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FAST AND FLEXIBLE DRUG SCREENING USING A PURE HYDRODYNAMIC DROPLET CONTROL

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KEY WORDS

Droplet-based microfluidics, drug screening, microfluidic networks, networked labs-on-chips, pure hydrodynamic droplet control

SHORT SUMMARY

In this work, we review the concept for a pure hydrodynamic droplet control in a microfludic bus network. With this approach, single or multiple droplets can be routed to a desired target. Moreover, multiple droplets can be routed to multiple targets, enabling parallel processing. In both cases, the destination address is encoded in the distance between the droplets. Afterwards, we apply this concept to realize a microfluidic design, which performs screening of different antibiotics and concentrations on lung cells infected with a bacteria. The resulting design allows for a fast and flexible drug screening and can be fabricated at low-cost.

EXTENDED ABSTRACT

Introduction

Droplet-based microfluidic systems [1] are a promising platform for the realization of *Labs-on-Chips* (LoCs). Two approaches have been established in the past: Two-phase flow microfluidics and digital microfluidics. In two-phase flow microfluidics, the droplets flow in closed channels inside an immiscible continuous phase, which is driven by a pressure or syringe pump at the chip boundary [2]. In digital microfluidics, the droplets are moved on a planar-surface using *Electrowetting On Dielectric* (EWOD) or *Dielectrophoresis* (DEP) [3]. However, digital microfluidics require a complex and costly fabrication process and lack in biocompatibility for some biological settings [4]. Hence, in this work, we focus on two-phase flow microfluidics, which exploit hydrodynamic effects to control the droplets and, thus, can be fabricated at low cost. The main design parameters are the channel geometries and the hydrodynamic forces (e.g., the volumetric flow rate).
Recently, microfluidic networks for two-phase flow microfluidics [5, 6, 7, 8] as well as corresponding design methods [9, 10, 11] have been introduced as new research field, which aims at realizing programmable and flexible LoC devices. In particular, microfluidic networking targets to dynamically assign the droplets' path through a microfluidic network in order to perform specific analyzes. In this work, we present a microfluidic design, which performs fast and flexible drug screening based on a pure hydrodynamic droplet control. To this end, we consider a microfluidic network with a bus topology as sketched in Fig. 1, which consists of a cascade of microfluidic control units (*switches*), each connected to a LoC device. The microfluidic switches control the path of the droplets and the LoC devices perform the drug screening.
In the following, we will first introduce the droplet control concept and then we use this concept for a microfluidic design, which is able to perform fast and flexible drug screening.

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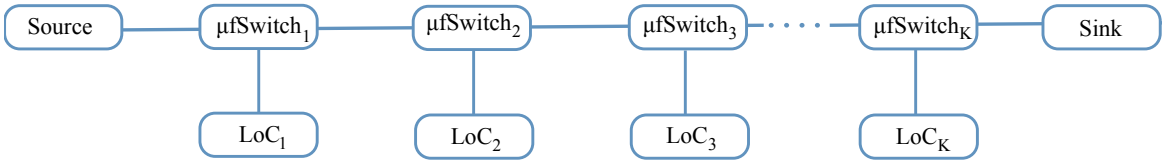
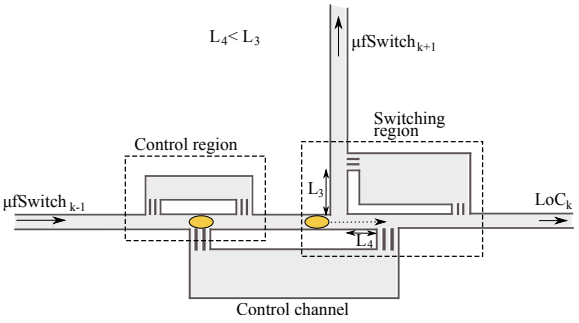


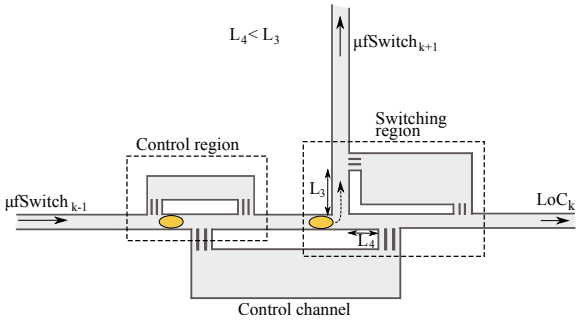
Figure 1: Microfluidic network with bus topology.

