

## Supplementary Information

### Microfluidic cell volume sensor with tunable sensitivity

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## S1. Fabrication Details

### Ag Electrodes

Ag electrodes were fabricated on glass using a common lift-off process. Electrodes were fabricated so as to have multiple independent leads into the fluidic channel, with a width and separation chosen to be similar to channel diameter (15-25  $\mu\text{m}$ ). While only two are required for current measurement, the others allow flexibility in measurement technique (4 point measurement), as well as backups in the case of damage. A separate grounding electrode provides electromagnetic shielding along the edges of the device. 1" x 3" x 1mm glass microscope slides were thoroughly rinsed with acetone, isopropanol and ethanol, then heated to 110°C for 10min and cooled for 2 min. They were then spin-coated with Shipley S1813 photoresist for 30s at 4000rpm, for a final thickness of 1.4 $\mu\text{m}$ . Slides were prebaked 60s at 90°C, UV exposed 60s, toluene soaked 60s, and postbaked at 90°C for 15s. Development was done in tetramethylammonium hydroxide (TMAH). 5nm Cr / 80nm Ag was deposited by sputter deposition, and lift-off was completed in 1-methyl-2-pyrrolidinone (NMP). Electrodes were rinsed with acetone, isopropanol and ethanol prior to bonding to PDMS (Sylgard 184 silicone elastomer kit).

### Photoresist Moulds

For the flow channel mould, two protocols were followed, one for rounded channels (P4620 photoresist – AZ Electronic Materials), a second for straight-walled rectangular channels (SU8-10 - MicroChem). A silicon wafer was cleaned using piranha etch, then baked at 200°C for 30 min, and let cool for 5 min. For the rounded P4620 channel mould, an adhesion layer of hexamethyldisilazane (HMDS) is first spin-coated. Two P4620 layers are deposited in turn. Spin speed and time were chosen depending on final thickness requirements. For example, a 20 $\mu\text{m}$  layer required the following spin settings (Table S.1):

**Table S1** Spin settings P4620

Layer 1	Layer 2
4s@500rpm	4s@500rpm
5s @ 0rpm	5s @ 0rpm
5s @ 500rpm	5s @ 500rpm
40s @ 1800rpm	40s @ 1150rpm
40s @ 400rpm	40s @ 400rpm
10s @ 900rpm	10s @ 900rpm

After each spin-coated layer, the wafer is prebaked at 90°C for 1 min, then 115°C for 1.5 min. The wafer is then exposed to UV for 60s, and immersed in AZ400K developer as required. The wafer is postbaked at 75°C, 95°C, 115°C and 200°C for 5 min each. For the SU-8 mould, photoresist is spin coated to required thickness. The wafer is then prebaked at 65°C for 2 min, then 95°C for 6 min, and exposed to UV for 45s. The wafer is postbaked at 65°C for 1 min, and 95°C for 3 min. SU-8 developer is then used to remove the photoresist. For the valve layer, a similar protocol is used.

### PDMS multilayer

All wafers are then coated with aminosilane to facilitate PDMS removal. PDMS for the flow layer was prepared with 7:1 base to curing agent ratio, and baked at 80°C for 24 min. Devices were cut out, inlet holes were punched, and devices were rinsed with soapy water, deionised water, ethanol and isopropanol, in that order. Fluidic layer PDMS was mixed at a 20:1 base to curing agent ratio. Using two PDMS ratios, one with a higher percentage of base, one with a higher percentage of curing agent, allows for cross-linking of both layers, and thus helps strengthen the bond. Further, a 20:1 ratio leads to more flexible PDMS to make the channel layer more elastic. The mixture was degassed, spin coated to required thickness, and baked for 20 min at 80°C. PDMS thickness was chosen to be above the height of the channel. For example, for a 20 $\mu\text{m}$  channel, a thickness of 25 $\mu\text{m}$  was desired, requiring the following spin settings:

**Table S2** Spin settings PDMS

Layer 1
10s@1000rpm
30s@3000rpm

Both PDMS pieces were exposed to oxygen plasma (Glow Research AutoGlow) at 30W for 30s, bonded using a mask aligner, and baked at 80°C for >12hrs. Devices are cut out, and flow channel inlets are punched. Devices were again cleaned using soap and water, deionised water, methanol and isopropanol. Electrodes and PDMS devices were exposed to oxygen plasma (30W, 30s), and aligned using a mask aligner. Devices were then baked at 80°C for 12hrs. Completed devices are then glued to PCB boards, where leads are connected with a wire-bonder to bonding pads leading to external electronics.

