

## Supplementary Information

Video S1 System validation under low flow speed.

Video S2 Cells separated by the channel.

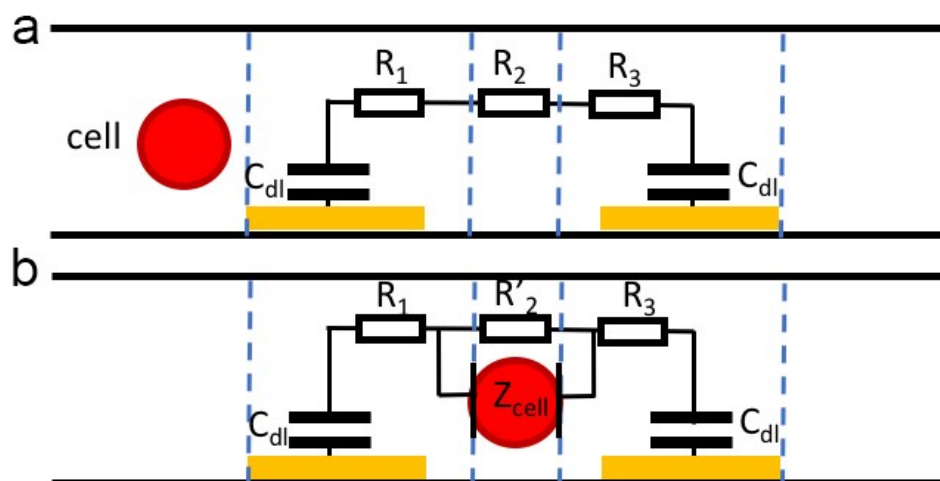
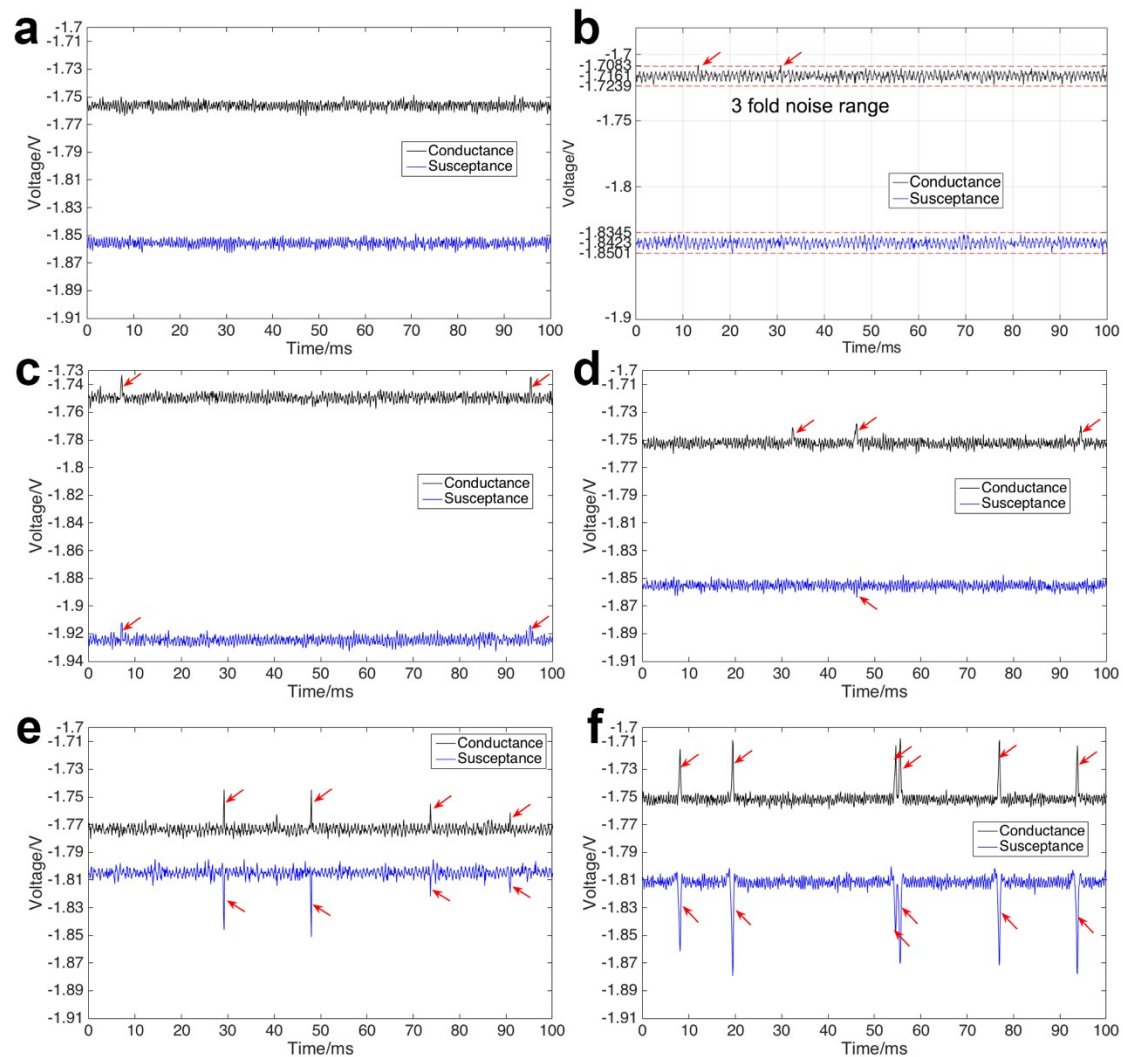


