

## Technical Notes

# Printed Circuit Technology for Fabrication of Plastic-Based Microfluidic Devices

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One of the primary advantages of using plastic-based substrates for microfluidic systems is the ease with which devices can be fabricated with minimal dependence on specialized laboratory equipment. These devices are often produced using soft lithography techniques to cast replicas of a rigid mold or *master* incorporating a negative image of the desired surface structures. Conventional photolithographic micromachining processes are typically used to construct these masters in either thick photoresist, etched silicon, or etched glass substrates. The speed at which new masters can be produced using these techniques, however, can be relatively slow and often limits the rate at which new device designs can be built and tested. In this paper, we show that inexpensive photosensitized copper clad circuit board substrates can be employed to produce master molds using conventional printed circuit technology. This process offers the benefits of parallel fabrication associated with photolithography without the need for cleanroom facilities, thereby providing a degree of speed and simplicity that allows microfluidic master molds with well-defined and reproducible structural features to be constructed in  $\sim 30$  min in any laboratory. Precise control of channel heights ranging from 15 to 120  $\mu\text{m}$  can be easily achieved through selection of the appropriate copper layer thickness, and channel widths as small as 50  $\mu\text{m}$  can be reproducibly obtained. We use these masters to produce a variety of plastic-based microfluidic channel networks and demonstrate their suitability for DNA electrophoresis and microfluidic mixing studies.

Microfluidic technology plays a key role in a myriad of miniaturized “lab-on-a-chip” analytical systems being developed for use in biological, chemical, and medical applications. Although a variety of techniques can be used to construct these devices (microcontact printing, selective etching, replica molding, injection molding), an ongoing need exists for improved fabrication processes capable of producing micrometer-scale features in a rapid and inexpensive manner with minimal dependence on specialized facilities and equipment. From a commercial standpoint, it is also highly desirable to shorten the time lag between

successive design, fabrication, and testing cycles in order to speed the development of new devices. Since many of the intermediate iterations of the design process are focused on testing individual fluidic components or sets of components, significant interest has emerged in exploring *rapid prototyping* approaches to accelerate and simplify the production of microfluidic devices at these critical stages.

Plastic-based substrates have become extremely popular for use in microfluidic applications owing to their favorable combination of low cost and relative ease of fabrication via *soft lithography* techniques that involve producing replicas of features patterned on the surface of a rigid mold or *master*.<sup>1</sup> These masters are typically constructed either by patterning dense photoresist materials<sup>2</sup> or by directly etching features into silicon substrates using conventional micromachining processes.<sup>3</sup> Although these techniques allow masters with micrometer-scale features to be reliably produced, the speed at which they are constructed can be relatively slow and access to cleanroom facilities is usually required for at least part of the process. Since each new device design requires the fabrication of a new master, the speed with which new masters can be fabricated often imposes severe limitations on the rate at which new fluidic designs can be built and tested.

Progress toward addressing the need for rapid prototyping technology has recently been made in several research groups. One approach, for example, involves fabricating masters by subjecting an epoxy-based substrate to a series of successive UV-curing steps.<sup>4</sup> In this process, an epoxy resin and photoinitiated cross-linker mixture is pipetted onto a brass block and covered with a polyester grating film pretreated with sodium dodecyl sulfate. A photomask is placed on top of the grating film and the epoxy is cured for 30 s, after which the un-cross-linked epoxy is removed by immersion in acetone at  $-20\text{ }^{\circ}\text{C}$  in a stream of  $\text{CO}_2$ . The patterned master is then dried under nitrogen, postcured under UV, and baked at  $50\text{ }^{\circ}\text{C}$  under vacuum for 2 h to generate features with heights ranging from approximately 30 to 60  $\mu\text{m}$ .

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A further variation on liquid-phase photopolymerization techniques is based on successive injection and removal of UV cross-linkable polymers.<sup>5</sup> Masters are fabricated by dispensing a small amount of prepolymer solution (containing a monomer and photoinitiator mixture) onto the surface of a silicon substrate and exposing it to UV illumination through a photomask. The uncured prepolymer in the masked areas is then removed with methanol, and the patterned master is baked for 1 h to harden the material. A similar method for fabrication of individual microfluidic devices consists of introducing the prepolymer solution into a glass or plastic cartridge attached to a microscope slide with an adhesive gasket. After exposure to UV light through a photomask, the uncured polymer mixture is extracted from the channels using a syringe and any remaining unpolymerized monomer is removed using methanol.

