Microfluidics combinatorial minilaboratories

Author: Krzysztof Churski
Supervisor: Piotr Garstecki

We report a droplet microfluidic system that accepts small liquid samples deposited directly on chip and automatically forms a sequence of droplets containing mixtures of these samples. This system i) minimizes consumption of reagents: in an exemplary demonstration we used 50 µL of suspension of bacteria and ~200 µL of three buffer solutions, ii) provides sterile conditions and avoids the flow of samples through the valves, and iii) provides the means for a rapid screen of a wide range of conditions of reaction or incubation.

Droplet microfluidic systems have been demonstrated to support bacterial cultures with the potential advantages in running several cultures in parallel in small and precisely metered volumes. Development of automated microfluidic systems for rapid screens of conditions of growth largely remains a challenge. The system that we finally present uses our earlier result on a system for high-throughput screening of compositions of droplets and introduces i) full automation of the high-throughput screen within a simple architecture of 8 external valves and ii) facile world-to-chip interface that allows to deposit samples directly on chip from a pipette and avoids transition of the solutions through the valves.

In an exemplary application we use the system to determine the minimal inhibitory concentration (MIC) of three antibiotics (ampicillin, chloramphenicol and tetracycline) on *Escherichia coli* and the interactions between the pairs of these compounds.

Our microfluidic system comprising i) ports and reservoirs for deposition of samples, ii) externally controlled T-junction droplet generators, iii) the merging chamber for formation of the incubation mixtures and iv) an outlet channel. We verified the precision of generation of compositions using fluorescent dyes. In the first set of experiments we deposited a suspension of *E.coli* (ATCC25922), two solutions (concentrated and diluted) of the antibiotic in the growth medium (LB) and neat medium. After deposition of samples and sealing the ports, a LabView routine executes a protocol that forms (at 0.5 Hz) a sequence of 49 or 121 (~2 µL) droplets containing different concentrations of the antibiotic and transfers it into a polyethylene tubing for 180 minutes of incubation. Then the sequence is purged through a spectrophotometer that reads out (at ~1 Hz) the intensity of fluorescence from a marker of metabolism (resorufin) to determine the MIC. We then used the same system to form sequences of droplets representing a range of combinations of concentrations of two antibiotics, revealing synergistic, additive and buffer interactions between them.

The system that we present allows to automatically generate a sequence of incubation mixtures within a very short time (~a minute) from small samples of liquid (~tens of microliters) without passing the solutions through valves and within a simple chip that comprises only channels and can be inexpensively manufactured. We believe that this system may provide an attractive platform for rapid screening of reaction conditions in chemical synthesis and of growth conditions in microbiology.