Figure S1 Electric model of the microfluidic channel (a) and a cell pass through the channel (b).

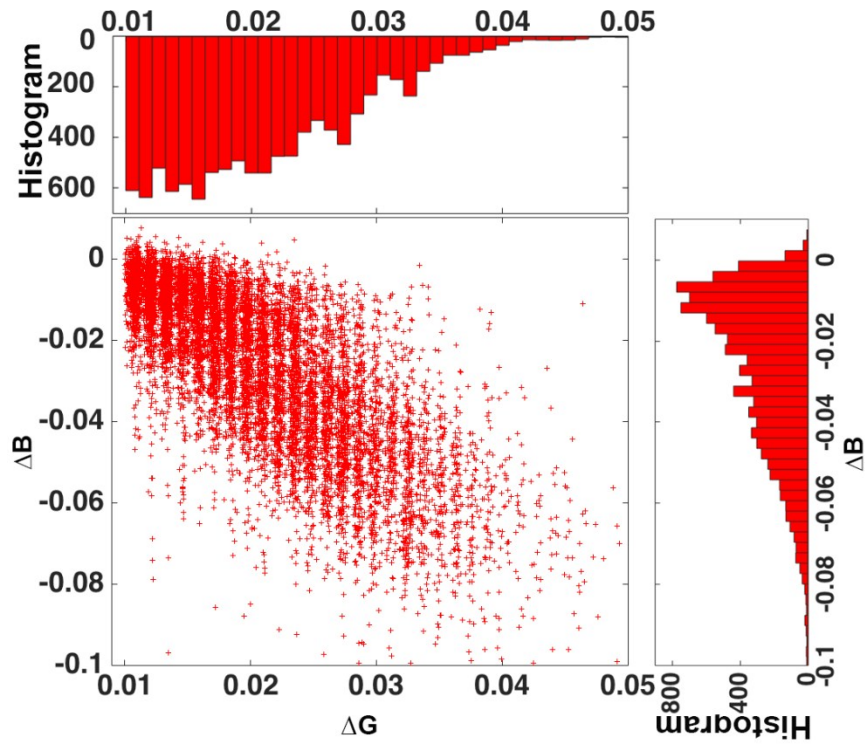
$C_{dl}$ ,  $R_{1-3}$ , and  $Z_{cell}$  represent electrode-solution interface capacitance, solution resistance of different parts ( $R_1$  and  $R_3$ , the constriction part of the channel;  $R_2$ , the narrowest part), and impedance of cell, respectively.



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12 Figure S2. Raw signals of different cells/particles. Red arrows indicate the pulses of the  
 13 cells/beads (channel height = 12 µm and excitation amplitude = 0.30 V). Impedance  
 14 signals (conductance and susceptance) without any particles (a), or with Ø5-µm polymer  
 15 beads (b), Ø10-µm polymer beads (c), necrotic HeLa cells (d), live Jurkat cells (e), and  
 16 live HeLa cells (f) passing through the detection area.

