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**Electronic Supplementary Information (ESI)** 

## High accuracy particle analysis using sheathless microfluidic impedance cytometry

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Figure S1: (a) Velocity and (b) signal amplitude (cube root of the impedance magnitude, 7  $\mu$ m diameter beads) as a function of the particle position in the channel cross-section.



Figure S2: Simulation results (7  $\mu$ m diameter beads). (a) Particle vertical position vs signal amplitude (cube root of the impedance magnitude). (b) Particle vertical position vs dimensionless ratio  $\Delta x_{\rm OBQ}/\Delta x_{\rm TSV}$  (see definition in Figure 2 of main text). The plot of  $\Delta x_{\rm OBQ}/\Delta x_{\rm TSV}$  (equivalent to  $\Delta t_{\rm OBQ}/\Delta t_{\rm TSV}$ ) vs signal amplitude is shown in Figure 3 of main text.



Figure S3: Simulation results (7  $\mu$ m diameter beads, 30  $\mu$ m electrode width, variable channel height h). Normalized vertical position (z/h) versus signal amplitude (cube root of the impedance magnitude) normalized by signal amplitude relevant to centered bead (z = 0).



Figure S4: Finite element geometric model of the microfluidic device. A typical mesh is shown.

## Finite element model

The device is modeled as the union of two homogeneous conducting regions  $\Omega_1$  and  $\Omega_2$ , representing the bead and the suspending fluid, respectively. Their complex conductivities  $\sigma_1^*$  and  $\sigma_2^*$  are given by  $\sigma_k^* = \sigma_k + i\omega\varepsilon_k\varepsilon_0$ , k = 1, 2, where  $\varepsilon_0$  is the permittivity of free space, and  $\sigma_k$  and  $\varepsilon_k$  are the conductivity and relative permittivity of the media, respectively;  $\omega$  denotes the circular frequency, and i is the imaginary unit. Continuity of electric potential and normal current flux density is enforced at the bead surface  $\Gamma$ . The boundary of the domain is divided into an insulating part ( $\partial\Omega_{ne}$ ), and a part covered by electrodes ( $\partial\Omega_e$ ) which generate the electric field.

In the Fourier domain, the electrical problem is stated as follows:

$$-\operatorname{div}(\sigma^*\nabla\Psi) = 0, \qquad \text{ in } \Omega_1 \cup \Omega_2 \text{ ;} \qquad (1)$$

$$\llbracket \sigma^* \nabla \Psi \cdot \boldsymbol{n} \rrbracket = 0, \qquad \text{ on } \Gamma; \qquad (2)$$

$$\llbracket \Psi \rrbracket = 0, \qquad \text{ on } \Gamma, \qquad (3)$$

where  $\Psi$  is the electric potential phasor,  $\sigma^* = \sigma^*_k$  in  $\Omega_k$ , k = 1, 2, div and  $\nabla$  respectively

Table S1: Electric parameters adopted for bead, electrodes, and extracellular fluid

$\sigma_1$ (S/m)	$\varepsilon_1$	$C_{e}$ (mF/m $^2$ )	$\sigma_{2}$ (S/m)	$\varepsilon_{2}$
$6.6 \times 10^{-4}$	2.5	144	1.6	80

denote the divergence and gradient operators,  $[\![\cdot]\!]$  is the jump of the enclosed quantity across  $\Gamma$ , and n denotes the outer unit normal vector. An insulating boundary condition is applied on the boundaries not covered by electrodes

$$\sigma^* \nabla \Psi \cdot \boldsymbol{n} = 0, \qquad \text{on } \partial \Omega_{\mathsf{ne}} \,.$$
 (4)

On the *i*-th electrode ( $\partial \Omega_{e_i}$ ), the following electrode equation holds

$$Y_{\mathbf{e}}(\Psi_i - \Psi) = \sigma^* \nabla \Psi \cdot \boldsymbol{n}, \quad \text{on } \partial \Omega_{\mathbf{e}_i},$$
 (5)

where  $Y_{e} = G_{e} + i\omega C_{e}$  is the double-layer admittance per unit area, expressed in terms of conductance  $G_{e}$  and capacitance  $C_{e}$  per unit area, and  $\Psi_{i}$  is the electrode potential. In the radio-frequency range,  $G_{e}$  is usually negligible with respect to  $\omega C_{e}$ .

Problem (1)–(5) is recast into the following weak formulation

$$\int_{\Omega_1 \cup \Omega_2} \sigma^* \nabla \Psi \cdot \nabla \Phi \, \, \mathrm{d}V + \sum_i \int_{\partial \Omega_{\mathbf{e}_i}} Y_{\mathbf{e}} \Psi \Phi \, \, \mathrm{d}A = \sum_i \int_{\partial \Omega_{\mathbf{e}_i}} Y_{\mathbf{e}} \Psi_i \Phi \, \, \mathrm{d}A \,, \qquad \text{(6)}$$

where  $\Phi$  is an arbitrary test function. Equation (6) is solved using the commercial finite element code COMSOL Multyphysics. In particular, Weak Form PDE Physics are adopted for the bead and fluid contributions, whereas the electrode terms are treated as Weak contributions on the relevant boundaries. Quadratic Lagrangian tetrahedral elements are used to interpolate the electric potential  $\Psi$ .

Figure S4 shows the geometric model of the microfluidic chip (36  $\mu$ m wide  $\times$  45  $\mu$ m high, with 30  $\mu$ m wide electrodes and 10  $\mu$ m spacing), along with a typical tetrahedral mesh. Parameter values in the simulations are shown in Table S1. A signal of 4 V at 1 MHz was applied to two top electrodes and the resulting currents through the bottom electrodes were calculated.