FINAL TEST REPORT

Detection of Human DNA Contamination and DNase in 14 Cotton, 14 Nylon Flocked and 14 Foam Swabs

Testing Facility

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Proprietary Information

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Experiment Start Dates:	September 22 and October 9, 2009	
Experiment End Dates:	September 25 and October 14, 2009	

DS09-01 and DS09-02

Study Numbers:

Objectives

The objectives of the study were to detect the presence of human DNA contamination and DNase presence on the test swabs.

Methods

Test Swabs

PSR# 296-09, Lot 2297 (cotton, n = 14)

PSR# 290-09, Lot 2288 (nylon flocked, n = 14)

PSR# 207-09, Lot 2198 (foam, n = 14)

Detection of Human DNA Contamination

Human DNA extraction on the test swabs was performed with an extraction buffer. An aliquout of the extract was added to the PCR buffer containing primers specific for human DNA. A positive control was prepared by adding 25 or 50 pg of human DNA to the PCR buffer. The negative control contained PCR buffer only. The reaction mixture was amplified for 50 cycles. The PCR products were subjected to gel electrophoresis. Gels were examined visually under ultraviolet illumination and photographed using a Gel Logic 100 Imaging system and Kodak MI SE software (v4.5.0).

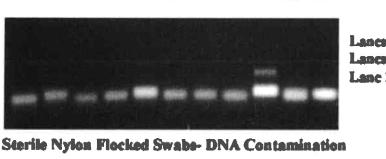
DNase Detection

A PCR reaction buffer containing 1 KB DNA ladder was prepared. Swabs were aseptically removed from packaging and placed in the reaction buffer. A positive control was prepared by adding DNase to the buffer. The negative control consisted of reaction buffer only. Swabs and controls were incubated for two hours at $37^{\circ}\text{C} \pm 1^{\circ}\text{C}$, centrifuged at 12,000 rpm for 5 minutes, and 5 μ l of tracking dye were added to 25 μ L of the reaction buffer. Samples were electrophoressed on agarose gel and photographed using a Gel Logic 100 Imaging system and Kodak MI SE software (v4.5.0). Degradation of DNA ladder by the presence of DNase was examined visually.

Results

10 11

Detection of Human DNA Contamination



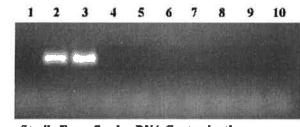
Lanes 1-8: Samples Lancs 9-10: Positive Controls (Mpg. 25pg) Lane 11: Negative Control



Lance 1-2: Positive Controls (1000, Med) Lane 3: Negative Control Lanes 4-11: Samples

Sterile Cotton Tipped Applicator- DNA Contamination

Figure 1. Gel electrophoresis pattern of amplified human DNA to detect human DNA contamination of nylon flocked swabs and cotton swabs after PCR amplification.



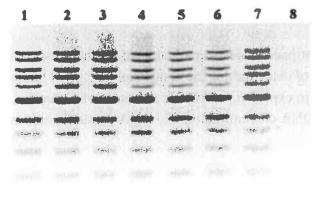
Lane 1: Negative Control Lanes 2-3: Positive Controls (1879, 80pg)

Lanes 4-10: Samples

Sterile Foam Swabs- DNA Contamination

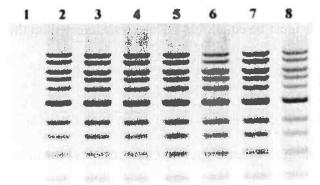
Figure 2. Gel electrophoresis pattern of amplified human DNA to detect human DNA contamination of foam swabs after PCR amplification.

DNase Detection



Lanes 1-6: Samples
Lane 7: Negative Control
Lane 8: Positive Control

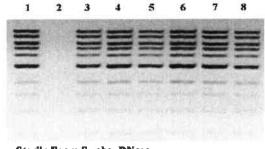
Sterile Nylon Flocked Swabs- DNase



Lane 1: Positive Control Lane 2: Negative Control Lanes 3-8: Samples

Sterile Cotton Tipped Applicator- DNase

Figure 3. Gel electrophoresis pattern of DNA ladder to detect the presence of DNase in nylon flocked swabs and cotton swabs.



Lane 1: Negative Control Lane 2: Positive Control Lanes 3-8: Samples

Sterile Feam Swabs- DNase

Figure 4. Gel electrophoresis pattern of DNA ladder to detect the presence of DNase in foam swabs.

Conclusions

Human DNA Detection

Fourteen cotton, 14 nylon flocked and 14 foam-tipped swabs were evaluated for the presence of human DNA contamination by PCR. The limit of detection for the assay was ≤ 25 pg of human DNA. Positive and negative controls were within expected parameters. Absence of a specific sized DNA amplicon indicated lack of human DNA contamination. No DNA contamination was detected on the swab samples tested.

DNase Detection

Fourteen cotton, 14 nylon flocked and 14 foam-tipped swabs were evaluated for the presence of DNase by gel electrophoresis. Positive and negative controls were within expected parameters. The presence of DNase was determined by comparison to the negative control. The relative intensities of the negative control and test bands must be equal. No DNase was detected on the swab samples tested.

Reference:

Butler, J.M. (2005) Forensic DNA Typing: Biology, Technology, and Genetics of STR Markers, 2nd Edn., Elsevier Academic Press, MA