# Electronic Supporting Information for

# Microfluidic Mixing of Nonpolar Liquids by Contact Charge Electrophoresis

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		St	d (µm)	f(Hz)	<i>U</i> (mm/s)	$V_{0}(V)$ *	$\sigma_{avg}$ **
Fig. 1	Diffusive	•			$0.8\pm0.04$		•
8	Orbital	$4.8\pm0.2$	35	41	$0.8\pm0.04$	800	•
Fig. 2	Linear (a)	$4.1\pm0.2$	30	25	$0.58\pm0.02$	800	•
	Orbital (b)	$4.8\pm0.2$	35	41	$0.80\pm0.04$	800	•
Fig. 3	Diffusive	•	·	•	$0.54\pm0.02$	•	0.95
8.0	Linear	$50.0 \pm 2.2$	34	230	$0.42\pm0.05$	900	0.68
	Orbital	$13.9\pm0.3$	35	70	$0.48\pm0.01$	800	0.25
Fig. 4a,b	1 (low <i>St</i> )	$0.6\pm0.0$	35	51	$7.5\pm0.3$	800	0.70
	2	$1.3 \pm 0.1$	35	55	$6.0 \pm 0.3$	800	0.35
	3	$2.8 \pm 0.3$	35	57	$1.9 \pm 0.2$	800	0.35
	4	$5.3 \pm 0.4$	35	55	$1.0 \pm 0.6$	800	0.39
	5	$13.9\pm0.3$	35	70	$0.48\pm0.01$	800	0.25
	6 (high <i>St</i> )	$26.9 \pm 1.6$	35	57	$0.20\pm0.01$	800	0.12
Fig. 4c	Small	$9.2 \pm 0.1$	27	47	$0.49 \pm 0.01$	950	0.68
	Large	$5.6\pm0.3$	38	34	$0.58\pm0.03$	800	0.26

## I. Supplemental Data and Videos

**Table S1:** Summary of several key parameters for the experimental data presented in the Figures of the main text: Strouhal number (*St*); particle diameter (*d*); oscillation frequency (*f*); maximum fluid velocity downstream of the mixing region (*U*); applied voltage ( $V_0$ ); and average mixing index ( $\sigma_{avg}$ ).

\*The polarity of the voltage had no effect on particle motion; therefore, we report only the magnitude of the applied voltage.

\*\*The mixing index  $\sigma_{avg}$  represents the average over the last 100 µm of channel visible in the images presented in the Figures.

## Supplemental Videos

**Video 1: Nonreciprocal ("Orbital") Mixing.** This video is representative of the entire operation of an orbital mixer (specifically the orbital mixer displayed in Figures 1 & 2). Initially the particle is held in place at -800 V by a dielectrophoretic force. During this time, mixing only occurs through diffusion. When the mixer is turned "on," (*i.e.* when the polarization is flipped from -800 V to 800 V), the particle begins to oscillate thereby mixing the fluid. Over time the particle begins to slow down, presumably due to the screening of the applied field by the slow accumulation (minutes) of charged species on the surface of the PDMS and/or the electrode surface. Occasionally (such as in this video), the field diminishes to the point where the particle can no longer complete its oscillations and stops. Importantly, particle motion can be maintained indefinitely by periodically reversing the polarity of the applied voltage (*e.g.*, each minute) to prevent charge accumulation.

**Video 2: Reciprocal ("Linear") Mixing.** This video shows a typical reciprocal ("linear") mixer immediately upon startup (Figure 2).

**Video 3: Diffusive, Linear and Orbital Mixing.** Brief clips are played of each of the three different mixers analyzed in Figure 3.

**Video 4: Orbital Mixers at Varying Strouhal Numbers.** Six clips are played of mixers operating at different flow rates (Figure 4).

**Video 5: Orbital Mixers with Different Particle Diameters.** A small diameter particle (27  $\mu$ m) is compared to a large diameter particle (38  $\mu$ m) in an orbital mixer (Figure 5).