Hydrodynamic Droplet Control

The realization of the microfluidic switch is shown in Fig. 2 [8]. It can be used to route single or multiple droplets (payload droplets) to a target LoC device. Payload droplets contain chemical or biological samples to be processed. To route K payload droplets to a certain destination, a single signaling droplet (trailer droplet) must be inserted after the payload droplets. The trailer droplet is only used for signaling and contains no sample. The address of the target LoC is encoded in the distance between the individual droplets*, where the droplets are equally spaced. The switch has a control region and a switching region. The switching region contains an asymmetric junction with branches 3 and 4 of length L_3 and L_4, respectively. In order to ensure that the hydrodynamic resistance of branch 4 is lower than of branch 3 their lengths are chosen such that $L_4 < L_3$. Branch 4 is connected to a particular LoC device (LoC_k) and branch 3 to the next microfluidic switch ($\mu\mathrm{fSwitch}_{k+1}$), respectively. Both branches are connected through a bypass channel to equalize the pressure between the connecting points, which makes the switching behavior independent of the remaining network [13].



(a) Control channel is clogged: Droplet flows towards the kth LoC device.



(b) Control channel is not clogged: Droplet flows towards the next switch.

Figure 2: Microfluidic switch.

The basic idea is that a droplet in the control region controls the path of a droplet currently in the switching region. More precisely, the switch toggles between two states depending on the presence of the droplet in the control region. A droplet arriving at a switching region flows into branch 4, if there is a droplet in the control region that clogs the flow through the control channel (ON state). This is because branch 4 has a lower hydrodynamic resistance than branch 3. On the other hand, a droplet arriving at the switching region flows into branch 3, if there is no droplet in the control region or it does not clog the control channel (OFF state). The reason for this behavior is that the flow through the control channel acts as a block for the incoming droplets. So to route K payload droplets to the kth LoC device, an equally spaced droplet train consisting of K payload droplets and a single trailer droplet must be generated. The distance must be set so that the kth switch is in the ON state for each droplet arriving at the switching region. The last payload droplet is correctly routed by the trailer droplet. It is important to note that this switch can also be used to route a train of payload droplets to different LoC devices. This can be accomplished through different distances between the individual droplets, where each distance indicates the target LoC device.

* Note that a similar addressing scheme has previously been proposed in [12].

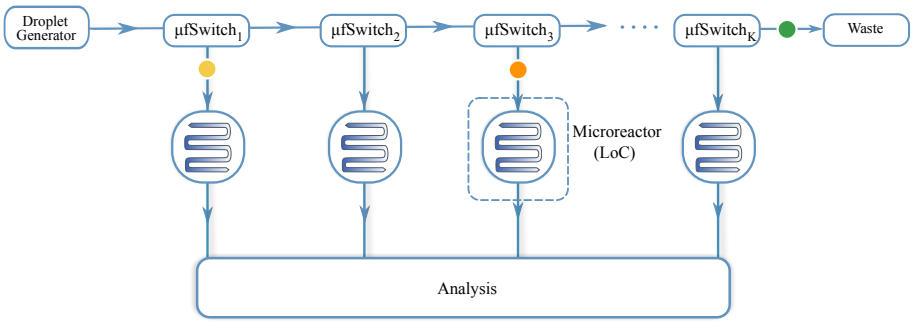


Figure 3: Microfluidic design for fast and flexible drug screening.

