

Video Article

# One-Step Approach to Fabricating Polydimethylsiloxane Microfluidic Channels of Different Geometric Sections by Sequential Wet Etching Processes

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## Abstract

Polydimethylsiloxane (PDMS) materials are substantially exploited to fabricate microfluidic devices by using soft lithography replica molding techniques. Customized channel layout designs are necessary for specific functions and integrated performance of microfluidic devices in numerous biomedical and chemical applications (e.g., cell culture, biosensing, chemical synthesis, and liquid handling). Owing to the nature of molding approaches using silicon wafers with photoresist layers patterned by photolithography as master molds, the microfluidic channels commonly have regular cross sections of rectangular shapes with identical heights. Typically, channels with multiple heights or different geometric sections are designed to possess particular functions and to perform in various microfluidic applications (e.g., hydrophoresis is used for sorting particles and in continuous flows for separating blood cells<sup>6,7,8,9</sup>). Therefore, a great deal of effort has been made in constructing channels with various sections through multiple-step approaches like photolithography using several photoresist layers and assembly of different PDMS thin sheets. Nevertheless, such multiple-step approaches usually involve tedious procedures and extensive instrumentation. Furthermore, the fabricated devices may not perform consistently and the resulted experimental data may be unpredictable. Here, a one-step approach is developed for the straightforward fabrication of microfluidic channels with different geometric cross sections through PDMS sequential wet etching processes, that introduces etchant into channels of planned single-layer layouts embedded in PDMS materials. Compared to the existing methods for manufacturing PDMS microfluidic channels with different geometries, the developed one-step approach can significantly simplify the process to fabricate channels with non-rectangular sections or various heights. Consequently, the technique is a way of constructing complex microfluidic channels, which provides a fabrication solution for the advancement of innovative microfluidic systems.

## Video Link

The video component of this article can be found at <https://www.jove.com/video/57868/>

## Introduction

Microfluidic techniques have drawn attention over the past decades because of their intrinsic advantages for a variety of biomedical and chemical research and applications. Several material usage options for constructing microfluidic chips are available nowadays, such as polymers, ceramics, and silicon materials. To the best of our knowledge, among the microfluidic materials, PDMS is the most common one due to its appropriate material properties for various microfluidics research and applications, including its optical and biological compatibilities with particles, fluids, and extremely small living organisms<sup>1,2,3,4,5</sup>. Furthermore, the surface chemical and structure mechanical properties of PDMS materials can be adjusted to facilitate microelectromechanical and mechanobiological studies by applying such polymer-based microfluidic devices<sup>10,11,12</sup>. Concerning the manufacturing of microfluidic devices with designed channel patterns, soft lithography replica molding methods are usually applied to create the microfluidic channels by utilizing their corresponding master molds which are composed of photolithography-patterned photoresist layers and silicon wafer substrates<sup>12</sup>. Owing to the nature of molding approaches using silicon wafers with patterned photoresist layers, the microfluidic channels commonly have regular cross sections of rectangular shapes with identical heights.

Recently, researchers have made significant progress in biomedical studies which deal with, for instance, sorting particles and cells using hydrophoresis, separating blood plasma, and enriching white blood cells by applying microfluidic chips with channels of different heights or geometric sections<sup>6,7,8,9</sup>. Such sorting and separating functions of microfluidics for biomedical applications are realized by customizing channels with different geometric sections. Several studies have been devoted to the manufacture of microfluidic channels with cross sections of different geometry features by fabricating master molds with specific surface patterns of various heights or non-rectangular cross sections. These studies on mold fabrication include such techniques as multi-step photolithography, photoresist reflow, and grey-scale lithography<sup>13,14,15</sup>. Inevitably, the existing techniques involve finely crafted photomasks or a precise alignment in multi-step manufacturing processes, which may substantially enhance the complexity levels of the corresponding fabrication of microfluidic channels. So far, several attempts have been made on single-

step manufacturing processes for microfluidic channels of various sections, but the respective techniques are highly restricted to specific cross-sectional shapes of channels<sup>16</sup>.

Over the past two decades, in addition to the molding approaches for fabricating PDMS microfluidic channels with various sections, etching techniques for patterning PDMS channels with geometric features have become the fabrication of choice in a variety of microfluidic applications. For instance, PDMS wet etching is exploited along with multi-layer PDMS bonding for constructing a pneumatic actuated cell culture device of microfluidics with reconstituted organ-level lung functions<sup>17</sup>. The PDMS wet etching technique is employed together with PDMS casting on cylindrical microwells machined by computer-aided control systems for fabricating 3D PDMS microneedle arrays<sup>18</sup>. PDMS dry etching is used to make PDMS microstructures as parts of micro-electromechanical actuators<sup>19,20</sup>. Porous PDMS membranes with designed pore layouts are also fabricated through dry etching processes<sup>21</sup>. Both the wet and the dry etching techniques can be integrated into patterning PDMS films with designated geometric shapes<sup>22</sup>.

However, the etching techniques for forming PDMS channel structures with complex section shapes have not been commonly applied because of their intrinsic limitations on microfluidic fabrication. First, while the techniques of PDMS wet etching utilizing laminar flows of chemicals for creating microfluidic channels of various sections have been established, the subsequent channel section formation is still restricted because of the basic characteristics of isotropic chemical etching processes<sup>23</sup>. Furthermore, even though there seems to be reasonable space for controlling the channel section geometries in a microfluidics fabrication using the PDMS dry etching techniques<sup>20</sup>, the required etching time is usually too long (in terms of hours) to be practical for manufacturing microfluidic chips. In addition, the etching selectivity between PDMS materials and the corresponding masking photoresist layers might be low in general, and the resulted etched depths for the channels are, thus, not acceptable<sup>20</sup>.

