

Print-and-Peel Fabrication of Microelectrodes

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Received June 5, 2008. Revised Manuscript Received July 6, 2008

We describe a facile and expedient approach for the fabrication of arrays of microelectrodes on smooth substrates. A sequence of print-and-peel procedures allowed for the microfabrication of capacitance microsenors using office equipment and relatively simple wet chemistry. Microfluidic assemblies with reversibly adhered elastomer components allowed for the transfer of patterns of metallic silver, deposited via Tollens' reaction, onto the substrate surfaces. Electroplating of the silver patterns produced an array of micrometer-thick copper electrodes. Capacitance sensors were assembled by placing nonlithographically fabricated flow chambers over the microelectrode arrays. Triangular-waveform current-voltage (I/V) measurements showed a linear correlation between the capacitance of the print-and-peel fabricated devices and the dielectric constant of the samples injected into their flow chambers.

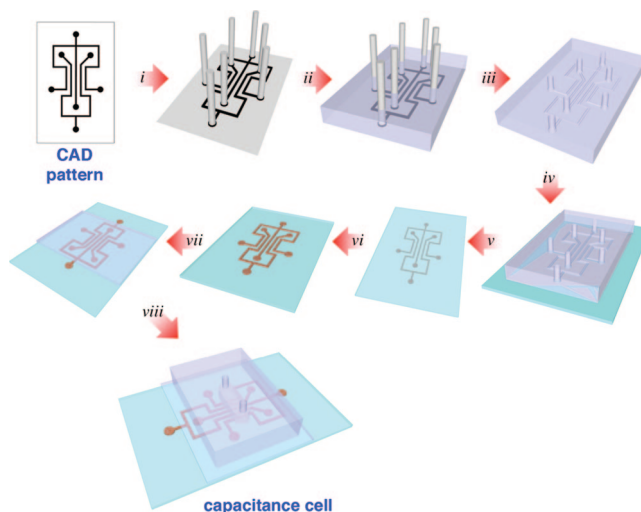
This letter describes a facile approach for fabrication of arrays of microelectrodes on smooth substrates. Microfluidic assemblies with reversibly adhered components allow for the transfer of patterns of conducting materials onto the substrate surface. Using this microfabrication approach, we prepared capacitance sensors and characterized their performance.

Nonlithographic, or print-and-peel (PAP), fabrication allows for facile and expedient prototyping of microfluidic devices.^{1–3} Unlike other fabrication techniques, PAP consists of direct printing of the masters, using regular office equipment, and attaching 3D elements to their surfaces.¹ Due to its simplicity and low cost, PAP offers valuable venues for research and development in an environment where microfabrication facilities are not readily available.

Impedance spectroscopy (IS) is an invaluable analytical tool for investigation of a broad variety of systems, such as complex liquids and biological samples.^{4–6} Electric interfaces are essential for microfluidic IS devices. A range of techniques, such as photolithography,⁷ sputtering,⁸ and microtransfer printing,⁹ are employed for the fabrication of microelectrodes for IS sensors. These fabrication methods, however, require specialized equipment and/or a clean-room environment.

Herein, we demonstrate the utilization of PAP for the microfabrication of IS sensors using an office printer and relatively simple wet chemistry. Direct printing of the computer-aided-design (CAD) pattern of microelectrodes to a polyester transparency film produced a master, to which we attached 3D elements for the inlet and outlet channels (Scheme 1i). To obtain

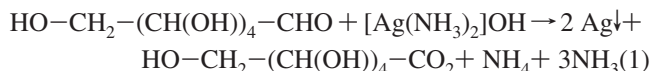
Scheme 1. Print-and-Peel Fabrication of a Capacitance Cell^a



^a (i) Master preparation; (ii) casting PDMS; (iii) curing of the PDMS "stamp"; (iv) reversible adhesion to glass; (v) consecutive flows of aqueous solutions of dextrose and $[\text{Ag}(\text{NH}_3)_2]^+$; (vi) electroplating with Cu; (vii) coating with PDMS; (viii) PDMS-to-PDMS permanent adhesion.

microchannels with improved contour smoothness, we utilized a solid-ink printer, instead of LaserJet printers, which we previously used for the PAP fabrication of microfluidic devices.¹ Casting polydimethylsiloxane (PDMS) over the master (Scheme 1ii) produced elastomer components with negative-relief imprints (Scheme 1iii). Reversible adhesion of the PDMS components to glass surfaces formed microfluidic assemblies (Scheme 1iv).

