



A58 Concurrent Internal Validation of Four Forensic STR DNA Profiling Kits Using a Single Genetic Analyzer

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After attending this presentation, attendees will be familiar with the validation study performed and how the results were used to set mixture interpretation guidelines and policy for the Department of Biology of the National Institute for Criminalistics and Criminology of the Algerian Gendarmerie Nationale (INCC).

This presentation will impact the forensic science community by presenting the results of a single combined internal validation of four STR kits: AmpFISTR® Identifiler™, PowerPlex 16™, AmpFISTR® SGM Plus™, and PowerPlex® ESI 16™ using the same analysis conditions and a single instrument throughout the entire study. With these concurrent validations, relative kit sensitivities, precision, analytical, and stochastic thresholds were determined. This study has allowed the authors to determine a hierarchy of kit use during analysis and has allowed us to create fixed interpretation guidelines for interpretation of DNA profiles in forensic casework at the biology department INCC-Algeria.

Forensic casework very often involves samples that may not produce DNA results. When results are obtained, the DNA profile information may not be interpretable. This may be due to the variable quality of biological material that can be obtained from forensic evidence exhibits, but this also may be due to the inherent variability of current STR DNA profiling technology. Every technology has a limit at which usable or reliable data can be obtained or cannot be interpreted for conclusive analysis. Forensic DNA profiling technology has two such limits.

Currently, forensic DNA profiling technology is centered upon the use of a very finite number of commercially available forensic human identity (HID) kits which produce amplified products via the polymerase chain reaction (PCR). PCR can fall prey to stochastic effects. An even more finite set of commercially available genetic analyzers are currently used in forensic DNA laboratories. The genetic analyzer is merely an analytical instrument used to observe the amplification products. These analytical instruments individually have their own limits of linearity and detection. A thorough comparative assessment of HID kit performance has not been truly executed, in the author's opinion, because the performance of these kits has been assessed under greatly varying analytical conditions, differing analysis equipment, and analysis conditions. Widely differing DNA interpretation methods are also used to evaluate the data generated, greatly exacerbating the already highly variable results that may have been obtained during the kit performance assessments.

Laboratory interpretation variability has been highlighted in recent years by inter-laboratory studies and surveys that have been performed. This interpretation variability appears to be the result of an oversimplification of observations on an analytical device without taking into account its limits and the disregard for an HID kit to produce non-stochastic DNA amplification during PCR depending upon input template concentration in PCR. In an effort to minimize the use of non-reliable PCR amplification results, several oversight committees have suggested new DNA interpretation guidelines which incorporate methods for determining and handling analytical and stochastic limits within DNA profiled data to ensure it is accurate analytically and reproducibly amplified.

These newly revised interpretation guidelines have recently been adopted by many accrediting bodies internationally in the last few years. In preparation for accreditation, the INCC was in the unique position to carry out validation studies on the four commercial STR HID kits currently in use using the new interpretation guidelines as part of the assessment of performance of each kit. To this end, a single combined internal validation of four HID STR kits (AmpFISTR® Identifiler and AmpFISTR® SGM Plus from Applied Biosystems and PowerPlex 16 and PowerPlex® ESI 16 from Promega) was designed specifically to observe and compare the performance abilities of these HID kits using a single dilution series of template DNA for amplification, the same post-amplification analysis conditions, and using the same multicapillary electrophoresis instrument throughout the entire study.

With the concurrent data generated by this validation study, it was determined that the relative sensitivities, precisions, and analytical/stochastic thresholds did not vary significantly among these four kits. However, due to the varying size of reporting amplicons within each kit, it was found that certain kits are able to complement each other to allow more complete results. The author's believe the use of new fixed interpretation guidelines for the interpretation of the DNA profiles in their validation study was key in determining that no single kit was able to out-perform the others. This allowed consolidation of methods for profiling forensic casework at the biology department of the INCC-Algeria.

DNA STR Kit Validation, DNA STR Kit Sensitivity, Analytical and Stochastic Thresholds of STR Kits