In this paper, we develop a one-step approach to fabricate microfluidic channels of different geometric cross sections by PDMS sequential wet etching processes (hereafter referred to as SWEP). The SWEP begin with a PDMS microfluidic device with single-layer channels. With assorted layout designs of the channels, fabricating microfluidic channels with different geometric sections of various kinds can be achieved through sequential etching processes. The sequential etching only needs an etchant to be introduced into specific channels of the planned single-layer layouts embedded in PDMS materials. Compared to conventional PDMS fabrication processes, the SWEP just require one further step to fabricate microfluidic channels of non-rectangular sections or various heights. The proposed SWEP provide a straightforward and simple way of fabricating microfluidic channels with various sections along the flow direction, which can significantly simplify the processes in the aforementioned methods.

## Protocol

### 1. Fabrication of Microfluidic Devices with Single-Layer Channel Layouts

NOTE: In this paper, the soft lithography method<sup>3</sup> is adopted for fabricating microfluidic devices made of PDMS materials, to demonstrate how to manufacture channels with various sections.

#### 1. Creation of master molds for a PDMS layer with designed topology features

- Design channel layouts on a PDMS layer for a single etching process or etching in sequence.
- Sketch the inverted topology features of the designed PDMS layer using a computer-aided drawing program.
- Deliver the sketch file to a photolithography facility to obtain a patterned photomask with the high-precision inverted topology features of the channel layouts printed on a transparency<sup>24</sup>.
- Use isopropyl alcohol (2-Propanol (IPA),  $\geq 99.9\%$ ), acetone (Propan-2-one,  $\geq 99.5\%$ ), and buffered oxide etch (BOE,  $\text{NH}_4\text{F}:\text{HF}$  (v/v) = 6:1) on the surfaces of a 4-inch silicon wafer to remove any dust or residuals and avoid contaminations.
- Use around 500 mL of deionized water to wash the silicon wafer for a final polishing, and then apply nitrogen gas to dry the rinsed wafer.
- Place a negative tone photoresist of about 20 g on the wafer. Then spin coat the wafer at 500 rpm for 15 s and 2,000 rpm for 30 s to produce a photoresist layer of around 75  $\mu\text{m}$  in thickness.  
NOTE: Different photoresist thicknesses can be achieved using negative tone photoresists with different product numbers and with different spin coating, baking, and development conditions, according to the user manuals<sup>25,26</sup>.
- Soft bake the wafer by heating it on a hotplate at 65 °C for 3 min and then at 95 °C for 9 min.
- Put the wafer into a photomask aligner machine along with the patterned transparency from step 1.1.3 as a mask.
- In the aligner machine, apply ultraviolet (UV) light at 300  $\text{mJ}/\text{cm}^2$  to expose the wafer covered by the transparency.
- After exposure to the UV light, place the wafer on a hotplate at 65 °C for 2 min and then at 95 °C for 7 min as the post-exposure bake (PEB).
- Following the PEB, strongly agitate the wafer immersed in a negative tone photoresist developer, or place the immersed wafer in an ultrasonic bath (37 kHz, effective power of 180 W) for 7 min.
- Clean the entire wafer again with isopropyl alcohol to eliminate any developer remaining on the wafer surface.
- To prevent undesired bonding, silanize the surface of the wafer by putting the wafer together with 100  $\mu\text{L}$  of 97% silane (1H,1H,2H,2H-perfluorooctyl-trichlorosilane) in a 6 cm Petri dish in a desiccator.
- Connect the desiccator to a vacuum pump and set the vacuum pressure at 760 mmHg.
- Next, turn the pump on for 15 min. Switch it off, and then leave the wafer to rest in a vacuum in the desiccator for 30 min.  
CAUTION: The evaporated silane is extremely harmful to humans; thus, the whole wafer surface passivation must be carried out in a fume hood.
- Fetch the silanized wafer, which was undergoing surface passivation. Fix the wafer in a 15 cm Petri dish for further use.  
NOTE: The patterned wafer is ready to be used as a mold to replicate the designed channel layouts inversely by PDMS materials.

#### 2. Fabrication of PDMS channel layouts by replicating the inverted topology on the molds

- Put the base PDMS (monomer) along with the corresponding catalyst (curing agent) at a volume ratio of 10:1 into a clean and single-use plastic cup.

2. Mix the PDMS prepolymer mixture (from step 1.2.1) homogeneously by using a power stirrer.
3. Put the cup in the desiccator connected to the vacuum pump for 60 min to remove any trapped bubbles in the PDMS mixture.
4. Pour 20 g (for Section 2) or 8 g (for Section 3) of the PDMS prepolymer mixture on top of the master mold (made in step 1.1) with the inverted topology features of the designed channel layouts, and then eliminate any possible bubbles embedded in the PDMS materials by using the desiccator (for 60 min).
5. Put the mold carrying the PDMS mixture in an oven at 60 °C for 4 h to cure the silicone-based liquid prepolymer materials.
6. After cooling the wafer together with the PDMS to room temperature for approximately 20 min, detach the cured PDMS from the mold with a scalpel and tweezers.
7. Tailor the detached PDMS layer to an area (approximately 6 x 6 cm<sup>2</sup> for Section 2 or 2 x 7.5 cm<sup>2</sup> for Section 3) covering the entire channel layouts using a scalpel.
8. Create channel access ports (inlets and outlets) by using a biopsy punch of 1.5 mm in diameter.  
NOTE: The numbers and the positions of the inlets and outlets are designed based on the etching processes for fabricating specific microfluidic channels.
9. Pour 30 g of the PDMS prepolymer mixture into a Petri dish, and then eliminate any possible bubbles embedded in the PDMS materials by using the desiccator (for 60 min).
10. Put the Petri dish carrying the PDMS mixture in an oven at 60 °C for more than 4 h to cure the liquid prepolymer materials.
11. After cooling the Petri dish together with the PDMS to room temperature for approximately 20 min, detach the cured PDMS from the dish with a scalpel and tweezers.
12. Using a scalpel, tailor the detached PDMS layer without any features to dimensions equal to those of the aforementioned PDMS layer (approximately 6 x 6 cm<sup>2</sup> for Section 2 or 2 x 7.5 cm<sup>2</sup> for Section 3).
13. Activate the surfaces of both PDMS layers (made in steps 1.2.7 and 1.2.12) with the designed channel layouts and without any features by exposing the top PDMS materials to oxygen plasma in a surface treatment machine at 90 W for 40 s.
14. Bond the 2 PDMS layers by making contact between their treated surfaces right after the oxygen plasma surface activation. Then, leave the bonded PDMS layers in an oven at 60 °C for more than 30 min.  
NOTE: There is no upper time limit for leaving the bonded PDMS layers in the oven.
15. After the 2 bonded PDMS layers have cooled, trim the excess PDMS materials away from the fabricated device for a later experimental set-up.

