CE-LIF Coupled with PDMS-interconnected Microfluidic Systems for Rapid Separations of Neurotransmitters

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Abstract. In vivo measurement of neurotransmitters in cerebrospinal fluidic (CSF) is an effective method to monitor neuronal activities. Microdialysis coupled with HPLC or capillary electrophoresis (CE) is a powerful approach for in vivo monitoring of neurotransmitters. HPLC requires relative large sample volumes (e.g. 5-15 □L), which is infeasible for limited availability of CSF [1], whereas CE is able to analyze nano- or pico-liter samples in a short time such as 20 seconds. We have developed an integrated CE system targeting on in vivo measurements of essential neurotransmitters. This CE system employs laser-induced fluorescence detectors, and online fluorogenic derivatization. Experimental results show that this system is capable of performing long-term monitoring with robustness, accuracy, high sensitivity and reproducibility in a real-time fashion.

Keywords: Flow gate, PDMS, Prototyping, Interconnect, Capillary electrophoresis

1. Introduction

The complexity of microfluidic systems is increasing rapidly as various functional parts are integrated in one system [2], while interfaces between these parts play a critical role in the performance of a microfluidic system [3]. Commercially available interfaces used in microfluidic systems are relatively expensive and difficult to further miniaturize. Reported here is a rapid prototyping method developed to fabricate poly(dimethylsiloxane) (PDMS) interfaces for a capillary electrophoresis (CE) system (Fig. 1). Owing to PDMS inherent properties: elasticity, optical transparency, and suitability for prototyping, the fabricated PDMS interfaces have the following advantages: ease of fabrication, ease of use, reduced dead volumes, and excellent performances.

In this paper, we report the method of rapidly prototyping PDMS interfaces for an integrated CE system. These interfaces have smaller channel diameters, offer easy capillary alignment and stabilization, and are reusable, inexpensive, and transparent. To demonstrate the performance of the PDMS-interconnected system, amino acid neurotransmitters were derivatized online with NDA （naphthalene-2,3-dicarboxaldehyde）at the presence of cyanide and then electrophoretically separated by the integrated CE system.

2. Experimental

Preparation of the flow gate and the 4-way mixer

A stainless steel wire was clamped to produce slots in the middle. A nylon line was pressed to produce a flat section, at the center where a hole was punched. Then, the wire was inserted through the hole until reached the slots. The assembly of the cross was suspended in a petri dish.

Preparation of the linear connector

A stainless steel wire was tapered and inserted into a silica capillary until it stopped. The linear assembly was suspended in a petri dish.

Prototyping.

PDMS prepolymer and curing reagent at the mass ratio of 10/1 were mixed, degased and poured into a petri dish with the mold suspended. The mixture was cured for >30 minutes at 80 ℃, then cured at 110 ℃ to enhance PDMS rigidity.

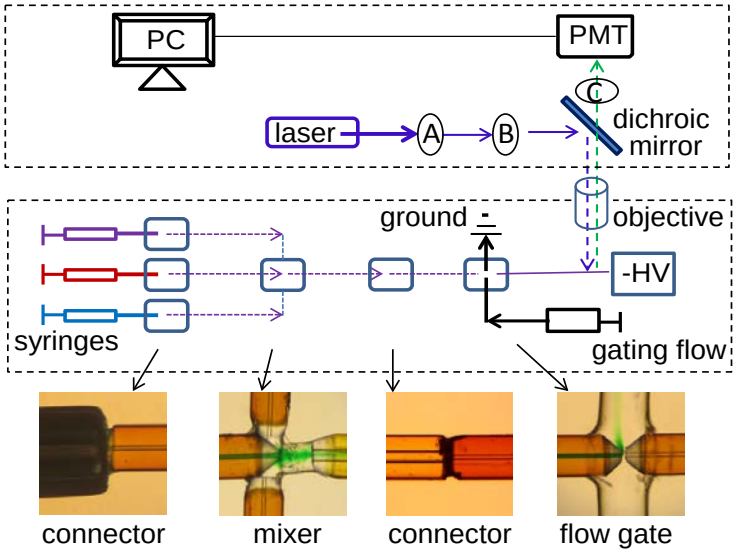

Fig. 1. Schematic diagram of the PDMS-interconnected CE system (see details in the text). A, pin hole filter; B, excitation filter; C, emission filter.