Another recently developed prototyping approach involves the use of a precision milling instrument to fabricate master molds by machining features into an aluminum alloy substrate.<sup>6</sup> An acrylic sheet is then pressed on top of the master and heated in an oven at 205 °C for 2 h to allow the material to sink into the machined areas in order to generate positive features on the acrylic surface. This process is capable of producing channels with heights ranging from 7 to 20  $\mu\text{m}$ ; however the entire process including fabricating devices in PDMS takes  $\sim 8$  h, largely due to the serial nature of the milling process, which scales with device complexity.

Finally, a fabrication technique involving direct imprinting by pressing a mold onto a thin polystyrene film spin-coated onto a glass or plastic substrate has recently been demonstrated.<sup>7</sup> Channel heights generated from this technique range from approximately 10 to 15  $\mu\text{m}$ . While this technique is capable of producing submicrometer-scale features, the pressing process is sensitive to both the mechanical characteristics of the polystyrene film and the degree of precision at which contact with the mold can be reproducibly achieved. The need to start with a preexisting mold also introduces additional complexity.

While each of these prototyping processes is useful for specific applications, they often involve multiple steps and are generally not capable of achieving high levels of parallel fabrication. In this paper, we address these issues by demonstrating that master molds can be easily constructed using photosensitized printed circuit board (PC board) substrates, thereby enabling plastic-based microfluidic components to be rapidly built and tested at minimal expense.<sup>8</sup> These PC board materials are readily available and allow construction of fluidic channel structures at discrete and well-controlled heights ranging from 15 to 120  $\mu\text{m}$  through selection of the appropriate copper layer thickness—a benefit that is not as easily available with other rapid prototyping techniques. Using this capability, and without the need for specialized laboratory equipment, we are able to execute a complete process cycle from master fabrication to device fabrication, assembly, and testing in under 1 h. These masters can be used repeatedly with minimal degradation, and the parallel nature of these techniques allows many devices to be cast simultaneously. We apply these techniques to construct a variety of plastic-based microfluidic channel networks suitable for DNA electrophoresis and mixing applications.

Table 1. Copper Weight and Thicknesses of Standard PC Board Substrates<sup>8</sup> Compared with Surface Profilometry Measurements

general terminology	area wt (oz/ft <sup>2</sup> )	foil thickness ( $\mu\text{m}$ )	measd thickness <sup>a</sup> ( $\mu\text{m}$ )
1/2 oz	0.5	17.2	15
1 oz	1	34.3	29
2 oz	2	68.6	58
4 oz	4	137.2	120

<sup>a</sup> Note that the measured foil thicknesses are  $\sim 85\%$  of the reported value. This deviation is attributable to factors influencing the precise composition of the copper foil layer, including the presence of impurities and details associated with the deposition and lamination processes. Excellent reproducibility was observed among feature heights in all PC boards used in our experiments.

## EXPERIMENTAL SECTION

**Master Fabrication.** PC boards are constructed by selective etching of an electrically conductive foil layer (usually copper) deposited onto the surface of a rigid insulating substrate, yielding a pattern of lines that allow interconnection of various electronic components. These circuit board layouts are generated using conventional photolithographic pattern transfer and wet etching processes. Since the standard copper foil layer thicknesses used in PC board manufacture are within the range of feature heights associated with many microfluidic channel network structures (Table 1), the possibility exists that PC board substrates could be used to rapidly and inexpensively produce master molds for soft lithographic fabrication of microfluidic devices.

To investigate this, we first created layouts incorporating a variety of microchannel geometries using Adobe Illustrator (Adobe Systems Inc.; San Jose, CA) and then produced transparency film photomasks with a 3166 dpi printer (Mika Color; Los Angeles, CA). PC boards were purchased precoated with a positive tone photoresist (1-oz copper foil; Circuit Specialists Inc., Mesa, AZ; 0.5-, 2-, and 4-oz copper foils; Injectorall Electronics Corp., Bohemia, NY) and exposed to UV illumination through the photomask for 90–120 s (flux  $\sim 4.5$  mW/cm<sup>2</sup>) to transfer the pattern onto the PC board. Note that, in circuit board nomenclature, foil thicknesses are typically specified in units of oz/ft<sup>2</sup>, or more commonly oz, representing the weight of copper foil per square foot of base material.<sup>9</sup> Following exposure, the PC boards were immersed for 90–120 s under gentle agitation in a developer solution prepared by mixing 3.5 mL of a 50% w/w aqueous sodium hydroxide solution (Fisher Scientific; Hampton, NH) with 500 mL of deionized water. Next, the PC boards were transferred to a vertical plastic tank containing an etching solution prepared by dissolving 150 g of ammonium peroxydisulfate crystals (certified ACS grade; Fisher Scientific, Hampton, NH) in 1 L of deionized water to etch away the exposed copper foil in the patterned areas. The etching tank was mounted on a hot plate in order to maintain the solution at a temperature of 40–55 °C, and an air pump was used to provide continuous agitation. Etching times increase with

