

Q1

Electroanalytical devices with pins and thread†

Q2 Cite this: DOI: 10.1039/c5lc00867k

 Ana C. Glavan,^a Alar Ainala,^a Mahiar M. Hamedj,^a M. Teresa Fernández-Abedul^{*ac} and George M. Whitesides^{*ab}

 Received 23rd July 2015,
Accepted 27th October 2015

DOI: 10.1039/c5lc00867k

www.rsc.org/loc

This work describes the adaptive use of conventional stainless steel pins—used in unmodified form or coated with carbon paste—as working, counter, and quasi-reference electrodes in electrochemical devices fabricated using cotton thread or embossed omniphobic R^F paper to contain the electrolyte and sample. For some applications, these pin electrodes may be easier to modify and use than printed electrodes, and their position and orientation can be changed as needed. Electroanalytical devices capable of multiplex analysis (thread-based arrays or 96-well plates) were easily fabricated using pins as electrodes in either thread or omniphobic R^F paper.

Introduction

Paper and thread have become increasingly attractive as substrates for the development of electroanalytical devices that are simple, portable, disposable and affordable.¹ Others^{2–8} and we^{9,10} have described the development of microfluidic paper-based electroanalytical devices (E μ PADs), and used this methodology to detect a wide variety of analytes, including small-molecule metabolites,^{6,9,11} metal ions,⁹ nucleic acids,² and serum proteins.¹⁰ Thread-based microfluidic analytical devices (μ TADs), first described by Shen,¹² and by us,¹³ have since been used for several bioassays with colorimetric detection.^{12,14–17} Fewer thread-based than paper-based analytical devices have been integrated using electrochemical detection.^{18–21}

One challenge in the development and accessibility of paper and thread-based electroanalytical devices is the ability to fabricate and integrate electrodes rapidly, efficiently and cost-effectively. The most widely used methods for fabrication of electrodes in paper-based^{2,4–11} and thread-based^{19,20} electroanalytical devices (*e.g.* screen printing, stencil printing, rotogravure, and chemical vapor deposition with shadow masking) require custom-patterned components such as screens, stencils, or masks to deposit conductive materials on the surface of a substrate.^{22–27} The fabrication of these custom-patterned components is time-consuming, and can be expensive. In the finished device, the position of the

electrode cannot be altered after fabrication has been completed.

Existing strategies for the fabrication of electrodes are particularly challenging on non-planar substrates such as embossed hydrophobic,^{28,29} and omniphobic paper,^{29,30} and thread.^{12,13,16,17,19,20} The deposition of conductive materials on porous media such as paper or thread can alter its interfacial energy, porosity, tortuosity, and wicking. Inhomogeneity in the distribution of fluid can also affect the stability of the liquid electrode interface and influence the performance of the electroanalytical devices.³¹ These effects are most significant in the case of thread, where the fluid flow is confined to one dimension, and might be among the reasons behind the few examples of electroanalytical devices that utilize thread^{19,20} (in contrast to the widespread interest in those that utilize paper^{2–11}).

We wished to develop a new class of electrodes that could expand the versatility of the paper and thread-based analytical systems developed by our group^{32,33} and by others.^{34–39} Adaptive use of prefabricated stainless steel pins—either unmodified, or coated with a thin layer of graphite ink—provides a simple solution to the problems of fabrication and integration of electrodes in a low-cost analytical device. Platinum, gold, copper and stainless steel wires have been used as electrodes in microfluidic electrochemical applications,^{3,40–43} but their integration with paper-based devices has so far been limited to the use of gold wires several microns in diameter reported by Fodick *et al.*³ These gold microwire electrodes might be difficult to manipulate due to their small size, and require treatment with highly oxidizing solution (piranha etch) immediately prior to use; these safety concerns might make them challenging to use by minimally trained personnel, or at the point of care.

We show that stainless steel pins can be used as electrodes in systems fabricated using either omniphobic R^F

^a Department of Chemistry and Chemical Biology, Harvard University, 12 Oxford Street, Cambridge, MA 02138, USA. E-mail: gwhitesides@gmwgroup.harvard.edu

^b Wyss Institute for Biologically Inspired Engineering, Harvard University, 60 Oxford Street, Cambridge, MA 02138, USA

^c Departamento de Química Física y Analítica, Universidad de Oviedo, Julián Clavería 8, 33006 Oviedo, Asturias, Spain. E-mail: mtfernandez@uniovi.es

† Electronic supplementary information (ESI) available. See DOI: 10.1039/c5lc00867k

paper or thread; as electrodes, pins are sensitive and can be used to quantify metabolites (*e.g.*, lactate in human plasma). Their macroscopic size and minimal requirements for cleaning and post-processing make them easy to handle in settings where resources are sparse. Surprisingly, because they offer readily accessible connection points to electrochemical readers and easily modifiable configurations, pin electrodes allow the fabrication of devices suitable for multiplexed analysis. We demonstrate the fabrication of thread-based arrays that can be used to detect different analytes in the same array, or to perform multiple measurements of the same analyte simultaneously, or in close succession. We also describe the fabrication of a 96-well plate in omniphobic R^F paper that can be used to perform independent measurements in each well.