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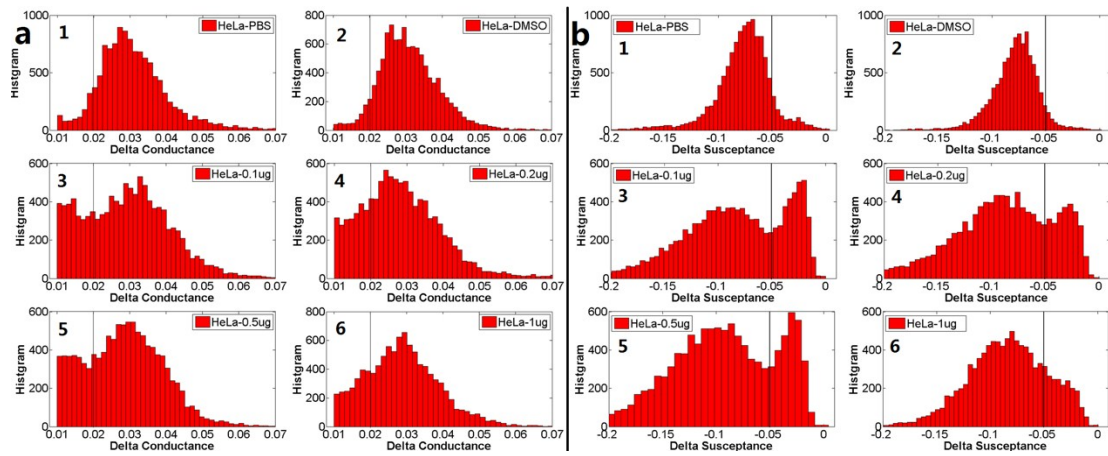


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19 Figure S3. The scatter and histogram plot of Jurkat cells

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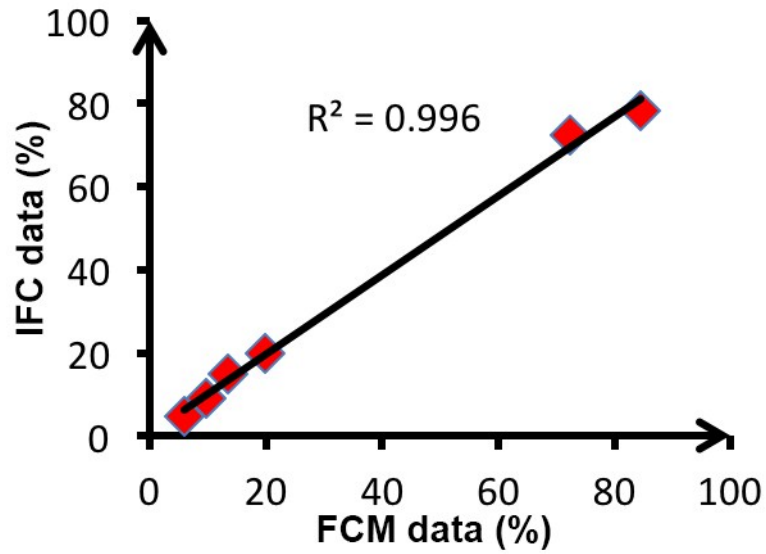


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23 Figure S4. Histograms of six samples. Black lines indicate the threshold for conductance

24 (a) and susceptance (b).

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27 Figure S5. Comparison of viability tests by IFC and traditional FCM.

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29 Table S1. Literature summary of IFC chip researches

