INTEGRATED MICROSYSTEM FOR FORENSIC DNA FINGERPRINTING

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DNA typing is widely accepted in the forensic community for human identification. However, the current methodologies for typing short tandem repeat (STR) DNA markers are slow and labor-intensive. Federal and state crime laboratories have a large number of DNA samples waiting for STR analysis. To address this backlog, and prevent a recurring backlog, the FBI Laboratory has recently sought new technologies that enable rapid STR typing. STR typing has been demonstrated on glass^[1-3] and plastic^[4, 5] microchip devices. However, sample preparation (DNA extraction and PCR) must still be done off chip. We are currently developing an integrated microfluidic system that integrates all of the necessary processing steps of forensic DNA STR marker analysis and performs them automatically. In this paper we report the development of an integrated device capable of allele PCR amplification and CE separation for STR typing. Functional devices for DNA extraction and the feasibility of further integration will also be discussed.

The design of the integrated device for the PCR and CE analysis of forensic samples is shown in Figure 1. Amplification of the STR loci in a forensic sample is followed by the addition of an internal size standard to the amplification products and to an allelic ladder. The sample amplification products and the allelic ladder are then separated sequentially by CE. The PCR requires thermal cycling between three temperature zones, ranging from 55 to 95°C, and the CE channel needs to be maintained at 60°C during STR analysis. Therefore, a printed circuit board (PCB) and a Peltier device were designed to provide all the thermal requirements of the integrated microsystem. This approach results in a compact operational device with an integrated control unit, which is crucial to the success of functional integration.

To minimize the cost, hot embossing has been used to produce the integrated plastic cartridge. The PCB is then attached to the microchip device to provide heating, temperature sensing, and electrical circuits for CE analysis. The portion of the PCB under the PCR chamber is replaced with a copper plate. A Peltier device is attached to the backside of the copper plate to provide the heating and cooling during the PCR. Figure 2 illustrates the design of a multilayer printed circuit board, which can be constructed using standard multilayer PCB processing and materials. Heating for CE channel is provided by graphite ink screen printing on the board. The resistive ink employed is a carbon-graphite ink in a phenolic resin rated at 20 ohm/square at 0.5 mil thickness. Desired temperatures have been achieved by power modulation to the resistor network with feedback provided by surface mounted temperature sensors or PTCR resistive inks. The performance of this integrated device is currently being tested. Results on allele resolution and detection sensitivity will be presented.

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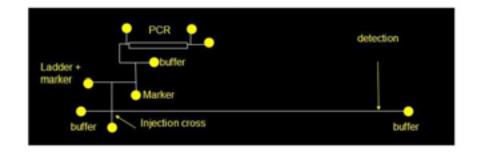


Figure 1. Schematic of an integrated DNA STR typing system.

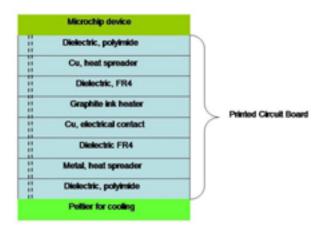


Figure 2. Design of printed circuit board layer structure.

References:

- 1. Schmalzing, D., et al., *DNA Typing in Thirty Seconds with a Microfabricated Device*. Proceedings of the National Academy of Sciences USA, 1997. **94**: p. 10273-10278.
- Schmalzing, D., et al., Two-color multiplexed analysis of eight short tandem repeat loci with an electrophoretic microdevice. Anal Biochem, 1999. 270(1): p. 148-52.
- 3. Mitnik, L., et al., *High-speed analysis of multiplexed short tandem repeats with an electrophoretic microdevice.* Electrophoresis, 2002. **23**(5): p. 719-26.
- 4. Sassi, A.P., et al., *Rapid, parallel separations of d1S80 alleles in a plastic microchannel chip.* J Chromatogr A, 2000. **894**(1-2): p. 203-17.
- 5. Shi, Y. and R.C. Anderson, *High-resolution single-stranded DNA analysis on 4.5 cm plastic electrophoretic microchannels.* Electrophoresis, 2003. **24**(19-20): p. 3371-7.