Results and discussion

Design of the R^F paper-based and thread-based devices incorporating pins as electrodes

Stainless-steel pins have several characteristics that make them attractive as candidates for adaptive use as electrodes in electrochemical devices. Stainless steel pins are inexpensive (less than \$0.001 per pin when purchased from commercial retailers, and much less if purchased wholesale) and available nearly all over the globe. Stainless steel is highly conductive and stable electrochemically in neutral or mildly acidic or basic aqueous solutions.^{47,48} Different parts of a pin (the head, the shaft and the sharp tip) can be used for different purposes: the head can serve as RE electrode in omniphobic paper-based devices, part of the stem can serve as CE electrode in thread-based devices, the stem can be used for connection to the potentiostat, and the sharp tip can be used to anchor the pins in a mechanical support. To generate an electroactive surface area of the working electrode that is sufficiently large to be useful for analysis, we coated a stainless-steel pin with carbon ink prepared by mixing graphite paste and solvent thinner with a multi-walled carbon nanotube powder⁴⁹ (details in the ESI[†]).

We have chosen paper and thread as a substrate for the fabrication of the electrochemical cells because they are widely available, inexpensive, lightweight, and flexible. We recently demonstrated that treatment with an organosilane in the gas-phase can be used to transform cellulose paper into a material resistant to wetting by a wide range of liquids, while leaving the characteristic properties of paper (*flexibility*, foldability, mechanical flexibility, porosity, large surface area, light weight, low cost) unaltered. The resulting “fluoro-alkylated paper,” or “ R^F paper” is omniphobic (*i.e.* both hydrophobic and oleophobic), and thus resists wetting by liquids spanning a broader range of surface tensions than paper treated with non-fluorinated alkyl trichlorosilanes (“non-fluorinated paper”, or “ R^H paper”). The combination of old and new properties allowed the use of R^F and R^H papers in a variety of new applications—open-channel microfluidics,^{45,46} printed electronics and MEMS,³⁰ and low-cost diagnostics.²⁸

Here, we took advantage of these properties of omniphobic R^F paper to develop a simple and flexible liquid storage system that provides sufficient mechanical support for the positioning of the pin electrodes, and can be manufactured inexpensively, using embossing⁴⁴ and gas-phase silanization.²⁹

We show in Fig. 1A the strategy used for the fabrication of an R^F paper-based electrochemical cell in which the pins—serving as working electrode (WE), reference electrode (RE), and counter electrode (CE)—were inserted in an embossed well. The electrodes were placed 0.1 in (~ 2.53 mm) away from one another, using a transparency with precut holes as an alignment tool. Fig. 1B shows the design of an electrochemical cell, in which pins (WE, RE, CE) are surrounded by helical turns of thread. To allow the solution of analyte to contact the electrodes, we used a micropipette to add a drop of liquid either to the embossed omniphobic well or to the thread.

In the omniphobic paper-based device, the liquid rests on the surface of the well, and forms an interface with the surfaces of the heads of the pins. Here the area between the head and the liquid determines the area of the electrode-fluid interface – which is the *effective* electrode area (Fig. 2A1 and A2).

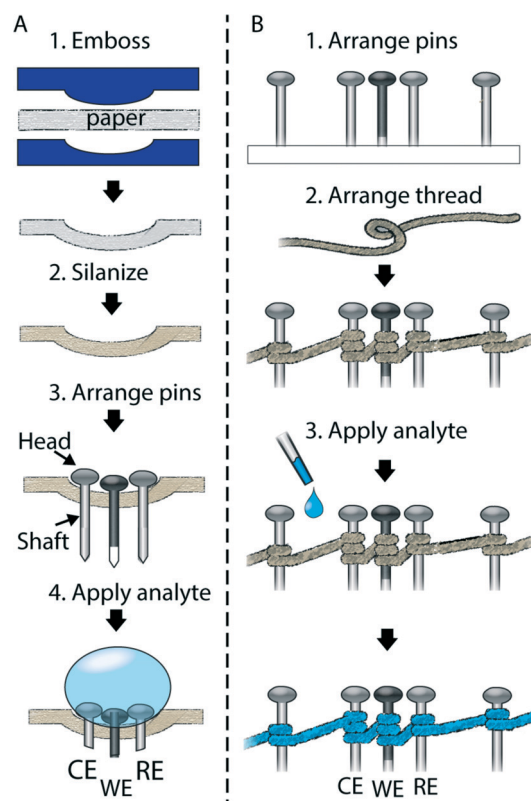


Fig. 1 A) Schematic representation of the process used for the fabrication of electrochemical cells in embossed omniphobic R^F paper. B) Schematic representation of the process used for the fabrication of an electrochemical cell with cotton thread. In both cases we use stainless steel pins as reference and counter electrodes (RE and CE), and a stainless steel pin coated with a graphite and carbon nanotube ink as working electrode (WE).