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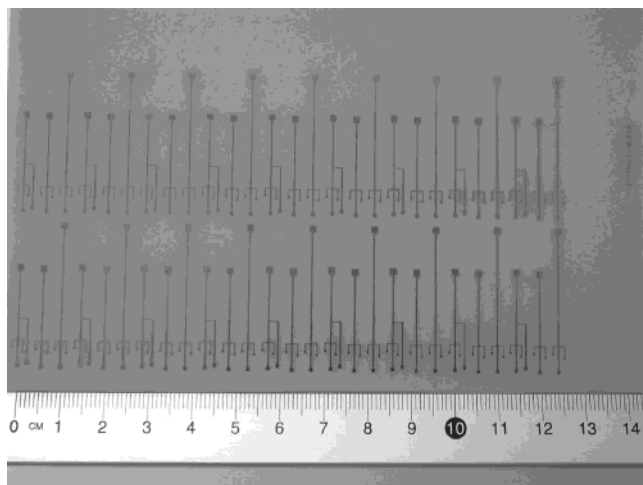


Figure 1. A 1-oz PC board master containing 54 individual electrophoresis channels, fabricated in  $\sim 20$  min. The main channels range from 2.3 to 3.3 cm in length with cross-sectional dimensions of  $400 \times 29.5 \mu\text{m}$ .

the thickness of the copper foil layer, ranging from 10 min for a 0.5-oz PC board to  $\sim 1$  h for a 4-oz PC board. After the etching process was completed, the remaining photoresist masking the channel structures was stripped by spraying the board surface with acetone from a squirt bottle for a few seconds. This setup allowed PC board masters with dimensions up to  $6 \times 6$  in. to be uniformly etched over the entire surface (Figure 1).

**DNA Electrophoresis.** Double-stranded DNA separations were performed using a 100-base pair standard ladder (Bio-Rad Laboratories, Hercules, CA) fluorescently labeled with YOYO-1 intercalating dye (Molecular Probes, Eugene, OR) at a dye-to-DNA ratio of 2:5. A  $1\times$  TBE solution (Bio-Rad Laboratories) was used as the running buffer. Fluorescence from the migrating DNA bands was detected using an Olympus SZX-12 fluorescence stereoscope with a mercury arc illumination source (Olympus America, Inc., Melville, NY) and imaged using a CCD camera (CCD-300; DAGE-MTI, Michigan City, IN). The camera output was recorded and digitized to allow intensity profiles corresponding to the migrating bands to be extracted by monitoring the variation in fluorescence intensity with time at a fixed on-screen location using our own MATLAB-based image analysis code (The MathWorks, Inc., Natick, MI).

Thermoreversible electrophoresis gels were formulated by dissolving 2.75 g of Pluronic-F127 powder (Sigma, St. Louis, MO) in 1 mL of  $10\times$  TBE (Bio-Rad Laboratories). Initially, a sufficient amount of deionized water was added to ensure that the powder was fully wetted. The mixture was then incubated in a refrigerator until the powder dissolved completely (usually 2–3 days), during which time additional water was added to the solution until a final volume of 10 mL was attained. Prior to loading, the electrophoresis devices were refrigerated at  $4^\circ\text{C}$  after which the gel solution was introduced by placing a drop at one end of the separation channel while simultaneously applying downstream vacuum. The vacuum was released once the liquid interface approached the injection tee, and the flow quickly stopped as the device temperature rose to room temperature. If the gel interface was not correctly positioned, the device was refrigerated for a few minutes, after which the reliquefied matrix could be aspirated away and a new

