discriminative, since the loci are physically linked, as autosomal kits with 16 loci. Additionally the Y-chromosome is shared in paternal lineages making the resulting profile less unique within families. The completion of these internal validation studies will allow the lab to start using the Yfiler® kit in casework. The laboratory can increase the efficiency of determining the male(s) associated with a crime.

#### **Yfiler®, Validation, Male**