Supplementary Material (ESI) for Lab on a Chip

Electronic Supporting Information

UV Polymerization of Hydrodynamically Shaped Fibers

*Abel L. Thangawng, Peter B. Howell Jr., Christopher M. Spillmann, Jawad Naciri, and Frances S. Ligler**

Center for Bio/Molecular Science & Engineering, Naval Research Laboratory, 4555 Overlook Ave SW, Washington, DC 20375-5348, *frances.ligler@nrl.navy.mil

Device Fabrication and Operation

The UV polymerization process dictates the materials that can be employed in the sheath-flow device downstream from the hydrodynamic focusing grooves. The primary requirement for UV polymerization is an integrated window through which the shaped core can be polymerized using a UV source. Possible options include the use of all PDMS parts, top and bottom, or a combination of PDMS and silicon/glass substrates. In both options, optical lithography can be utilized to make the mold for PDMS or to add the chevron features onto the substrate with SU-8 resist. However, both options have one major drawback; since the top and bottom are permanently bonded, clogging anywhere in the device would necessitate replacement. Thus, a modular device design with PMMA base and PDMS top was chosen to enable easy assembly and cleaning processes (see Figure 1S). The device was sandwiched between two aluminum clamps and held together by machine screws. The PMMA-PDMS device assembly technique provided for repeated cleaning without any problem.

The sheath flow device used in this work was fabricated in two halves utilizing a combination of a computer numerical controlled (CNC) milling system (Mini Mill, Hass Automation Inc, Oxford, CA) and standard soft lithography. Figure 1S shows an overview of the sheath flow device assembly. The base piece was machined in transparent polymethylmethacrylate (PMMA) (Plexiglas®; Arkema Inc, Philadelphia, PA) with chevron-shaped grooves recessed below the surface of the channel. The top piece was made of polydimethysiloxane (PDMS; Dow Corning's Sylgard 184, Ellseworth Adhesives, Germantown, WI) to enable the UV transmission (350 nm) necessary for the polymerization. The mold for the PDMS, which has only the chevrons corresponding to those in the base, was machined in the PMMA using the same process as the base PMMA piece. The chevrons were recessed features on the PMMA mold and thus produced protruded chevron features on the molded PDMS piece. The top PDMS piece was aligned and assembled on the base PMMA part and screw holes were punched out using a coring tool. The assembled device was then sandwiched between two aluminum clamps with a thin PMMA interface on top of the PDMS. The PMMA interface layer had a small narrow slot, a little wider than the width of the channel) cut out to allow the UV to be transmitted through the PDMS. The slot is necessary as PMMA would absorb the UV and inhibit the polymerization. There were two primary functions of the PMMA interface. First, it provided a flat and smooth support for the PDMS window which might otherwise be distorted if assembled directly against the aluminum clamp. Second, the window provided a means for inspection of the channel after assembly and during the experiment. Note that the slot on the PMMA interface layer only exposes that portion of the channel beyond the chevron features for polymerization of the shaped acrylate solution. If the chevron features were exposed to the UV source, the polymerization could be inadvertently initiated while the acrylate solution was still being shaped by the chevrons. Prior to tightening the screws to secure the assembly, the device was lightly clamped by hand or with a vice to ensure uniform distribution of pressure on the PDMS. The screws were then tightened just enough to maintain the applied pressured. Over tightening of the screws would distort the PDMS piece and cause variation in the cross-section of the fibers.

The sheath flow device had two inlets, one on either side of the channel, for the sheath fluid that focused the sample laterally toward the center of the channel while the sample was introduced from an inlet directly in line with the main flow channel. The overall dimension of the device was 2 inches wide and 3 inches long (5.1 cm by 7.6 cm), and the channels were 1000 μm x 750 μm (width x height). The width and depth of the chevron grooves were 250 μm and 250 μm, respectively. The leading edges of the chevrons were rounded to a radius of 125 μm to accommodate for the 250 μm endmill used in the CNC machining process.

