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Department of Forensic Biology is Implementing New and/or Updated Technologies – a New Genetic Analyzer, Updated Versions of STR Analysis and Probabilistic Genotyping Software, a New YSTR DNA Typing Kit, and a New Platform for Mitochondrial DNA Typing

The OCME will implement two sets of new technologies. The first suite of products is for STR analysis and includes the Applied Biosystems® 3500xL Genetic Analyzer, SoftGenetics® GeneMarker® HID v3.0.0 analysis software, STRmix™ v2.7 probabilistic genotyping software, and Promega PowerPlex® Y23 YSTR amplification kit. The second suite of products is for massively parallel sequencing (MPS) for mitochondrial DNA which uses the Verogen MiSeq FGx® Sequencing System, Promega PowerSeq® CRM (control region mitochondrial) Nested System, Qiagen CLC Genomics Workbench, and AFDIL-QIAGEN mtDNA Expert (AQME) toolkit.

New Technology	Retiring Technology
Applied Biosystems® 3500xL	Applied Biosystems® 3130xL
SoftGenetics® GeneMarker® HID v3.0.0	SoftGenetics® GeneMarker® HID v2.9.0
STRmix™ v2.7	STRmix™ v2.4.08
Promega PowerPlex® Y23	Applied Biosystems® Yfiler
Verogen MiSeqFGx® Sequencing System	Applied Biosystems® 3130xL
Promega PowerSeq® CRM Nested System	ABI Big Dye Terminator Kit
Qiagen CLC Genomics Workbench and AFDIL-QIAGEN mtDNA Expert (AQME) toolkit.	ABI Sequencing Analysis Software and GeneCodes Sequencher

New Genetic Analyzer
Applied Biosystems® 3500xL is replacing the 3130xL genetic analyzer. This is a newer version of the previous instrument with the ability to process more samples per run (24 from 16), has a larger dynamic range (peak heights from 0 – 30,000rfu), and has a slight increase in sensitivity. There are other cosmetic updates to the design of the instrument that allow for easier operation but do not affect the output. The main difference that will be seen by the customer is that the raw data output file is now a .hid file versus the previous .fsa output file.

Updated STR Analysis Software
OCME will be upgrading to GeneMarker® v3.0.0. (from the previous v2.9.0) for analysis of autosomal and Y-chromosomal STRs. For autosomal STR analysis (we will continue to use Promega's PowerPlex® Fusion 5C amplification kit), this upgrade will include the implementation of allele-specific stutter filters which should assist analysts in differentiating

stutter peaks from true minor peaks in mixtures. Once the samples are processed through the program, the output in the form of an electropherogram will be essentially the same with the exception of the aforementioned larger dynamic range of peak heights.

Updated Probabilistic Genotyping Software
OCME will be upgrading from STRmix™ v2.4.08 to v2.7. This updated version of the generally accepted and widely used STRmix™ software will include mostly cosmetic changes seen in the results printout. The general functions of assisting the analyst in the deconvolution of mixtures and calculating a likelihood ratio (LR) remain the same. The software continues to use the fundamentals of probabilistic theory and incorporates biological parameters such as peak heights, drop-in/drop-out, and stutter.

This updated suite of products brings the ability to interpret more samples. With the increased sensitivity of the 3500xL genetic analyzer, incorporation of allele specific stutter filters within GeneMarker® HID v3.0.0, and the inclusion of five person mixtures during the validation of STRmix™ v2.7, OCME will be interpreting autosomal DNA single source samples and mixtures up to and including four contributors.

New STR Kit
Promega PowerPlex® Y23 is replacing Applied Biosystems® AmpF*l*STR Yfiler™. This new YSTR amplification kit increases the number of loci typed per run to 23. Therefore, we will generate more genetic information for each sample we analyze. YSTR interpretation will continue to be done on single source and mixtures of two or three males.

New Platform for Mitochondrial DNA Typing
The OCME has validated and received approval by the New York State Commission on Forensic Science to use Verogen's MiSeqFGx® to perform massively parallel DNA sequencing (MPS) for the typing of mitochondrial DNA samples. This will further the mission of identifying missing persons using the newest technology available. The use of the MiSeqFGx® for MPS will replace the fluorescent-based Sanger DNA sequencing performed on the Applied Biosystems® 3130xL and will cover the same areas of the mitochondrion as previously tested. With the changeover to MPS, we will see an increase in sensitivity as well as an update to the type of raw data generated. The reported results, however, will remain the same.

The OCME is fully committed to staying on the cutting edge of new technology to best serve the City of New York and will continue to evaluate new techniques for implementation in the laboratory.