# **II. Additional Experimental Details**

CCEP micromixers were fabricated in polydimethylsiloxane (PDMS) by soft lithography<sup>1</sup> as detailed below:

# Photolithography

- 1. Pretreat silicon wafer (University Wafer 100mm ID#1196)
  - a. Rinse with isopropyl alcohol.
  - b. Rinse with deionized (DI) water.
  - c. Bake at 175 °C for 5 min.
- 2. Spin coat photoresist (SU8-50)
  - a. Dispense ~6 mL of SU8-50 onto wafer.
  - b. Spin at 500 rpm for 10 s (ramp 100 rpm/s) to spread.
  - c. Spin at 2500 rpm for 40 s (ramp 300 rpm/s) to produce ~50µm thick film of photoresist.
- 3. Soft bake coated wafer
  - a. Prebake at 65°C for 6 min.
  - b. Soft bake at 95°C for 20 min.
- 4. Hard Contact Exposure
  - a. Expose wafer under UV light (8 mW) for 7 on/off cycles with 12 s on and 10 s off.
    Note: An optical filter (Omega Optical Inc. PL-360LP 170618 1PC) was used to improve the vertical aspect ratio of the SU8-50; actual exposure was therefore ~70% of that applied.
- 5. Post-Exposure Bake
  - a. Bake at 65°C for 1 min.
  - b. Bake at 95 °C for 5 min.
  - c. Bake at 65°C for 30 s.
  - d. Cool to room temperature.
- 6. Develop
  - a. Soak wafer in SU8 developer for 6 min.
- 7. Rinse & Dry
  - a. Rinse wafer with isopropyl alcohol.
  - b. Dry under nitrogen stream.
  - c. Check for residue; repeat steps 6-7 as needed.
- 8. Hard Bake
  - a. Bake at 65°C for 30 s.
  - b. Bake at 95°C for 30 s.
  - c. Bake at 175°C for 2 min.
  - d. Bake at 95°C for 30 s.
  - e. Bake at 65°C for 30 s.
  - f. Cool to room temperature.

### **PDMS** Patterning

33 g of polydimethylsiloxane (PDMS, Sylgard 184; 10:1 ratio of silicone base to silicone curing agent) was poured onto the patterned silicon wafer in a petri dish and hardened at 65 °C for 2 hr. The cured PDMS was peeled off and bonded to a glass slide (cleaned with 20 vol% ethanol in DI water and dried in nitrogen stream). Prior to bonding, both the PDMS and the glass were oxidized in 10:1 O<sub>2</sub>:He plasma for 30 s (Harrick Plasma PC-32G). Channel inlets were cut using a Harris Unicore device. To mitigate the sticking of the particles to the PDMS/glass surfaces, the channels were functionalized with n-octyltriethoxysilane by flowing a 1 wt% solution in ethanol for 60 min and drying under nitrogen<sup>2</sup>.

#### **Gallium Electrodes**

Liquid gallium was heated to ~60  $^{\circ}$ C in an oven and injected into the electrode channels using a syringe. Copper leads were inserted into the liquid gallium at the channel inlet, and the device was cooled to room temperature to solidify the electrodes<sup>3</sup> (crystallization of supercooled liquid gallium was initiated by contact with a piece of solid gallium). The small openings, which separate the electrode channels from the central mixing channel, prevented the flow of liquid gallium into the mixing channel due to the large Laplace pressure.

### Mixing Liquids

To minimize particulate matter and debris from entering the channel, cleanliness was essential. All storage containers were cleaned with ethanol and dried with nitrogen gas. All fluids were filtered prior to use (VWR sterile syringe filter, 0.45  $\mu$ m pores) and aspirated into syringes. The first syringe (SGE syringe 250  $\mu$ L) was filled with mineral oil (Sigma Aldrich M5904); the second syringe (1 mL BD) was filled with a 0.05 wt% dispersion of silver-coated hollow glass microspheres (Cospheric M-40, 27-32  $\mu$ m diameter); the third syringe (SGE) was filled with mineral oil saturated with *ca*. 0.25 g/L Sudan IV dye (Sigma Aldrich).

### **Mixer Operation**

The mixer was positioned under an optical microscope (Carl Zeiss Axio Imager 10x) and connected to two programmable syringe pumps (New Era NE-500) via polyethylene tubing (Intramedic Clay Adams 427415). The copper leads were connected to a high voltage amplifier (Trek 20/20C) controlled by a sourcemeter (Keithley 2612A). A single mixing particle was positioned near the electrodes by aspirating/dispensing the particle dispersion from the second syringe. Application of a constant voltage across the channel caused the dielectrophoretic capture of the particle onto the electrode surface. Once the particle was in place, the second syringe was locked to prevent undesired flows into/from the particle channel. The syringe pumps then delivered the two mineral oil streams at flow rates ranging from 2.5 to 80  $\mu$ L/hr.

Once the desired flow was achieved, the voltage was increased above the liftoff voltage, and the polarity was reversed repeatedly until the particle began to oscillate. Videos were captured by a high speed camera (Phantom v310) at 5000–10000 fps.