We used the Tollens' reaction to generate silver patterns on the glass surfaces. This process was named after Bernhard Tollens¹⁰ and involves the formation of metallic silver from an aqueous solution of diamminesilver(I) hydroxide, $[\text{Ag}(\text{NH}_3)_2]\text{OH}$, via a two-electron oxidation of aldehydes, such as dextrose (open form), $\text{HO}-\text{CH}_2-(\text{CH}(\text{OH}))_4-\text{CHO}$:¹¹



The reaction between the aqueous solutions of the reducing agent, dextrose, and of the Tollens' reagent, which were

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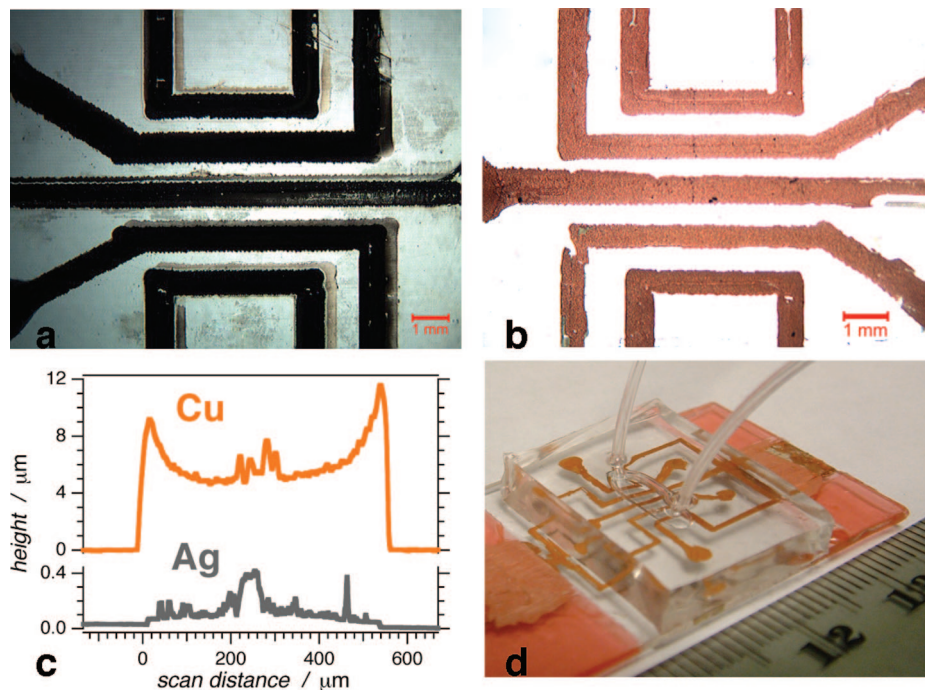


Figure 1. Print-and-peel fabricated capacitance cell. Reflection/transmission optical microscopy images of (a) silver and (b) copper electrode arrays. (c) Profilometry traces across silver (three consecutive dextrose/[Ag(NH₃)₂]⁺ treatments) and copper electrodes. (d) Photograph of a cell with a 1.5 mm center-to-center electrode separation.

consecutively flown through the microchannels, resulted in the deposition of silver metal onto the channel walls. To ensure continuous coverage with conductive material, the procedure of passing [Ag(NH₃)₂]⁺ solution through the microchannels prewet with dextrose mixture was repeated three to five times (Figure 1a).