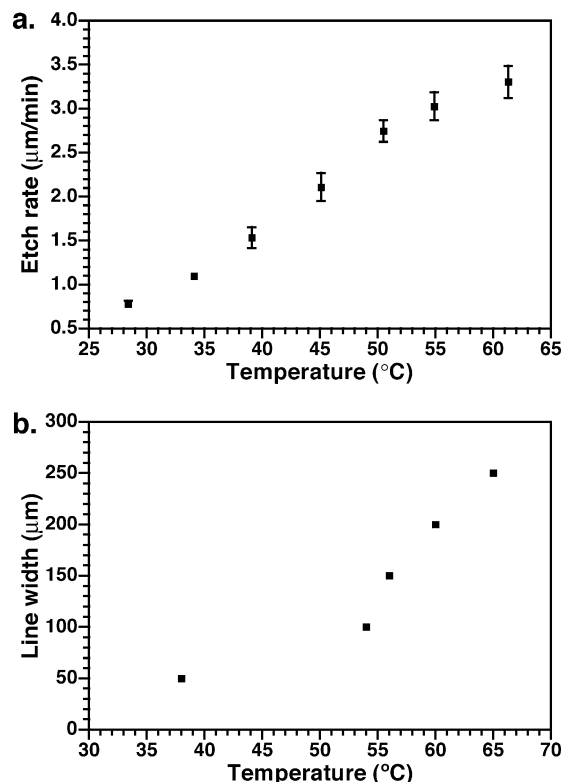


Figure 2. Characterization of the etching process in a 1-oz copper clad PC board. (a) Variation of etching rate with etchant solution temperature. Each data point is the average of five experiments, and the error bars indicate one standard deviation. (b) Minimum resolvable line widths as a function of etching temperature.

gel reloaded. The gel-loaded devices were then allowed to equilibrate under ambient conditions for at least 10 min before performing electrophoresis experiments. This approach allowed the separation matrix to be reproducibly positioned within the fluidic channel.

## RESULTS AND DISCUSSION

**Effect of Etching Conditions.** The etching rate is a critical factor governing both the achievable resolution and the surface profile of the copper features. Etching rates were measured over a wide temperature range by comparing the weight of  $7.5 \times 5.0$  cm bare copper (i.e., stripped of all photoresist) PC boards before and after incubation in the etchant solution (Figure 2a). While the highest etching rates were obtained in the vicinity of  $55\text{--}65^\circ\text{C}$ , it was not possible to reliably produce structures less than  $100 \mu\text{m}$  in width under these conditions. Finer structures can, however, be resolved at lower etching temperatures (Figure 2b). For example, line widths as small as  $50 \mu\text{m}$  could be fabricated using a 1-oz copper clad board at an etching temperature of  $38^\circ\text{C}$ , even though the same features experienced severe undercut or were etched away completely at higher temperatures. Line widths of  $100 \mu\text{m}$  or more could be consistently patterned at all temperatures studied. We found that it was difficult to reproducibly achieve line widths narrower than  $50 \mu\text{m}$ , even in the thinnest foil layers. This limitation arises as a consequence of the undercut associated with the isotropic etching process that, in the case of extremely narrow lines, severely erodes the photoresist and exposes a substantial portion of the patterned features to direct



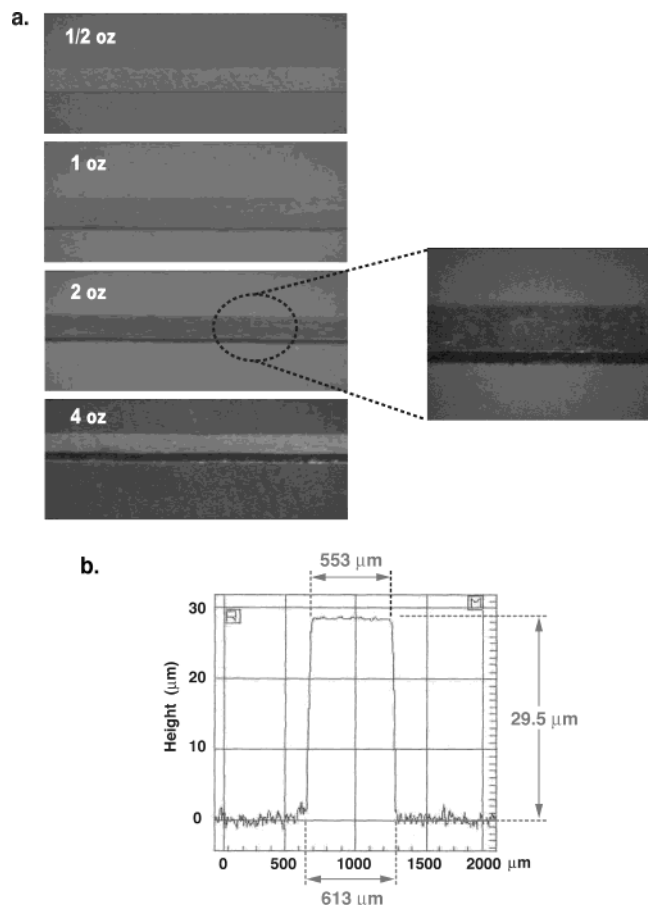


Figure 3. (a) Photographs of 600- $\mu\text{m}$ -wide etched copper lines on the surface of PC board masters incorporating various foil thicknesses. (b) Surface profile of a 600- $\mu\text{m}$ -wide line patterned on the surface of a 1-oz PC board at an etching temperature of 38  $^{\circ}\text{C}$ .

etching. Although it is possible that careful optimization of etching process parameters could allow narrower features to be produced, we note that the 50- $\mu\text{m}$  line widths achieved here satisfy the needs of numerous microfluidic applications and are compatible with the resolution limits of inexpensive transparency film-based photo-masks.