## **III. Image Analysis**

## Image Acquisition

Prior to imaging, a current session reference (CSR) was performed on the Phantom v310 camera to correct for noise in the CMOS sensor. For each experiment, we captured two reference images along with video of interest: (i) an image of the mixer filled with clear mineral oil, and (ii) an analogous image of the mixer filled with the dyed mineral oil. These images were used to correct for spatial inhomogeneties in the illumination of the mixer as described below. All images were collected at a resolution of 1.26  $\mu$ m/px.

## Image Processing

The images were first cropped to obtain a common region of interest and filtered using a  $3 \times 3$  median filter (MATLAB) to reduce noise. A mask was added to remove any unwanted features – namely, anything outside of the channel. The concentration of dye C(x,y) within the channel was approximated using the Beer-Lambert law. Specifically, we computed the logarithm of the ratio between the image of interest and the reference image of clear mineral oil,  $C \propto \ln(I / I_{ref})$ , where I(x,y) and  $I_{ref}(x,y)$  are the greyscale intensities of the image and the reference, respectively. This ratio approximates that between the transmitted light captured by the camera and the incident light through the mixer.

Using the resulting concentration "image", we computed the standard deviation of each pixel column spanning the width of the channel to obtain a "raw" mixing index. This index was then adjusted by a multiplicative factor such that the unmixed region upstream of the mixing zone corresponded to a mixing index of  $\sigma \approx 1$ . This is the mixing index reported in the main text.

We also computed the mixing index of a homogeneous fluid using the reference image for the dyed mineral oil stream. Ideally, a homogeneous fluid should result in a mixing index of  $\sigma = 0$  (*i.e.*, no variation in concentration across the channel). In practice, we obtain mixing indices of  $\sigma \approx 0.1$  for the dyed reference stream due to uncontrolled variations in the images (*i.e.*, noise). For these calculations, there was no "unmixed" region by which to scale the "raw" mixing index; we instead used the multiplicative factor obtained for another image taken on the same day. This approach should be effective as the scaling factors were relatively constant over the entire data set.

#### **IV. Power Requirements**

To determine the electrical power required to operate a single CCEP mixer, we measured the current supplied to the mixer during its operation. Briefly, a microfluidic mixer was prepared as describe in Section II, and a 30  $\mu$ m particle was positioned in the channel. The mixer was surrounded by a Faraday cage to reduce external electrical noise, and the electrodes were connected to a preamplifier (DL Instruments 1211) to measure the current. A voltage of 800V was supplied by sourcemeter (Keithley 2410), and the motion of the particle was captured by a high speed camera (Phantom v310). Figure S1 shows the measured current and the position of the particle (across the channel) as a function of time. Each time the particle contacted an electrode, there was a sharp spike in the measured current, and the particle upon contact. The average electrical current delivered to the mixer during its operation was 0.16 nA as compared to 0.09 nA in the absence of the particle. Multiplying this current by the applied voltage gives the electrical power needed to operate the mixer, 130 nW. By further reducing any parasitic currents (here 0.09 nA), it should be possible to power mixer operation using only ~60 nW.



**Figure S1.** Synchronized current and position measurements. The **red** curve shows the position of the particle between the two electrodes as a function of time (right axis). The **blue** curve shows the measured current through the external circuit (see Figure 1) as the particle oscillates between the two electrodes (left axis). Each time the particle changes direction, there is a brief "spike" in the current corresponding to the rapid transfer of charge between the particle and the electrode on contact. The **black** curve shows the measured current through the circuit in the absence of particle motion (e.g., due to the finite conductivity of the fluid).

# V. Estimation of Dye Diffusivity

The diffusivity D of the dye in mineral oil was estimated using the Stokes-Einstein equation,

$$D = \frac{k_B T}{6\pi\eta a},\tag{S1}$$

where  $k_BT$  is the thermal energy,  $\eta = 0.027$  Pa·s is the viscosity of mineral oil, and *a* is the hydrodynamic radius of the dye molecule. The size of the dye *a* was estimated from the molar volume *v* as

$$a \approx \left(\frac{3\nu}{4\pi N_A}\right)^{1/3} = 5.6 \dot{A} , \qquad (S2)$$

where  $N_A$  is Avogadro's number. The resulting diffusivity is  $D = 1.4 \times 10^{-7} \text{ cm}^2/\text{s}$ .

