IN SITU MICROFLUIDIC PARTITION WITH SEMI-PERMEABLE MEMBRANES FOR STATIC GRADIENT GENERATION

X.L. Luo[1*] and G.W. Rubloff[2]
[1]*Catholic University of America, USA and*
[2]*University of Maryland, USA*

ABSTRACT

We report an *in situ* biofabrication strategy that partitions a microfluidic network into multiple microchannels with semi-permeable biopolymer membranes. The biofabrication of parallel biopolymer membranes was initiated with trapped air bubbles in hydrophobic polydimethylsiloxane (PDMS) microfluidic devices, followed by tunable membrane growth with time. Static gradients were generated and well maintained in the partitioned microfluidic channels by pure diffusion through the semi-permeable membranes. The *in situ* biofabrication provides a simple approach to generate static gradients and an ideal platform for biological applications where flow-free static gradients are indispensable.

KEYWORDS: Static Gradient, Semi-Permeable Membranes, Biofabrication, Air Bubbles

INTRODUCTION

Microfluidic gradient generation roughly falls into two categories: (1) steady gradients that are established within laminar flow streams; and (2) static gradients that are generated purely by diffusion through porous hydrogel/membrane structures or flow-free perfusion channels. While steady in-flow gradient generators normally have faster response time and enable sharper and profound gradient profiles, diffusion-based static gradients are preferred in cellular studies to minimize shear stress, and in chemotaxis studies to decouple cell motion from flow. Here, we report a new static gradient generator composed of biopolymer membranes in PDMS microfluidics. Microchannels were partitioned *in situ* into two source/sink side channels and a middle static gradient chamber established by the air bubble-induced biofabrication of parallel biopolymer membranes. Static gradients were generated and well maintained by pure diffusion through the semi-permeable membranes.

EXPERIMENTAL

The devices were fabricated with soft lithography techniques. The microchannels in Fig. 1 were 50 µm deep with apertures of 50 µm × 50 µm connecting the adjacent microchannels. Air-filled tubing was inserted into the outputs to balance fluidic pressure in the microchannel. During the membrane biofabrication, a chitosan solution of pH 5 and alginate solutions of pH 11 were introduced into the middle and two side channels, respectively, at 1 µL/min flow rate (Fig. 1(c)).

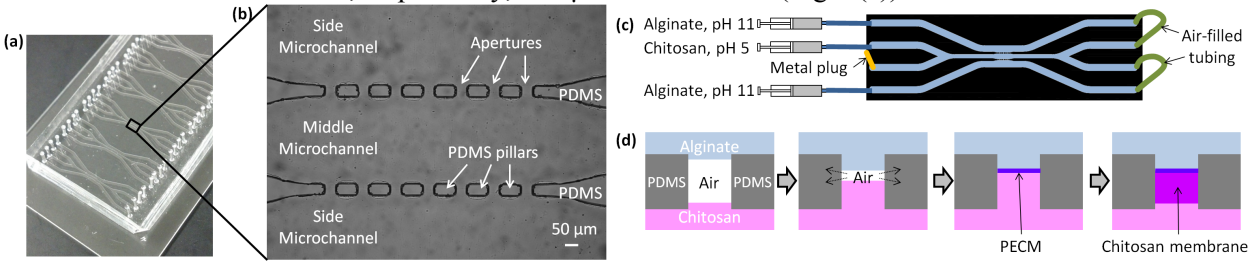

Figure 1: Device and schematic of the biofabrication of parallel membranes. (a) Overall device on a 3" × 1.5" glass slide; (b) apertures connecting three microchannels; (c) solution introduction and tubing connection to balance fluidic pressure; (d) schematic of membrane formation process.

RESULTS AND DISCUSSION

The results of air bubble-initiated membrane biofabrication are shown in Fig. 2. Thanks to the high surface tension of hydrophobic PDMS, air bubbles were trapped within the apertures after the chitosan

and alginate solutions were introduced (Fig. 2(b)). As pressure in the microchannels increased, air bubbles dissipated through the gas permeable PDMS and induced the formation of a polyelectrolyte complex membrane (PECM) layer in each aperture (Fig. 2(c) & (d)). Chitosan grew upon PECM to the desired thickness as hydroxyl ions in alginate solutions continued to diffuse through the semi-permeable membranes. As a result, the thickness of the membrane was tunable with time (Fig. 2(e) & (f)). As separately demonstrated [1], a single membrane withstands fluidic pressure gradients up to 1 atm.

