## **Supporting Information**

## Surface Enhanced Raman Scattering (SERS) Based Microfluidics for Single Cell Analysis

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<u>Instrument</u>	<u>Setup</u>	<u>Objective</u>	<u>NA</u>	<u>Wavelength</u>	<u>δx (μm)</u>	<u>δy (μm)</u>	<u>δz (μm)</u>	Volume (µm <sup>3</sup> )
Renishaw	Inverted	20	0.40	633	0.97	0.97	7.91	7.37
Renishaw	Inverted	50	0.75	633	0.51	0.51	2.25	0.60
WITec	Upright	10	0.3	633	1.29	1.29	14.1	23.30
WITec	Upright	20	0.4	633	0.97	0.97	7.9	7.37
WITec	Upright	50	0.7	633	0.55	0.55	2.6	0.79

Table S1. Microscope Objective Descriptive Details



**Figure S1**. Optical image of a single PC3 cell encapsulate in a droplet. The red box corresponds to the XY mapping area and the blue line corresponds to the location of XZ map (A). XY SERS map represented by the red box processed in Project FOUR 4.1 (B). Raw XZ SERS map represented by the red box box processed in Project FOUR 4.1 (C).



**Figure S2**. Demonstration of the efficacy of Matlab (A & C) baseline correction processing and map rendering by direct comparison to proprietary Wire 4.2 (B & D) processing. A and B show the rendered SERS maps scaled with same look up table (LUT). Image B inset shows the data from a single pixel and C and D show the spectra from the representative pixel. Images C and D show minor variation in the peak of interest as expected with different baseline correction algorithms.

mages processed in mages and Manab	<b>Coefficient of Variation</b>			
Experiment	Image J	Matlab		
1	0.42	0.40		
2	0.49	0.36		
3	0.50	0.28		
4	0.45	0.35		

Table S2. Coefficient of variation for the SERS maps within each experiment as calculated for the images processed in ImageJ and Matlab

Table S3. Coefficient of variation	for the fluorescence images as calculated in ImageJ
	<b>Coefficient of Variation</b>

Experiment	Image J
Experiment A – Replicate 1	0.51
Experiment A – Replicate 2	0.56
Experiment A – Replicate 3	0.42
Experiment B – Replicate 1	0.55
Experiment B – Replicate 2	0.46



**Figure S3**. Fluorescence Images of PC3 cell tagged with wheat germ agglutin (WGA) functionalized with fluorescein (FITC; Sigma,UK and Sigma, US). A) Large view image of tagged PC3 cells adhered to a 96-well plate. B) Fluorescence image of single cell encapsulation in the optofluidic platform. C) Image of a single cell encapsulation in the optofluidic device with the fluorescence overlaid on the bright field image.