#### **VI. Estimation of Fluid Velocity**

The fluid velocity was measured by tracking the movement of some fluid inhomogeneity (e.g., variations in the dye concentration) down the center of the channel. For example, at intermediate Strouhal numbers ( $St \sim 5$ ; see Figure 4), we tracked the leading edge of one "band" of dyed fluid between two time points as it moved down the channel; the fluid velocity was estimated by the distance traveled divided by the time interval. This measurement gave the maximum fluid velocity in the center of the channel, which is related to the average velocity as  $v_{max} \approx 1.991 v_{avg}$  for the experimental aspect ratio of W/H = 2 (see below). This "direct" approach to measuring the velocity was found to be considerably more reliable than estimating the average velocity from the fluid flow rate Q and the channel cross-section ( $v_{avg} = Q/HW$ ) due to deviations in Q from its specified value.

When there were no clear fluid inhomogeneities by which to estimate the fluid velocity (*e.g.*, in the absence of active mixing or at high Strouhal numbers), the velocity was instead measured by tracking the movement of small particles (*e.g.*, dust or debri) down the channel. We assumed that the particles moved at the local fluid velocity and were centered midway between the floor and ceiling of the channel (z = H/2). Based on the particle's velocity and its position within the channel (0 < y < W), we estimated the maximum fluid velocity (at the center of the channel) using the calculated velocity profile for steady, laminar flow in a rectangular channel (see below). As particles would move slower at other depths within the channel, this estimate should be treated as a lower bound on the true fluid velocity.

### Velocity Profile

The velocity profile  $\mathbf{v}(\mathbf{x})$  within the channel is governed by the Navier-Stokes equations for an incompressible fluid,

$$\rho\left(\frac{d\mathbf{v}}{dt} + \mathbf{v} \cdot \nabla \mathbf{v}\right) = -\nabla p + \eta \nabla^2 \mathbf{v} \text{ and } \nabla \cdot \mathbf{v} = 0, \qquad (S3)$$

where  $\rho$  and  $\eta$  are the fluid density and viscosity, respectively, and p is the pressure. Assuming steady unidirectional flow in the *x*-direction, these equations simplify to

$$0 = -\frac{dp}{dx} + \eta \left( \frac{\partial^2 v_x}{\partial y^2} + \frac{\partial^2 v_x}{\partial z^2} \right), \tag{S4}$$

where  $v_x = v_x(y, z)$  and dp/dx = constant. For flow in a rectangular channel of width W and height H, the no-slip boundary conditions on the channel walls are given by

$$v_x(y,0) = v_x(y,W) = v_x(0,z) = v(H,z) = 0.$$
 (S5)

Solving equations (S4) and (S5), the velocity profile is given by

$$v_x(y,z) = \frac{4H^2}{\eta\pi^3} \left(-\frac{dp}{dx}\right) \sum_{n \text{ odd}}^{\infty} \frac{1}{n^3} \left[1 - \frac{\cosh\left(n\pi(y - \frac{1}{2}W)/H\right)}{\cosh\left(n\pi W/2H\right)}\right] \sin\left(\frac{n\pi z}{H}\right)$$
(S6)

Integrating over the channel cross-section, the fluid flow rate is

$$Q = \frac{8H^4}{\eta \pi^5} \left( -\frac{dp}{dx} \right)_{n \text{ odd}} \frac{1}{n^5} \left( \frac{n\pi W}{H} - 2 \tanh\left(\frac{n\pi W}{2H}\right) \right)$$
(S7)

For the experimental aspect ratio W / H = 2, the solution above implies that

$$v_{\rm max} \approx 0.1138 \frac{H^2}{\eta} \left(-\frac{dp}{dx}\right)$$
 (S8)

$$v_{avg} = \frac{Q}{WH} \approx 0.05717 \frac{H^2}{\eta} \left(-\frac{dp}{dx}\right)$$
(S9)

The velocity profile is illustrated in Figure S2.



**Figure S2.** Velocity profile v(y,z) scaled by  $v_{avg}$  for  $H = 50 \ \mu\text{m}$  and  $W = 100 \ \mu\text{m}$ .

#### VII. Estimation of Hydrodynamic Drag in a Channel

At low Reynolds numbers, the hydrodynamic drag on a sphere of radius *a* in an unbounded quiescent fluid is given by Stokes law,  $F_{vis} = 6\pi a\eta u$ , where  $\eta$  is the fluid viscocity, and *u* is the velocity of the particle. In the experiments described in the main text, the fluid bounded by the walls of the PDMS channel, which act to increase the drag force resisting particle motion. The most significant contributions come from the floor and ceiling of the channel which are separated by a distance  $H \approx 50 \,\mu\text{m}$  not much larger than the particle diameter  $d \approx 34 \,\mu\text{m}$ . The enhancement in the drag force for a sphere moving between two parallel walls has been calculated previously by Brady and Swan<sup>4</sup>. For a sphere moving parallel to the walls along the center of the channel, the drag enhancement is estimated to be 3.5 from Figure 9 of reference <sup>4</sup> using the experimental aspect ratio,  $H/a \approx 3$ . With this correction, the drag force becomes  $F_{vis} \approx 11\pi d\eta u$  as quoted in the main text.