## 2. The One-Step Approach to Fabricating PDMS Microfluidic Channels of Different Sections

NOTE: To characterize the PDMS wet etching rate, a microfluidic device with a single-layer and straight channel of rectangular shapes is suggested to be exploited for identifying specific etching rates corresponding to certain experimental settings.

### 1. Experimental characterization of PDMS wet etching

1. Prepare an etchant solution by mixing tetra-n-butylammonium fluoride (TBAF, a 1 M solution in tetrahydrofuran (THF)) with 1-Methyl-2-pyrrolidinone (NMP) at a rate of v:v = 1:10.  
NOTE: NMP is capable of efficiently dissolving chemical residuals induced by the etchants. In general, PDMS materials are swollen marginally by the NMP, and the PDMS microfluidic devices are still able to preserve their shapes, volumes, and seal conditions.
2. Draw the mixed TBAF/NMP etchants into a 10 mL syringe connected to a stainless blunt needle (16 G).
3. Set up a syringe pump as a controller of the pressure-driven fluids in the channels.
4. Connect the blunt needles of the syringes filled with the etchant solution to the channel port of the abovementioned simple device and guide the respective port from the outlet tubing to a waste container as shown in **Figure 1**.
5. Run the syringe pump carrying the syringes containing the mixed TBAF/NMP etchant solution at a 150 µL/min flow rate for characterizing the PDMS wet etching.
6. Use bright-field microscopic views and make sure that the etched channel along the flow direction has a uniform width, to consequently confirm that the volume mixing ratio of the etchants and the etchant flow rate are adequate.
7. Capture the time-series images of the channel cross section under an inverted microscope with a 4X magnification during the PDMS etching process.
8. Analyze the stored images by applying the basic measurement function in a 2D analysis of the imaging processing program to collect a time sequence of numbers for the channel width during the wet etching process of PDMS materials.
9. Evaluate the time-series etching rates through the equation shown in **Figure 2**, which is dividing 50% of the channel width change ( $\Delta W / 2$ ) by the duration of the PDMS etching ( $t$ ).
10. Perform a linear regression of the collected data points to estimate an overall etching rate of the mixed TBAF/NMP etchants with the specific volume mixing ratio of 1:10 for the PDMS materials as shown in **Figure 2**.

### 2. PDMS sequential wet etching for fabricating microfluidic channels of different geometric sections

1. Design an arrangement of etchant inlets for the single-layer PDMS channel layout serving the corresponding etching processes in sequence, so that a specific channel type of different cross-sectional shapes as shown in **Figure 3** can be fabricated.
2. Follow the procedures described in steps 2.1.1 - 2.1.7 for the PDMS wet etching approach.  
NOTE: The flow rate is set as 50 µL/min.
3. While the TBAF/NMP etchants are flowing, inspect the etched channels under the microscope to see if there exist significant problems such as a noticeable amount of bubbles, a remaining of several chemical residuals induced by the etchants, a leakage of the etchants, or a flow of etchants on an inclined plane.
4. Observe the microfluidic channel wall thickness variation by inverted microscopy, and time the wet etching process to ensure the proper channel geometries are achieved.

### 3. The Design of a Microfluidic Mixer

NOTE: A design of the microfluidic mixer which can efficiently mix 2 dissimilar fluids is demonstrated here to show an advantageous application of microfluidic channels with different sections.

#### 1. Fabrication of a microfluidic mixer with different channel sections

1. Make a PDMS device with a single-layer microfluidic channel of the design shown in **Figure 4** by the soft lithography replica molding technique (section 2).
2. In the single-layer microfluidic channel layout, introduce the TBAF/NMP etchant solution prepared by following the procedures described in step 2.1.1 from the port marked as "outlet" at a 20  $\mu\text{L}/\text{min}$  flow rate in **Figure 4**.
3. Observe the microfluidic channel wall thickness variation under the microscope, and time the wet etching process to ensure the proper channel geometries as represented in **Figure 5** are achieved.

#### 2. Experimental characterization of the microfluidic mixer

1. After the microfluidic channel with sections of different shapes in an alternate pattern is realized, pump 2 dissimilar fluids including a solution of fluorescein sodium salt having a 50  $\mu\text{g}/\text{mL}$  concentration and distilled water into 2 separate channels at a 20  $\mu\text{L}/\text{min}$  flow rate.
2. Take fluorescence microscope images of the channel in top view at the positions marked as A, B, C, and D under an inverted microscope (4X magnification) for the 2 mixers with uniform (before etching) and different geometric sections (after 2 h of SWEP), respectively (**Figure 6**).

NOTE: The fluorescence microscope images are taken while the stable flows occur, at the time point of 5 min, counted from the beginning moments of mixing through the mixer channels.

