

Supplementary Information for

Multiphasic droplet microfluidics platform for controlled bacteria and mammalian cell coculture

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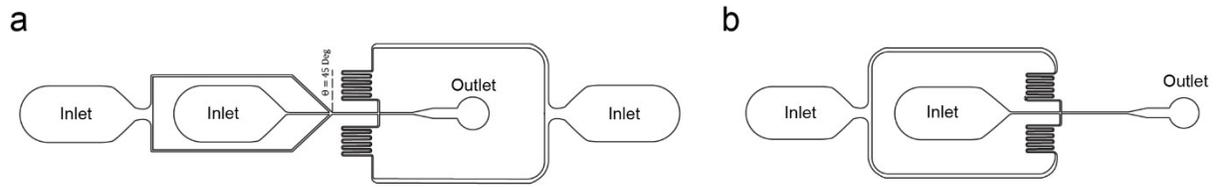


Figure S1. Design of microfluidic chips for phase separation-based co-encapsulation **(a)** and sequential encapsulation **(b)**.

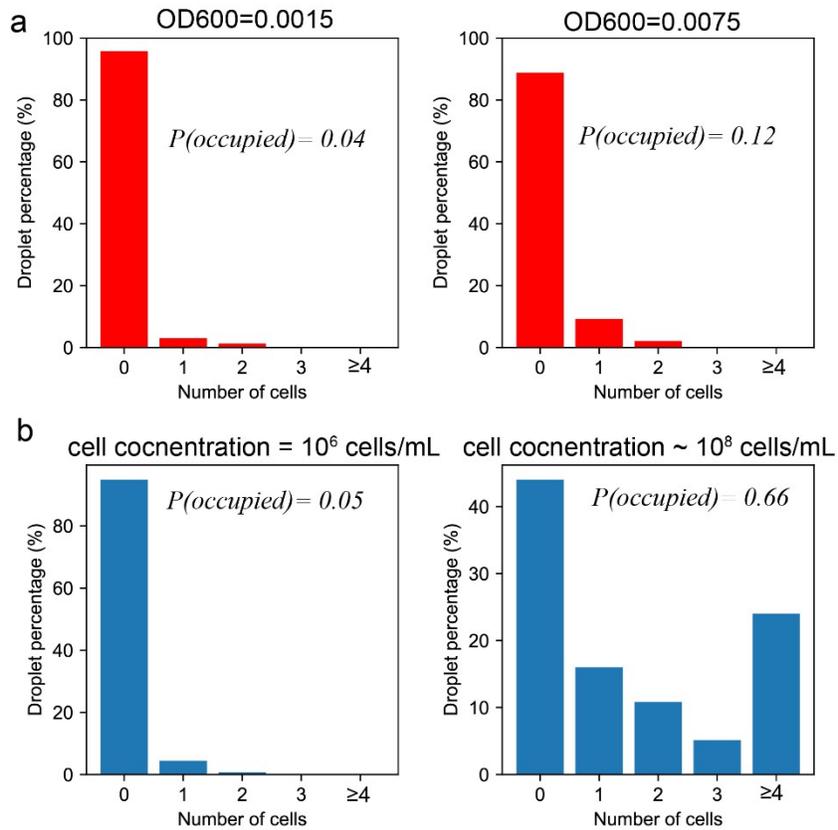


Figure S2. Droplet occupancy for bacteria and human cells. (a) the percentage of droplets containing 0,1,2,3, and $4 \leq$ bacterial cells for two samples that were generated via co-flow focusing device. The left plot shows the droplet occupancy percentage for an input bacterial concentration in the dextran phase equivalent to an optical density of 0.0015. The right plot shows the same information for another sample with an optical density of 0.0075. **(b)** Droplet occupancy percentage for human cells in core-shell droplets. The concentration of human cells in the channel is 1 million cells/mL (left) and ~ 15 -20 million cells/mL (right). Note that the right sample flown to generate one compartment droplets.