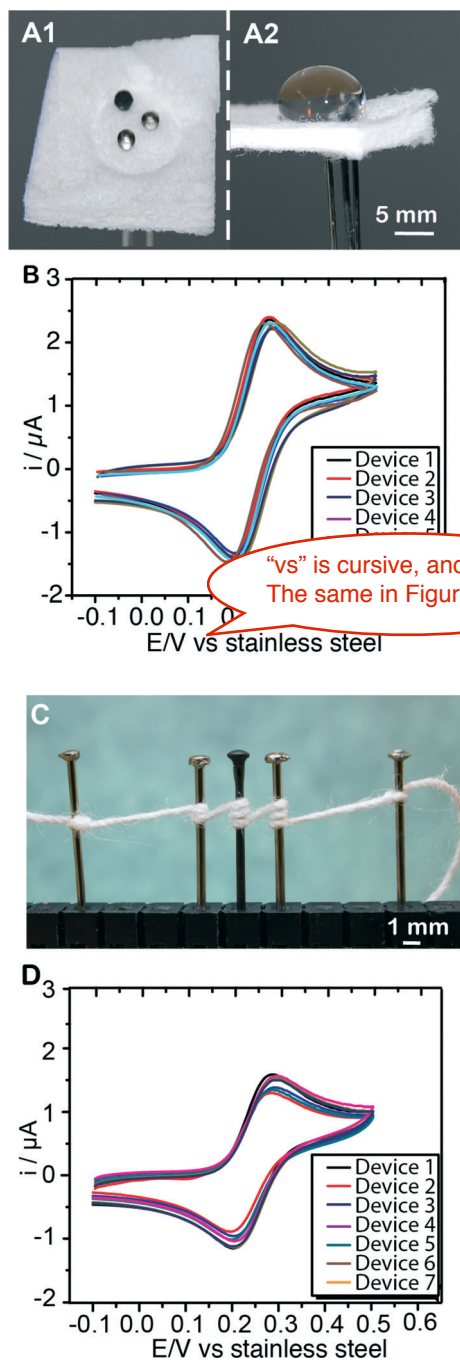


Fig. 2 (A1, A2) Photographs of an electrochemical cell fabricated using embossed omniphobic R^F paper and stainless steel pins as reference and counter electrodes (RE and CE), and a stainless steel pin coated with a graphite and carbon nanotube ink as working electrode (WE). The electrodes are placed at a distance of ~ 0.1 inch (2.53 mm) away from one another. (C) Photograph of an electrochemical cell fabricated using cotton thread and stainless steel pins as reference and counter electrodes, and a stainless steel pin coated with a graphite and carbon nanotube ink as working electrode. The electrodes are placed at a distance of ~ 0.1 inch (2.53 mm) away from one another. Cyclic voltammograms recorded in a $500 \mu\text{M}$ solution of FcCO_2H in $1\times$ -PBS, pH 7.6 at a scan rate of 100 mV s^{-1} using: (B) seven independent embossed omniphobic R^F paper devices, and (D) seven independent thread-and-pin arrays.

On thread, the liquid wicks along the thread and forms a cylindrical interface with the shaft of each pin (Fig. 2C), and the area of the thread in contact with the shaft determines the area of the electrode–fluid interface. The approximate geometrical areas of the paper and thread interfaces are 5 mm^2 and 4 mm^2 , respectively.

Evaluation

We evaluated the performance of the pins as electrodes in thread-based and omniphobic R^F paper-based electrochemical cells by recording the cyclic voltammograms (CVs) at a scan rate of 100 mV s^{-1} of a solution of a redox probe with well-characterized electrochemical behavior (ferrocene carboxylic acid, FcCO_2H). Fig. 2B and D show the variation in the CVs of the solution of FcCO_2H (at $100 \mu\text{M}$ in $1\times$ -PBS, pH 7.6) for paper-based and thread-based cells, each recorded using seven different devices. The device-to-device variation in the performance of the electrodes, measured from the anodic peak current i_{pa} , was in both cases, less than 10% as indicated by the relative standard deviation, RSD, (defined as the percentage ratio of the standard deviation to the mean of the distribution) of 6.3% (for an average current $\overline{i_{\text{pa}}} = 2.2 \mu\text{A}$) and 9.4% ($\overline{i_{\text{pa}}} = 1.6 \mu\text{A}$), for paper and thread-based cells, respectively.

We found that by controlling the number of helical turns, and the spacing between the turns, of the thread surrounding each pin electrode, we were able to control the area of the electrode–electrolyte interface and reduce the device-to-device variation (see the ESI† for details). For the thread-based cells we did not find the results to vary with the tension of the thread, at least within the range of tensions that allowed the formation of stable electrode–electrolyte interfaces between the thread and the pin electrodes and allowed the measurement of a stable current (although we did not explicitly measure this tension). We believe that the electrolyte interface between the thread and the pin, an area that is defined largely by the wetting properties of the two surfaces, determines the device-to-device variation.

To determine whether the electrochemical processes at the pin electrode–liquid interface in R^F paper and thread-based devices are diffusion-controlled, we recorded cyclic voltammograms of $500 \mu\text{mol L}^{-1}$ FcCO_2H in PBS, pH 7.6, at scan rates between 10 and 300 mV s^{-1} . The anodic and cathodic peak currents (i_{pa} and i_{pc}) were linearly proportional to the square root of the scan rate in both R^F paper ($R^2 = 0.991$ and 0.992 , respectively) and thread-based cells ($R^2 = 0.986$ and 0.989 , respectively) (see Fig. 3C). These results indicate that, in both cases, the rate of the electrochemical reaction at the surface of the pin electrode is governed by the diffusion of FcCO_2H to the surface of the electrode. These results are in agreement with results of finite-element simulations confirming that, for short diffusion distances and high scan rates, the geometry of the electrode (cylindrical or hemispherical, depending on whether the shaft or the head of the

