B116 Increasing DNA Mixture Analysis Quality and Efficiency

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After attending this presentation, attendees will better understand the challenges of distinguishing low-level artifacts from low-level alleles and how expert software can help distinguish between them to increase analyst efficiency and mixture profile interpretation reproducibility.

This presentation will impact the forensic science community by increasing awareness of how free, open-source software can be used to improve the significant problem of reproducibility in the interpretation of mixed DNA profiles.

Forensic DNA laboratories are attempting to find analytical thresholds that utilize the greatest amount of information possible in their Short Tandem Repeat (STR) mixture profiles without mistaking small artifacts and noise for alleles. Simultaneously, backlogs create pressure to use analyst time efficiently, while increased sensitivity increases the need for more reproducible interpretation.[1] The Scientific Working Group on DNA Analysis Methods (SWGDAM) Interpretation Guidelines for autosomal STR typing indicate that laboratories should not use their analytical threshold to avoid artifacts, but rather should apply analytical thresholds that take the laboratory's empirical noise levels into account. This allows laboratories to utilize the maximum information in STR profiles, and not "leave data on the table." As laboratories begin to use analytical thresholds below 30 Relative Fluorescence Units (RFU) to analyze complex mixed profiles, it becomes critical to be able to distinguish actual alleles from small STR artifacts and excursions in the baseline noise. STR analysis noise arises from Polymerase Chain Reaction (PCR) amplification artifacts and noise in the capillary electrophoresis analyzer. With more sensitive analytical thresholds, analysts spend more time determining whether low-level peaks in mixtures are alleles or artifacts. Software can employ various mathematically determined metrics to identify and discriminate low-level artifacts from alleles.

The Open Source Independent Review and Interpretation System (OSIRIS), downloadable from the National Center for Biotechnology Information (NCBI) OSIRIS homepage, was created in response to recommendations arising from the World Trade Center victim identification effort. OSIRIS is in use as an expert system for Combined DNA Index System (CODIS) samples and in clinical and forensic caseworking laboratories to analyze complex and low-level mixtures. OSRIS implements unique STR analysis metrics and a variety of artifact signatures that give the software exceptional capabilities when analyzing low-level STR profiles.

OSIRIS computes novel metrics for peak shape, peak shifting, sample-to-ladder fit, and channel-specific baseline noise. OSIRIS also matches various mathematical artifact signatures to different peak shapes and applies these metrics and signatures to discriminate allele peaks from artifacts and noise. Low-level artifacts include pull-up, non-specific peaks and random deviations in baseline noise that can appear to be alleles. Low-level alleles include those masked as shoulders on larger alleles and can be mistaken for artifacts or noise. Low-level artifact peaks can be difficult to visually distinguish from allele peaks. OSIRIS uses its calculated metrics, peak signatures, and its expert knowledge base to automatically identify and annotate these and other artifacts, saving significant analyst time and enhancing reproducibility among analysts.

Accurately and robustly identifying these artifacts increases the assurance of quality profiles and reduces both the editing burden and the number of conflicting analyst/reviewer calls that require resolution. In this way, OSIRIS improves the efficiency of analysts that are interpreting profiles. This gives analysts more time to do the important work of case interpretation.

Reference(s):

1. Butler J.M. (2015) The future of forensic DNA analysis. *Philos Trans R Soc Lond B Biol Sci*. 2015 Aug 5;370(1674). pii: 20140252.

DNA, Mixture, Reproducibility