

08/12/24

<sup>1</sup> Virginia Department of Forensic Science Forensic Biology Procedures Manual, Extraction of DNA.

### ***Non-probative casework***

Seventeen casework samples that were potentially good candidates for Y-STR analysis were retrieved from the Normalization Wizard normalization plate. Candidacy was determined by the presence of sufficient DNA extract volume left in Normalization plate and Y-DNA quantitation data. If there was no Y-DNA quantitation data, the samples were not collected for this test. These samples were then manually set up for quantitation and analyzed on the QS5 platform. The samples had been normalized based upon the autosomal target of 0.1 ng/μL, therefore, the expected autosomal quantitation values for the PQ/QS5 analysis were the same. The concentration of expected Y-DNA was estimated for the PQ/QS5 system based upon the initial Y-DNA concentration measured using PlexorHY™ and the Stratagene MX3000P qPCR instrument (PLX) and the DNA dilutions performed by the Normalization wizard.

### ***DNA quantitation***

Reaction setup was performed using the PQ system following the manufacturer's recommendations.<sup>2</sup> Amplification used 2μL of DNA extract added to the 18μL amplification cocktail. Samples were then quantified using the QS5. The dye filters used on the QS5 were: FAM for autosomal target (84-base-pair amplicon), CAL Fluor Gold 540 (CFG540) for the male DNA targets (81bp and 136bp), TMR for the internal positive control (IPC) (435bp), Quasar 670 for degradation (294bp), and CXR for the passive reference dye. The raw data were collected with ABI QuantStudio Design & Analysis Software v 1.4.3 and analyzed using Promega's PowerQuant™ Analysis Tool software, version 1.0.0.0. Values are reported in ng/μL. Undetermined means the DNA value could not be detected.

### ***Contamination assessment***

Contamination assessment was performed by tallying all associated reagent blanks and no template control (NTC) samples associated with the sensitivity, excess female to male ratio and non-probative sample studies.

## **RESULTS**

A total of 15 reagent blanks and 20 NTC samples were assessed for any signs of contamination for the PQ system using the autosomal, Y-DNA, and degradation markers. No contamination was observed.

## **CONCLUSIONS**

The PQ/QS5 system setup and process did not introduce contamination into the reagent blank and NTC controls. Therefore, it is suitable to use for casework samples since it has a low probability of introducing contamination into casework samples.

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<sup>2</sup> Promega's Technical Manual for PowerQuant® System, 1/2020.