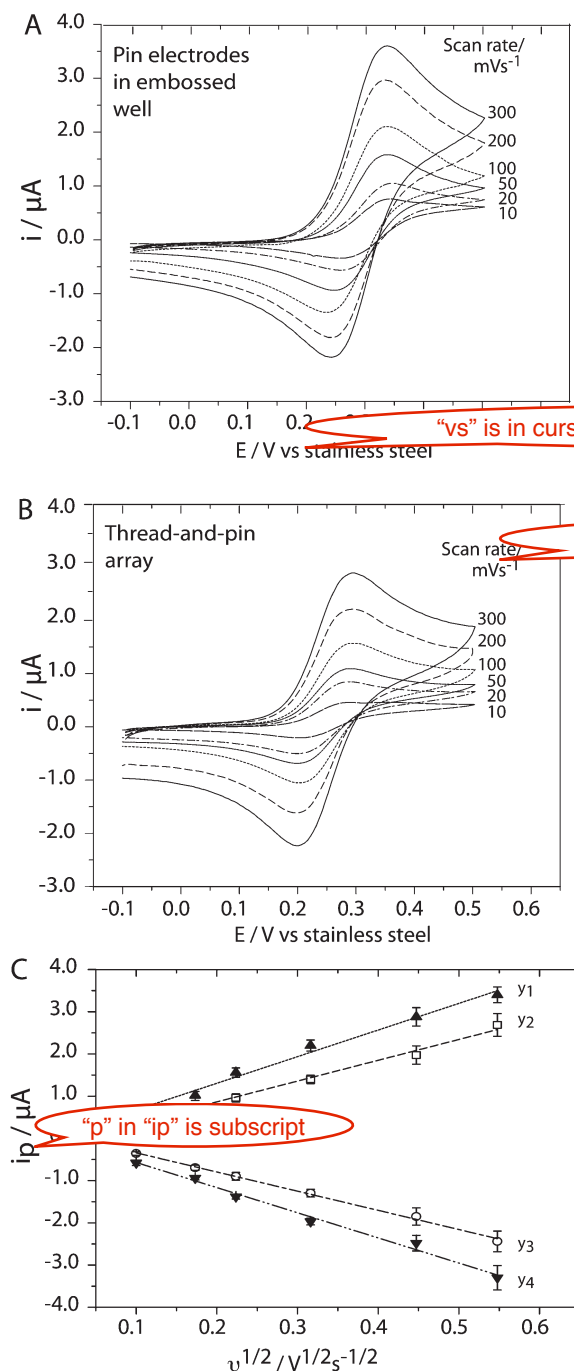


Fig. 3 Cyclic voltammograms of 500 μM FcCO_2H in 1 \times -PBS (pH 7.6) in electrochemical cells fabricated using (A) embossed omniphobic R^{F} paper and (B) thread, at various scan rates ascending along y-axis: 10, 20, 50, 100, 200, 300 mV s^{-1} . (C) The plot of anodic and cathodic peak currents vs. the square root of the scan rate ($v^{1/2}$). CV experiments conducted on an omniphobic R^{F} paper device (up-pointing triangle \blacktriangle , anodic peak current, down-pointing triangle \blacktriangledown , cathodic peak current) and in a thread device (circles \circ : anodic peak current, squares \square : cathodic peak current). The dashed lines represent linear regressions with equations: $y_1 = 6x \mu\text{A V}^{-1/2}/\text{s}^{1/2} + 0.1 \mu\text{A}$ [$R^2 = 0.991$], $y_2 = 5x \mu\text{A V}^{-1/2} \text{s}^{1/2} - 0.05 \mu\text{A}$ [$R^2 = 0.986$], $y_3 = -4x \mu\text{A V}^{-1/2} \text{s}^{1/2} + 0.05 \mu\text{A}$ [$R^2 = 0.989$], $y_4 = -6x \mu\text{A V}^{-1/2} \text{s}^{1/2} - 0.05 \mu\text{A}$ [$R^2 = 0.992$]. Error bars correspond to standard deviation of measurements performed using seven independent devices.

pin forms an interface with the solution of analyte) does not influence the process. Thus, under our experimental conditions, the electrochemical cells fabricated using pin electrodes and either thread or R^{F} paper can reproduce the classical diffusion-limited electrochemical processes reported in $\text{E}\mu\text{PADs}$ incorporating screen-printed electrodes.

These results also confirmed that the stainless steel was electrochemically stable and did not make any noticeable contribution to the electrochemical processes that occurred at the working electrode. We chose stainless steel pins, rather than pins made from less inert materials such as copper or silver, because they were highly stable under our experimen-

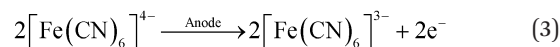
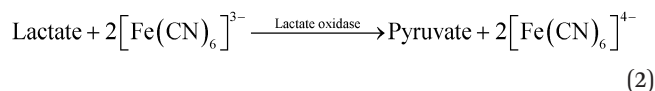
electrochemical processes typical of the oxidation of iron). The stability of stainless steel electrodes has also been reported by others.^{47,48} In addition, the surface of the stainless steel pins was coated with three layers of carbon ink, further reducing the risk of oxidation.

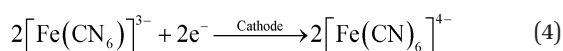
Applications in clinical diagnostics: analysis of L-lactate in human serum

We evaluated the feasibility of using R^{F} paper-based and thread-based devices to measure the concentration of a clinically relevant analyte, L-lactate, in human serum. The range of L-lactate concentrations relevant for diagnosis is between 0.5 and 15–20 mM in serum. We used chronoamperometry to perform this demonstration of principle because it is a simple and frequently used technique that provides a quantitative result. Cyclic voltammetry (CV) is less useful for accurate quantitation of electroactive species than chronoamperometric or pulse voltammetric techniques, because the correction for the capacitive current in CV is typically ambiguous.⁵⁰ Chronoamperometry measures current as a function of time at constant applied voltages, and starts with a large capacitive current that decays within the first few seconds. Faradaic current, which is proportional to the concentration of the analyte, becomes dominant, and decays according to the Cottrell equation (eqn (1)), where n is the number of electrons, F is Faraday's constant, A is the area of the electrode, D is the diffusion coefficient of analyte, C is the concentration of analyte, and t is time.

$$i = \frac{nFAD^{1/2}C}{\pi^{1/2}t^{1/2}} \quad (1)$$

In the lactate assay, potassium ferricyanide, $\text{K}_3[\text{Fe}(\text{CN})_6]$,





For each mole of lactate that is oxidized, two moles of $[\text{Fe}(\text{CN})_6]^{3-}$ are reduced to $[\text{Fe}(\text{CN})_6]^{4-}$; the latter can be quantified using chronoamperometry at an applied voltage of 0.4 V vs. a stainless-steel quasi-reference electrode. Fig. 4 shows the calibration curves for the measurement of L-lactate, for values between 1.1 mM (the value initially present in the serum) and 20 mM (with additional lactate spiked in the serum). The sensitivity is $0.08 \mu\text{A mM}^{-1}$ for the R^{F} paper- and $0.06 \mu\text{A mM}^{-1}$ for the thread-based device.^S

Approaches to multiplexing: electrochemical 96-well plate.

