Fluorescence Detection on a Droplet-Based Microfluidic Device

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In droplet-based microfluidics, liquid droplets in contact with hydrophobic dielectric surfaces are moved, combined, and/or split. This is done by applying AC or DC potentials across electrodes patterned beneath the dielectric layers on the microfluidic devices. These devices can be used for a variety of applications on the micro-scale. Microfluidics can be utilized in proteomics, fluorescence detection, and other micro-scale reactions. Fluorescence assays on these devices can be employed as an efficient and easy technique for micro-scale sample analysis or diagnostic tests, where samples or reagents are limited. The goal of this research is to demonstrate real-time monitoring of reactions on a droplet-based microfluidic device. Optimization and calibration experiments were done to test the efficacy of a new experimental apparatus for fluorescence detection. Fluo-3, a calcium-dependent fluorescent indicator, was mixed with varying concentrations of calcium. Experiments showed that there was a reproducible linear trend for measured fluorescence intensity as a function of calcium concentrations in the range of 0µM to 0.225µM. Dynamic fluorescence intensity changes as a function of calcium concentration were then measured with fluorescence quenching experiments. Changes were detected when a droplet containing calcium was merged with a droplet containing Fluo-3, and then quenched with a droplet containing EDTA. The final stage of the project was to use the fluorescence detection to quantify protein degradation rates using dye-quenched proteins that fluoresce upon digestion. Fluorescently labeled casein was digested with proteinase K, and fluorescence intensity was measured as a function of time. These experiments show that reactions can be performed and monitored using droplet-based microfluidics.