Detachment of the PDMS components left patterns of electroconducting silver layers on the glass surfaces (Scheme 1v, Figure 1a). The silver patterns, however, were rough and prone to mechanical scratching (Figure 1c). Therefore, using electroplating, we deposited a 10-μm-thick copper layers over the silver patterns, producing microelectrodes with improved mechanical stability (Scheme 1vi, Figure 1b,c).

In the middle of the patterns, the copper strips formed arrays of five parallel electrodes, which we utilized for impedance measurements. Electrical connections between every other electrode resulted in a “multilayer” capacitor, the characteristics of which were dependent on the dielectric properties of the surrounding media. IS allowed us to obtain information about the frequency dependence of the complex impedance, Z , of the microelectrode assemblies¹²

$$Z = Z_0 e^{i\varphi} = Z_S + Z_P + (R^{-1} + i2\pi fC)^{-1} \quad (2)$$

where Z_0 and φ are the amplitude and the phase of Z , respectively; f is the frequency of the sinusoidal wave of the applied voltage; Z_S and Z_P are the source (e.g., cable) and polarization impedance, respectively; and R and C are the resistance and the capacitance, respectively, of the microelectrode assembly. The capacitance, C , is directly dependent on the dielectric properties of the media surrounding the electrodes.

Placing water and other electroconductive samples directly into contact with the electrodes causes a considerable decrease in the resistance, R , of the capacitance cells. Hence, it results in an overwhelming contribution from R^{-1} in the last term of eq

2, making the contribution from C in the measured signals negligible. To prevent direct electrical contact between the microelectrodes and the samples, we coated the electrode arrays with 70 (±10) μm PDMS layers (Scheme 1vii). Over the coated electrodes we placed PDMS flow chambers, which were also fabricated using PAP techniques (Scheme 1viii). We prepared capacitance flow cells with 1 and 1.5 mm center-to-center electrode separations that correspond, respectively, to about 450 and 950 μm separations between the edges of neighboring electrodes.

Because we are interested in the properties of the media in the flow chambers above the electrodes, we modeled each cell as two capacitors connected in parallel with total capacitance, C , of

$$C = C_C + C_S = C_C + \alpha\epsilon \quad (3)$$

The representation of the cells as parallel-connected capacitors is the simplest model that allows for a linear relation between the measured capacitance, C , and the dielectric constant of the sample, ϵ . C_S accounts for capacitance resulting from the penetration of the electric field through the sample flow chambers. Through C_S , therefore, C is linearly dependent on the dielectric constant of the sample. The proportionality parameter, α , depends on the shape, the size, and the position of the flow chamber over the electrodes. C_C accounts for the capacitance of the rest of the device and remains independent of the properties of the sample flown through the chamber of the capacitance cell.

For the PAP-fabricated capacitance cells, IS revealed phase values close to -90° and a nearly linear dependence of the amplitude on the frequency on a double logarithmic scale (Figure 2). These features indicate that the capacitance of the cells comprises most of the measured impedance. Fitting the impedance spectra with the $Z(f)$ function (eq 2) yielded the total capacitance, C , of the cells filled with different fluids. Consecutive linear fits of C versus the dielectric constant of the fluid (eq 3) produced the characteristics of the PAP-fabricated microelectrode capaci-

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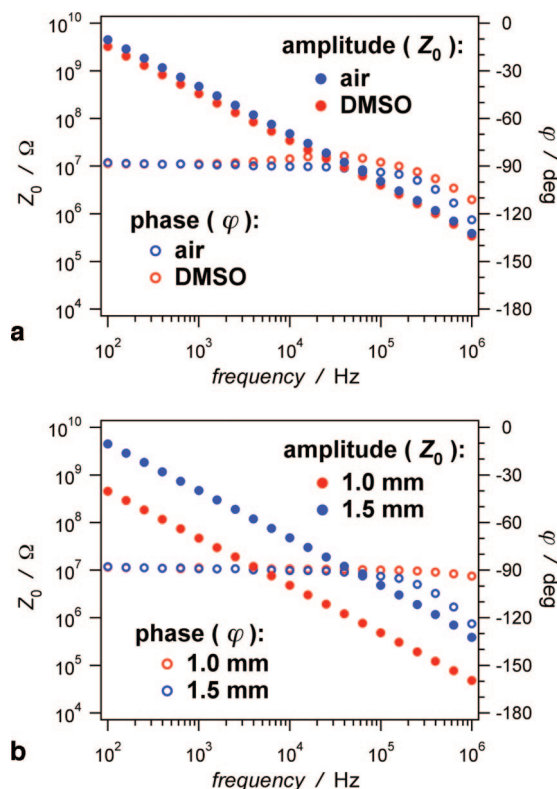


