B98 SRM 2372: How the Human DNA Quantitation Standard Was Characterized at NIST and How it Can be Used to Calibrate qPCR Measurements in Your Laboratory

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After attending this presentation, attendees will understand the certification process for SRM 2372 and know how to calibrate Human DNA quantitation materials currently used in their laboratories using SRM 2372.

This presentation will impact the forensic community and/or humanity by educating of the forensic DNA community to the intended

use of SRM 2372; specifically SRM 2372 is intended to enable the comparison of DNA concentration measurements across time and place. Manufacturers can use SRM 2372 to validate the values assigned to their own reference materials. Individual forensic laboratories can use SRM 2372 to validate new DNA quantitation methods as well as to verify the assigned DNA concentration of their in-house calibration standards.

The National Institute of Standards and Technology (NIST) is the National Metrology Institute for the United States of America. NIST's mission is to develop and promote measurement, standards, and technology to enhance productivity, facilitate trade, and improve the quality of life. To help achieve these goals, NIST develops and distributes Standard Reference Materials (SRMs) for many different measurement systems. SRMs allow laboratories to establish the traceability of a measurement to internationally recognized scales and units.

Approximately 1300 SRMs are currently available for use in basic research and the applied industrial, chemical production, environmental, fire safety, health, and law enforcement communities. NIST SRMs for the forensic and paternity DNA typing communities include SRM 2390 DNA Profiling Standard for RFLP typing, SRM 2391b PCR-Based DNA Profiling Standard, SRM 2392 Mitochondrial DNA Sequencing Standard (Human), and SRM 2395 Human Y-Chromosome DNA Profiling Standard.

This presentation will introduce SRM 2372 Human DNA Quantitation Standard, which has been produced to support the need for human-specific DNA quantitation in forensic casework and calibration of a variety of new quantitative polymerase chain reaction (qPCR) assays now available. Each unit of SRM 2372 consists of three well- characterized DNA extracts; each delivered in a separate screw-capped micro-centrifuge tube. Component A is a single-source human male material derived from blood Buffy coats. Component B is a multiple- source human female material derived from blood Buffy coats. Component C is a multiple-source male and female material derived from Sigma-Aldrich (St. Louis, MO) freeze-dried human placentas. Approximately 120 µL of 50 ng/µL double stranded DNA is provided in each tube.

The conventional DNA concentration of all three of these materials has been assigned with the U.S. National Reference UV/Visible Spectrophotometer, where an absorbance of 1.0 at 260 nm equals 50 ng/µL of double stranded DNA. Additional absorbance readings have been made at 230 nm, 270 nm, 280 nm, and 330 nm in order to help characterize the purity of the extracted DNA materials. In addition, an interlaboratory study has been conducted, to verify that the SRM 2372 materials perform well in currently used DNA quantitation assays by the forensic DNA community. A description will be provided for how the information gathered from the interlaboratory study and the detailed homogeneity and stability tests performed at NIST with SRM 2372 are used to define the certified values found on the Certificate of Analysis.

SRM 2372 is intended to enable the comparison of DNA concentration measurements across time and place. Manufacturers can use SRM 2372 to validate the values assigned to their own reference materials. Individual forensic laboratories can use SRM 2372 to validate new DNA quantitation methods as well as to verify the assigned DNA concentration of their in-house calibration standards.

The availability of SRM 2372 provides a Quality Assurance tool for those laboratories that desire to make their DNA quantitation measurements traceable to a National Standard.

Human DNA Quantification, qPCR, Quantitative DNA Standard

 ** Presenting Author*