B95 Quantitation of Male/Female Mixed DNA Samples Using the ABI PRISM® 7000 Sequence Detection System

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Attendees will learn how the ABI PRISM® 7000 Sequence Detection System, in conjunction with the Quantifiler™ Human and Quantifiler™ Y Human Male DNA Quantification Kits, can be used to determine DNA concentration in male/female mixed source samples.

This presentation will impact the forensic community and/or humanity by describing a relatively new technology that can be implemented in a forensics laboratory.

Mixed Male/Female DNA samples are often encountered in the forensic laboratory. Quantitation of each DNA source in a sample can be an important tool in interpreting the final STR profile. The Quantifiler™ Human and Quantifiler™ Y Human Male DNA Quantification kits, in conjunction with the ABI PRISM® 7000 Sequence Detection System use real-time PCR to amplify human and human-male DNA, respectively. Male/female DNA mixtures of known ratios will be amplified with both kits to determine the concentrations of total human and total male DNA present in each sample. Theoretically, the difference between total human DNA and total male DNA should equal the amount of total female DNA in each sample. This project will determine if the ABI system can accurately predict the amount of male/female DNA in a mixture.

Thirty-eight mixtures of male/female DNA were quantitated with both the Quantifiler™ Human DNA and Quantifiler™ Y Human Male DNA quantification kits developed by Applied Biosystems. When the female contribution in a male/female mixture was the minor component and the total DNA concentration was greater than approximately 1.48 ng/1l, the female DNA concentration could not be accurately determined by subtracting the total male DNA concentration from the total human DNA concentration. When the male contribution in a male/female mixture was the minor component, the presence of an excess contribution of female DNA did not interfere with the performance of the Quantifiler™ Y Human Male DNA quantification assay.

The Quantifiler™ Y Human Male DNA Quantification kit did not produce linear results at higher DNA concentrations (greater than 20 ng/1l). This non-linearity was reproducible, but varied in severity from assay to assay. In addition, it cannot be understated how essential correct pipetting techniques and properly calibrated pipettes are to accuracy and precision. Small errors in pipetting can translate to substantial deviation in the final quantitation results.

Quantifiler, Quantification, DNA

 ** Presenting Author*