COMPARISON OF POWERPLEX[®] 16 BIO WITH DNA IQ™ OR CHELEX[®] 100 ISOLATION PROCEDURES

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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 represents some of the 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. We have evaluated the performance of PowerPlex[®] 16 BIO (FGA, TPOX, D8S1179, vWA, Penta E, D18S51, D21S11, TH01, 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[™].

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

For DNA IQ™ samples, 10ul of whole blood were used following manufacturer's recommendations (Promega Corporations). Samples were re-suspended 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 TH01 loci with Chelex isolated samples when compared to the signals obtained with DNA IQ^{TM} . 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^{TM} isolated samples (4/700).

Our results underline the importance of 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.