



## **B121 The Development of a Five-Dye Insertion/Deletion (INDEL) Panel for Ancestry Determination**

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**Learning Overview:** After attending this presentation, attendees will understand the benefits of INDEL and Ancestry Informative Markers (AIMs). This presentation will focus on their application with low-quantity DNA and their ability to provide investigative leads. This research developed an INDEL panel for ancestry prediction followed by a validation according to the Scientific Working Group on DNA Analysis Methods (SWGDM).

**Impact on the Forensic Science Community:** This presentation will impact the forensic science community by providing a novel INDEL panel for ancestry prediction. Additionally, the biogeographic prediction power of this panel was demonstrated with degraded and challenging samples.

The use of Short Tandem Repeat (STR) markers to find individualizing features in the human genome in conjunction with the national Combined DNA Index System (CODIS) database has streamlined human identification. DNA evidence continues to evolve as improvements in sensitivity allow for utilization with many forensic samples. As effective as individual identification markers are, other marker types have been shown to be complementary in forensic cases. INDEL markers are similar to STRs in that they provide genetic identification but differ in other ways. INDELs are bi-allelic, which limits human identification, but two important benefits were exploited in this study. INDELs can be genotyped using small amplicons, which allows for greater success with degraded/low quantity sample types. Additionally, ancestry prediction is possible because INDEL markers with high Fst values and low mutation rates can be targeted for analysis.

This research focuses on the use of INDEL polymorphisms that were selected for their ability to differentiate ancestry. These markers, known as AIMs, can distinguish biogeographic differences as these markers exhibit different allele frequencies between populations. Additionally, INDEL amplicons were chosen because they can be designed to be differentiated by size and integrated into a Capillary Electrophoresis (CE) platform. An advantage INDELs have over STRs is the ability to amplify many markers (25–30) using amplicons less than 200bp. INDELs have the added benefit of no stutter artifacts allowing for easier interpretation from degraded samples.

This presentation will discuss the limitations of current individual identification testing and how the developed panel can serve as a complement to forensic cases. The panel consists of 25 ancestry informative INDEL markers configured across five dye channels and validated according to SWGDAM guidelines. Stability, mixtures, sensitivity, and reproducibility were evaluated to ensure the effectiveness of the panel. This panel assessed biogeographic separation of samples from African American, Hispanic, Caucasian, and East Asian groups. PCA plots were formed to determine separation of individuals based on self-identified ancestry and INDEL results. In conclusion, a 25 marker INDEL panel was developed, and preliminary results indicate the assay can separate populations based on AIMs and effectively recover full profiles down to 32 pg and partial profiles at 16pg.

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### **Insertion Deletion Polymorphisms, Capillary Electrophoresis, Ancestry Informative Markers (AIMs)**