B90 Comparison of Powerplex® 16 Bio With DNA IQ or Chelex 100 Isolation Procedures

Juan J. Yunis, MD, Cielo R. Pineda, MSc, Sandra Moreno, BS, Claudia Rodriguez, BS, and Emilio J. Yunis, MD, Universidad Nacional de Colombia and Servicios Medicos Yunis Turbay y Cia., Ciudad Universitaria, Calle 45, Universidad Nacional de Colombia., Ave 22 # 42-24, of 102, Bogotá, Columbia*

The goal of this presentation is to compare the efficiency and reproducibility of Powerplex□ 16BIO with DNA isolated samples by Chelex 100 and DNA IQ.

Different DNA isolation methods are used worldwide in forensic and paternity testing laboratories. The amount of template and the absence of PCR inhibitors are crucial factors in order to obtain efficient and reproducible results.

The isolation of DNA by Chelex 100 is one of the most popular methods for DNA isolation. However, the presence of PCR inhibitors and the difficulty to determine the amount of DNA in a sample represent some drawbacks for this method. The DNA IQ method has been recently introduced. The virtual absence of PCR inhibitors and a known quantity of DNA at the end of the extraction procedure represent major advantages for this method. The performance of Powerplex□ 16 BIO (FGA, TPOX, D8S1179, vWA, Penta E, D18S51, D21S11, THO1, D3S1358, Penta D, CSF1PO, D16S539, D7S820, D13S317, D5S818, and Amelogenin, Promega Corporation, Madison, WI) on DNA isolated samples by either Chelex 100 or by DNA IQ has been evaluated.

For Chelex samples, 10 ul of whole blood were incubated in 1 ml of distilled water at room temperature for 30 minutes, spun down 3 minutes at 13000 rpm, and the pellet resuspended in 180 ul of Chelex 100 at 5%, vortexed, boiled for 8 minutes, vortexed and spun down before use.

For DNA IQ samples, 10 ul of whole blood were used following manufacturers recommendations (Promega Corporations). Samples were resuspended at 1 ng /ul according to the instructions.

The amplification of Powerplex□ 16 BIO was carried out as recommended and the PCR amplification products were resolved in Long Ranger Gels (Biowhittaker) for 1 hr. 50 min. in a 20 x 43 SA32 gel electrophoresis box at 2000 volts (50 Watts) in 1X TBE.

The results showed a decrease in the intensity of signals for the TPOX and THO1 loci with Chelex isolated samples when compared to the signals obtained with DNA IQ. In addition, the amplification failure rate was higher in Chelex isolated samples (2.3%, 11 out of 480 samples analyzed) compared to 0.57% with DNA IQ isolated samples (4/700).

Results underline the importance for the proper amount of DNA template and the absence of PCR inhibitors in order to obtain balanced and reproducible results with PCR megaplex systems such as Powerplex□ 16 BIO.

DNA, Paternity Testing, Powerplex□

 ** Presenting Author*