3. Analyze the captured fluorescent images by using an imaging processing program to estimate the corresponding mixing efficiency numbers which are defined by the mixing residual (MR, 0.5 = unmixed, 0 = fully mixed) in the following equation<sup>27,28</sup>:

$$\text{MR}(t) = \frac{1}{L} \int_S \left| I(x, t) - \frac{1}{2} \right| dx$$

Here,

$t$  is the etching time,

$L$  is the channel width at a certain position of interest,

$S$  is a line segment across the channel at the position, and

$I$  is the fluorescence intensity distribution over  $S$  at  $t$ .

4. Plot the fluorescence intensity distribution over  $S$  across the channel at the positions marked as A, B, C, and D for the 2 mixers with uniform (before etching) and different geometric sections (after 2 h of SWEP), respectively. Estimate the corresponding MR as depicted in **Figure 6**.

### Representative Results

Recently, a large number of studies have been made on the fabrication of microfluidic devices with channels of different sections by lithography replica molding<sup>13,14,15</sup> and PDMS etching techniques<sup>17,18,19,20,21,22</sup>. However, there still exist considerable limitations of patterning shapes and difficulties with manufacture operations<sup>16,23</sup>. In this paper, a one-step approach to fabricating PDMS microfluidic channels of different geometric sections by SWEP is proposed.

**Figure 1** schematically shows the microfluidic single-layer channel layouts for creating PDMS channels of different sections by SWEP and displays the experimental setup of the associated tubing system. NMP is a buffer used for the SWEP experiments, as shown in **Figure 1a** and **1b**. In the SWEP experiments, it is important to choose a proper solvent to eliminate the etching products in the channels for maintaining laminar flows exploited by the etching processes. Consequently, the NMP buffer is chosen as the solvent to effectively dissolve the products of the SWEP<sup>22,23</sup>.

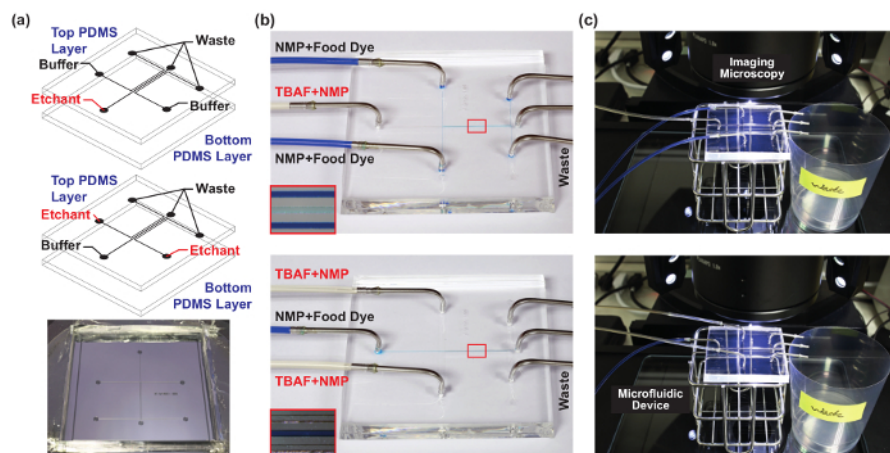
The etched channels are also filled with blue food dyes to demonstrate the evolution of the channel sections inside the microfluidic device. By arranging etchant inlets of the designed single-layer channel pattern, microfluidic channel sections with various geometry features of different kinds can be obtained through the SWEP as demonstrated in **Figure 3**.

To characterize the PDMS wet etching, a microfluidic device with a single-layer and straight channel of rectangular shapes is exploited for identifying an overall etching rate of the mixed TBAF/NMP etchants with a specific volume-mixing ratio for the PDMS materials. By the linear regression of the collected data points of the channel width variations with respect to certain etching times, the overall etching rate of the etchant solution is experimentally estimated as 2.714  $\mu\text{m}/\text{min}$  (**Figure 2**).

In microfluidic channels with uniform cross sections, fluids mostly flow along channel walls, which suppress random contacts between substance particles; therefore, fluid mixing driven by diffusion is usually achieved through particularly long channels. As a result, microfluidic channels of different geometric sections are anticipated to facilitate fluid mixing with the help of lateral fluid motions over channel sections. In this study, a design of the microfluidic mixer (**Figure 4**) where two dissimilar fluids are efficiently mixed is demonstrated here for presenting one advantageous application of microfluidic channels with different sections. **Figure 5** presents the time-series images of the microfluidic mixer channel fabricated by the SWEP using PDMS materials in top view at etching stages of 0 h, 0.25 h, 0.40 h, 0.55 h, 0.70 h, 1.00 h, and 2.00 h in sequence.

