Electronic Supplementary Material (ESI) for Lab on a Chip.

Sorting by Interfacial Tension (SIFT): Label-Free Selection of Live Cells Based on Single-Cell Metabolism

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Supplemental Information

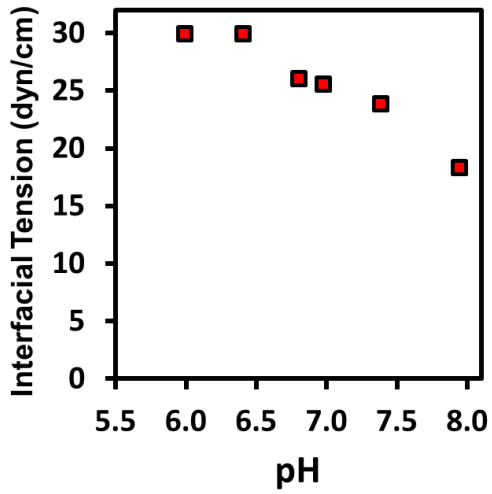
Table of Contents:

Video Caption:

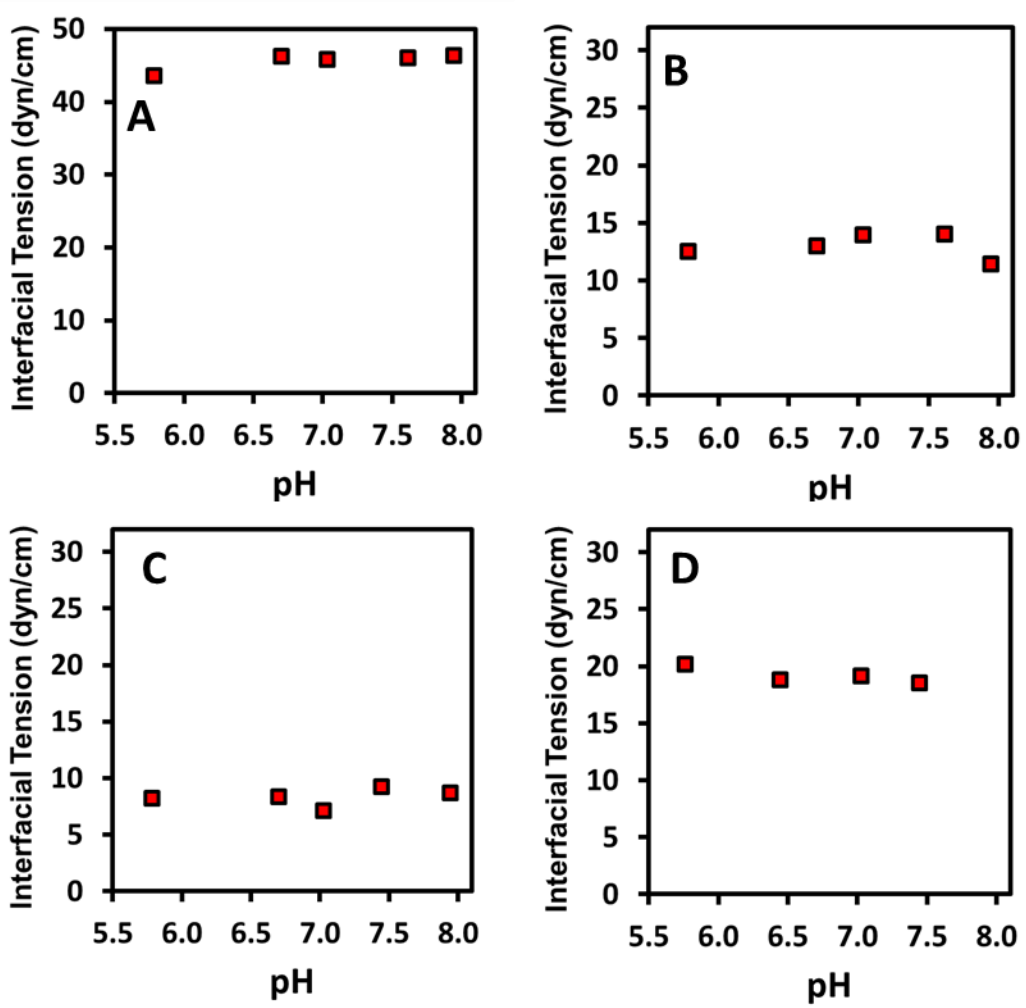
Video S-1. Sorting of droplets of different pH by SIFT. Oil Flow is from left to right. Droplets enter from bottom left of image. White droplets at higher pH (pH = 7.48) are only slightly deflected by the rail. Clear droplets at lower pH (pH = 7.01) of higher interfacial tension follow the rail upwards. A small amount of fluorescein is added to the droplets at pH = 6.90 to identify the two droplet populations. Video speed is 10 times slower than actual run speed.