Separation conditions

Separation buffer was 20 mM tetraborate buffer at pH 9.2 and electric field 1560 V/cm.

3. Results and Discussion

To demonstrate the performance of the CE system interconnected through PDMS interfaces (Fig. 1), NDA/cyanide/amino acids were used to perform online derivatization and separations. The performance of the PDMS-interconnected system was tested through separating amino acids. Fig. 2a shows a typical electropherogram showing well-resolved 5 standard amino acids which were directly delivered through a sample syringe followed by online mixing, derivatization, injection, separation, and detection. Overlap injection was also performed as shown in Fig. 2b which doubled the analysis throughput. To test its long-term performance, more than 300 consecutive injections were performed and the results were plotted in Fig. 2c, and an excellent reproducibility in peak height was demonstrated by the %RSD (relative standard deviation) of 1.6; high separation efficiency was also obtained with the theoretical plates of 250 k. Further, amino acids at various concentrations of 1.0, 2.0, 4.0, and 8.0 □M were sequentially dialyzed for 5 minutes through a microdialysis probe and the results of glutamate are summarized in Fig. 2d, which suggests that the integrated system was able to dynamically monitor sample concentration variation with the response time of ~40 s at 90% of the maximum signal. These results demonstrate that the PDMS-interconnected system is potentially valuable for in vivo neurotransmitter monitoring [4, 5].

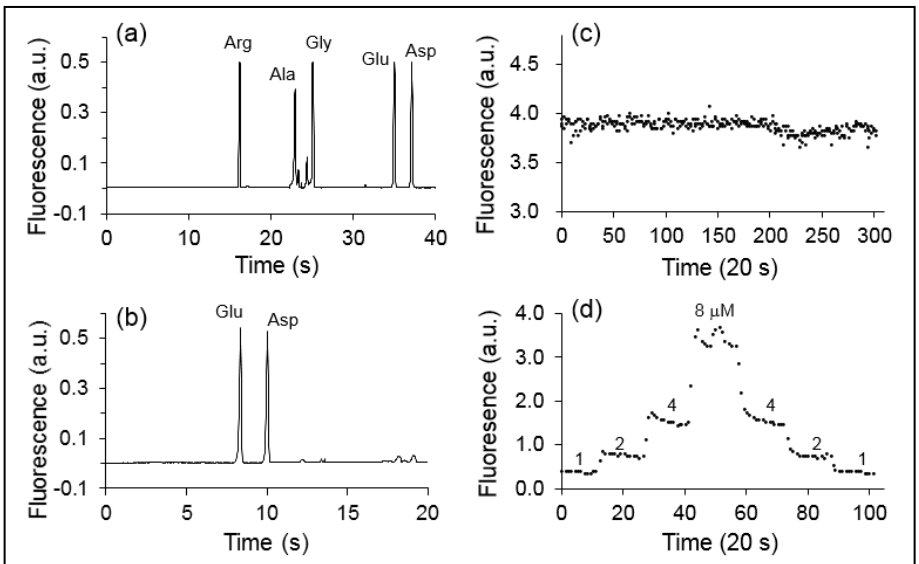

Fig. 2. (a) Electropherogram of 5 amino acids, 1.0 Electropherogram of glutamate and aspartate at 1.0 □M each with overlap injections; (c) Glutamate peak heights of repeated overlap injections. Glutamate at 5.0
a microdialysis probe.

As a summary, we have developed the method for fabricating PDMS interfaces used for flow-gated injection, multiple-reagent online mixing, and tube-to-tube connection. Our studies have shown that these miniaturized interconnectors accommodated tubing connection and provided visible trouble shooting. Overall, the rapidly prototyped PDMS interconnectors are reusable, inexpensive, convenient for connection, and reliable when integrated with the CE detection system. Therefore, these robust interconnectors are suitable for rapid separations in microfluidic systems and could be further miniaturized in an integrated device.

4. Acknowledgements

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5. References

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