The inlets for sample and sheath fluids were integrated on the base aluminum clamp (connected with ¼-28 flat bottom Peek nut (P-283) with a corresponding ferrule, Upchurch Scientific, Oak Harbor, WA) and a viewing window was integrated on the top aluminum clamp. The upper left inset in Figure 1S shows the complete device assembly with the PMMA and aluminum windows.

Syringe pumps were used for both the sheath and the sample fluid. The sheath solution was pumped through a home-made pulse dampener to reduce instability in the system. Since a single-phase system was desired, the acrylate and sheath solutions (glycerol diluted with water and methanol) were prepared to have the same viscosity and to be entirely miscible. Methanol in the sheath solution was utilized to match the phases between the acrylate solution and sheath solution. The microfluidic device was first flushed with an ethanol/water mixture to aid in eliminating bubbles in the system. The flow of sheath solution was initiated first in order to minimize contact of the acrylate with the channel walls. Into this flow, a steady stream of the acrylate solution was introduced and focused by the seven pairs of chevrons; the acrylate stream was completely surrounded by the sheath solution by the time it passed the chevron grooves.

Once the proper sheathing process was confirmed under a stereomicroscope, the device was placed on a holder, with the outlet submerged inside a beaker filled with water, such that the microchannel was in vertical position and both fluids exited the microchannel into the water. The dual 3 mm diameter lightguides of the UV lamp [Bluewave® 200 UV Light Curing Spot Lamp, Dymax, Torrington, CT] were bundled and positioned approximately 1 inch away from the surface of the PDMS window. The lamp power was set at 4.5 W/cm^2. This provided approximate intensity of 270 mW/cm^2 from each of the two lightguides on the channel surface with an overall exposure length of 40 mm (length of window on PMMA clamp). The cured fibers were then manually wound on a glass slide as they exited the device. The sheath surrounding the fibers drained into the water bath as the fibers were pulled up. The samples were then stored in air for characterization.

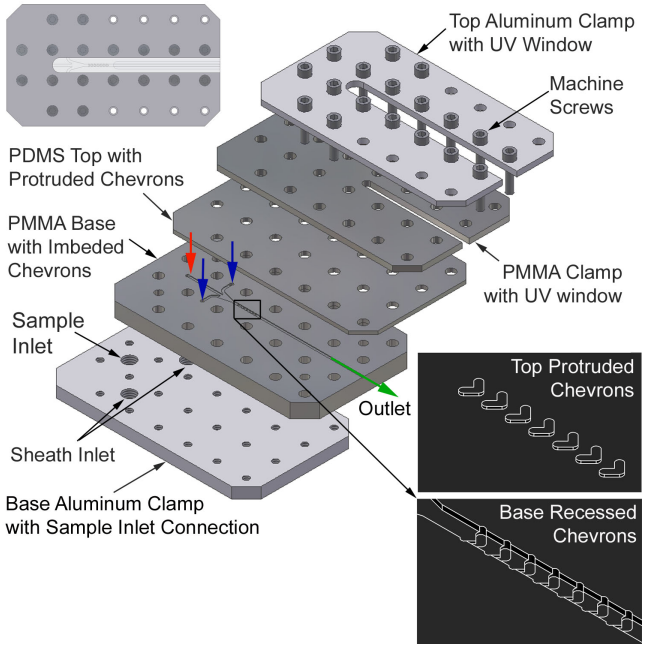

Figure 1S: An overview of the sheath flow device. Lower right insets show the protruded and recessed chevrons integrated to the top PDMS and base PMMA pieces, respectively. The upper left inset shows how the PMMA window and the aluminum viewing windows are assembled. (Figure not to scale.)

Fluid Flow Simulations

The combination of chevrons recessed into one surface of the channel and protruded from the opposite surface has not been previously explored. Simulations were implemented to determine how to use such structures to produce a sheathed core with predictable dimensions. Fluid flow simulations were carried out using COMSOL Multiphysics®

finite element package (COMSOL Inc., Burlington, MA). As shown in Figure 2, both the recessed and protruded chevrons provided a mechanism by which hydrodynamic lifts were generated, displacing the sample stream successfully in the vertical direction. This, along with the lateral focusing accomplished by the introduction of sheath fluid on both sides of the acrylate solution inlet manipulated the core stream to the desired shape.