**Surface Characterization.** Figure 3a shows images of the etched structures on PC board masters fabricated using different copper foil thicknesses. Images were obtained using a Hirox 3-D microscope (Hirox-USA, River Edge, NJ). The geometries of the etched copper structures on PC board masters with line widths ranging from 50 to 600  $\mu\text{m}$ , fabricated using 1-oz copper clad boards at an etching temperature of 38  $^{\circ}\text{C}$ , were further characterized using a stylus profilometer (Dektak 3; Veeco Instruments, Woodbury, NY) (Figure 3b). The isotropic nature of the copper etching process yields features that exhibit a characteristic trapezoidal sidewall profile (Table 2). These data indicate that feature heights are reproducible within 1%, while feature widths are reproducible within 3–5%. Adjustment of additional parameters including etchant formulation and degree and uniformity of agitation could provide further optimization of the etching process.

**Microfluidic Device Fabrication.** We tested the PC board prototyping process by using PC board masters to cast replicas of microfluidic devices in poly(dimethylsiloxane) (PDMS) (Sylgard 184; Dow Corning, Midland, MI). The base and curing agent were

Table 2. Feature Sizes Produced on a 1-oz PC Board Determined Using Surface Profilometry (38  $^{\circ}\text{C}$  Etching Temperature)

on mask	line width ( $\mu\text{m}$ )		line ht ( $\mu\text{m}$ )
	on master (base)	on master (top)	on master
50	58.7	22.4	26.9
100	111.3	61.5	26.9
150	156.4	106.2	28.2
200	207.9	156.4	30.2
250	257.0	206.7	28.7
300	307.3	244.4	28.8
350	360.1	284.9	28.4
600	613.4	553.1	29.5

measured in a 10:1 ratio by weight, mixed thoroughly, and degassed under vacuum for 10 min, after which the mixture was poured over the master mold and cured for 1 h at 90  $^{\circ}\text{C}$ . Following curing, the devices were released by peeling from the master and fluidic access holes were punched using syringe needles. The devices were then mounted on glass microscope slides and filled with a colored dye to test for leaks in the channel network. Surface analysis using X-ray photoelectron spectroscopy indicated that no residual copper was transferred to the PDMS surface during the casting process.<sup>8</sup>

Although this process yielded working devices, a slight degree of surface roughness on the PC board's insulating substrate layer (at most  $\pm 1$   $\mu\text{m}$ , based on surface profilometry measurements) sometimes prevented sufficient adhesion of the PDMS device to the glass surface. These problems could be avoided by bonding the PDMS channels to a PDMS surface using the same techniques employed in the construction of complex multilayered structures.<sup>10</sup> A flat mounting surface for the devices was constructed using a 20:1 ratio of base to curing agent while the devices were fabricated using a 5:1 ratio of base to curing agent. The excess curing agent in the channel layer is sufficient to promote cross-linking with the excess resin in the base layer, so that a permanent bond can be formed between the two partially cured surfaces. The two mixtures were partially cured for 45 min at 80  $^{\circ}\text{C}$ , after which the device was mounted on the flat surface and cured for an additional 45 min at the same temperature. This process consistently yielded uniformly bonded microfluidic channels (Figure 4a,b).

To demonstrate the versatility of the PC board masters, we also used them to construct microfluidic devices using a melt-processable thermoplastic elastomer synthesized by dissolving a polystyrene-(polyethylene/polybutylene)-polystyrene (SBS/SEBS) triblock copolymer resin in a hydrocarbon oil for which the ethylene/butylene midblocks are selectively miscible.<sup>11</sup> The resulting gels are elastic solids at room temperature and share many desirable features of PDMS (e.g., biocompatibility, electric neutrality, optical transparency) but incorporate the additional advantage of melt processability when heated above a critical temperature ( $\sim 95$   $^{\circ}\text{C}$ ). Devices were fabricated by placing a slab of elastomer on top of a master mold that had been preheated to 120  $^{\circ}\text{C}$  on a hot plate. Once the elastomer began to soften, a glass plate was placed on top of the slab and gentle pressure was applied by hand to ensure complete contact with the structures on the

