TITLE: Development of Poly(ethylene oxide) as a Polymeric Sieving Matrix for Forensic STR Analysis

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TEXT:

As the forensic community moves towards fully-integrated microdevices capable of sample preparation and full genetic analysis, the DNA separation matrix chosen will play a major role in device design, analysis time, and ease of use. Though current commercial products accurately and reproducibly separate fragments with high resolution in lengthy capillary systems, their cost and separation efficiency over short distances become prohibitive when transitioning from capillary-based separations to microchip-based systems. Polymer evaluation using capillary electrophoresis requires that attention be paid to the feasibility of the sieving matrix for microchip based analysis, the next-generation of technology for rapid DNA analysis. Critical evaluation of polymer viscosity, resolution over short distances, capillary/microchannel pretreatment, reproducibility of separations, ease of polymer preparation, and cost of analysis must occur when developing a polymer for separation of STR fragments for forensic genetic analysis with the goal of translation to the microdevice.

The work presented here describes the evaluation of poly(ethylene oxide) (PEO), an inexpensive, commercially-available polymer, for separation of STR fragments in capillaries and for potential inclusion in the microchip platform. Accurate, repeatable separations of STR fragments with single base pair differences are reported using a denaturing, low-viscosity, low-molecular weight solution of PEO. The effects of buffer composition, capillary temperature, polymer weight and concentration are described. A variety of polymer weights are explored at varying concentrations to determine the polymeric conditions that result in the highest resolution separation. Reproducibility of the separation is evaluated, as well as the use of epoxy-poly(dimethylacrylamide)(EPDMA)-coated capillaries for minimization of electroosmotic flow (EOF). Urea concentration and capillary temperature are also evaluated and optimized conditions for high-resolution separations are presented.

Key Terms: DNA, capillary electrophoresis, microchip