## S2. Flow Control

Fig.S1 illustrates our pressure control scheme, used for most measurements featured in this work.

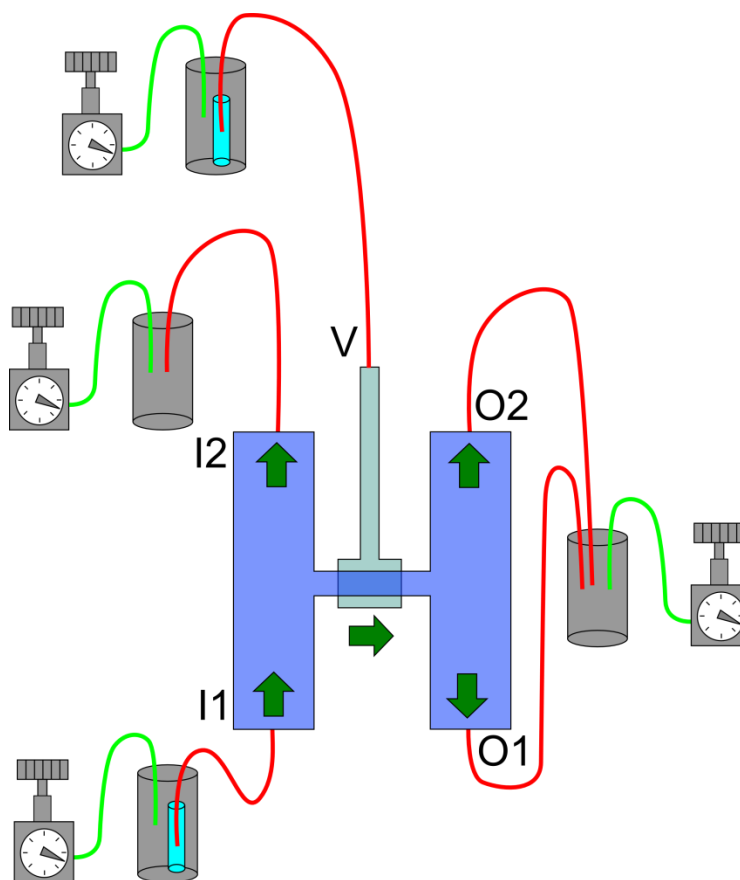


Figure S1: Flow control diagram. Pressures at I1, I2, O1 and O2 are set to about 3 psi, with I1 slightly higher. This drives a flow from I1 to I2, and from left to right through the sensor. Solution is driven through the device by pressurizing the corresponding vial with a regulator. The valve is filled with water, and controlled via a separate regulator. Green: Air Tube. Red: Flow Tube.

## S3. Translocation Videos

These videos show the flow path of particles (top view) through the device.

### Supplementary Video 1: Transiting Microspheres - Valve OFF.wmv

Microspheres of 6 $\mu$ m diameter transit through a volume sensor, with PDMS valve unpressurized.

6 $\mu$ m diameter microspheres flow along the 50 $\mu$ m wide PDMS channel. Transiting particles cause a drop in current, and thus a measure of their volume. Valve is unpressurized; sensitivity is low.

**Keywords: Coulter, volume sensing, microfluidic, microspheres, ionic current, impedance**

**Supplementary Video 2: Transiting Microspheres - Valve ON.wmv**

Microspheres of 6µm diameter transit through a volume sensor, with PDMS valve pressurized.

6µm diameter microspheres flow along the 50µm wide PDMS channel. Transiting particles cause a drop in current, and thus a measure of their volume. Valve is pressurized; sensitivity is high.

**Keywords: Coulter, volume sensing, microfluidic, microspheres, ionic current, impedance**