B96 The Application of Ultraviolet Irradiation to Exogenous Sources of DNA in Plasticware and Water for the Amplification of Low Copy Number DNA

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The goal of this presentation is to describe a technique to eliminate amplification of exogenous DNA from plastic ware and water used in PCR applications without compromising the detection of Low Copy Number DNA.

This presentation will impact the forensic community and/or humanity by providing a fast and cost effective means for sterilization of contaminants present in plastic ware used for PCR detected under Low Copy DNA amplification Methods.

Using High Sensitivity Forensic STR PCR DNA typing methods, it was determined that contamination of presumably sterile plastic ware and water can be present in low concentrations not previously detected by standard PCR methods. One technique commonly used to eradicate the presence of DNA is ultraviolet irradiation; the authors optimized such a protocol used in the treatment of water, tubes, plates, and tips for Low Copy Number DNA (LCN) amplification.

Ultraviolet light from a Stratalinker 2400 (Stratagene, La Jolla, CA) was administered to 0.2mL tubes, 1.5mL tubes, and PCR plates containing up to 500pg of DNA. They were subsequently quantified with an ALU based real time PCR method using the Rotorgene 3000 (Corbett Research, Sydney, Australia) (1, 2). Overall, there was a decrease in concentration of DNA recovered as the duration of treatment increased. Nonetheless, fol- lowing 45 minutes of irradiation of a PCR plate with 500 pg of DNA, 5.7 pg was still apparent. However, when the plate was raised within an inch of the UV source, less than 0.24 pg of DNA was detected. Additionally, lining the area around the samples with aluminum foil reduced the amount of time necessary for irradiation, as only 30 minutes was necessary to eliminate the presence DNA in the raised PCR plate. Similar experiments were con- ducted with respective concentrations of DNA in water for 50mL tubes, 15mL tubes, and 1.5mL tubes with comparative results. It is plausible that the aluminum foil increased the amount of reflection in the area thereby enhancing penetration of ultraviolet rays through the walls of the plastic ware.

This protocol was tested for the possibility of inhibitors produced from irradiation of plastic tubes (3). Since these protocols require less irra- diation time than previous studies, PCR sensitivity was not affected. Moreover, the lifespan of the ultraviolet lamps was extended. The findings demonstrate that this method is useful as an additional precautionary measure to prevent amplification of extraneous DNA from plastic ware and water without compromising the sensitivity of LCN DNA amplifications.

1. Buel E, Nicklas JA. Development of an Alu-based, QSY 7 labeled primer PCR method for quantitation of human DNA in forensic science. *J Forensic Science* 2003; 48(2): 282-291.

2. Buel E, Nicklas JA. Development of an Alu-based, real-time PCR method for quantitation of human DNA in forensic samples. *J Forensic Science* 2003; 48(5): 936-944.

3. Burgess LC, Hall JO. UV light irradiation of plastic reaction tubes inhibits PCR. *Biotechniques* 1999 Aug; 27(2):252,254-4,256

Contamination, Ultraviolet Irradiation, Low Copy Number DNA

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