Video S-2. Sorting of droplets containing cells. Droplets that do not contain cells are only slightly deflected by the sorting rail and exit the rail near the bottom. Droplets containing a cell ride the rail laterally up and leave the rail near the top. Cells have been labelled with a fluorescent viability marker. Fluorescent excitation is localized in a hexagonal central region of the imaging field and thus cells appear non-fluorescent in the periphery of the imaging field. Video speed is 10 times slower than actual run speed.

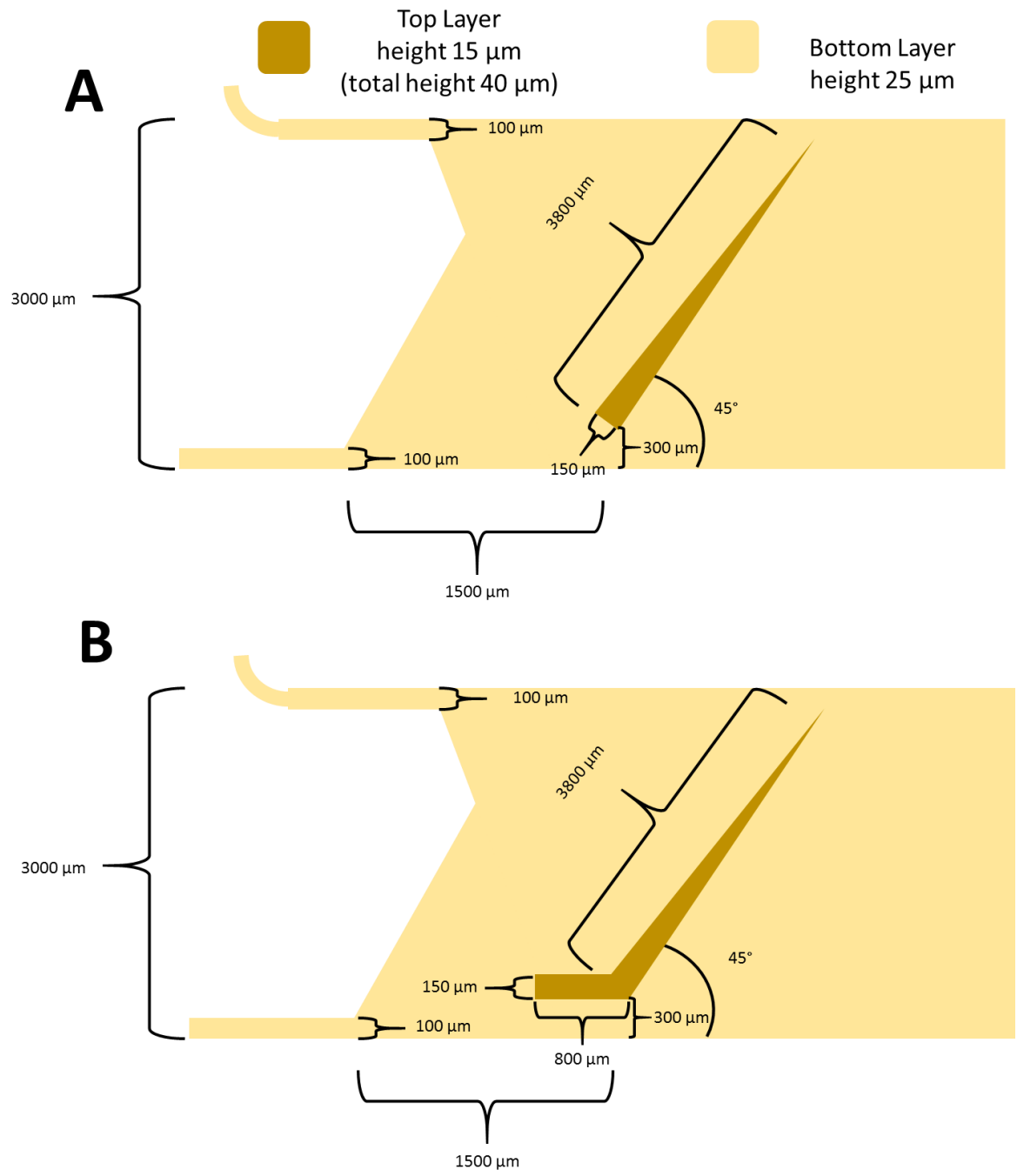
Video S-3. Sorting of droplets containing live and dead cells. Most droplets contain no cells and are only slightly deflected by the sorting rail and exit at the bottom of the rail. A droplet containing a live cell (labelled with fluorescent viability marker) rides the rail laterally up and leaves the rail near the top. This is followed soon after by a droplet containing a dead cell that exits the rail near the bottom. Fluorescent excitation is localized in the central region of imaging field and thus all cells appear non-fluorescent in the periphery of the imaging field. Video speed is 10 times slower than actual run speed.