We prepared a 96-well plate capable of carrying out parallel analyses of different analytes, using embossed omniphobic R^{F} paper as a substrate, and pins as electrodes. Fig. 5 shows that different wells can be used to perform independent

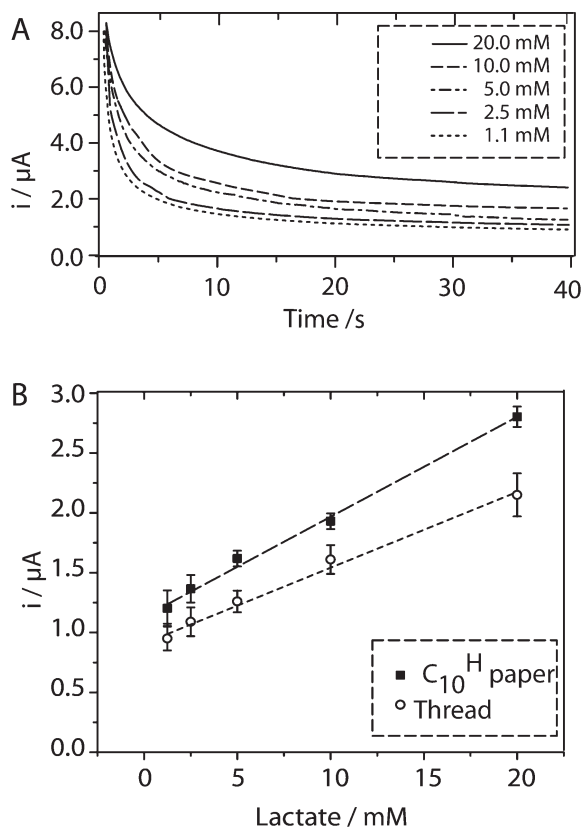


Fig. 4 (A) Chronoamperograms recorded on thread for the mixture of the enzymatic assay for the determination of lactate, at concentrations between 1.1–20 mM. The chronoamperograms were recorded at 0.4 V versus a stainless steel quasi-reference pin electrode. (B) The calibration plots of the currents recorded after 40 s as a function of concentration of lactate on thread-and-pins arrays and in wells embossed in omniphobic R^{F} paper. The dashed black line represents a fit to the equation: $y = 0.08x + 1.13$ ($R^2 = 0.994$) whereas the red line represents a fit to the equation: $y = 0.06x + 0.91$ ($R^2 = 0.987$), for concentrations of lactate between 1.1–20 mM. Error bars correspond to standard deviation of measurements performed using 7 independent devices.

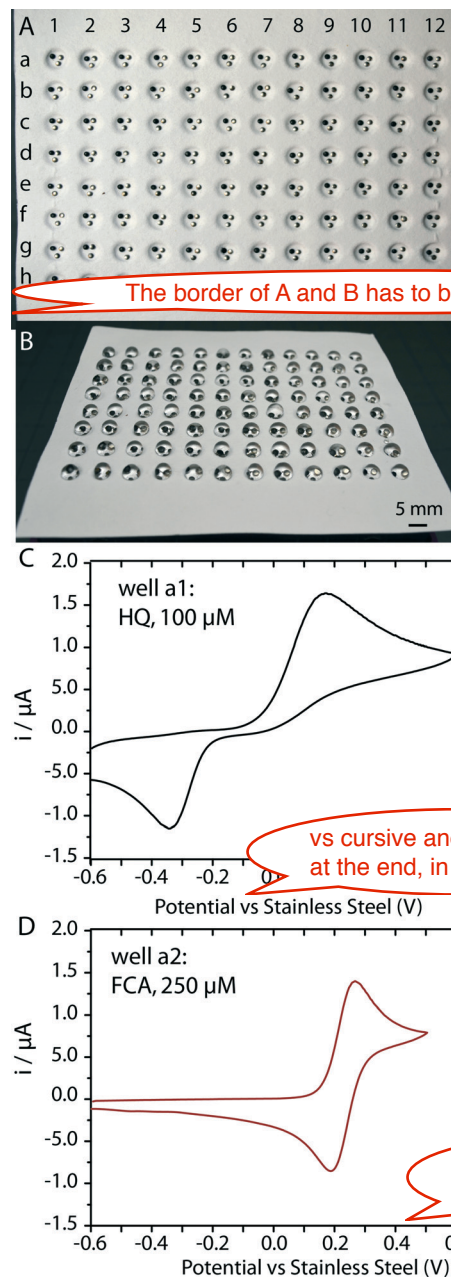


Fig. 5 Photography of a 96-well plate fabricated using embossed omniphobic R^{F} paper and pin electrodes. (A) before and (B) after 50 μL drops of an aqueous solution are added to each well. (C) Independent voltammograms recorded in different wells (a1 and a2) of the plate: a 100 μM solution of hydroquinone (HQ), and a 250 μM solution of FCA, both in $1\times$ PBS, pH 7.6, recorded at a scan rate of 100 mV s^{-1} ; the presence of a different analyte in a neighboring well does not interfere with measurements.

analyses—cyclic voltammetry for the analysis of solutions of FcCO_2H and hydroquinone, respectively.