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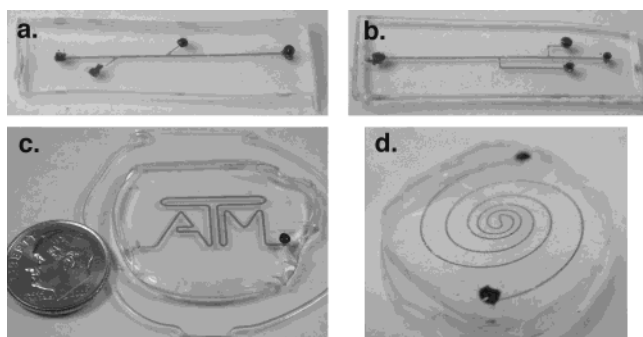


Figure 4. Examples of microfluidic devices fabricated from 1-oz PC board masters. Channels are filled with blue dye to enable visualization of the fluidic network. (a, b) PDMS-based devices incorporating 2.3-cm-long horizontal channels with cross-sectional dimensions of  $400 \times 29.5 \mu\text{m}$ . (c, d) Microfluidic channels constructed using a melt-processable elastomer incorporating cross-sectional dimensions of  $400 \times 29.5 \mu\text{m}$ .

mold. After cooling and release, the solidified gel incorporates the shape of the structures on the master (Figure 4c,d). For melt-processable materials, the slight degree of roughness on the PC board surface does not pose a problem because strong uniform bonds can be easily achieved, either with glass or elastomer surfaces, by briefly heating the material to a temperature just below its softening point using a handheld heat gun. Moreover, the entire fabrication process can be completed in  $\sim 5$  min using these elastomeric materials, compared to the hour required to cure PDMS. The devices can also be cleaned and remelted for subsequent reuse.

**DNA Electrophoresis.** The ability to perform size-selective fractionation of DNA fragments using gel electrophoresis is a key component of many biomedical and genomic analysis assays. We used 2-oz PC board masters to construct a series of electrophoresis microdevices from thermoplastic elastomer substrates incorporating fluidic channels  $400 \mu\text{m}$  in width by  $58 \mu\text{m}$  in depth arranged in a flow network consisting of an upstream sample injection region intersecting with a straight 1.7-cm-long separation section (Figure 5a). Electric fields within the channel were applied by using a dc power supply to induce a potential across platinum wire electrodes inserted into access holes located upstream and downstream of the separation channel.

To demonstrate separation of double-stranded DNA fragments in these devices, we selected sieving gels based on thermoreversible Pluronic-F127. These gels belong to a family of  $(\text{EO})_x(\text{PO})_y$   $(\text{EO})_x$  copolymers, which can be synthesized with different length poly(ethylene oxide) or  $(\text{EO})_x$  and poly(propylene oxide) or  $(\text{PO})_y$  blocks. Above a critical concentration and temperature, Pluronic solutions associate into micelles as hydrophobic interactions drive  $(\text{PO})_y$  segments into a nearly water-free central core surrounded by hydrated  $(\text{EO})_x$  tails.<sup>12,13</sup> Consequently, aqueous solutions of Pluronic copolymers are free flowing liquids at low temperatures ( $0$ – $5^\circ\text{C}$ ) and concentrations below 32% but self-organize into a transparent liquid-crystalline gel at room temperature. This thermoreversible behavior makes Pluronic copolymers attractive