Figure 2. Comparison between impedance spectra for capacitance cells with different electrode separation filled with fluids with different dielectric constants, represented as the frequency dependence of the amplitude, Z_0 , and the phase, ϕ , of the measured complex impedance, $Z = Z_0 e^{i\phi}$ (eq 2). (a) Spectra for air and DMSO measured with a capacitance cell with a 1.5 mm center-to-center electrode separation. The capacitance values for air and DMSO are 0.34 and 0.48 pF, respectively. (b) Spectra for air measured with cells with 1.0 and 1.5 mm center-to-center electrode separations. The capacitance values for 1.0 and 1.5 mm electrode separations are 3.5 and 0.34 pF, respectively.

tance cells. From the intercept of the C versus ϵ linear fits, we determined that $C_C = 3.4$ and 0.34 pF for cells with 1 and 1.5 mm center-to-center electrode separations, respectively. The slopes of the linear fits gave values of 60 and 3.0 fF for the parameter α (eq 3) for cells with 1 and 1.5 mm center-to-center electrode separations, respectively.

Despite the successful application of IS for sensing, the relative complexity of the impedance model (eq 2) and the relatively long time required to collect a spectrum (seconds to minutes) pose a demand for an analytical technique in which the capacitance, modulated by the examined samples, can be directly and reliably extracted from the measured signals in an expedient manner. Triangular waveform (TW) dielectric measurements (i.e., application of TW voltage bias to the capacitance cells) yield current signals that are superpositions of two waves: (1) a triangular waveform resulting from the resistance component of the impedance, R , in eq 2 and (2) a rectangular waveform (RW) resulting from the capacitance component of the impedance, C , in eq 2.^{13,14} The height, h , of RW is linearly proportional to the capacitance, C , of the cell, and hence, to the dielectric constant, ϵ , of the sample. (The height, h , represents plateau-to-plateau current difference for RW; i.e., h is twice the value of the RW amplitude.) The lower limits of the duration of the measurement of TW dielectric response signals are compatible to the periods

