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Micro/Nano/Bio
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Homework Micro-scale Engineering #4, Due Date: February 20, 2014

1. Please read the paper on "Ultrahigh-throughput screening in drop-based microfluidics for directed evolution," and fill in right answers of the following statements taken from the paper. To save time, you only need to read the first two pages (pp. 4004-4005), Fig. 2, and Materials and Methods on page 4008. (7 points)

- To achieve full control and flexibility, current state-of-the-art high-throughput screening methods employ microtiter plates and sophisticated **robots** to process up to 100,000 assays a day, or ~1/s.

- Compared to state-of-the-art robotic screening, this is **1000**-fold faster and uses **10-million**-fold less volume of reagent, representing a cost savings estimated to be about 10-million-fold.

- (Fig. 1) A suspension of yeast cells displaying the HRP on their surface **(aq1)** is combined with a second aqueous stream containing the fluorogenic substrate **AUR** (aq2).

- (Fig. 2). The wild-type HRP gene is encoded on a plasmid as a C-terminal fusion to the **Aga2** gene to allow surface display ……create two libraries for each generation. Each library has **~10^7** variants …… each cell displays on its surface **~10,000** copies of a single mutant HRP protein (μHRP).

- (Materials and Methods) We design **electrodes** as channels in the **PDMS** device and fill them with a low melting point metal alloy.

- The drop-making geometry and flow rates have both been optimized to produce a high volume fraction (67%) emulsion of **23** μm drops at a rate of 2 kHz. This is achieved by flowing each of the three streams (two aqueous and one oil) at 20 μLh^{-1} through a **10 μm** square nozzle.

- The relative fluidic resistance of the top and bottom branches is set by the **length** of the narrow part of the channels at the sorting junction (Fig. 1).

- The sorted drops move up the field gradient created by the electrodes by **dielectrophoresis** (18) and are pulled into the keep channel.

2. In addition to the above-mentioned paper, we also studied "High-throughput microfluidic single-cell RT-qPCR." Both microfluidic devices demonstrate how to manipulate single cells. Please conduct a literature search and find another microfluidic device with single-cell manipulation. Write one paragraph to explain one single-cell manipulation feature of this device that is different from those covered by the two papers studied. Provide the link of the paper or the web site of the device. (3 points)

In the article "Microfluidics Technology for Manipulation and Analysis of Biological Cells," the primary method of cell manipulation for their miniaturized cell manipulator is achieved through their use of magnetic particles that selectively attach to cells (super-paramagnetic beads that are 10-100 nm). Due to the beads small size, they do not affect the cell's function, thus permitting the manipulation of certain rare cell types. This separation and manipulation is performed through the application of a magnetic field gradient, which captures the bead that is attached to the cell, effectively trapping the cell as well. Once this cell is captured, optical tweezers can be used in order to position the cell in a specific channel within the microfluidic device.

Citation:

Yi, Changqing, Cheuk Wing Li, Shenglin Ji, and Mengsu Yang. "Science Direct." *Microfluidics Technology for Manipulation and Analysis of Biological Cells*. (2006): n. page. Web. 14 Feb. 2014. <http://research.vuse.vanderbilt.edu/srdesign/2006/group19/Microfluidicstechnologyformanipulationandanalysisofbiologicalcells.pdf>.