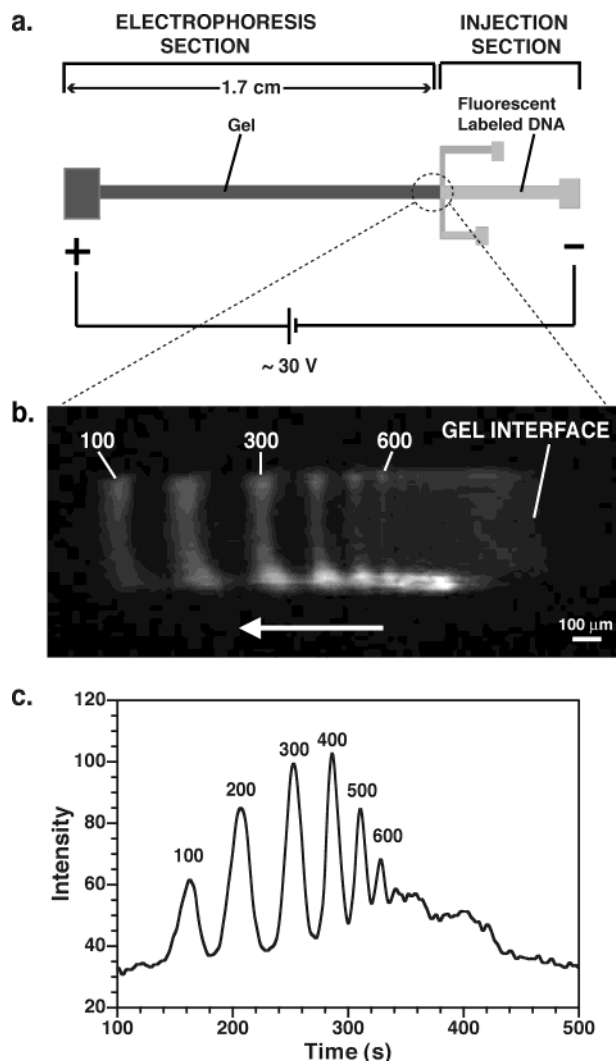


Figure 5. Illustration of the use of devices fabricated from PC board masters for DNA electrophoresis. (a) Schematic of the channel network consisting of an electrophoresis section loaded with a Pluronic-F127 gel matrix and an injection section loaded with a fluorescently labeled DNA solution. The width of the main horizontal channel is  $400 \mu\text{m}$ , the electrophoresis section is 1.7 cm long, and the injection section is 0.6 cm long. (b) Digitized image of a 100-base pair ladder separation ( $E = 15 \text{ V/cm}$ ). The arrow indicates the direction of DNA migration. (c) Electropherogram corresponding to the separation shown in (b) obtained at a distance of 2 mm downstream from the gel interface. Fragment sizes corresponding to each peak are indicated on the plot. All electrophoresis runs were performed at room temperature.

gel matrix candidates for DNA electrophoresis applications.<sup>14–18</sup> We selected Pluronic gels for initial electrophoresis experiments in part due to the ease of injecting the gel matrix into the separation channel and in part because of difficulties associated

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with casting in situ-polymerized cross-linked polyacrylamide gels, most likely as a consequence of inhibition of the polymerization reaction at the microchannel surfaces due to oxygen permeability of the plastic substrate material.<sup>19</sup>

Separation of a 100-base pair double-stranded DNA ladder was performed using a 27.5% Pluronic F-127 gel matrix. Electrophoretic injection was accomplished by applying an electric field of 15 V/cm between the ends of the separation channel for 10–20 s, resulting in compaction of the sample at the gel interface as a consequence of the discontinuity in electrophoretic mobilities inside and outside the gel. After a sufficient level of compaction had been achieved, the field was switched off and the injection ports were flushed and refilled with buffer solution, after which the field was reactivated at 15 V/cm for the duration of the separation run. Using this procedure, we were able to resolve dsDNA fragments in the 100–600-base pair range in a distance of 2 mm using low electric fields (Figure 5b,c). This level of separation performance is consistent with that previously demonstrated in microfabricated electrophoresis devices using Pluronic gels and is sufficient for a variety of applications including characterization and recovery of PCR products or restriction digestion fragments.<sup>20</sup>

**Diffusive Transport in Laminar Flow.** Under normal operating circumstances, fluid flow in microchannels is laminar and mixing occurs purely by the action of molecular diffusion. Consequently, it is nearly impossible to achieve adequate residence times for mixing to occur between parallel fluid streams without the use of extremely long channels. To study this phenomenon, we fabricated 600- $\mu\text{m}$ -wide microfluidic channels with various features to create obstructions in the flow using thermoplastic elastomer substrates and 1-oz PC board masters. These obstructions consisted of rectangular barriers (325  $\times$  400  $\mu\text{m}$ ) and circular poles (diameter, 300  $\mu\text{m}$ ) spaced at regular intervals of 3 mm (Figure 6a). Flow studies were carried out by imaging parallel streams of blue and yellow food coloring diluted to 0.01 g/mL (Adams Extract, Austin, TX), which became green at the interface where the two streams mixed. The degree of mixing could then be determined by measuring the increase in width of the green intermixed region with downstream distance. All experiments were carried out at flow rates of 0.05 mL/min generated using a syringe pump (PHD 2000; Harvard Apparatus, Holliston, MA).