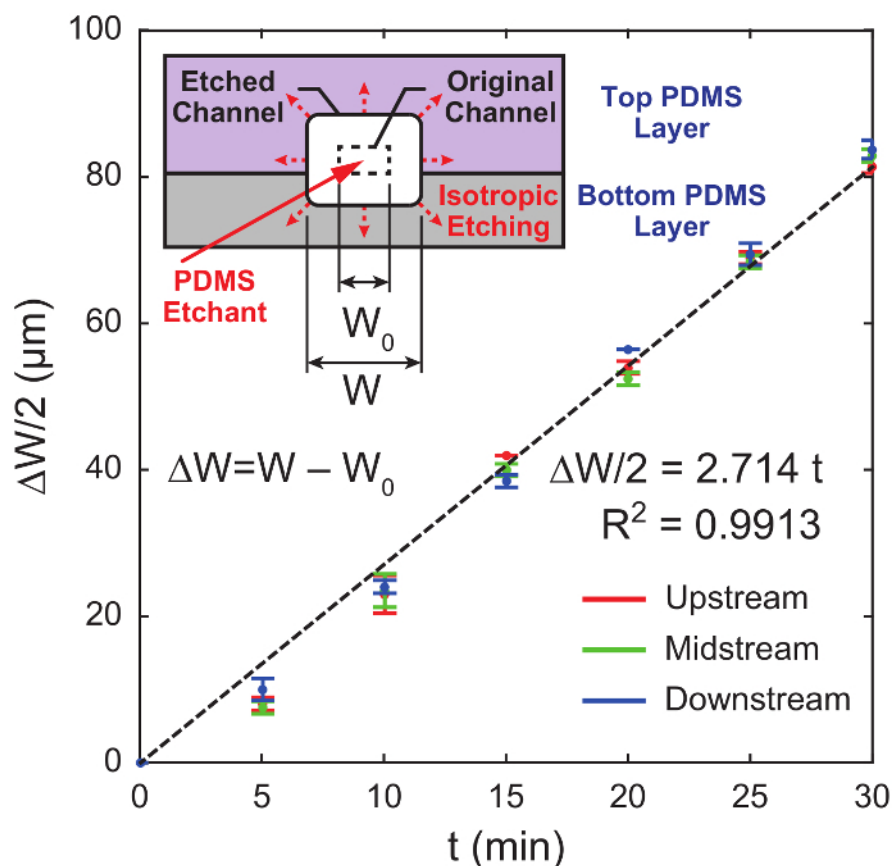
After the microfluidic channel with sections of different shapes in an alternate pattern is realized and two dissimilar fluids including a solution of fluorescein sodium salt and distilled water are subsequently pumped into two separate channels, fluorescence microscope images of the channel in top view at the positions marked as A, B, C, and D are captured under an inverted microscope for the two mixers with uniform (before etching) and different geometric sections (after 2 h of SWEPE), respectively (**Figure 6**). These images are taken while the stable flows occur, at the time point of 5 min, counted from the beginning moments of mixing through the mixer channels. Then, these fluorescence microscope images are delivered to an automated program developed in this study to extract the corresponding MR numbers representing the mixing efficiency of the mixer.

Before the etching process, the channel of the mixer with a serpentine channel layout had identical cross sections of rectangular shape. Due to the sufficient channel length necessary for diffusion mechanisms, the microfluidic mixer has an essential mixing efficiency represented by 0.4607, 0.3403, 0.2450, and 0.1940 MR numbers at A, B, C, and D positions, respectively. After 2 h of SWEPE, with an equal overall channel length to the original one, the microfluidic mixer has channel sections of different shapes in an alternate pattern. It is important that the mixer with the different channel sections provides a marked rise in mixing efficiency, represented by noticeably decreasing MR numbers of 0.3875, 0.1915, 0.1336, and 0.0680 at A, B, C, and D positions, respectively, because of lateral fluid motions leading to advection in addition to diffusion mechanisms. Besides, from position B - D, such advection mechanisms occurring over channel sections result in an apparent and uniform increase in the mixing efficiency of the mixer fabricated by the SWEPE.

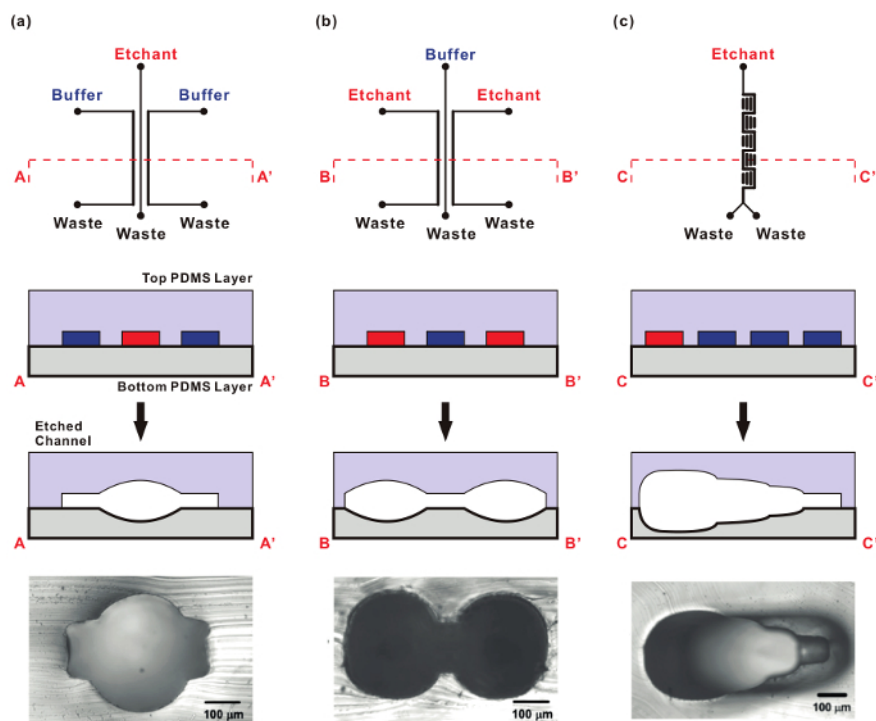


**Figure 1: Tubing set-up on microfluidic single-layer channel layouts for creating PDMS channels of different geometric cross sections by sequential wet etching processes (SWEPE).** (a) This schematic shows the microfluidic devices with single-layer channels. The top layer is fabricated using PDMS of multiple channel designs for wet etchant inlet arrangements. The bottom layer is made of PDMS with a blank pattern. (Top: one etchant inlet; middle: two etchant inlets.) The bottom is the mold for the top layer fabrication. (b) These panels show the assembled device for the fabrication of channels of different sections. The width of the channels and the thickness of the walls are 50  $\mu\text{m}$  and 100  $\mu\text{m}$ , respectively. (c) These panels show the experimental photos of the tubing set-up on the microfluidic single-layer channel layouts for the SWEPE. (Top row: one etchant inlet; bottom row: two etchant inlets.) [Please click here to view a larger version of this figure.](#)