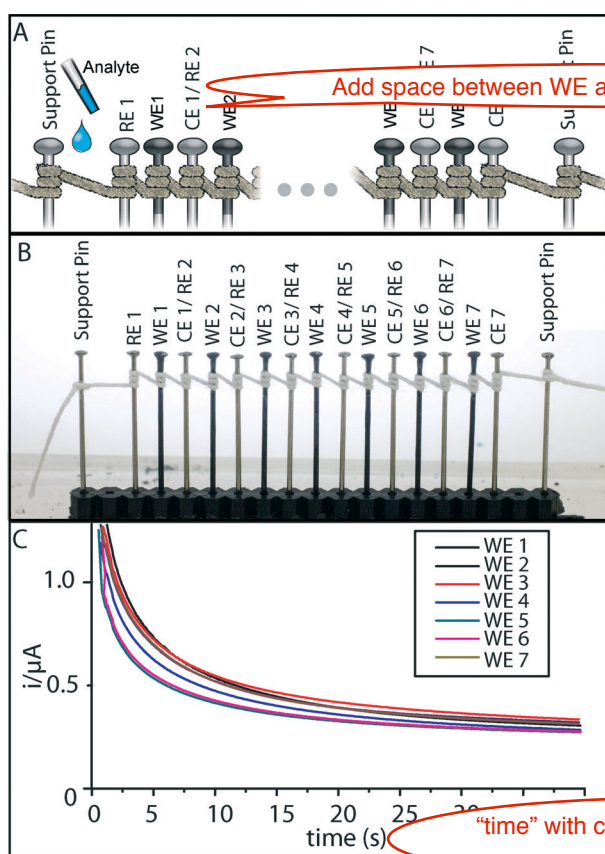
Approaches to multiplexing: thread-based arrays of pin electrodes

We produced linear arrays of electrodes (carbon-coated stainless steel pins as working electrodes, and stainless steel pins

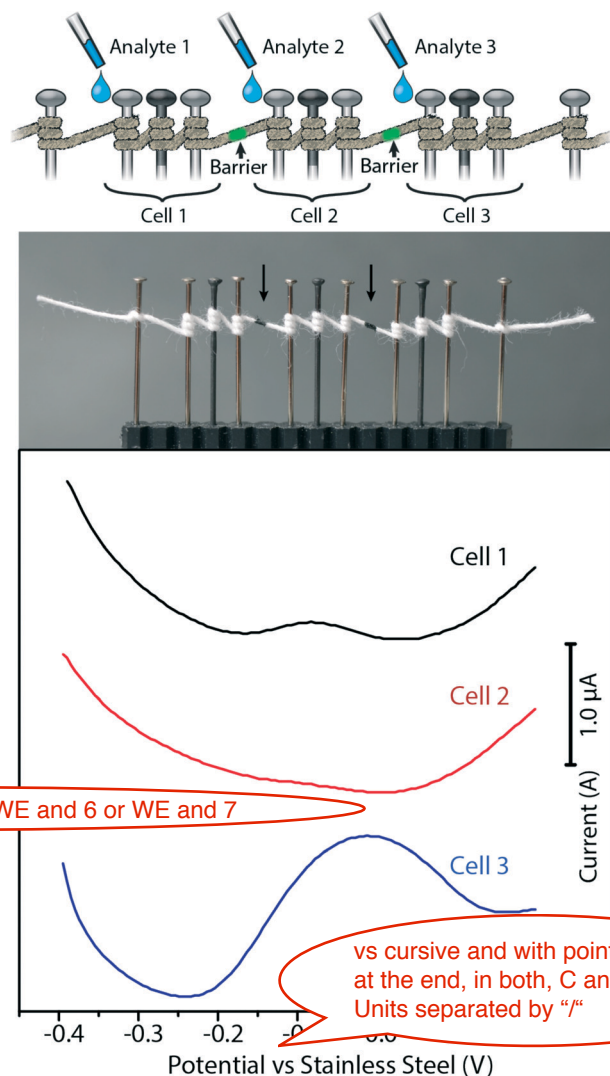
as either counter or quasi-reference electrodes) that form interfaces with the liquid wicking along the same thread. The electrochemical cells within the thread-based arrays can be either linked or independent, such that each cell in a multiplex device can be used to perform, in rapid succession or simultaneously, independent measurements for one or several solutions of analyte along the same thread.

Fig. 6 shows chronoamperograms for the same solution of analyte, recorded, in succession, using each of the seven WEs positioned along a single thread. In each measurement, the two adjacent stainless steel pins served as CE and RE, such as each two successive cells share one stainless steel pin that serves as a counter electrode in the former and as reference electrode in the latter.

By introducing hydrophobic barriers along the thread, we can form independent electrochemical cells capable of performing different analyses along the same thread. Fig. 7 shows square-wave voltammograms recorded with each of the three electrochemical cells along a single thread, for three solutions with different concentrations of analyte: 10



Q7 Fig. 6 (A) Schematic and (B) photograph of a device comprising multiple alternating stainless steel pins and carbon-coated stainless steel pins and a single thread that forms three helical turns around each pin. The device can be used for multiple measurements (in rapid succession) of the same analyte on the thread. (B) Chronoamperograms at 0.4 V recorded with each of the seven cells along the thread, using in each case the two adjacent stainless steel pins as CE and RE. Solution: 100 mM potassium ferricyanide in 1× PBS pH 7.6.