As the flow encounters a barrier or pole, a sudden increase in the degree of mixing occurs, followed by a resumption of the behavior observed in an unobstructed straight channel (Figure 6b). It can also be seen that the streamlines associated with the laminar flow field are affected for some distance upstream and downstream of each obstruction. The rate at which the width of the green interface increases satisfies the following differential equation

$$\frac{\partial c}{\partial t} = D \frac{\partial^2 c}{\partial x^2} \quad (1)$$

where  $c$  is the concentration and  $D$  is the diffusion coefficient. Assuming a Gaussian concentration profile within the mixed

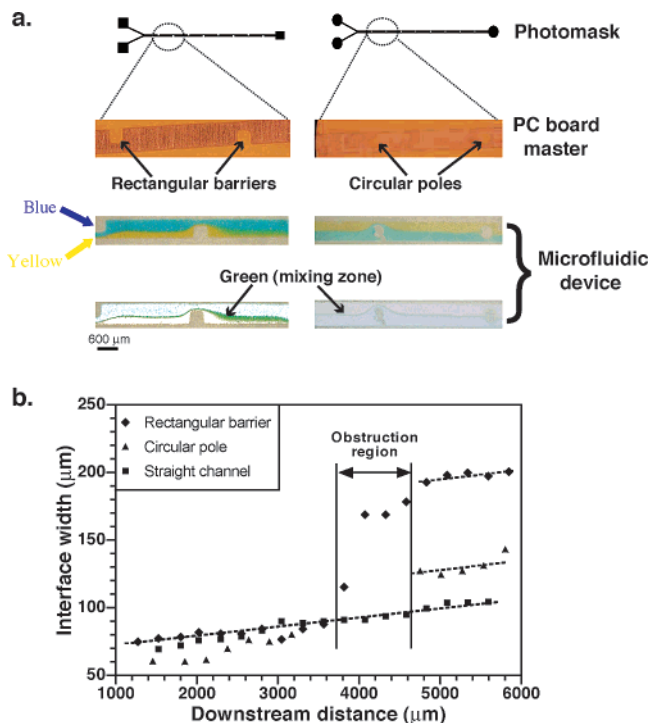


Figure 6. Illustration of the use of devices fabricated from PC board masters for studies of diffusive transport and mixing in microfluidic systems. (a) Device designs incorporating rectangular barriers along the channel sidewalls and circular poles along the channel centerline. Diffusive mixing was studied using 600- $\mu\text{m}$ -wide channels fabricated from a 1-oz PC board master by imaging the size of the green interface between parallel streams of blue and yellow dye. (b) Degree of mixing between blue and yellow streams as determined from the width of the green intermixed region as a function of downstream distance (measured with respect to an arbitrary fixed reference point). A sudden increase in the size of the mixing zone is observed when the flow encounters a barrier or pole. The zone in which the streamlines associated with the laminar flow field are influenced by the presence of the obstruction is indicated on the plot. Data were obtained at a flow rate of 0.05 mL/min. The dashed lines denote unobstructed regions of the respective fluidic channels and are intended to guide the eye.

interface, the effective width of the mixing zone,  $w$ , can be expressed in terms of the variance,  $\sigma$ , as follows.

$$w = 4\sigma \quad (2)$$

The diffusion coefficient can then be obtained from the rate of increase in the width of the mixing zone using the relationship

$$\sigma^2 = 2Dt \quad (3)$$

Applying this analysis to the unobstructed straight channel yields a value of  $D = 7.1 \times 10^{-6} \text{ cm}^2/\text{s}$ , close to the reported value of  $6.55 \times 10^{-6} \text{ cm}^2/\text{s}$  for the diffusion coefficient of tartrazine (yellow food color).<sup>21</sup> These results demonstrate the suitability of devices

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fabricated using PC board masters for use in studies of transport and mixing in microfluidic systems.

## CONCLUSIONS

In this paper, we show how conventional printed circuit technology can be used to quickly and reproducibly fabricate master molds for construction of microfluidic channel networks in a wide range of lengths, widths, and channel heights under normal laboratory conditions without the need for expensive equipment or cleanroom facilities. This process yields masters incorporating features as small as 50  $\mu\text{m}$  in width that can be used repeatedly with minimal surface degradation. These feature sizes satisfy the needs of numerous microfluidic applications and are compatible with the resolution limits of inexpensive transparency film-based photomasks. Master fabrication times depend on the thickness of the copper foil used and range from 10 min (0.5-oz PC boards; 15- $\mu\text{m}$  feature heights) to  $\sim 1$  h (4-oz PC boards; 120- $\mu\text{m}$  feature heights). The use of inexpensive copper clad boards that are precoated with photoresist offers a degree of simplicity unavailable with other rapid prototyping techniques. Potential future improvements include further optimization of the etching

process to allow fabrication features less than 50  $\mu\text{m}$  in width and the use of smoother PC board substrate materials (e.g., polyimides used in the construction of flexible circuit connectors) to reduce surface roughness. Recent work, for example, has shown that surface roughness can be reduced by employing a combination of sequential etching and polishing steps. It is also possible to produce channel features of arbitrary height by partial etching of the copper surface, provided that sufficient control can be exerted over the etching process to ensure adequate surface uniformity.

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