COMSOL Multiphysics® software was utilized to simulate the fluid flow and design the device. Flow in the inlets was assumed to be laminar. Taking advantage of the device's symmetry, only half of the fluid was modeled. A non-slip boundary condition was applied to the channel walls, and the symmetry condition was applied at a horizontal symmetry plane through the center of the channel. The simulation was carried out using a model with a set number of chevron pairs, and cross-sectional images were obtained at desired positions. The images were then processed with Photoshop to render the full cross section of the channel. Steady-state solutions of incompressible Navier-Stokes flows were coupled with the convection and diffusion module to model the profile of the acrylate solution. In all cases, the diffusion coefficient of 1 x 10^{-9} m^2 s^{-1} was used, and the fluid was modeled as water.

Simulation was carried out for flow-rate ratios ranging from 25:1 (300:12) to 300:1(900:3). The simulation results indicated that the seven-chevron pair design should produce rectangular fibers with slightly rounded edges. The sizes of the sample stream were estimated by measuring the width and height of the concentration at approximately 1 mm away from the last chevron feature. Fibers with estimated size (width x height) of 188 µm x 52 µm to 70 µm x 28 µm were predicted from the simulation results. Boundaries for dimensions included concentrations >85% of that in the input stream (red region in Figure 2). The estimated dimensions are presented in Table 1, column 5. The simulation was not optimized to obtain maximum accuracy due to the large device used in the work, which would require intensive computing power, and thus produced the thick diffusion layers in the concentration plots. Also, the simulation was carried out as if both the sheath and sample fluid were water, ignoring the actual higher viscosity and density of the fluids, and again generating an overestimate of the diffusion rate. In general, the concentration plots showed that the core streams were flat with rounded edges. Nevertheless, the transition to a flatter core-stream shape was obvious (Figure 2) as the flow-rate ratio increased and more sheath fluid was directed through the chevrons to vertically compress the core stream.

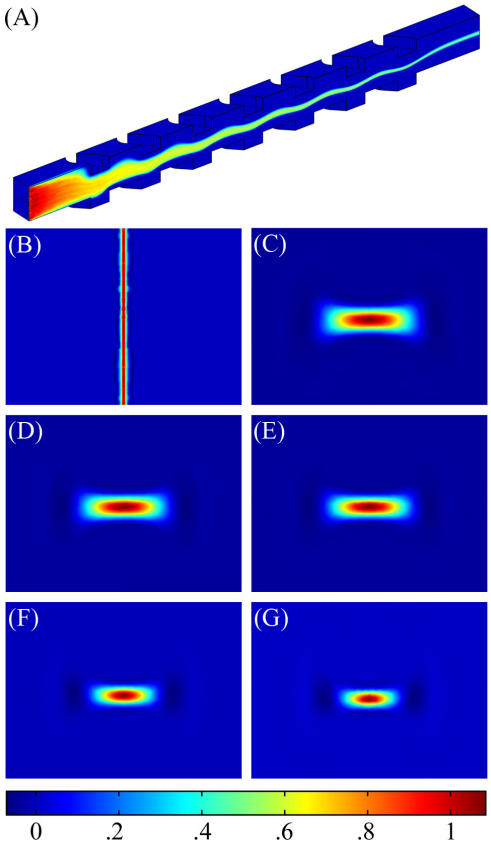

Figure 2S: COMSOL Simulation of sheath flow microfluidic system with recessed chevrons on the bottom wall and corresponding protruding chevrons features on the opposite wall. The sample stream is red and the sheath fluid is blue;

intermediate colors reflect the concentration gradient. (A) Side view showing the sample stream sheathed by hydrodynamic forces, (B) cross section before the first chevron, and (C-G) cross sections after the seventh chevron at a sheath-to-sample flow-rate ratio (μL/min units and absolute ratio) of (C) 300:12 [25:1], (D) 300:6 [50:1], (E) 300:3 [100:1], (F) 600:3 [200:1], (G) 900:3 [300:1]. The diffusion coefficient was 1×10^{-9} m^2 s^{-1} for all cases.