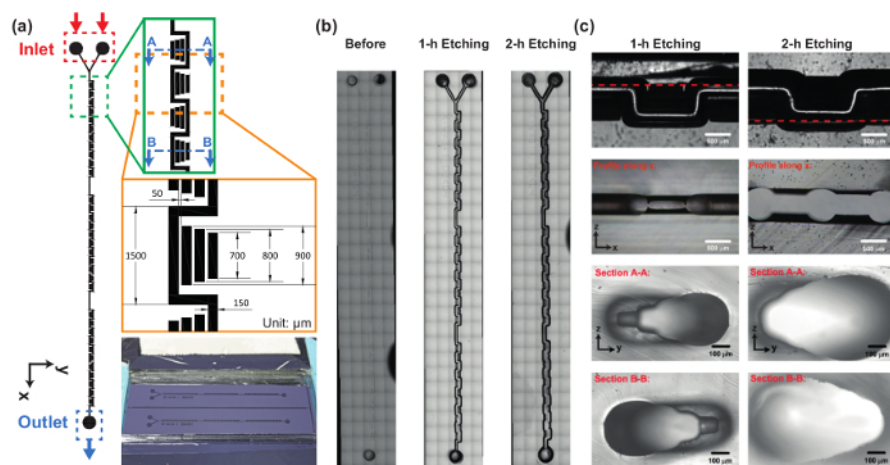




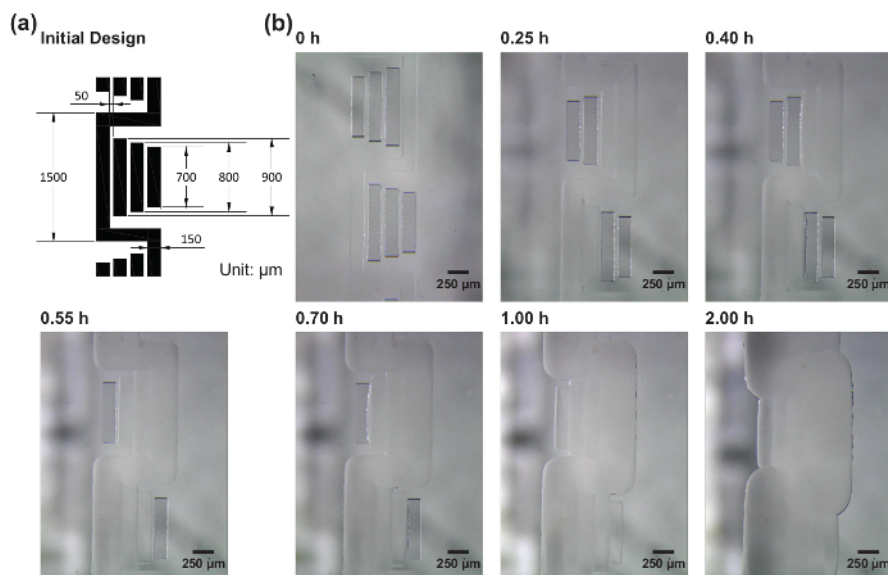
**Figure 2: Characterization of PDMS wet etching.** This figure shows the linear regression of the collected half channel width changes with respect to the etching times for estimating an overall etching rate of the mixed TBAF/NMP etchants with a specific volume-mixing ratio for the PDMS materials. [The inset is a schematic of the cross-sectional geometry of a simple and straight channel pattern for characterizing the wet etching rates of PDMS materials. The overall etching rate of TBAF/NMP (v:v = 1:10) is 2.714  $\mu\text{m}/\text{min}$  and the corresponding  $R^2$  (coefficient of determination) is 0.9913.] [Please click here to view a larger version of this figure.](#)



**Figure 3: Fabricated microfluidic channels of different geometric sections by sequential PDMS wet etching.** These panels show various arrangements of etchant inlets for single-layer PDMS channel layouts serving the corresponding etching processes in sequence for fabricating particular channel types of different cross-sectional shapes such as (a) cross-shaped, (b) dumbbell-shaped, and (c) bell-shaped cross-sectional geometries. [Please click here to view a larger version of this figure.](#)