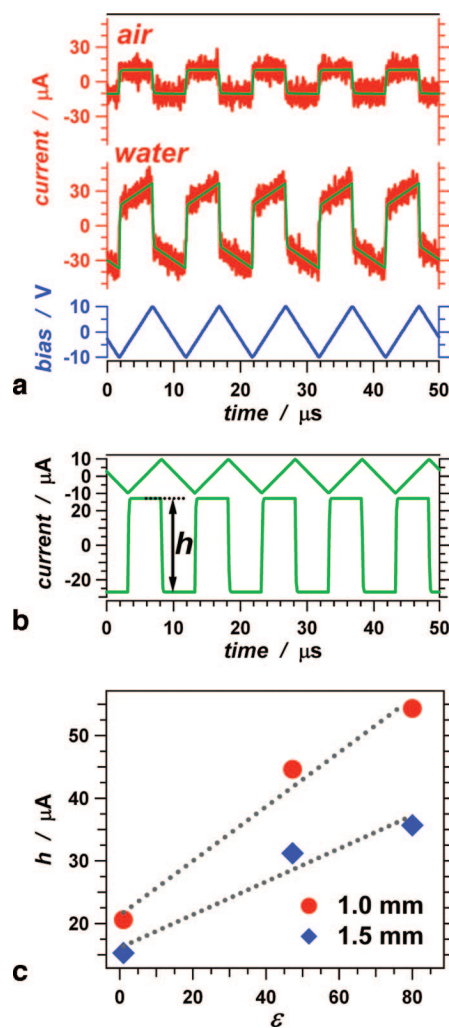


Figure 3. Triangular waveform (TW) current-voltage measurements. (a) Applied voltage (in blue) and current signals (in red) for air and water in a capacitance cell with a 1 mm center-to-center electrode separation. The data fits are shown in green. (b) Deconvolution of the current signal for water (from b) into triangular and rectangular waves. (c) Linear correlation between the height of the rectangular current waveforms and the dielectric constant of the fluid (air, DMSO, and water) in the sample chambers, measured with devices with 1 and 1.5 mm center-to-center electrode separations.

(i.e., f^{-1}) of the waves used (e.g., 10 μs for measurements using 100 kHz TW). This relatively high temporal resolution makes the TW technique an excellent choice for analytical and sensing applications, especially if real-time monitoring is required.

Using the TW technique, we characterized the performance of PAP-fabricated capacitance sensors. Measurements of air, dimethyl sulfoxide (DMSO), and water (i.e., fluids that do not swell PDMS)¹⁵ produced current waves that we deconvoluted into TW and RW components (Figure 3a,b). We observed a linear correlation between h and ϵ (Figure 3c), supporting our hypothesis that, despite its simplicity, the two-parallel-wired capacitor model (eq 3) adequately represents the investigated PAP-fabricated capacitance cells.

The described PAP technique offers a facile and inexpensive alternative for the fabrication of components for electrical interfaces of microfluidic devices. In addition to silver and copper, the procedure can be readily expanded to utilize the electroless deposition of other conducting and semiconducting materials, followed by electroplating of a wide range of metals.

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Acknowledgment. This work was funded by the U.C. Energy Institute, the NSF (for C.H. (REU)), and the U.S. Department of Education (for M.S.T. (GAANN)).

Supporting Information Available: Experimental details. This material is available free of charge via the Internet at <http://pubs.acs.org>.

LA801752K

Print-and-Peel Fabrication of Microelectrodes (Supporting Information)

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Materials

Pre-polymer of PDMS (Sylgard 184 silicone elastomer base kit) was purchased from Dow Corning Corporation. Microscope glass slides (75×25×1mm) and polyethylene tubing (ID 0.38 mm, OD 1.09 mm) were purchased from Fisher Scientific. Polyethylene line cords (0.64 mm diameter) and hot glue were purchased from a hardware distributor. Transparency films for a solid-ink printer were obtained from Xerox. Silver nitrate and dextrose were purchased from Sigma-Aldrich. Potassium hydroxide, ammonium hydroxide, sulfuric acid and hydrochloric acid were obtained from Fisher Scientific.

Print-and-peel fabrication

Fabrication of the PDMS components. The CAD patterns of the microelectrodes were created using Adobe Illustrator. Using a solid-ink printer (Xerox Phaser 8550), the designed patterns were printed on overhead transparency films to form positive-relief masters. Upon immobilization of the masters to the bottoms of polystyrene Petri dishes, three-dimensional (3D) elements (posts of polyethylene line cords for the inlet and outlet channels) were glued to the circles at the termini of the patterned lines (Scheme 1,i).

PDMS prepolymer was vigorously mixed with the curing agent for at least five minutes and degassed under vacuum. The resultant mixture was poured over the printed masters with the 3D posts attached to them (Scheme 1,ii) and allowed to cure for a few hours at 40 °C. The cured PDMS slabs were detached from the master and the posts were removed from the slabs. The elastomer microfluidic components were cut from the PDMS slabs and cleaned with adhesive tape (Scheme 1,iii).

Preparation of silver patterns. Tollens' reaction was used for generating silver patterns on glass surfaces.

