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Active or Passive On-Demand Droplet Merging in a Microfluidic Valve-Based Trap

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Abstract

A microfluidic valve-based trap enabling controlled capture, release, and temporary immobilization of droplets together with on-demand merging of selected droplets is presented in this paper. The microfluidic trap technology can merge droplets passively or in active manner via a pneumatically actuated membrane. A microchip is developed with two functional units of droplet generator and merging mechanism to implement the passive or active merging performance of the microfluidic valve-based trap using a low and high surfactant concentrated continuous oil-phase.

I. INTRODUCTION

Two-phase flow microfluidics represents a powerful technique for a broad range of applications such as biochemical analysis, biomedical assays, and microbiosynthesis with aqueous-phase droplets inside an immiscible fluid serving as picoliter-scale carriers enabling rapid and high-throughput analyses. Merging of droplets is a key procedure in this approach, providing functionality such as initiating a chemical reaction or combining the encapsulated contents of individual droplets.

To merge multiple droplets, they must be brought into contact with each other to overcome the stabilizing effects of surface tension and remove the thin lubrication film of immiscible fluid between adjacent droplets. Generally, there are two main approaches to merge droplets, namely passive merging and active merging. In the case of passive merging, the droplets are merged naturally with no external force or control. Droplets are brought together by the designs of channel geometries to control the droplet motion by decelerating or stopping them. One common approach for passive droplet merging is to employ channels with an expansion zone or trifurcating junction [1–3] to reduce droplet speed. Droplet merging in an expanded channel is limited to a range of rates and sizes of droplets. In these designs, the

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separating fluid film between the droplets is drained at a fixed rate depending on the width of the expansion [2]. On the other hand, a trifurcating junction allows a controllable rate of fluid draining providing a wider range of rates and sizes of merging droplets [3].

Alternatively, droplets can be merged passively in a channel with incorporation of a constriction or an array of pillars [4–7]. The fluid resistance in these designs causes the droplets to stop and merge. Surface-induced passive merging method in which a surface energy pattern is introduced on the channel wall for entrapment, merging and releasing of droplets has also been reported [8]. In all these passive approaches, transportation and synchronization of pairs of droplets concurrently in the merging zone is challenging. Unsuccessful or undesired multiple merging events are common. Furthermore, surfactant-stabilized droplets cannot be readily merged using the passive techniques.

In contrast, active merging approaches such as electrocoalescence, dielectrophoresis, thermo-capillary, acoustic wave, and magnetic field rely on external force or control for droplet merging. Electrocoalescence is the most popular method of active merging by applying an electric field generated by an embedded electrode at the merging zone. It causes an electric-field-induced instability in the interface between the droplets leading to coalescence. Several studies have shown controlled electrocoalescence of droplets in a microfluidic channel [9–11]. Dielectrophoresis (DEP) is another method for active merging of droplets in which the droplets are subjected to a non-uniform electric field [12]. The DEP force causes the droplets to stretch toward each other and merge by applying a relatively high voltage (i.e. kV) [13]. Thermo-capillary effects by a focused laser beam on the interface of two adjacent droplets can also be used to achieve active merging of droplets [14, 15]. Finally, surface acoustic waves [16] and magnetic field [17] are two novel approaches to induce active merging of the droplets. Although active merging methods have wide applicability, the techniques can add significant complexity to the system.

To address the deficiencies of current merging methods, here we describe an effective approach requiring minimal extra equipment that provides highly controllable manipulation of the rates, sizes, and motion of the droplets using an on-demand passive or active droplet merging technique.

II. DESIGN AND FABRICATION

A. Microchip Design

A schematic diagram of the microchip composed of the fluidic and control layers and two functional units of droplet generator and merging mechanism is shown in Fig. 1a. The fluidic layer includes two independent inlet ports for the aqueous-phase and oil-phase liquids and two outlet ports for waste and merged droplets collection. In this design, the width of the main flow channel is 60 μm and the perpendicular channels are 100 μm wide. The control layer includes three membrane micro-valves for droplet generator, waste line, and merging mechanism. Each membrane micro-valve is individually addressable by a separate control channel.

B. Microfluidic Valve-based Trap Design

A detailed illustration of the microfluidic valve-based trap with the labelled parts is shown in Fig. 1b. The microfluidic trap in the fluidic layer is composed of a micro-chamber followed by a narrow path and two bypasses connecting two sites of the micro-chamber. The microfluidic trap is integrated with three membrane micro-valves as shown in Fig 1b. The micro-valve #1 is designed to block the merging mechanism while the undesired droplets are shunted to the waste line. The micro-valve #2 functions to control the direction of incoming flow into the micro-chamber or bypasses for controlled trapping and releasing of droplets. To entrap the droplets, the micro-valve #2 is switched on and the incoming continuous-phase flow bypasses into the lateral capillaries. It provides a dead volume inside the micro-chamber for trapping and merging of the droplets. To release the combined droplet, the micro-valve #2 is switched off and the incoming flow runs into the micro-chamber. For active merging of the droplets, a pneumatically-controlled membrane is incorporated underneath the micro-chamber. Fluctuation of this underlying membrane can increase the draining rate of the oil film between the entrapped droplets.