Structural and Mechanical Characterization

For scanning electron microscopy, the fibers were coated with ~50nm of gold/palladium using a sputtering machine (Cressington Auto 108 Sputter Coater, Ted Pella Inc, Redding, CA) and imaged using a LEO Supra 55 (Carl Zeiss SMT Inc, Peabody, MA).

Following UV polymerization, fibers synthesized at a flow ratio of 300:12 were wrapped length-wise around a standard glass microscope slide to quantify the tensile modulus and the viscous loss of the material. To obtain an adequate cross-sectional area for the measurements, over twenty fibers strands greater than 1 cm in length were removed from the microscope slide and mounted between two pieces of tape. Fibers cut to lengths of ~11 mm for mechanical measurements exhibited average dimensions of 0.210 μm x 40 μm (W × H) as determined by electron microscopy. The pieces of tape with the fibers between them were mounted in a dynamic mechanical analyzer (DMA, Q800 series, TA Instruments) and held in a tension clamp consisting of a fixed upper clamp and a floating lower clamp positioned on an air bearing. A high-resolution linear optical encoder in the DMA allowed for precise monitoring of sample displacement as a function of applied force.

The tensile modulus and viscous losses were conducted under isothermal conditions at 30 ºC. The tensile modulus was determined by ramping the force on the fibers from 0.001 N to 0.1 N at 0.001 N/min and calculating the slope from a plot of the stress versus strain. The initial force of 0.001 N was the minimum force applied by the DMA to hold the sample in position. The viscous loss was measured using similar conditions, except that the force was repeatedly cycled from 0.001 to 0.1 N to form a passive work loop. The raw data of several successive passive work loops is shown in Figure 3S. The area under the curve, which reflects the hysteresis of the material in response to an applied stress, was then used to calculate the viscous loss, W, based on:

$$W = \int F dl$$

where the force, F, was integrated over the length of the sample, dl. Converting the expression to the variables collected during a passive work loop results in an expression of the viscous loss scaled by the mass, m, of the material:

$$\frac{W}{m} = \frac{1}{100\rho}\int \sigma \, d(\%\varepsilon)$$

where the integral, equal to the area under the curve of a passive work loop, now consists of the measured parameters stress, σ, and strain, $\%\varepsilon$. The variable ρ is the density of the material equal to ~1.04 g/cm^3 and the factor of 100 accounts for the strain reported as a percentage. The end result is a measure of the viscous loss if the fibers reported in units of *J/kg*.

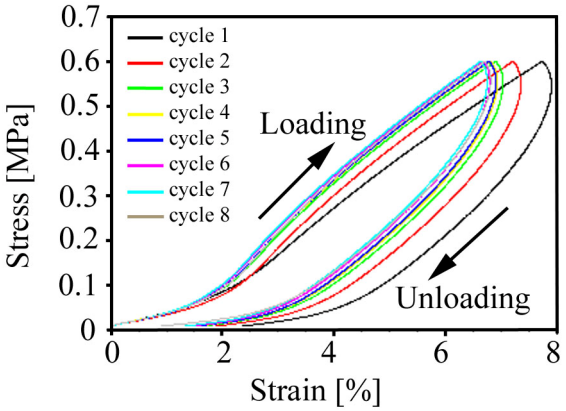

Figure 3S. Raw data showing eight passive work loops of the bundled fibers synthesized with a flow ratio of 300:12. With each successive loop, there was an overall decrease in the elastic modulus from ~6 to ~3MPa accompanied with a decrease in the

viscous loss of the fibers. Both of these observations are evidence of polymer training or annealing as the mechanical stress is applied.