Preparation of the solutions for the Tollens' reaction. A drop of aqueous solution of ammonium hydroxide (20%) was added to 1 mL aqueous solution of silver nitrate (0.10 M), resulting in the formation of a brown precipitate. Upon vigorous shaking the solution became clear. 0.5 mL aqueous solution of potassium hydroxide (0.80 M) was added to the silver nitrate mixture resulting in a dark brown precipitate. Ammonium hydroxide solution was added dropwise until the precipitate dissolved. (Excess of NH_3 should be avoided.)

Caution: Aging of Tollens' reagent leads to the formation of silver azide, which is violently explosive. To avoid the formation of such hazardous salts, always freshly prepare only small amounts of the ammonical silver reagent. After use, quench the excess reagent with dilute acid and dispose it properly. Never store the leftover Tollens' reagent.

For the silver deposition, a reducing reagent is required (eq 1). For this purpose we prepared an aqueous solution of dextrose (0.25 M).

Deposition of silver patterns. The surfaces of glass slides were cleaned and scratched with sand paper to increase the adhesion area for the silver. The surfaces of the PDMS microfluidic components (with the negative-relief patterns) were cleaned with an adhesion tape and without any further treatment, pressed against the glass surfaces resulting in reversible adhesion between the PDMS and the glass. The PDMS-glass assemblies were kept under vacuum at 60 °C for several hours prior to use (Scheme 1,iv).

Using a syringe pump (Harvard Apparatus Pico Plus), dextrose solution was flown through each channel at a rate of 1 $\mu\text{L}/\text{min}$. Immediately after the dextrose solution reached the outlets of the channels, the inlet tubing was disconnected. Tubing primed with diamminesilver Tollens' reagent was connected to the same inlet. The Tollens' reagent was flown through the same channels (at rate of 1 $\mu\text{L}/\text{min}$) causing formation of dark depositions onto the channels walls. To assure the deposition of continuous silver layers, the procedure of consecutive runs of dextrose and diamminesilver solutions was repeated several times. At certain occasions, the PDMS components had to be detached from the glass slides and cleaned. The glass slides were rinsed with MilliQ water and blown dry with nitrogen. The PDMS components were cleaned with adhesive tape, realigned with the silver patterns and reversibly readhered to the glass slides for more dextrose/diamminesilver treatments. Repeating the silver-deposition procedure for about three to five times resulted in continuous and electroconducting patterns (Scheme 1,v). Misalignments during the readhesion of the PDMS component to the glass with partially deposited silver pattern, resulted in thin layers of silver along the edges of the patterned strips (Figure 1a).

Electroplating copper. For the copper electrodeposition, the silver patterns were connected to copper tape coated with silver paint to assure a good electrical connection. The copper tape was covered with nonconducting tape leaving only the silver patterns exposed. Each of the glass slides with exposed silver patterns, along with copper plates, were wedged into a plate holder (with the silver patterns facing the copper plates). The slides and the plates were immersed into a bath containing copper sulfate (0.25 M), sulfuric acid (0.015 M) and hydrochloric acid (0.0014 M). The silver patterns on the slides were wired as cathodes and the copper plates as anodes. The current was set based on the area of the silver patterns, exposed to the solution (11.25 mA/cm^2 for thickness deposition rate of 10 nm/s). When the electrodeposition was completed, the slides with the formed copper strips on them were taken out of the bath, washed with copious amounts of D.I. water and blow-dry with nitrogen.

Assembly of capacitance cells. The arrays of copper microelectrodes were covered with PDMS prepolymer (mixed with curing agent and degassed). The slides were immobilized vertically, allowing the extra prepolymer mixture to flow off

their surfaces. Curing of the remaining prepolymer at 50 °C produced ~70- μ m-thick PDMS coatings covering the copper microelectrodes (Scheme 1,vii). The thickness of the PDMS coatings was determined from profilometry measurements across edges of spots where we peeled off the polymer.