Q8 Fig. 7 (A) A schematic and (B) a photograph of a device comprising three distinct electrochemical cells formed from a single thread; arrows indicate the presence of a small amount of polymer (cyanoacrylate) that serves as a boundary between consecutive cells. The device can be used for measurement of three different analytes (in rapid succession or simultaneously). (C) Square-wave voltammograms recorded with each electrochemical cell, for: (1) left: 10 μM FCA in PBS, pH 7.6; (2) middle: buffer only, PBS, pH 7.6; (3) right: 100 μM FCA in PBS, pH 7.6. The center cell, where the thread is wet with buffer only, has no observable interference with neighboring cells.

μM FcCO_2H in PBS, pH 7.6; buffer only, PBS, pH 7.6; and 100 μM FcCO_2H in PBS, pH 7.6. There is no observable interference between neighboring cells.

“time” with capital at the beginning

One unmet need in the development of low-cost electroanalytical devices is a method to assemble and reconfigure devices “on-the-fly”, to meet the needs of specific applications and settings. The strategy we describe in this paper is based on electrodes that are by themselves nearly as ubiquitous, portable, inexpensive, and easily storable as are paper

1 and thread. The combination of stainless-steel pins—
 untreated or coated with a thin layer of carbon ink—and
 embossed omniphobic R^F paper or cotton thread, provides
 the basis for the fabrication of simple, versatile and low-cost
 5 electroanalytical **Capital** pin-based devices have four
 advantages: i) they are inexpensive, lightweight and portable.
 ii) They rely on stainless steel pins are mass-produced at very
 low cost and readily available. iii) ^{that} They are compatible with
 biological samples. iv) They eliminate the need for patterning
 10 electronic vias through the microfluidic substrate. The pins
 go through the paper, allowing three-dimensional electronic
 connections to a potentiostat or another instrument. This
 property is especially useful for multi-well electrochemical
 plates/substrates, where we believe the devices can inter-
 15 face with existing instrumentation for multiplexed electro-
 chemical readout.

With further development, the paper- and thread-based
 devices fabricated using this method have the potential to
 provide new functional options in clinical diagnostics, environ-
 20 mental monitoring, and microfluidic and electronic systems.

Acknowledgements

M. T. Fernández-Abedul thanks the Spanish Ministry of Econ-
 25 omy and Competitiveness (project MICINN CTQ2011-25814)
 for funding her stay at Harvard University. A. Ainla thanks the
 Swedish Research Council (VR) for a postdoctoral fellowship.
 A. C. Glavan acknowledges funding by the Bill & Melinda
 Gates Foundation, award # 51308 and DTRA award #
 HDTRA1-14-C-0037. M. M. Hamedí acknowledges support
 from Marie Curie IOF FP7 for project nanoPAD (Grant Agree-
 30 ment Number 330017).

References

- 1 E. J. Maxwell, A. D. Mazzeo and G. M. Whitesides, *MRS Bull.*, 2013, **38**, 309–314.
- 2 J. C. Cunningham, N. J. Brenes and R. M. Crooks, *Anal. Chem.*, 2014, **86**, 6166–6170.
- 3 S. E. Fosdick, M. J. Anderson, C. Renault, P. R. DeGregory, J. A. Loussaert and R. M. Crooks, *Anal. Chem.*, 2014, **86**, 3659–3666.
- 4 R. F. Carvalhal, M. S. Kfourí, M. H. Piazzetta, A. L. Gobbi and L. T. Kubota, *Anal. Chem.*, 2010, **82**, 1162–1165.
- 5 C. K. Tang, A. Vaze and J. F. Rusling, *Anal. Methods*, 2014, **6**, 8878–8881.
- 6 W. Dungchai, O. Chailapakul and C. S. Henry, *Anal. Chem.*, 2009, **81**, 5821–5826.
- 7 A. Apilux, W. Dungchai, W. Siangproh, N. Praphairaksit, C. S. Henry and O. Chailapakul, *Anal. Chem.*, 2010, **82**, 1727–1732.
- 8 M. Santhiago, C. S. Henry and L. T. Kubota, *Electrochim. Acta*, 2014, **130**, 771–777.
- 9 Z. Nie, C. A. Nijhuis, J. Gong, X. Chen, A. Kumachev, A. W. Martinez, M. Narovlyansky and G. M. Whitesides, *Lab Chip*, 2010, **10**, 477–483.