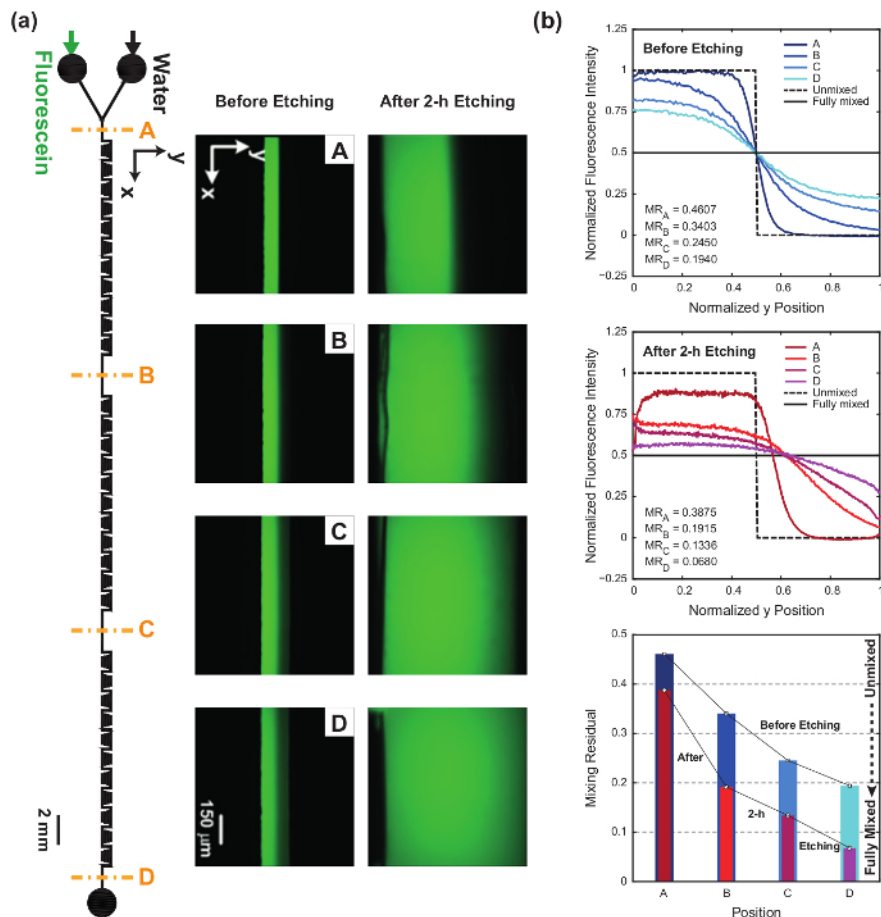


**Figure 4: Fabricated microfluidic mixers utilizing channels with different sections.** (a) This panel is a design drawing of a single-layer channel layout for the fabrication of a microfluidic mixer utilizing channels with different sections. The bottom shows the mold for the single-layer channel fabrication. (b) These panels show the tile scan microscope images of the whole mixer channel before and after 1 and 2 h of PDMS wet etching. (c) These panels show experimental bright field images of the mixer channel sections which are fabricated by 1 and 2 h of PDMS wet etching in a top view (top row), in a cutting view perpendicular to the flow direction along the x-axis (second from the top), and in a section view at the A-A cut (third from the top) and B-B cut (bottom row) positions. [Please click here to view a larger version of this figure.](#)



**Figure 5: Time-series images of microfluidic mixer channels of different sections fabricated by the sequential wet etching of PDMS materials.** (a) This panel shows the schematic of a single-layer channel layout for the fabrication of a microfluidic mixer with different channel sections. (b) These panels show microscope images of the mixer channel in a top view at each etching stage in sequence. [Please click here to view a larger version of this figure.](#)





**Figure 6: Characterization of the microfluidic mixer fabricated by sequential PDMS wet etching.** (a) These panels show fluorescence microscope images of the mixer channel at the positions marked as A, B, C, and D before introducing etchants and at 2 h wet etching of PDMS materials. (b) These panels show measured fluorescence intensity fields presented in a normalized coordinate across the mixer channel at the A, B, C, and D positions before (top) and at 2 h of PDMS wet etching (middle). It also shows the analyzed MR representing the mixing efficiency of the mixer (0.5: unmixed, 0: fully mixed) at various channel positions before and at 2 h of etching (bottom). [Please click here to view a larger version of this figure.](#)

## Discussion

Over the past decades, microfluidics has offered promising means by which experimental platforms for chemical and biomedical research can be constructed systematically<sup>1,2,3,4,5</sup>. The platforms have also presented their capabilities of investigating several cellular functions *in vivo* under physiological microenvironment conditions via *in vitro* cell studies<sup>6,7,8,9</sup>. In experimental research and related applications, most of the channel cross sections of microfluidic devices are uniform and rectangular-shaped. In such microfluidic devices, the channel structures play an important role in the microenvironment conditions. For example, while using microfluidics as an apparatus for drug delivery, a passive control over such chemical transport is modulated by tuning flow rate in the rectangular channel of standard cross-section geometry<sup>29</sup>. For a desired flux distribution of the substance transport over the channel along the flow direction, microfluidic channels with different geometric sections under an overall volumetric flow rate set-up may be needed. A considerable number of studies have taken some important steps to fabricate such chips with desired channels with different sections, including the construction of master molds with particular surface patterns of various heights or non-rectangular cross sections<sup>13,14,15</sup> and PDMS etching techniques for creating surfaces with geometric features<sup>17,18,19,20,21,22</sup>. However, these endeavors not only involve complex manufacturing processes but also are restricted to specific cross-sectional shapes of channels<sup>16,23</sup>.

In this paper, a one-step approach to creating PDMS channels with various sections is advanced by introducing etchant into specific channels of planned single-layer layouts embedded in PDMS materials in a straightforward and consistent way. Furthermore, the isotropic sequential wet etching processes of forming channels with different cross-sectional shapes are verified by using iterative numerical calculation<sup>30</sup>. Apparently, it is difficult to fabricate channel section geometries with sharp angles because of the isotropic removal of the PDMS material during the sequential wet etching processes. In practical applications, the precise control over the fabricated section geometries of microfluidic channels requires an accurate characterization of PDMS wet etching rates and careful arrangements of the associated tubing system set-up. Compared to the existing methods for manufacturing PDMS microfluidic channels with different geometries, the developed one-step approach can significantly simplify the processes of fabricating channels with non-rectangular sections or various heights. Consequently, the developed technique provides a way of constructing complex microfluidic channels which may lead to the development of innovative microfluidic systems for various applications.

## Disclosures

The authors have nothing to declare.