### VIII. Dependence of Orbital Mixing on Particle Size

To better assess the role of particle size on the efficacy of orbital mixing, we simulated the movement of a spherical particle (radius a) about a circular orbit (radius R) oriented perpendicular to the interface between two viscous fluids (Figure S3). Using the Stokes flow field for a translating sphere<sup>5</sup>, we integrated the dynamics of the interface (neglecting diffusion) as the particle completed multiple orbits. Briefly, the interface was seeded uniformly with tracer "particles" (infinitesimal points), which moved along at the local fluid velocity determined by the movement of the sphere. The position of these tracer particles was integrated in time using a variable order Adams-Bashforth-Moulton PECE solver (MATLAB's ode113). The tracer particles were periodically redistributed along the interface by linear interpolation to maintain a roughly uniform distribution.

Figure S3 shows the effect of particle size on mixing kinematics. Here, the images correspond to planar slices oriented perpendicular to the initial interface and parallel to the circular orbit of the particle. These planar slices are displaced from the z = 0 plane by a distance of 0.1*R*. For small particles (a < 0.1R), the fluids remain poorly mixed even after n = 7 orbits of the particle. By contrast, larger particles create disturbances that extend farther from the plane of their orbit to stretch and fold the two fluids – thereby reducing the length scale over which diffusion must act. Interestingly, as the size of the particle becomes commensurate with the size of the orbit ( $a \sim R$ ), the particle becomes less effective at stretching and folding the two fluids. These observations suggest that there may be an optimal geometry (*i.e.*, optimal ratio a / R) that maximizes the efficacy of orbital mixing for a prescribed number of orbits.



**Figure S3**. Mixing kinematics of a spherical particle moving in a circular orbit between two fluids (blue and white). The image at the upper left shows the initial position of the interface and the sphere. The panel below shows the distribution of the two fluids within the plane z = 0.1R for different particle radii *a* and number of revolutions *n*.

### IV. Chaotic vs. Non-Chaotic Mixing

The most effective mixers rely on chaotic flows the cause neighboring fluid elements to diverge in time at an exponential rate. To investigate whether or not the present orbital mixers satisfy this criterion, we examined the kinematic model presented in the previous section, which describes the Stokes flow due to a single sphere moving in a circular orbit within an unbounded fluid. We integrated the motion of two fluid elements and examined how the distance between these two points varied in time; this process was repeated for several different initial locations. The results of this analysis are illustrated in the figure below. Neighboring points do diverge in time but at a constant rate – not exponentially as expected for chaotic mixing. By contrast, if the particle is made to follow a figure eight-type orbit, mixing becomes chaotic as evidenced by the exponential divergence of neighboring elements.



Figure S4. (a) Mixing due to a spherical particle of radius *a* moving through a viscous fluid along a circular orbit in the *xy* plane. The position of the sphere as a function of time is given by  $\mathbf{r}_s = [R \sin \omega t, R \cos \omega t, 0]$ , where R = 5a and  $\omega$  is the frequency of the sphere's orbit. The blue curve traces the motion of a fluid element in time. The plot on the right shows the distance

between two neighboring elements,  $d(t) = |\mathbf{r}_1(t) - \mathbf{r}_2(t)|$ , initially separated by a distance of  $d(0) = 10^{-6}a$ . This distance increases linearly in time. (b) Chaotic mixing due to a sphere moving along a figure eight orbit  $\mathbf{r}_s = [R \sin \omega t, R \sin 2\omega t, 0]$ , with R = 5a. Two fluid elements (red and blue) are initially separated by a distance  $d(0) = 10^{-8}a$  but diverge exponentially in time at a rate of 0.2 $\omega$  as shown in the plot on the right (note that logarithmic spacing on the *y*-axis).

In sum, this simple model suggests that orbital particle motions at zero Reynolds number do not lead to chaotic mixing in an unbounded fluid. The experimental system, however, is complicated by the presence of the channel walls which strongly influence the fluid disturbance due to the moving particle. Therefore, it remains unclear whether or not the mixing observed in experiment is chaotic. Visually, it does appear that the fluid is being continuously folded and expanded as illustrated in Figure 2 (e.g., the "interface" between the two streams becomes larger and larger as the particle completes its orbits), which is characteristic of chaotic mixing flows.

# **Supplemental References**

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