C. Microchip Fabrication

The microchip was fabricated by multi-layer soft-lithography. It is composed of polydimethylsiloxane (PDMS) (Sylgard 184, Dow Corning, USA) top fluidic and bottom control layers bonded on a glass slide, as shown in Fig. 1c. The fluidic layer master mold was obtained by spin-coating the negative photoresist SU-8 2075 (Microchem Corp, Newton, MA) at 3500 rpm for 60 s on a silicon wafer to obtain a thickness of 40 μm . The control layer master mold was obtained by laminating a 30 μm dry film photoresist (Think & Tinker Ltd., Palmer Lake, CO) on a silicon wafer. To form a 3 mm thick fluidic layer, the PDMS prepolymer and the curing agent mixture (10:1 ratio) was poured on the master mold, partially cured at 80 $^{\circ}\text{C}$ for 30 min, peeled off and punched to access holes in the fluidic layer. A 36 μm membrane control layer was formed by spin-coating the PDMS prepolymer and the curing agent mixture (20:1 ratio) on the master mold at 1500 rpm for 1 min and partially curing at 80 $^{\circ}\text{C}$ for 11 min. After that, the fluidic layer was aligned on the membrane control layer and fully cured at 80 $^{\circ}\text{C}$ overnight. The two-layer PDMS device was peeled off, punched to introduce pneumatic access ports into the control layer. The assembly was then bonded to a glass slide using oxygen plasma treatment for 60 s. The hydrophobicity of the PDMS surfaces was restored after oxygen plasma treatment which enhances temporary hydrophilicity by incubating the microchip in 115 $^{\circ}\text{C}$ oven overnight [18].

D. Microchip Operation

For introducing liquids into the fluidic layer, both continuous oil-phase and dispersed aqueous-phase reagents were independently stored and driven from custom plastic containers [19]. The oil-phase and aqueous-phase liquids are light mineral oil with Span-80 surfactant at a low (i.e. 0.01%) and high (i.e. 1%) concentration (Sigma-Aldrich, St. Louis, MO) and deionized water with dissolved green food dye, respectively. The driving gas pressure applied on each oil-phase and aqueous-phase container was adjusted independently by a 0–10 psi range precise pressure regulator (Marsh Bellofram Co., Newell, WV). The

liquids were transported into the microchip using flexible Tygon microbore tubing (0.51 mm ID, 1.52 mm OD, Cole-Parmer, Vernon Hills, IL) connected to 22 gauge needle segments (Hamilton, USA) inserted in the fluidic access holes.

The elastomer membrane micro-valves in the control layer were also actuated by the valve manifold, using an Arduino microcontroller board to control the switching of each 3-way solenoid valve. Custom code was developed and executed to actuate the individual on-chip micro-valves. High-pressure nitrogen gas was controlled through a 5–50 psi range pressure regulator (McMaster-Carr, Elmhurst, IL) and connected to a solenoid valve manifold (Clippard Instrument Lab Inc., Cincinnati, OH). The solenoid valves were interfaced with the microchip through urethane tubing (0.06 inch ID, 0.13 inch OD, Clippard) and 22 gauge needles inserted into the pneumatic access holes. The experimental pressures of 20 psi were applied to the control micro-valves. The microchip was monitored under an inverted microscope (AZ100, Nikon Co) and the images were captured with a high-resolution camera (Digital Sight DS-Fi1, Nikon Co).

III. RESULTS AND DISCUSSION

To conduct the experiment, picoliter aqueous-phase droplets are generated at T-junction structure integrated with the pneumatic micro-valve to trigger the droplet generation as shown in [20–23]. The generated droplets are moved by the continuous oil-phase downstream and selected droplets are transported to the merging zone while undesired ones are shunted to the waste line. In the merging unit, the incoming droplets can stop and merge with each other in a controlled passive or active manner as discussed in the following sections.

A. Passive Merging

The proposed merging mechanism is in principle a microfluidic trap to accommodate and immobilize multiple droplets. The droplets are driven sequentially into the merging zone of the microfluidic trap to be merged. Within the merging zone, an incoming droplet collides with the previously stored droplet and the selected droplets may be merged passively by the geometry constrains of the merging zone. In the passive approach, the merging eventually happens when the thin oil film between two successive droplets are drained out naturally after a certain period of time. It was observed that for a low level of surfactant concentration in the continuous-phase (e.g. mineral oil + 0.01% surfactant Span 80), the droplets are merged immediately after collision. A recorded merging sequence of three droplets is shown in Fig. 2.

B. Active Merging

Depending on the experimental conditions, active merging may be desired, for example to control the simultaneous merging of surfactant-stabilized droplets. It was observed that for a high level of surfactant concentration in the continuous-phase (e.g. mineral oil + 1% surfactant Span 80), the fusion time significantly increases. To shorten the merging process time, active merging was tested in which the underlying membrane was fluctuated with a low frequency (e.g. 1 Hz). In this design, the micro-chamber is connected to the bypasses

through lateral capillaries which help to flush the continuous-phase oil at the event of membrane fluctuations. In this case, merging occurred shortly after initiating the fluctuations. Sequential images of trapping, stimulating, merging of three droplets, and then releasing are shown in Fig. 3. The

IV. CONCLUSION

Here, we presented the design and development of a microfluidic valve-based trap as a merging mechanism enabling the selected droplets to be merged, either passively or actively, to produce a combined droplet with desired volumes and compositions.

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Figure 1.

(a) Schematic diagram of the microchip including two layers of fluidic and control and two functional units of droplet generator and merging mechanism. (b) The detailed design of the microfluidic valve-based trap. (c) Cross-sectional view of the fluidic and control layers at a single membrane micro-valve.

Figure 2.
Sequential views of passive merging process of three droplet.

Figure 3.
Close view of trapping and physical stimulation of underlying pneumatically-actuated membrane (i.e. images 3 & 6) for an active merging of three droplets, and then realising.