- 10 Z. Nie, F. Deiss, X. Liu, O. Akbulut and G. M. Whitesides, *Lab Chip*, 2010, **10**, 3163–3169.
- 11 H. Liu, Y. Xiang, Y. Lu and R. M. Crooks, *Angew. Chem.*, 2012, **124**, 7031–7034.
- 12 M. M. Hamedí, C. S. Henry, *ACS Appl. Mater. Interfaces*, 2009, **2**, 1–6. **Separated by commas, not points**
- 13 M. Reches, K. A. Mirica, R. Dasgupta, M. D. Dickey, M. J. Butte and G. M. Whitesides, *ACS Appl. Mater. Interfaces*, 2010, **2**, 1722–1728.
- 14 X. Li, D. R. Ballerini and W. Shen, *Biomicrofluidics*, 2012, **6**, 011301–011313.
- 15 D. R. Ballerini, X. Li and W. Shen, *Biomicrofluidics*, 2011, **5**, 014105.
- 16 G. Zhou, X. Mao and D. Juncker, *Anal. Chem.*, 2012, **84**, 7736–7743.
- 17 D. R. Ballerini, X. Li and W. Shen, *Anal. Bioanal. Chem.*, 2011, **399**, 1869–1875.
- 18 M. Hamedí, R. Forchheimer and O. Ingnas, *Nat. Mater.*, 2007, **6**, 357–362.
- 19 N. C. Sekar, S. A. Mousavi Shaegh, S. H. Ng, L. Ge and S. N. Tan, *Electrochem. Commun.*, 2014, **46**, 128–131.
- 20 Y.-C. Wei, L.-M. Fu and C.-H. Lin, *Microfluid. Nanofluid.*, 2013, **14**, 583–590.
- 21 T. Choudhary, G. P. Rajamanickam and D. Dendukuri, *Lab Chip*, 2015. **Q9**
- 22 J. P. Hart and S. A. Wring, *TrAC, Trends Anal. Chem.*, 1997, **16**, 89–103.
- 23 X. Xu, S. Zhang, H. Chen and J. Kong, *Talanta*, 2009, **80**, 8–18.
- 24 F. Eder, H. Klauk, M. Halik, U. Zschieschang, G. Schmid and C. Dehm, *Appl. Phys. Lett.*, 2004, **84**, 2673–2675.
- 25 D. Tobjork and R. Osterbacka, *Adv. Mater.*, 2011, **23**, 1935–1961.
- 26 O. Dominguez Renedo, M. A. Alonso-Lomillo and M. J. Arcos Martinez, *Talanta*, 2007, **73**, 202–219.
- 27 D. M. Cate, J. A. Adkins, J. Mettakoonpitak and C. S. Henry, *Anal. Chem.*, 2014, **87**, 19–41.
- 28 A. C. Glavan, D. C. Christodouleas, B. Mosadegh, H.-D. Yu, B. S. Smith, J. Lessing, M. T. Fernández-Abedul and G. M. Whitesides, *Anal. Chem.*, 2014, **86**, 11999–12007.
- 29 A. C. Glavan, R. V. Martinez, A. B. Subramaniam, H. J. Yoon, R. M. D. Nunes, H. Lange, M. M. Thuo and G. M. Whitesides, *Adv. Funct. Mater.*, 2014, **24**, 60–70.
- 30 J. Lessing, A. C. Glavan, S. B. Walker, C. Keplinger, J. A. Lewis and G. M. Whitesides, *Adv. Mater.*, 2014, **26**, 4677–4682.
- 31 C. Renault, M. J. Anderson and R. M. Crooks, *J. Am. Chem. Soc.*, 2014, **136**, 4616–4623.
- 32 A. W. Martinez, S. T. Phillips, M. J. Butte and G. M. Whitesides, *Angew. Chem., Int. Ed.*, 2007, **46**, 1318–1320.
- 33 A. W. Martinez, S. T. Phillips, E. Carrilho, S. W. Thomas, H. Sindi and G. M. Whitesides, *Anal. Chem.*, 2008, **80**, 3699–3707.
- 34 E. M. Fenton, M. R. Mascarenas, G. P. Lopez and S. S. Sibbett, *ACS Appl. Mater. Interfaces*, 2009, **1**, 124–129.
- 35 H. Liu and R. M. Crooks, *J. Am. Chem. Soc.*, 2011, **133**, 17564–17566.

- 1 36 H. Liu, X. Li and R. M. Crooks, *Anal. Chem.*, 2013, **85**, 4263–4267. 44 H. Becker and C. Gartner, *Electrophoresis*, 2000, **21**, 12–26. 1
- 37 D. Zang, L. Ge, M. Yan, X. Song and J. Yu, *Chem. Commun.*, 2012, **48**, 4683–4685. 45 A. C. Glavan, R. V. Martinez, E. J. Maxwell, A. B. Subramaniam, R. M. D. Nunes, S. Soh and G. M. Whitesides, *Lab Chip*, 2013, **13**, 2922–2930.
- 5 38 X. X. Yang, O. Forouzan, T. P. Brown and S. S. Shevkoplyas, *Lab Chip*, 2012, **12**, 274–280. 46 M. M. Thuo, R. V. Martinez, X. Liu, M. B. J. Atkinson, J. F. Bloch and G. M. Whitesides, *Chem. Mater.*, 2014, **26**, 4230–4237. 5
- 39 P. Yager, T. Edwards, E. Fu, K. Helton, K. Nelson, M. R. Tam and B. H. Weigl, *Nature*, 2006, **442**, 412–418. 47 H. Ayoub, V. Lair, S. Griveau, P. Brunswick, F. Bedioui and M. Cassir, *Electroanalysis*, 2012, **24**, 1324–1333.
- 10 41 M. Castaño-Díaz, M. T. Fernández-Abedul and A. Costa-García, *J. Chromatogr. A*, 2006, 291–299. 48 A. U. Malik, P. C. Mayan Kutty, N. A. Siddiqi, I. N. Andijani and S. Ahmed, *Corros. Sci.*, 1992, **33**, 1809–1827. 10
- 42 M. Wojciechowski and J. Balcerzak, *Anal. Chim. Acta*, 1990, **237**, 127–133. 49 M. T. Fernández-Abedul and A. Costa-García, *Anal. Bioanal. Chem.*, 2008, **390**, 293–298.
- 15 43 M. T. Fernández-Abedul and A. Costa-García, *Anal. Chim. Acta*, 1996, **328**, 67–71. 50 A. J. Bard and L. R. Faulkner, *Electrochemical Methods*, John Wiley & Sons, New York, 2001. 15

20

20

25

25

30

30

35

35

40

40

45

45

50

50

55

55