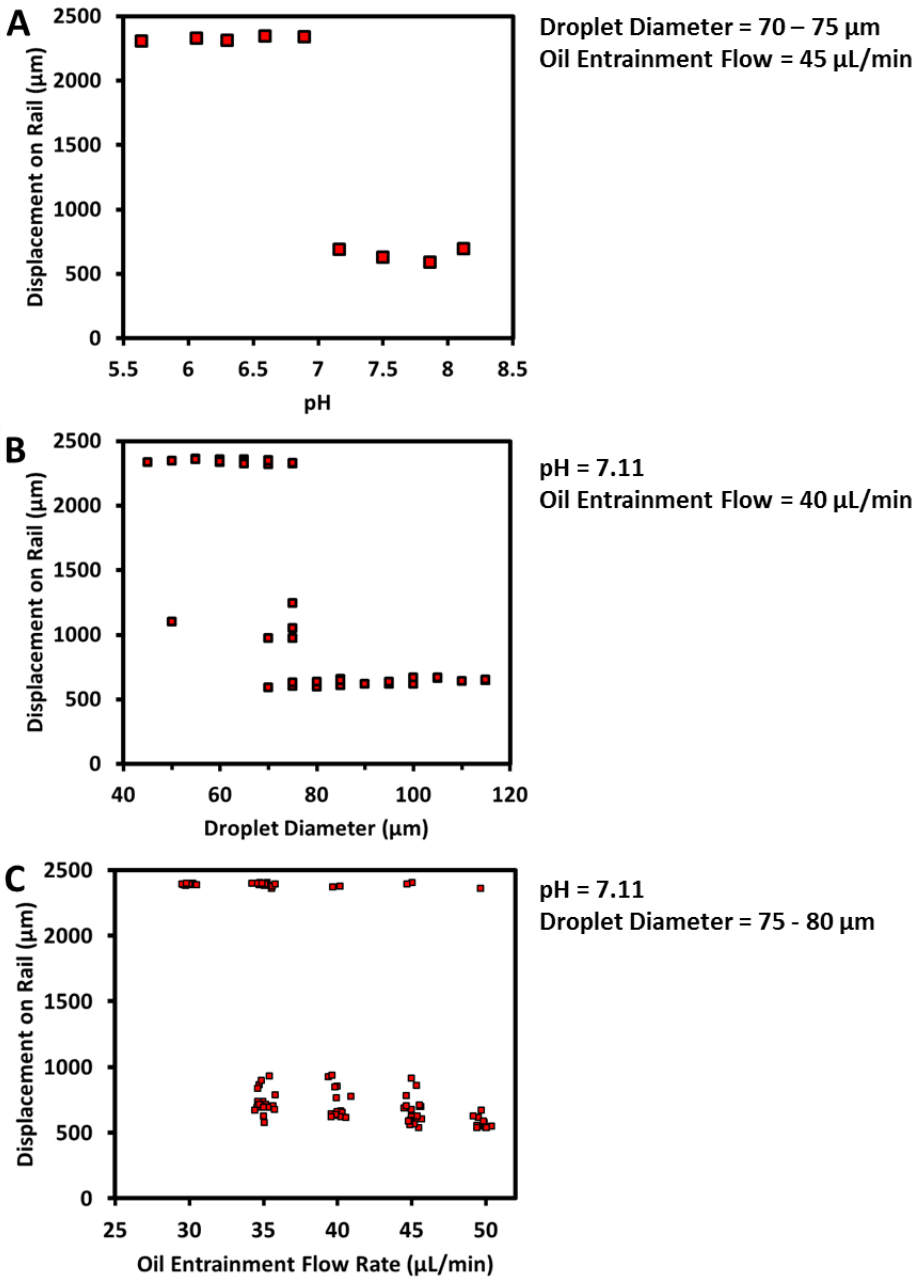
Supplemental Figure S1. a) Interfacial Tension at 37 °C for buffer droplets (137 mM NaCl 10mM Phosphate) in QX100 diluted 100 fold by mass with Novec 7500.