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## References

1. Tung, Y.-C *et al.* Optofluidic Detection for Cellular Phenotyping. *Lab on a Chip*. **12**, 3552-3565 (2012).
2. Lu, Y., Yang, L., Wei, W., Shi, Q. Microchip-based Single-cell Functional Proteomics for Biomedical Applications. *Lab on a Chip*. **17**, 1250-1263 (2017).
3. Jensen, K. F., Reizman, B. J., Newman, S. G. Tools for Chemical Synthesis in Microsystems. *Lab on a Chip*. **14**, 3206-3212 (2014).
4. Chang, C.-W. *et al.* A Polydimethylsiloxane-polycarbonate Hybrid Microfluidic Device Capable of Generating Perpendicular Chemical and Oxygen Gradients for Cell Culture Studies. *Lab on a Chip*. **14**, 3762-3772 (2014).
5. Mosadegh, B. *et al.* Integrated Elastomeric Components for Autonomous Regulation of Sequential and Oscillatory Flow Switching in Microfluidic Devices. *Nature Physics*. **6**, 433-437 (2010).
6. Choi, S., Park, J.-K. Tuneable Hydrophoretic Separation Using Elastic Deformation of Poly(Dimethylsiloxane). *Lab on a Chip*. **9**, 1962-1965 (2009).
7. Choi, S., Song, S., Choi, C., Park, J.-K. Microfluidic Self-Sorting of Mammalian Cells to Achieve Cell Cycle Synchrony by Hydrophoresis. *Analytical Chemistry*. **81**, 1964-1968 (2009).
8. VanDelinder, V., Groisman, A. Separation of Plasma from Whole Human Blood in a Continuous Cross-Flow in a Molded Microfluidic Device. *Analytical Chemistry*. **78**, 3765-3771 (2006).
9. VanDelinder, V., Groisman, A. Perfusion in Microfluidic Cross-Flow: Separation of White Blood Cells from Whole Blood and Exchange of Medium in a Continuous Flow. *Analytical Chemistry*. **79**, 2023-2030 (2007).
10. Duffy, D. C., McDonald, J. C., Schueller, O. J., Whitesides, G. M. Rapid Prototyping of Microfluidic Systems in Poly(dimethylsiloxane). *Analytical Chemistry*. **70** (23), 4974-4984 (1998).
11. Xia, Y., Whitesides, G. M. Soft Lithography. *Annual Review of Material Science*. **28**, 153-184 (1998).
12. Mello, A. Plastic Fantastic? *Lab on a Chip*. **2**, 31N-36N (2002).
13. Choi, S., Park, J.-K. Two-step Photolithography to Fabricate Multilevel Microchannels. *Biomicrofluidics*. **4**, 046503 (2010).
14. Zhong, K., Gao, Y., Li, F., Zhang, Z., Luo, N. Fabrication of PDMS Microlens Array by Digital Maskless Grayscale Lithography and Replica Molding Technique. *Optik*. **125**, 2413-2416 (2013).
15. Brower, K., White, A. K., Fordyce, P. M. Multi-step Variable Height Photolithography for Valved Multilayer Microfluidic Devices. *Journal of Visualized Experiments*. **119**, e55276 (2017).
16. Lai, D. *et al.* Simple Multi-level Microchannel Fabrication by Pseudo-grayscale Backside Diffused Light Lithography. *RSC Advances*. **3**, 19467-19473 (2013).
17. Huh, D. *et al.* Reconstituting Organ-Level Lung Functions on a Chip. *Science*. **328**, 1662-1668 (2010).
18. Deng, Y.-L., Juang, Y.-J. Polydimethyl Siloxane Wet Etching for Three-Dimensional Fabrication of Microneedle Array and High-Aspect-Ratio Micropillars. *Biomicrofluidics*. **8**, 026502 (2014).
19. Tung, Y.-C., Kurabayashi, K. Nanoimprinted Strain-controlled Elastomeric Gratings for Optical Wavelength Tuning. *Applied Physics Letters*. **86**, 161113 (2005).
20. Tung, Y.-C., Kurabayashi, K. A Single-Layer PDMS-On-Silicon Hybrid Microactuator with Multi-Axis Out-Of-Plane Motion Capabilities-Part II: Fabrication and Characterization. *Journal of Microelectromechanical Systems*. **14**, 558-566 (2005).
21. Chen, W., Lam, R. H. W., Fu, J. Photolithographic Surface Micromachining of Polydimethylsiloxane (PDMS). *Lab on a Chip*. **12**, 391-395 (2012).
22. Balakrishnan, B., Patil, S., Smela, E. Patterning PDMS Using a Combination of Wet and Dry Etching. *Journal of Micromechanics and Microengineering*. **19**, 047002 (2009).
23. Takayama, S. *et al.* Topographical Micropatterning of Poly(dimethylsiloxane) Using Laminar Flows of Liquids in Capillaries. *Advanced Materials*. **13**, 570-574 (2001).
24. Friend, J., Yeo, L. Fabrication of Microfluidic Devices Using Polydimethylsiloxane. *Biomicrofluidics*. **4**, 026502 (2010).
25. *NANO SU-8 2000 Negative Tone Photoresist formulations 2002-2025*. MicroChem Corporation, Newton, MA. (2018).
26. *NANO SU-8 2000 Negative Tone Photoresist formulations 2035-2100*. MicroChem Corporation, Newton, MA. (2018).
27. Hardt, S., Schönfeld, F. Laminar Mixing in Different Interdigital Micromixers: II. Numerical Simulations. *AIChE Journal*. **49**, 578-584 (2003).
28. Hessel, V., Löwe, H., Schönfeld, F. Micromixers-A Review on Passive and Active Mixing Principles. *Chemical Engineering Science*. **60**, 2479-2501 (2005).
29. Damiani, S., Kompella, U., Damiani, S., Kodzius, R. Microfluidic Devices for Drug Delivery Systems and Drug Screening. *Genes*. **9**, 103 (2018).
30. Wang, C.-K. *et al.* Single Step Sequential Polydimethylsiloxane Wet Etching to Fabricate a Microfluidic Channel with Various Cross-Sectional Geometries. *Journal of Micromechanics and Microengineering*. **27**, 115003 (2017).