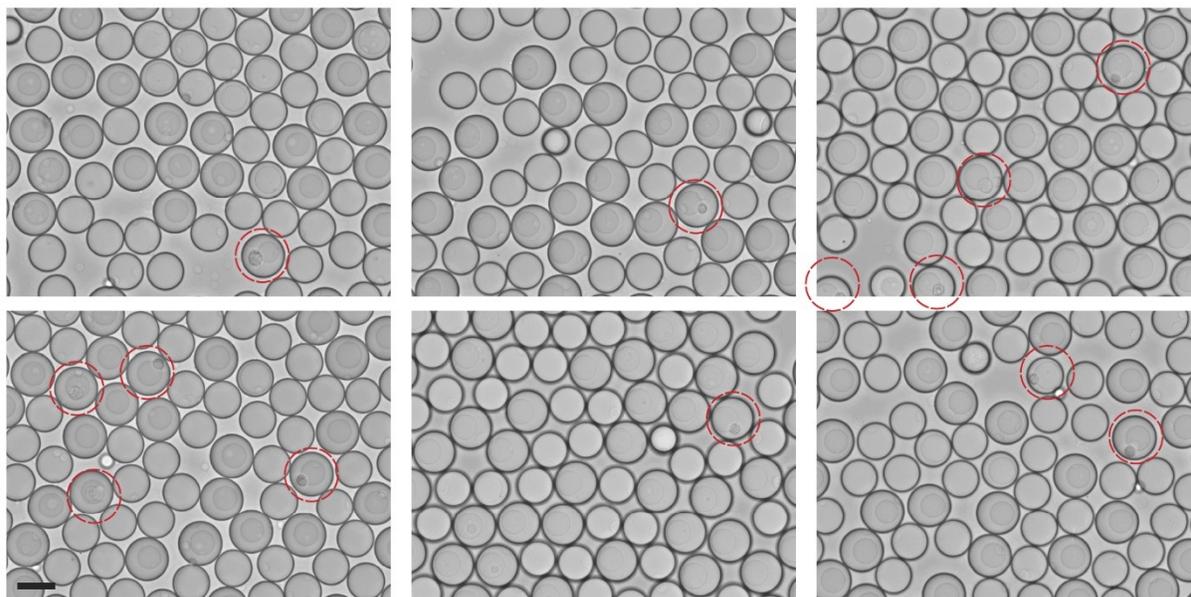


Figure S3. Encapsulation of cells into microdroplets. Bright field images showing the frequency of cell-laden multiphasic droplets as highlighted by the red circles. The fraction of cell-laden MMDs can be increased by increasing the initial cell concentration in the gel precursor solution prior to encapsulation. Scale bar is 50 μm . The shown images are representative of a larger set of images of the sample.

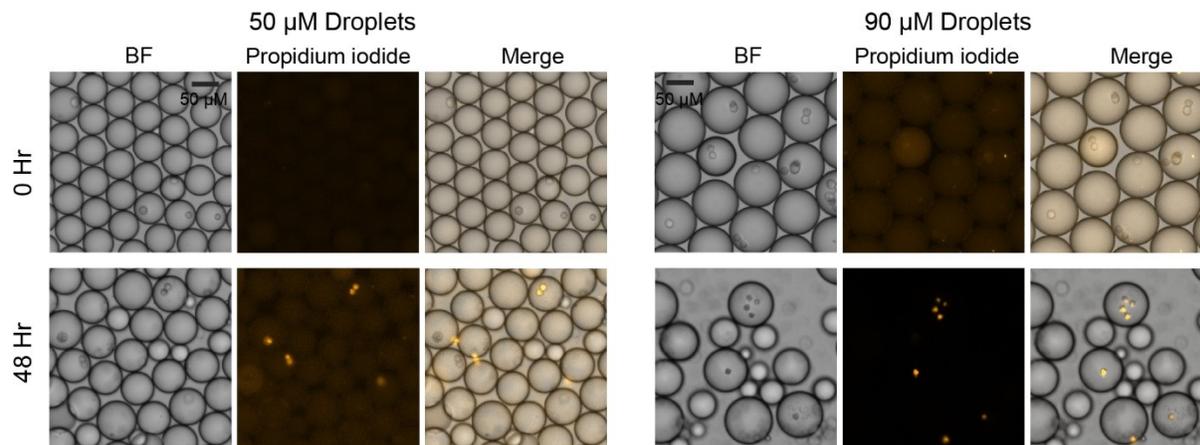


Figure S4. Cell viability in microfluidic droplets. Representative images of cells stained with propidium iodide to assess cell viability. The bright field and fluorescent images are of droplets suspended in oil containing cells at different incubation times (0 and 48 Hours). Two sets of droplets are shown: 50 μm and 90 μm. Dead cells show high fluorescence intensity of propidium iodide, while live cells show negligible intensity. Scale bars are 50 μm.

