Applying Droplet Fluidics to Mycobacterial Drug Discovery

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Mycobacterium tuberculosis is the causative agent of the human pulmonary infection tuberculosis (TB). Drug resistance is a growing issue that threatens TB care worldwide. It is estimated that a third of the global population has an asymptomatic, non-replicating persistent, infection which can rapidly progress to a difficult to treat drug resistant active infection upon immunosuppression. Currently, there are no sufficient *in vitro* models which capably mimic all physiological conditions of non-replicating persistent infections. There is an urgent need to design drug screening platforms which reduce animal models and replicate the human infection environment. Droplet microfluidics have promising future applications for bioengineered models and the miniaturisation of mycobacterial antibiotic susceptibility testing.

Water-in-oil monodisperse droplets were generated using a pressure-driven device and droplet generating chips made from PDMS following standard soft lithography procedures. Two immiscible phases of oil and aqueous bacterial culture are pumped into the chip by pressurised air controlled by an imaging feedback system.

Stable monodisperse droplets were generated at the pico-litre scale with no significant difference in droplet volume or diameter over time when either dH_20, nutrient rich media, or cholesterol minimal media was used as the aqueous phase.

The pressurised feedback device used is sufficient for generating monodisperse and stable droplets despite the presence of detergents such as polysorbate 80 and tyloxapol in culture media. This ensures biological assays using droplets will have monodisperse volumes and each droplet can be an experimental replicate for drug screening. Future work includes encapsulating mycobacteria in droplets and studying proliferation dynamics.