NOVEL SEPARATION METHOD ON A CHIP USING CAPILLARY ELECTROPHORESIS IN COMBINATION WITH DIELECTROPHORESIS

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Abstract

We propose a new methodology for the separation of the constituents of complex chemical and biological mixtures using electrophoresis combined with dielectrophoresis on a chip and laser induced fluorescence detection. We use a network of closely spaced interleaved microelectrodes to facilitate the use of dielectrophoresis as an electrical gradient "stationary phase" for bulk separations. Our new method combines the potential of dielectrophoretic separations with integrated chip electrophoreis, which offers the advantages of high throughput and of precise control of a small amount of sample volume using electroosmotically driven, high-efficiency separations of a number of chemicals and molecules.

Keywords: capillary electrophoresis, dielectrophoresis, chromatography, laser induced fluorescence

1. Introduction

Conventional chromatographic separations depend upon differential mobilities of analytes between stationary and moving phases. The stationary phase is often a poorly defined matrix of particles, aggregates, polymers, or monolithic sol-gel [1], whose characteristics are largely unknown. Chromatographic separations of the future might be different in that the separation mechanism is under direct control of the analyst. We propose a new methodology for the separation of the constituents of complex chemical and biological mixtures by integrated chip electrophoresis (ICE) combined with dielectrophoresis (DEP) [2] on a chip using laser induced fluorescence (LIF) detection.

We utilize dielectrophoresis to effect separation. An electric field gradient acting on an analyte induces a dielectrophoretic force depending on the analyte's polarizability. In such a scheme, the separating power depends on the electric field gradient and hence can be controlled externally. In previous work, Washizu [3], Fuhr [4], and Gascoyne [5] have investigated biopolymer separation, cell sorting, and DNA handling in a hydrodynamic flow using dielectrophoresis; however, these methods did not make use of electrophoresis. Our new method combines the potential of DEP separations with ICE, which offers the advantages of precise control of a small amount of sample volume using an electroosmotically driven flow and offers high-throughput, high-efficiency separations of chemicals mixtures.

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2. Theory

Dielectrophoresis, which arises from the dielectric force evoked by an electric field gradient [2], has been used primarily to form "traps" to manipulate individual micron-sized particles [3-5] depending upon their polarizability. DEP, in principle, should act on molecules in solution as well particles. We propose to use a network of closely spaced interleaved microelectrodes to facilitate the use of DEP as an electrical gradient "stationary phase" for bulk separations (Figure 1). Figure 2 illustrates a schematic design of a microchip using electrophoresis with dielectrophoresis. We can control and inject a small volume of sample in a microchannel utilizing electroosmotic flow as a means of transport in a chip [6-9]. The analytes travel along an array of interdigitated microelectrodes where their motion is retarded according to their size by DEP. The separation of a mixture is accomplished by combining ICE with DEP with LIF detection. We anticipate that this method will be valuable for a variety of analytical applications, allowing "tunable" separations based on the dielectric properties of the analytes.

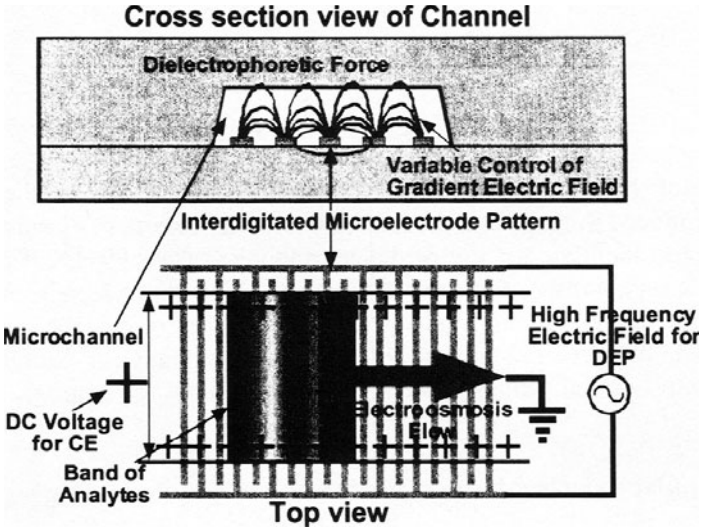

Figure 1. Schematic of separation with DEP and ICE. The separation efficiency for analytes with different dielectric properties can be manipulated by changing the frequency and magnitude of the AC field.