Fast and Flexible Drug Screening

Human lung infections can be caused by various bacteria and if not treated properly, and on time, can leave severe damages [14]. However, pharmaceutical industry lacks insight into which bacteria are antibiotic resistant, or what exact amount of antibiotics is required for treating specific bacteria. Standard laboratory procedures are time demanding, expensive and lack in flexibility. For this reason, it is essential to have a platform, which allows fast delivery of different antibiotics and concentrations to the infected lung cells and allows to observe the reaction. The concept reviewed above allows to realize a microfluidic design, which serves as a disease model for human lung infections and is able to perform fast and flexible drug screening.
The design, as shown in Fig. 3, includes a droplet generator, microfluidic switches, microreactors and an analysis stage. The droplet generator is able to generate droplets at a desired frequency. For the purpose of drug screening, different antibiotics or different antibiotic concentrations are isolated inside the payload droplet (e.g., water droplet), which are carried by a continuous phase (e.g., oil). The microfluidic switch is realized as described above and it directs the payload droplets with antibiotics towards the desired microreactor. It can route single/multiple payload droplets to a certain microreactor and multiple payload droplets to different microreactors using only a single trailer droplet (as sketched in Fig. 4). It is important to note that the routing of multiple droplets to different microreactors, enables fast delivery and, thus, allows for fast drug screening (parallelization).
The microreactor is shown in Fig. 5. We assume that *Human Lung Epithelial* (HPAEpiC) cells, lining more than 99% of the internal surface area of human lungs [15], are infected with the *Acinetobacter baumanii* bacteria* and incubated inside the microreactor. The *A. baumanii* [16] is thought to be a multidrug-resistant bacteria, and correct drug treatments are still needed in order to fight this bacteria. Therefore, lung cells are held in place by electrospun polymer nanofibers. Once a payload droplet enters the microreactor, the antibiotics inside the droplet initiate a biochemical reaction with the infected lung cells incubated inside the microreactor (as sketched in Fig. 5). Since the droplet volume is very small, antibiotics inside the droplets are able to initiate the reaction with lung cells and bacteria quickly and efficiently. The result of this reaction can be either observed inside the microreactor or at the analysis stage, using, e.g., high resolution microscopy tools. The proposed microfluidic design can be fabricated at low cost, since no active elements are required on the chip.

Practical Considerations

The droplets are generated inside a T-junction. In order to guarantee a stable droplet formation it is recommended that the width, w_d, of the dispersed phase channel is smaller than the width, w_c, of the continuous phase channel and that a low capillary number ($C_a << 1$) is ensured [7]. The latter condition depends on the channel geometries and the input flow rates, which should be in the range $2 - 20\,\mu$L/min. Moreover, it is recommended that the dispersed flow rate, Q_d, is lower than the continuous flow rate, Q_c, to achieve an optimal polydispersity index. We ensure rectangular channels by satisfying the condition $h < w_c$, where h denotes the uniform height of the channels. The proposed design can be fabricated in *Polydimethylsiloxane* (PDMS) polymer using the standard soft lithography process.

* It is also possible to include different cells and bacteria.

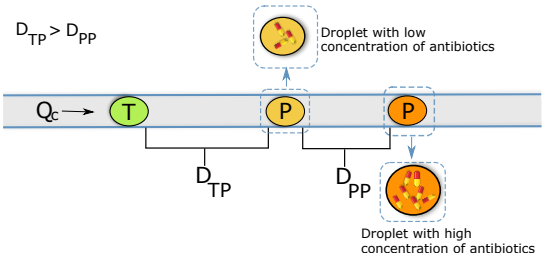


Figure 4: Droplet train with payload droplets carrying different concentrations of antibiotics and a single trailer droplet.

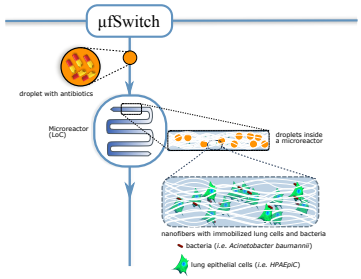


Figure 5: Microreactor with immobilized human lung cells infected by bacteria

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