



## Pathology/Biology Section - 2015

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### H116 A Multiplex PCR Assay for Simultaneous Analysis of 13 Rapidly Mutating Y-STRs

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After attending this presentation, attendees will better understand the new tool that can be used in forensic DNA analysis for casework samples, specifically for sexual assault cases and population study.

This presentation will impact the forensic science community by adding a useful new tool for forensic DNA analysis.

Y-chromosome Short Tandem Repeat (Y-STR) profiling has been broadly applied in forensic casework in sexual assault cases where male/female or male/male mixtures are expected and also for population studies, genealogical research, and kinship analysis. Recently, rapidly mutating Y-STRs were described. These loci are expected to help with investigating inbred populations and also differentiating closely related males. A multiplex panel has been developed comprised of 13 Rapidly Mutating Y-STRs (RM-Yplex) that can be amplified simultaneously. The multiplex will aid investigating the human genetic structure of United Arab Emirates populations and would also be used to investigate unresolved forensic cases in the Department of Forensic Sciences and Criminology at Dubai Police.

Thirteen simultaneously amplified markers included in multiplex are: DYF387S1, DYF399S1, DYF403S1ab, DYF404S1, DYS449, DYS518, DYS526I/II, DYS547, DYS570, DYS576, DYS612, DYS626, and DYS627. Four primer sets for DYF387S1, DYS570, DYS576, and DYS612 loci have been redesigned to accommodate the loci within the multiplex using 5-dye chemistry. An allelic ladder was developed and sequenced using alleles found in United Arab Emirates populations. A developmental validation was conducted for the RM-Yplex assay to investigate the robustness of the multiplex assay including sensitivity, specificity, male/male mixtures, and male/female mixtures studies. A sensitivity study resulted in a sensitivity of the assay in amplifying as low as 62.5pg of male DNA template. In the specificity study, full male profile was detected using 1ng of male DNA template in the presence of 2,000 ng of female DNA template. A stability study using three different common inhibitors encountered in DNA profiling analysis including hematin, humic acid, and tannic acid has been demonstrated. RM-Yplex has shown great stability by resisting a concentration of 100ng/ $\mu$ l from each of hematin and humic acid, whereas it has shown more stability with tannic acid by resisting as much as 200ng/ $\mu$ l of such inhibitor. In the male/male mixture study, a complete unique minor profile has been identified successfully at 1:3 and 3:1 ratios. The RM-Yplex multiplex assay was used to amplify non-probative casework samples which gave significantly more probative information than normal autosomal amplification kits being used in DNA profiling analysis and currently used Y-STR Polymerase Chain Reaction (PCR) amplification kit results.

This study illustrates that RM-Yplex multiplex is extremely sensitive, does not exhibit cross-reactivity with female DNA, and successfully types male DNA in the presence of overwhelming amounts of female DNA. The assay was successful in typing various forensic casework samples. Thirteen RM Y-STR markers have been analyzed in 600 male samples from United Arab Emirates populations. Allelic frequencies, haplotype diversity, haplotype frequencies, and discrimination capacity were determined for the 13 RM Y-STRs. Mutations pattern analysis of the RM Y-STR loci in a typical United Arab Emirates family has been carried out and will be presented.

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#### Rapidly Mutating, Y Chromosome, STR Multiplex