PDMS flow chambers were fabricated using PAP approach. Masters for elliptic chambers were prepared from hot glue. Polyethylene posts were placed at the opposite ends of the chamber for inlets and outlets. PDMS prepolymer (mixed with curing agent and degassed) was cast over the chamber masters and allowed to cure under ambient conditions.

The PDMS-coated glass slides (with microelectrode arrays) and the rectangular elastomer slabs (with the chamber cavities imprinted onto their surfaces) were treated with oxygen plasma (40 mBar, 50 W RF power) for 40 seconds using a capacitively-coupled-discharge system (FEMTO, Diener Electronics). The elliptic chambers of the plasma treated elastomer slabs were aligned to span across the PDMS coated microelectrodes and the polymer surfaces were pressed against each other to form capacitance flow cells (Scheme 1,viii). To assure the permanent adhesion between the PDMS surfaces, the capacitance cell assemblies were kept at 120 °C under vacuum for about 12 hours.

Profilometry

A bench-top surface profilometer (Dektak 8, Veeco Systems), with 12.5- μ m radius stylus, was employed for the analysis of the silver and copper deposition onto the glass substrates. Each of the electrode strip patterns was placed perpendicularly to the direction of movement of the stylus in order to obtain transverse measurements. Several 2-mm scans separated 0.5-1 mm along the strip were performed for each sample.

Impedance measurements

Impedance spectroscopy (using sinusoidal current/voltage, I/V, wave forms) allowed us to characterize the intrinsic capacitance, C_C , and the parameters, α , of the cells (eq 3).

Triangular-waveform (TW) measurements were employed for measurements involving relatively polar samples. TW measurements allow for facile deconvolution of the capacitance component of the impedance from the current signal and hence, for fast dielectric measurements.^{13,14}

For the measurements, liquids that do not cause PDMS to swell were chosen.¹⁵

Impedance spectroscopy. Impedance measurements were conducted using a Reference 600™ Potentiostat/Galvanostat/ZRA (Gamry Instruments, PA, U.S.A.). The microelectrode capacitance cells were wired to the instrument through probes clipped to the copper tapes electrically connected to the electrodes.

Air and organic solvents (perfluoromethyldecalin, decamethyl-cyclopentasiloxane and DMSO) were injected in the flow chambers and the impedance spectra were recorded at frequency from 100 Hz to 1 MHz.

The values for the capacitance were extracted from frequency regions, throughout which the measured phase was near $-\pi/2$ by fitting to eq 2. (Negative-90-degree phase indicates that the capacitance is the principal component of the measured impedance. An increase in the solvent polarity or frequency resulted in significant change in the phase.) Linear fits of the measured capacitance vs. the dielectric constants of the samples, for each cell, yielded the intrinsic cell capacitance, C_C , and the flow-chamber parameter, α .

Triangular-waveform measurements. The voltage triangular waveform (10 MHz, 10 V) was supplied by a 30-MHz Synthesized wave function generator (DS345, Stanford Research Systems). The applied voltage bias and the current signal were measured using a 500-MHz digital oscilloscope (54616C, Hewlett Packard) connected to a PC via a GPIB interface. 1 M Ω termination, connected in parallel with the capacitance cell, was used for monitoring the voltage wave applied to the cell. 50 Ω termination, connected in sequence with cell (i.e., between the cell and the ground), was used for recording the signal from the current passing through the capacitance cell. The voltage response signal (recorded with the oscilloscope) was divided by 50 Ω to yield the values for the current waves.

The recorded waves (current vs. time) for each sample measurement were fit to a function, composed of weighed sum of in-phase triangular and rectangular waveforms (TW and RW, respectively). The RW function was composed of sequential monoexponential decays and rises with identical time constants. The obtained time constants were orders-of-magnitude smaller than the inverted frequency (i.e., 10^{-5} s) resulting in rectangularly-appearing waveform (Figure 2b). The pre-exponential parameters, obtained from the deconvolution data fits, were ascribed to the heights of the waves, h .