Refs.	Impedance technology	Experimental samples	Materials & channel (width × height) & flow velocity	Demonstrated detection rate & statistical number	Focusing methods & hardware
Our methods	Constriction channel with two coplanar electrodes	3, 5, 7 and 10 $\mu\text{m}$ microspheres Hela, Jurket cell lines	PDMS $25 \times 12 \mu\text{m}^2$ 100 mm/s	172 cells/s 10000 events	No focusing
	1MHz				Lab-build lock-in amplifier
Cheung et al. (2005) Cytometry A 65A:124–132	Wheatstone bridge	4, 5, 14 and 6 $\mu\text{m}$ polystyrene beads RBCs, fixed RBCs, ghosts	Polyimide $40 \times 20 \mu\text{m}^2$ 10 mm/s	16.7 cells/s 800 events	nDEP forces
	350 kHz to 20 MHz				SR-844, Stanford Research Systems
Benazzi et al. (2007) IET Nanobiotechnology 1:94–101	Wheatstone bridge	Phytoplankton (Isochrysis Galbana, Rhodospirillum rubrum, Synechococcus sp.)	Polyimide $11 \times 20 \mu\text{m}^2$ 39 mm/s	100 cells/s 2500 events	No focusing
	327kHz and 6.03MHz				SR-844, Stanford Research Systems
Rodriguez-	Impedance	20 $\mu\text{m}$ beads	PDMS	20 beads/s	Two/three sheath

Trujillo et al. (2008) Biosens Bioelectron 24:290–296	analyser		$190 \times 50 \mu\text{m}^2$ < 5.3 mm/s	< 200 events	flow
	120 kHz and 1 MHz				Agilent 4294A
Wang et al. (2008) Lab chip 8:309-315	MOSFET drain current	CD4+ T cells	PDMS $16 \times 30 \mu\text{m}^2$ NA	8 cells/s 1166 events	No focusing
	DC				SR 850, Stanford Research Systems
Holmes et al. (2009) Lab Chip 9:2881–2889. (2010) Anal Chem 82:1455–1461	Parallel facing electrodes	Protein coated 5.6 $\mu\text{m}$ microspheres, whole blood, CD4 T-Lymphocytes	Polyimide $20 \times 20 \mu\text{m}^2$ 60 mm/s	100 cells/s 5000 events	nDEP forces
	500 kHz to 30 MHz				SR 844, Stanford Research Systems
Bernabini et al (2011) Lab Chip 11:407-412	Parallel facing electrodes	1, 2 $\mu\text{m}$ polystyrene beads and <i>E. coli</i> .	Polyimide $200 \times 30 \mu\text{m}^2$ 38 mm/s	100 cells/s 3000-5000 events	hydrodynamic focusing by oil
	503 kHz and 5 MHz				SR 844, Stanford Research Systems
Chen et al. (2011) Lab Chip 11: 3174-3181. (2011) Biomicrofluidics, 5:014113	Constriction channel with external Ag/AgCl electrodes	MC-3T3 (osteoblasts), MLO-Y4 (osteocytes), EMT6, EMT6/AR1.0	PDMS $6 \times 6 \mu\text{m}^2$ $8 \times 8 \mu\text{m}^2$ NA mm/s	<2 cells/s 770 events	No focusing
	10 kHz and 100 kHz				Agilent-4294A Impedance Analyzer
Song et al. (2013) Lab Chip 13: 2300-2310.	Constriction channel	20 $\mu\text{m}$ polystyrene beads, P19 stem cells	PDMS $40 \times 27 \mu\text{m}^2$ 3 mm/s	< 2 cells/s 200 events	No focusing
	50 kHz, 250 kHz, 500 kHz and 1 MHz				HF21S, Zurich Instruments
Hassan et al. (2014) Lab Chip 14:1469-1476. (2014) Lab Chip 14:4370-4381	Three coplanar electrodes in constriction channel	Lymphocytes, granulocytes, monocytes	PDMS $15 \times 15 \mu\text{m}^2$ 4400 mm/s	50 cells/s ~ 3000 events	No focusing
	303 kHz				HF21, Zurich Instruments
Nguyen et al. (2015) Lab Chip 15:1533-1544	external Ag/AgCl electrodes	6.2-8.2 $\mu\text{m}$ RBC, 10-15 $\mu\text{m}$ WBC	PDMS $15 \times 15 \mu\text{m}^2$ NA mm/s	~8.3 cells/s ~ 10000events	No focusing
	10, 100, 400, 990 kHz				NA
Watkins et al. (2011) Lab Chip 11:1437-	Three coplanar electrodes in constriction	7.32 mm polystyrene beads, CD4+	PDMS $15 \times 15 \mu\text{m}^2$ 2200 mm/s	2236 cells/s 26054 events	No focusing

1447.	channel	and CD8+ T lymphocytes			
	303 kHz, 1.1 MHz and 1.7 MHz				HF21, Zurich Instruments

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