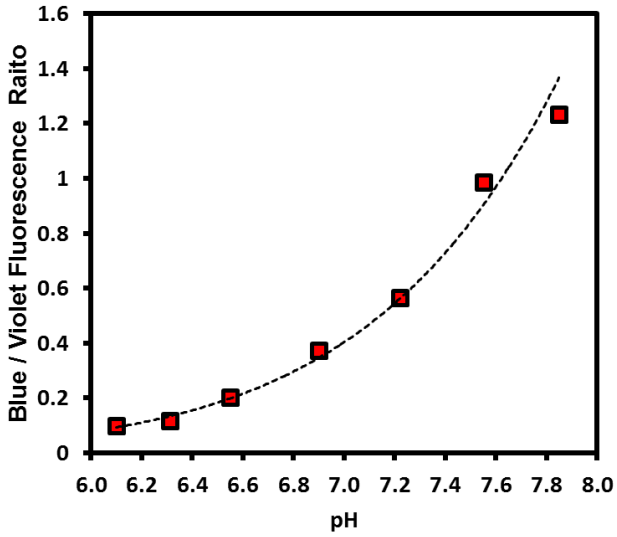
Supplemental Figure S2. Interfacial Tension vs. pH at room temperature for aqueous buffer droplets (137 mM NaCl 10mM Phosphate) in **a)** Novec 7500 **b)** 0.02% w/w 008 fluorosurfactant in Novec 7500 **c)** 0.002% w/w Picosurf 1 in Novec 7500 **d)** 0.002% w/w Picosurf 2 in Novec 7500.



Supplemental Figure S3. Sorting rail dimensions and position for two different rail geometries **a)** straight **b)** with horizontal section. Exact position of rail is approximate as layers are positioned by eye.



Supplemental Figure S4. Displacement on rail of droplets in the vertical direction. Droplet composition is identical to those used in cellular experiments. Zero displacement is defined as the bottom of the rail. Fixed experimental parameters are provided to the right of each graph. **a)** Displacement on rail of a train of droplets as a function of pH. **b)** Displacement on rail of individual droplets as a function of droplet diameter **c)** Displacement on rail of individual droplets as a function of oil entrainment flow rate at the discrete flow rates of 30, 35, 40, 45, 50 µL/min.



Supplemental Figure S5. Blue to violet fluorescence intensity ratio of droplets containing the pH sensitive ratiometric fluorescent probe, pyranine, as a function of pH. Droplets of known pH were produced in the microfluidic device under the same solution and temperature conditions as cellular experiments. Fluorescence intensity, average fluorescence of droplets in image, was determined for subsequent excitation with violet (395 nm) and blue (440 nm) light. For the purposes of determining pH of droplets containing cells, the ratio was fit to a power series (no physical model is associated to this fit). The calibration curve was found to be consistent across experimentation days.

Supplemental Table S1. Typical flow parameters used in experiments. Channel geometry is provided below for reference.

Inlets and Outlets	Flow Rates (µL/min)
Aqueous Inlet (only one inlet used)	0.1 - 0.5
Oil Inlet	3 – 5
QX100 Inlet	10 – 15
Oil Entrainment Inlet	40 - 60
Oil Outlet	- 2.5 to -4.5 (NEGATIVE FLOW)
Oil and Droplet Outlet	-0.25 to -0.5 (NEGATIVE FLOW)