B135 The Recovery of Full Single Source DNA Profiles From Contributors to Complex Mixtures by Direct Single Cell Subsampling (DSCS)

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Learning Overview: After attending this presentation, attendees will better understand a novel DNA mixture deconvolution tool that relies on subsampling of individual cells by physical capture, subsequent high-sensitivity DNA typing, and quantitative computer interpretation to extricate fully probative single source genotypes.

Impact on the Forensic Science Community: This presentation will impact the forensic science community by informing attendees how the DSCS approach could permit forensic scientists to reduce genotype information loss in standard mixture analysis caused by excessive numbers of overlapping alleles and/or the presence of low-level minor contributors, with some of the latter not even being detectable by standard mixture analysis.

DNA mixtures are often difficult to deconvolute and interpret due to the presence of overlapping alleles from multiple contributors and stutter artifacts. Therefore, standard mixture interpretation now requires the use of Probabilistic Genotyping (PG) software. However, even with the use of PG software, genetic information loss is often seen in some complex mixtures by decreased likelihood ratios in comparison to single source genotype profiles. In order to aid in mixture deconvolution and genotype information loss in complex mixtures, a simple micromanipulation technique referred to as DSCS has been developed. This technique allows for manual separation of individual cells or subsets of cells (typically 2–3) from mixtures prior to DNA typing paired with a subsequent Low Copy Number (LCN) technique in order to attempt to obtain single source DNA genotype profiles from all contributors to a mixture.

The feasibility of this approach will be presented by first using a simple two-person 1:1 buccal cell mixtures that were not fully resolvable by PG. Single and multiple cells were recovered and analyzed via DSCS and full single source profiles were achieved for each donor. The DSCS approach was then applied to more complex equi-proportional 3–6 person mixtures (i.e., 1:1:1, 1:1:1:1, 1:1:1:1:1, and 1:1:1:1:1:1). PG software, STRmix™, and EuroForMix were utilized to compare the information loss (in bans) of the standard "bulk" mixtures to that of the single source reference profiles. Comparison to the analyzed DSCS samples obtained indicated instances in which the DSCS method resulted in a substantial gain of probative information. The gain of information was maximized in some instances by co-inferring genotypes utilizing a joint likelihood function. The mixture and DSCS analysis conditions under which this occurred will be described. In addition to obtaining single source profiles, the DSCS sampling method can also result in subsample "mini mixture" byproducts. PG methods can then be utilized to analyze these mini mixtures. For example, a three-cell subsample could, depending upon the number of donors, result in cells originating from a single donor, two donors, or three donors. These artificial mini mixture subsamples may be significantly reduced in complexity compared to the original mixture by reducing the number of contributors present or creating more unbalanced weight ratios (e.g., in a three-person mixture, some of the three-cell mini mixtures recovered comprise only two of the three donors at weight ratios of 2:1 rather than 1:1). The PG results from these mini mixtures can be used to provide additional support to the inferences made from the single source DSCS results. The mini mixture analyses for each of the complex mixtures studied will be presented.

The DSCS approach could permit forensic scientists to reduce genotype information loss in standard mixture analysis caused by excessive numbers of overlapping alleles and/or the presence of low-level minor contributors, with some of the latter not even being detectable by standard mixture analysis.

Mixtures, Micromanipulation, Single Cell

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