MICROPARTICLE-ASSISTED CONTINUOUS 2-DIMENSIONAL GRADIENTS OF THERAPEUTIC AGENTS IN MICROCHANNEL FOR DRUG TESTS

Matthew Estes¹ and Chong H. Ahn¹,²

Microsystems and BioMEMS Lab
¹Department of Biomedical Engineering
²Department of Electrical and Computer Engineering
University of Cincinnati, Cincinnati, Ohio 45221 USA

ABSTRACT

In this work, a new microparticle-assisted 2-dimensional gradient generator for therapeutic agents in microchannel has been proposed, designed, fabricated and characterized, which uses continuous flow to simultaneously generate two overlapping gradients, one biomolecular gradient perpendicular to the direction of flow and a second gradient parallel to flow. The generated two simultaneous and perpendicular gradients show good promise towards adaptation in the drug testing field. This system is a simple, low cost, polymer, disposable design that can be integrated with other combinatorial chemistry processes.

KEYWORDS: Gradient Generation, Drug Testing, Combinatorial Chemistry

INTRODUCTION

The proliferation of therapeutic agents for diseases ranging from AIDS to cancer has provided new promise for living with and overcoming such conditions. Recent work has shown that frequently the most efficacious treatment is a combination of factors [1,2]. Combining treatments is not without its risks and all new medicines must be examined thoroughly to determine how they interact with other therapeutics. This paper has proposed and explored a new device capable of generating continuous and perpendicular gradients of two different therapeutic agents inside a microchannel.

The two-step 2-dimensional gradient generator presented in this work makes it capable of using first mixing to generate a gradient of low diffusivity drug perpendicular to the direction of flow and second diffusion to generate a gradient of high diffusivity drug parallel to flow.

DESIGN AND FABRICATION

The first gradient across the channel is generated using a repeating system of Tesla mixers [3] as illustrated in Figure 1. By recombining two separate parallel solutions of high and low micro particle concentration, a smooth gradient perpendicular to the direction of flow is created. The second gradient along the channel is generated using diffusion from areas of high concentration of high diffusivity nano-particles to areas of low concentration, steadily increasing the concentration along the path of the microchannel [4].

The gradients are generated one at a time, using particles of varying diffusivity because the stacking of multiple gradients generates some difficulties if the two therapeutic drugs are of similar diffusivity as illustrated in Figure 2. Two separate
Gradients of similar diffusivity mean that the low flow rate required to develop the second gradient would allow enough time for the first gradient to diffuse away, as seen in Figure 2(a). If the device is operated at a high flow rate to preserve the first gradient, then there is not enough time for the second gradient to develop fully by means of diffusion, as seen in Figure 2(b). An optimal balance can be found if the diffusivity of the first gradient is substantially reduced, such that the device can be operated at a low flow rate, allowing enough time for the second gradient to develop without losing the first gradient, as seen in Figure 2(c). This is accomplished by coupling the first drug to a micro-scale bead carrier to slow its rate of diffusion.

The device was fabricated simply by ordering a mold from the ECShaw company, creating an inverse mold using Polydimethylsiloxane, and using this mold to hot emboss the pattern into a Cyclic-Olefin-Copolymer wafer. This wafer was bonded to a second to create the device as shown in Figure 3.

![Diagram](image)

**Figure 1.** Device design combining a mixer-based gradient generator and a diffusion-based gradient generator to create two perpendicular and continuous gradients.

![Images](images)

**Figure 2.** Conceptual illustration of the problems of perpendicular drug gradient generation in continuous flow from left to right: (a) Two drugs of high diffusivity at low flow rate (b) Two drugs of high diffusivity at high flow rate, and (c) Ideal result obtained by using an upper drug of low diffusivity and lower drug of high diffusivity both at low flow rate.

![Images](images)

**Figure 3.** Photograph of functioning device showing bead gradient (circled in black) at start and end of diffusion bead gradient generator.
EXPERIMENTAL RESULTS AND DISCUSSION

For the characterization of the device, three solutions were prepared: a buffer solution, a bead solution to model a drug bound to a micro-scale bead carrier, and a dye solution to model a drug free in solution.

The buffer and bead solutions were flowed through the two inlet ports of the mixer array to generate a bead gradient perpendicular to the direction of flow. The dye solution was added via the last two inlet ports to provide a source for the parallel to flow gradient. This was done at a variety of flow rates to characterize the device performance as seen in Figures 4 and 5.

![Figure 4. Characterization result of mixer based gradient perpendicular to direction of flow with regard to flow rate.](image)

![Figure 5. Characterization result of diffusion based gradient parallel to direction of flow with regard to flow rate.](image)

The device shows good generation of gradients perpendicular to the direction of flow at a variety of flow rates. The flow rate of 1.0 µL/min results in the best gradient for this design. The ideal flow rate can be altered by adjusting the number of repeating Tesla subunits. For the gradient parallel to the direction of flow a low flow rate provided the best gradient. This is a result of the time needed for diffusion to take place.

CONCLUSION

This paper has demonstrated for the first time to the author’s knowledge a Micro Total Analysis System (µTAS) capable of generating two simultaneous and perpendicular gradients and this work shows good promise towards adaptation in the drug testing field.

REFERENCES:


