Electronic Supplementary Material (ESI) for Analytical Methods.

Supplementary Information

Dielectrophoretic Assembly and Separation of Particles and Cells in Continuous Flow

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Section 1. Fabrication process of microfluidic device

Fabrication process of the microfluidic device for investigation of dielectrophoretic Assembly and Separation of Particles and Cells include following three steps:

(1) **Fabrication of PDMS channel**. We passed two layers of dry films on the clean glass (Fig.S1a1), and they were exposed to the ultraviolet light for 12 s (Fig.S1a2). Dry films were processed into channel shapes using 1.5 wt% Na_2CO_3 solution for about 3 min (Fig.S1a3). Afterwards, the base and curing agent of PDMS with a weight ratio of 10:1 were mixed, and then we conducted vacuum treatment to remove the air bubbles (Fig.S1a4). Then, the mixture was poured to dried channel mould, and it was put in the oven at 80 ℃ for 2 h to obtain PDMS channel (Fig.S1a5). It was worth noting that channel moulds were proceeded to reduce the surface energy using 1H, 1H, 2H, 2H-perfluorooctyltrichlorosilane (Fig.S1a6).

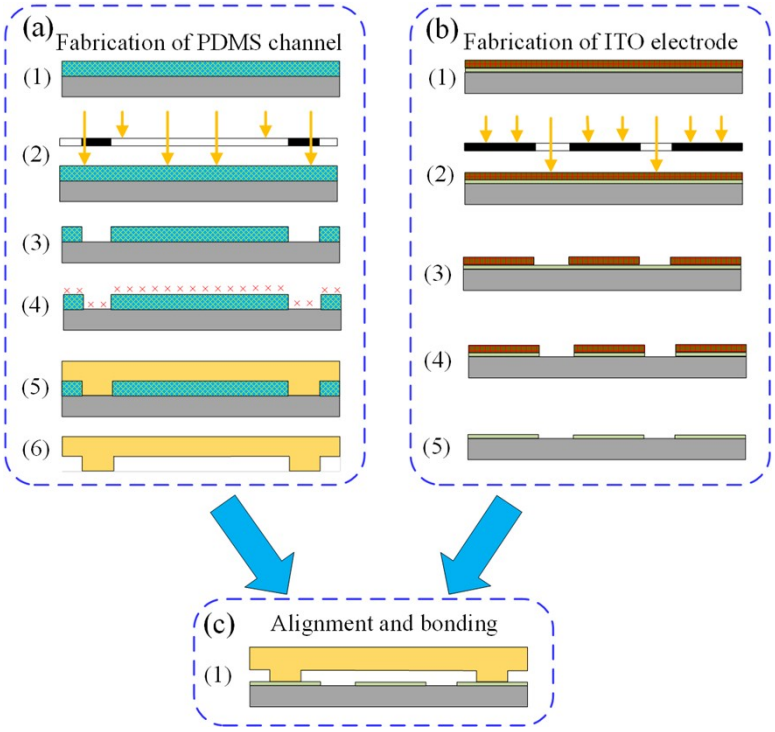

Figure S1 Fabrication processes of the device

(2) **Fabrication of ITO electrode**. Transparent ITO film on the glass substrate was processed to desired structures by the standard photolithography and chemical etching. Firstly, photoresist (AZ4620) was spin coated at on the ITO layer at the low speed of 1000 rad/min for 35 s, and then at the high speed of 3000 rad/min for 100 s (Fig.S1b1). After exposure of photoresist under the UV light (Fig.S1b2), we performed the development treatment in the 25 wt% developer (A400K) (Fig.S1b3). Then, we put the ITO glass on the hot plate to dry and reinforce the photoresist films at the 110 ℃ for 6 min. Afterwards, we removed the ITO layer without the photoresist films through the chemical etching treatment for about 15 min in 60 wt% HCl solution with the catalyzer of FeCl3 powders (Fig.S1b4). Finally, we removed the rest of the photoresist on the ITO electrode with the 98 wt% alcohol (Fig.S1b5).

(3) **Alignment and bonding.** Under the microscope (CKX53, Olympus, Japan), PDMS channel and ITO electrode were aligned and bonded after oxygen plasma treatment (Fig.S1c1).

Section 2. The electrode connection

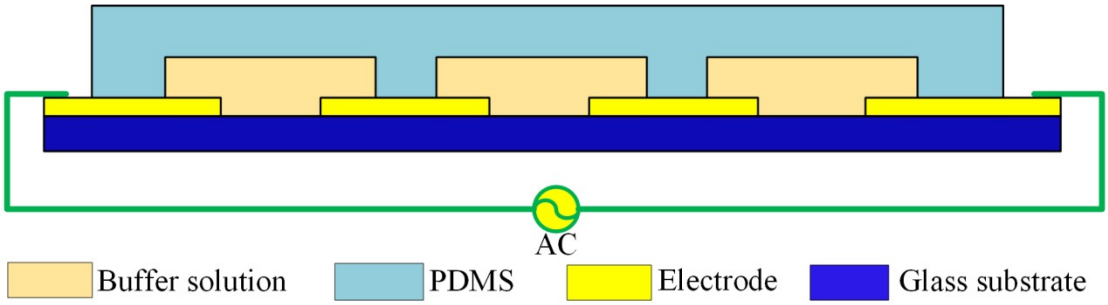

Figure S2 The side view of the device

Section 3. The electric field and flow field distribution

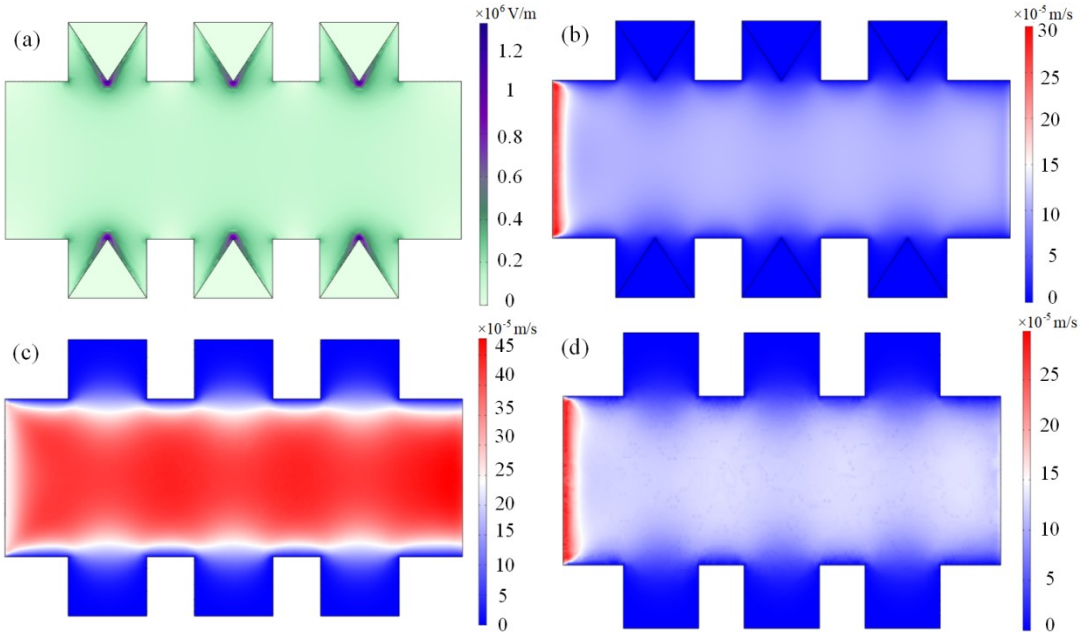

Figure S3 Simulation results at A=45 V, f=10 kHz, and u=250 μm/s. (a) Electric-field distribution in the channel. (b/c/d) Flow velocity distribution at the height of 5/38/70 μm.

Section 4. The assembly of particles with different electric characterizations

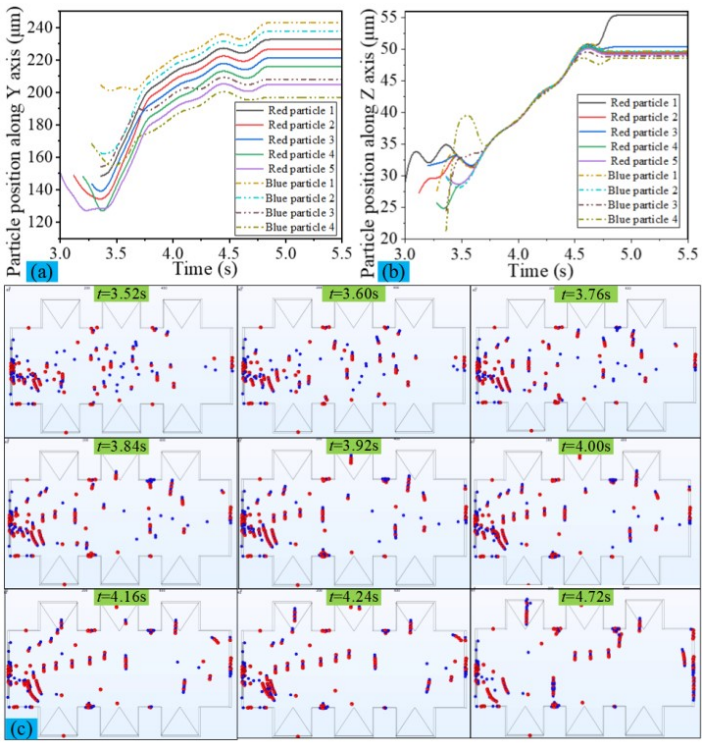

Figure S4 nDEP-force induced assembly of particles with different electric characterizations. (a) Particle positions along Y coordinate versus time. (b) Particle positions along Z coordinate versus time. (c) Particle trajectories of electrically similar particles under nDEP force.

Section 5. Effect of cell chains on the cell separation

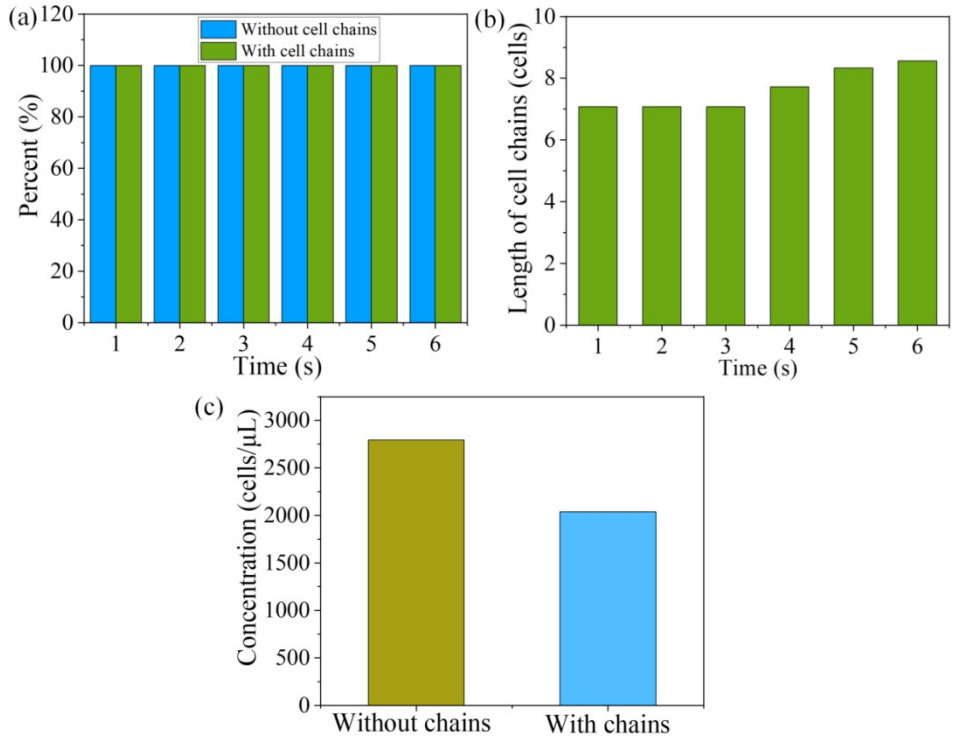

Figure S5 Effect of cell chains on the cell separation. (a) The purity of *C. vulgaris* with/without chains. (b) The average particle numbers of chains with time. (c) The concentration of injected cells without/with chains.