Mixed Sample Evaluation Using AmpF*l*STR™ Profiler Plus™ on the ABI Prism® 310 Genetic Analyzer and ABI 377 DNA Sequencer

Jeanne M. Willard, MFS, Demris A. Lee, MSFS, James P. Ross, BS, Mitchell M. Holland, Ph.D.,
Colonel Vernon W. Armbrustmacher, USAF, MC
The Armed Forces DNA Identification Laboratory, Office of the Armed Forces Medical Examiner,
Armed Forces Institute of Pathology, Rockville, Maryland, 20850-3125

Multiple source DNA specimens are often encountered during the analysis of forensic samples. These specimens are often observed in rape cases where the suspect' s DNA can be mixed with the victim' s DNA or in cases of commingled remains from aircraft accidents which contain DNA from multiple individuals. Before a new Short Tandem Repeat (STR) system can be used in forensic case work it should be extensively validated to ensure the reliability of the results obtained. TWGDAM (Technical Working Group on DNA Analysis Methods) recommends that DNA analysis systems be evaluated to determine if mixtures can be detected and the limits of the system. Evaluation of multiple source DNA specimens prior to use in case work will assist in the ability to recognize and interpret these samples.

According to ABI, heterozygous alleles within a locus should produce peak heights of equal intensity and therefore peak height intensities can be used as a possible indicator of a mixture. In order to assist in the evaluation of mixed DNA specimens, the mean peak height ratios for heterozygous genotypes and percent stutter for all loci in the Profiler Plus™ (D3S1358, vWA, FGA, D8S1179, D21S11, D18S51, D5S818, D13S317 and D7S820) STR system were determined. Once this information was obtained, purified DNA from known individuals was combined in defined ratios (1:1, 1:2, 1:5, 1:10, 1:20) and amplified with a constant input DNA of 1 ng. Whole blood mixtures were also combined in the same ratios, spotted onto filter paper, Chelex extracted and evaluated. In addition to the known ratio samples, several mixed specimens from non-probative cases were evaluated. All of the specimens were analyzed on both the ABI Prism® 310 Genetic Analyzer and ABI Prism® 377 DNA Sequencer to compare the limits of detection. The results of this study will be presented.

The opinions and assertions expressed herein are solely those of the authors and are not to be construed as official or as the views of the United States Department of Defense or the United States Department of the Army.