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A MICROFLUIDIC DROPLET SERIAL DILUTOR WITH ENHANCED MIXING

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ABSTRACT

We present the development of an automated microfluidic platform that is capable of creating logarithmic serial dilution by adapting pneumatically actuated microvalves. A valve-assisted droplet manipulator integrated with a peristaltic mixer allowed the accurate formation of droplets with controlled sizes and the dilution of reagent sequentially. Hence, serial dilution with flexible dilution factors was obtained in a series of nanoliter-scale droplets. We validated the mixing efficiency and droplet generating performance of the chip at various operating conditions and demonstrated an example of logarithmic serial dilution with two different dilution factors. The microfluidic droplet serial dilutor could be used as an analytical tool to evaluate various complex chemical and biochemical reactions.

KEYWORDS: Microfluidics, Droplets, Serial dilution, microvalves

INTRODUCTION

Serial dilution is one of the most common and basic processes used in chemical and biological laboratories for sample preparation and experiments, and a fundamental operation for microfluidic high-throughput screening devices. Therefore, several strategies have been introduced for the implementation of serial dilution on a chip [1-4]. Although previous systems successfully achieved on-chip serial dilution based on channel networks [1-2], parallel mixers [3], and droplets [4], accurate metering and enhanced mixing of extremely small volumes still remain challenges for rapid and reliable characterization of target systems. Here, we report an automated microfluidic droplet serial dilutor, which generates a series of droplets with a controllable size to form flexible logarithmic concentration gradients of a target molecule.

EXPERIMENTAL

The device consists of a mixer unit and a droplet generator (Figure 1 A). Solutions were introduced in the channels in a fluidic layer by applying a pressure to the backside of solutions, and fluid flows were controlled by operating pneumatic valves in a control layer. To start the dilution process, the sample solution was introduced in the mixing unit (Figure 1 B). Next, the dilution solution was injected into the mixer, and a droplet of the same volume was dispensed into a carrier fluid (Figure 1 C). The dispensed volume of sample was determined by controlling the valve opening time at specific flow-rates of the aqueous phase and the carrier fluid. After generating a droplet, the three mixing valves were operated sequentially to enhance mixing of the sample and dilution buffer. The cycles of droplet formation and solution mixing were automatically repeated to obtain a series of droplets with a concentration gradient.

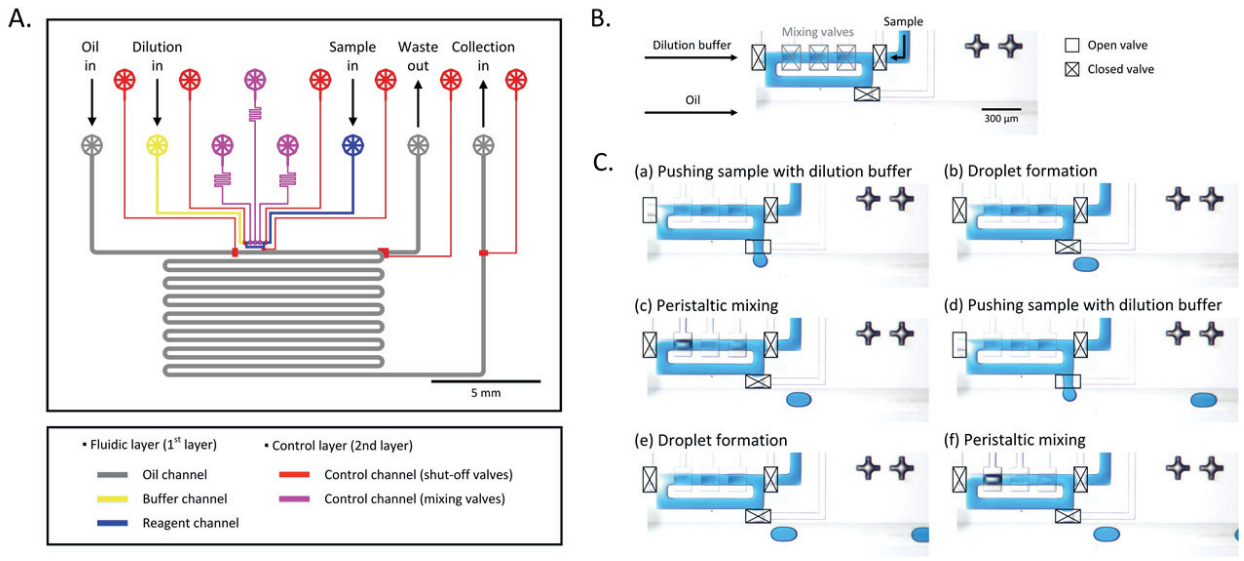

Figure 1: Design and operation of the droplet-based microfluidic device for serial dilution. (A) AutoCAD design of the device, (B) Microscopic image of a fabricated device, and (C) Process flow of on-chip serial dilution.