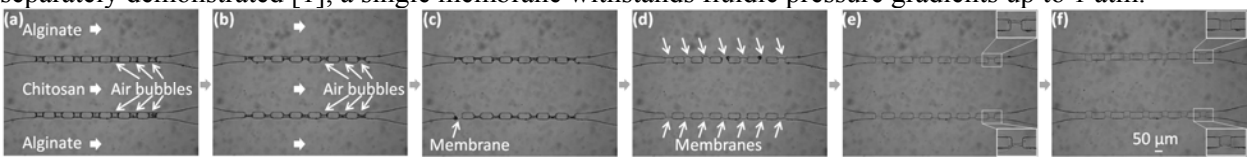

Figure 2: Air bubble-induced biofabrication of parallel membranes. (a) Air bubbles trapped in apertures; (b) air bubbles shrunk due to air dissipation through PDMS; (c) the first membrane formed; (d) parallel membranes formed in each aperture; (e) membrane grew thicker; (f) membranes with desired thickness.

The partitioned microchannels by semi-permeable biopolymer membranes were then used to generate static gradients, analyzed by ImageJ. Fig. 3(a) shows the flow setting (top left) and the fluorescence gradients in the middle microchannel at 20, 60 and 600 sec after the introduction of fluorescein (1mM) and pure PBS solutions into the side microchannels at 10 µL/min, establishing linear gradients (Fig. 3(b)) across stopped flow in the middle microchannel, which remained static after 10 minutes.

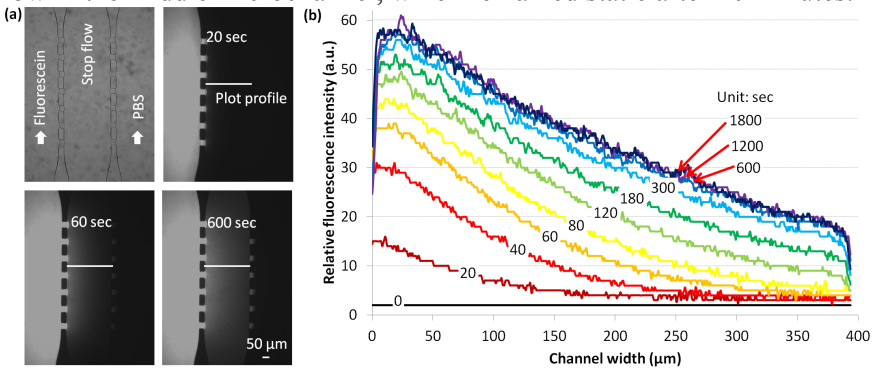

Figure 3: Static gradients generated in the membrane-partitioned microchannel. (a) Flow setup and fluorescent profiles at 20, 60 and 600 sec after the introduction of fluorescein and PBS solutions; (b) plot profiles of gradients vs. time showing that static gradients were generated and maintained after 10 min.

CONCLUSION

We demonstrated the *in situ* partition of microchannels with the biofabrication of semi-permeable biopolymer membranes that were initiated with air bubbles trapped in hydrophobic PDMS device. The membranes were biofabricated in a single-step flow process, creating membranes that were robust and semi-permeable to allow free diffusion of small molecules to generate static gradients in the partitioned microfluidics in 10 minutes. We believe that the *in situ* membrane biofabrication will provide an ideal platform for biological applications where flow-free static gradients are indispensable.

REFERENCES

[1] X.L. Luo, H.C. Wu, J. Betz, W.E. Bentley and G.W. Rubloff, "Air bubble-initiated biofabrication of freestanding, semi-permeable biopolymer membranes in PDMS microfluidics," Biochemical Engineering Journal, 2014, DOI:10.1016/j.bej.2013.12.013.

CONTACT

* X.L. Luo; phone: +1-202-319-6952; luox@cua.edu