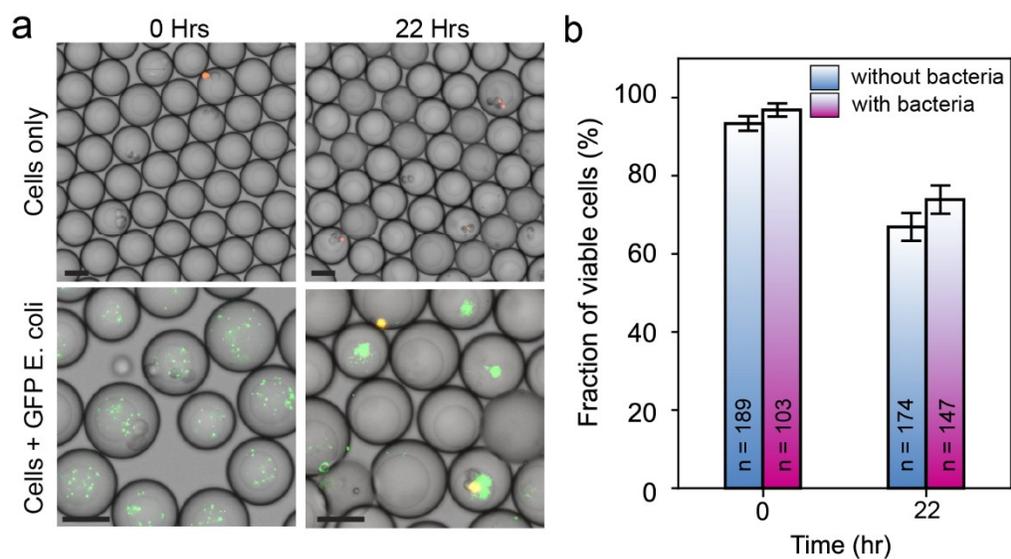


Figure S5. Cell viability in presence and absence of *E. coli*. (a) Representative images of cells stained with propidium iodide (orange) to assess cell viability in presence and absence of GFP-labeled *E. coli* (green). Two time points are shown at 0 Hrs and 22 Hrs. Scale bars are 50 μm . (b) Bar plot showing the fraction of viable cells at 0 Hrs and 22 Hrs within multiphase droplets in the presence and absence of *E. coli* cells. The fraction of viable cells was calculated by dividing the number of cells not stained with propidium iodide (dead cell stain) by the total cell count for each imaged sample.

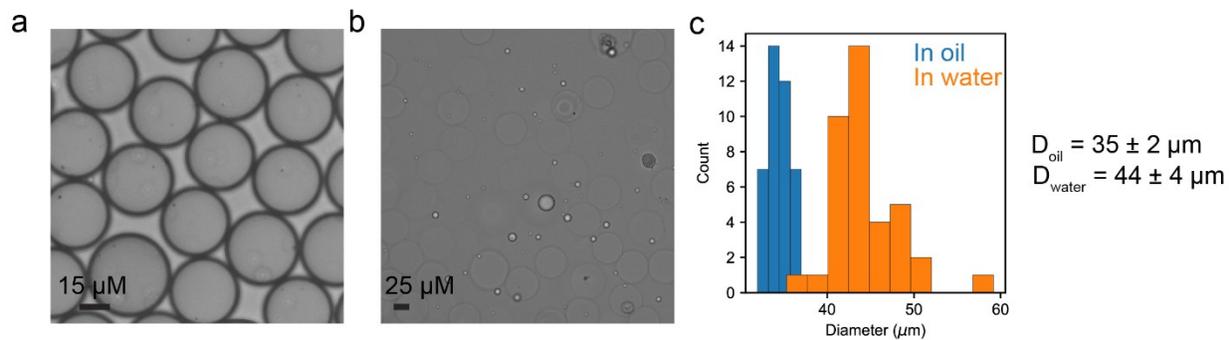


Figure S6. Effect of water hydration on the swelling of PEG-based hydrogel particles. (a) PEG hydrogels suspended in oil. **(b)** PEG hydrogel droplets suspended in aqueous media. **(c)** histogram of measured diameters of PEG hydrogel particles in oil and in aqueous media.