RESULTS AND DISCUSSION

The two key-elements of the device, the mixer and droplet generator, were calibrated at various operating conditions for robust on-chip serial dilution. Figure 2 A presents the mixing efficiency at various operating frequencies. Since mixing solutions could be obtained within 1 s at the optimized operating frequency of 20 Hz, one cycle of droplet manipulation and mixing could be completed within 1.1 s. Figure 2 B presents the calibration of the dispensing time and the flow rates of the water and oil phases to create a specific droplet size. The linear relationships and small standard deviation in Figure 2 B demonstrate robust and reliable operation of the system as well as droplet monodispersity.

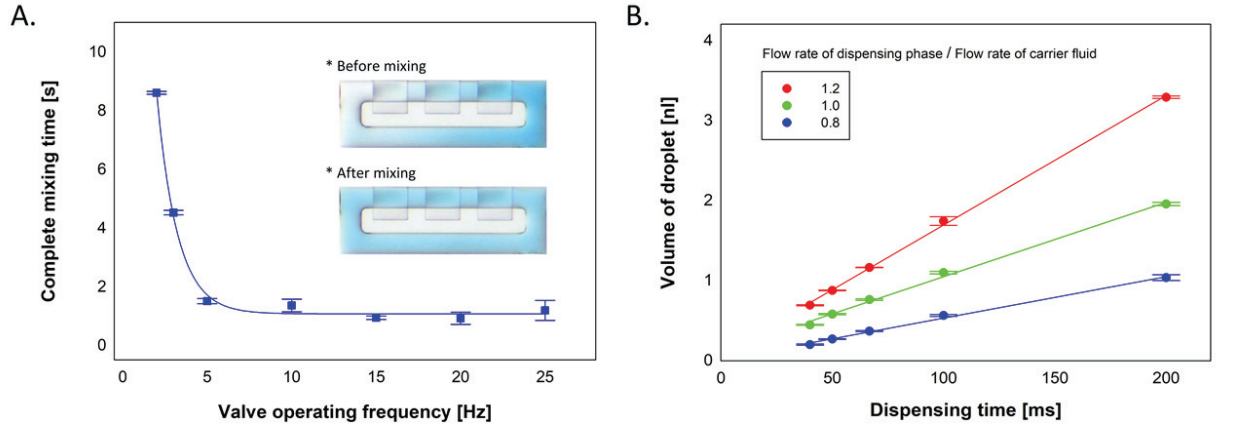

Figure 2: Calibration of the microfluidic chip. (A) Mixing efficiency at various mixing valve operating conditions, and (B) Droplet volume according to various dispensing times and the ratio of oil and water flow rates.

Figure 3 presents an example of serial dilution with two different dilution factors. A 6 nL volume of food dye solution was diluted by adding 0.25 nL (50-µs dispensing, red dots) and 0.5 nL (100-µs dispensing, blue dots) of Milli-Q water. By controlling the dilution dispensing time, logarithmic serial dilution with various log-bases could be achieved.

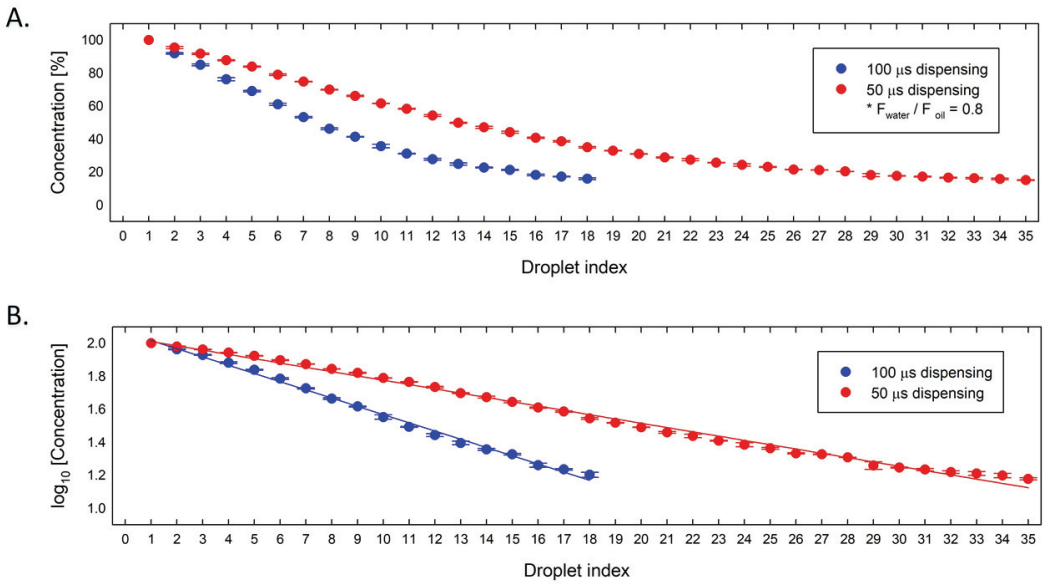

Figure 3: Logarithmic serial dilution with two different dilution factors (n=3).

CONCLUSION

We established an automated droplet-based microfluidic platform for performing logarithmic serial dilution. Integrating a peristaltic mixer and a valve-assisted droplet generator allows the generation of serial dilution with flexible dilution factors on a chip. The present work has great potential in sequential and combinatorial studies of complex systems in chemistry and biology.

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