3. Experimental

Glass wafers are patterned using standard microfabrication and lithographic techniques [6-9], using two photomasks. The main separation channel has a width of 80μm, and traverses an interdigitated array (1.5μm spacing) of microelectrodes. After wafer fabrication, access holes are drilled, another glass wafer is thermally bonded, and reservoirs are affixed. We control and inject a small sample volume of analyte using cross-injection. The analytes travel near the microelectrode array, which is isolated from the ICE voltage with an insulation layer silicon dioxide. Electroosmotic flow along the channel is impeded by DEP, which is generated by a high-frequency (kHz-MHz) electric field. A HeCd laser beam is focused into the channels for laser induced fluorescence (LIF) detection; the fluorescence signal passes through spectral and spatial filters before detection with a PMT.

4. Results and discussion

We have developed an interdigitated microelectrode array inside a microchannel on a glass chip. Figure 3 shows microscopic images of a top view of the microchannel and interdigitated microelectrode. We will first test this system with micron-size particles, including solutions of cells and organelles. Figure 4 shows that in preliminary experiments, we trapped 1.4 μm diameter polystyrene beads between interdigitated microelectrodes in a buffer-filled channel when applying high frequency electric field. After applying our technique to individual biological particles, we will attempt to extend it to the molecular level, to separate and detect small quantities of chemicals with varying dielectric properties.

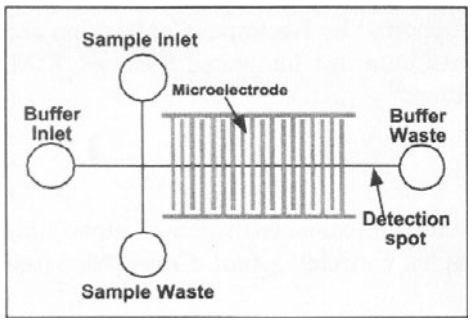

Figure 2. Schematic design of a microchip using CE with DEP

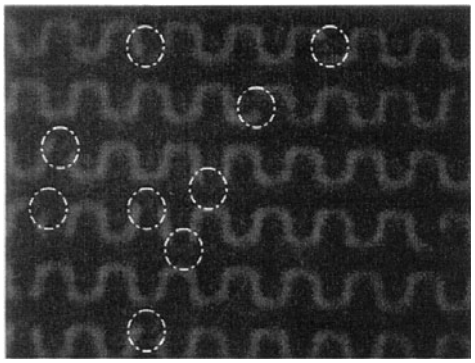

Figure 4. Microscopic images of trapping 1.4 μm diameter polystyrene beads (inside white circles) between interdigitated microelectrodes in a channel while applying high frequency electric field.

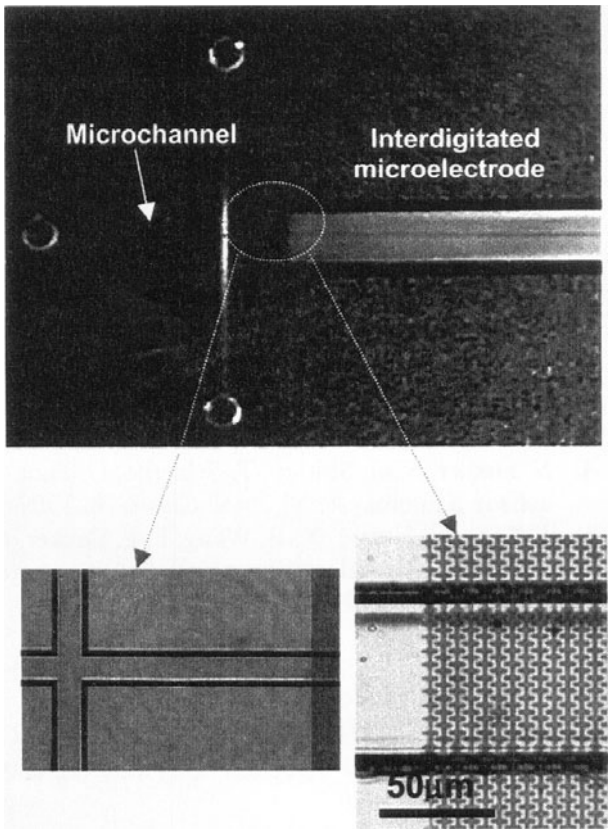

Figure 3. CCD image of top view of our prototype DEP chip. Inset: CCD image of the intersection of channel and the interdigitated microelectrodes.

5. Conclusions

We have fabricated a new chip-based separation system, using dielectrophoresis as an electrical gradient "stationary phase" for bulk separations. Our new method combines the potential of DEP separations with integrated chip electrohoresis. We will use this method to separate and detect anlytes with varying dielectric properties. This new technique has the potential to be easily adaptable to a wide variety of chemical separations, and has the novel advantage of separation "tunability."

Acknowledgements

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