Microfluidic reflow pumps

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A new microfluidic pump, termed a reflow pump, is designed to operate with a sub- μl sample volume and transport it back and forth between two pneumatically actuated reservoirs through a flow channel typically containing one or more sensor surfaces. The ultimate motivation is to efficiently use the small sample volume in conjunction with convection to maximize analyte flux to the sensor surface(s) in order to minimize sensor response time. In this paper, we focus on the operational properties of the pumps themselves (rather than the sensor surfaces), and demonstrate both two-layer and three-layer polydimethylsiloxane reflow pumps. For the three-layer pump, we examine the effects of reservoir actuation pressure and actuation period, and demonstrate average volumetric flow rates as high as 500 $\mu\text{l}/\text{min}$. We also show that the two-layer design can pump up to 93% of the sample volume during each half period and demonstrate integration of a reflow pump with a single-chip microcantilever array to measure maximum flow rate.

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I. INTRODUCTION

Microfluidic biosensor development^{1,2} frequently aspires to create a system that can detect small ($\sim\text{pM}$) concentrations of target analyte in a limited sample volume ($\sim\mu\text{l}$) in a short time ($\sim\text{min}$) using surface-tethered receptors that define a specific sensing region. The combination of these performance goals, however, imposes important constraints. For example, convection can be used to increase the flux of target molecules to the sensor surface in an effort to achieve the desired sensor response time (assuming the response time is mass-transport limited). However, it is well-known that this results in many target molecules never reaching the sensor surface, because they are too far away to diffuse to the surface before they flow past (see, for example, the discussion of channel and sensor Peclet numbers in Ref. 3). Never reaching the sensor surface is typically unacceptable, because the combination of small sample volume and small concentration implies that there are relatively few target molecules available in the sample and it is imperative to use them efficiently. A straightforward approach to avoid this problem and still gain the benefits of convective flow is to locate the sensor region in a flow channel configured as a loop and pump the sample around the loop.⁴ An even simpler setup is to pass the small sample volume back and forth between two reservoirs connected by a flow channel containing the sensor surface. In this paper, we report the design, fabrication, and characterization of a pneumatically actuated pump that performs this function, which we call a reflow pump. It is ultimately intended to enable small sample volumes to be efficiently used in microfluidic devices with surface-based sensor regions in order to minimize the sensor response time. We demonstrate that high average flow rates (500 $\mu\text{l}/\text{min}$) are readily achievable for sub- μl fluid volumes and illustrate integration of a reflow pump with a single-chip microcantilever array.

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II. DESIGN AND OPERATION

Our new reflow pump is fabricated with polydimethylsiloxane (PDMS), which is optically transparent, gas permeable, low cost, and relatively easy to work with.^{5–8} The basic pump design is shown in Fig. 1. It comprises three layers that are individually fabricated and then aligned and bonded together to form a complete device. As shown in Fig. 1(a), two reservoirs are connected with a fluid channel that is $200\ \mu\text{m}$ wide by $20\ \mu\text{m}$ tall and $4\ \text{mm}$ long. Each reservoir is $2\ \text{mm}$ in diameter and $80\ \mu\text{m}$ tall, with a calculated volume of approximately $0.25\ \mu\text{l}$ if the membrane at the top of the reservoir is undeflected. In our case, the membrane is typically deflected up into the actuation chamber when the reservoir is filled such that the capacity of the reservoir is approximately $0.30\ \mu\text{l}$. On-chip valves V1 and V2 are used to enable the reservoir/ fluid channel/reservoir structure to be isolated from the fluid input and output ports.

Figure 1(b) shows a cross sectional schematic diagram of the three layers of the device, with a total thickness of $2.5\ \text{mm}$. The two reservoirs, labeled R1 and R2, are the only features that span more than one layer, the purpose of which is to increase the reservoir volume by increasing its height. The actuation chamber above each reservoir is $31\ \mu\text{m}$ tall. The membrane thickness between the actuation chambers and reservoirs is $50\ \mu\text{m}$.

(a)

(b)

(c)

FIG. 1. Reflow pump schematic diagrams. (a) Top view with fluid channel in green, control channels and valves in red, and actuation channels and chambers in orange. (b) and (c) Cross section views along dashed lines indicated in (a). See text for details.

Figure 1(c) shows the connection of an actuation chamber to an input port (AC2). During operation, a switchable external pressure source is attached to AC2 such that when the external pressure source is activated, the membrane is deflected down into the reservoir, thereby expelling fluid from the reservoir. The valve V2 is shown on the right, consisting of a control layer above a rounded fluid channel. The control channels are $30\text{ }\mu\text{m}$ tall and are separated from the fluid channel by a membrane approximately $30\text{ }\mu\text{m}$ thick at the thinnest point. When the control channel is pressurized, it completely collapses the fluid channel underneath and prevents and fluid flow.⁹

In the pump's initial state, one of the reservoirs, say, R1, is filled with sample fluid and the other, R2, is collapsed by applying pressure through AC2 to the membrane. The channel connecting the reservoirs is also filled with fluid (either buffer or sample fluid), and the valves V1 and V2 are closed. To operate the pump, pressure is applied to the membrane of the filled reservoir, R1, while simultaneously being released from the collapsed reservoir R2. This expels fluid from R1, which flows through the connecting fluid channel and fills reservoir R2. By reciprocally actuating the reservoirs, fluid moves back and forth between them through the fluid channel, which, in an actual sensor device, would contain one or more functionalized regions to act as sensor surfaces. A reflow pump, therefore, offers the advantages of convective mass transport to sensor surfaces while recycling a small sample volume to enable efficient use of target molecules in the sample.

Note that when fluid flows from one reservoir to the other, some fluid is left behind in the connecting flow channel. In our device, the volume of the flow channel is 16 nL, which is only 5% of the reservoir volume. For sensing applications, it is important to ensure that the flow channel volume is significantly smaller than the reservoir volume in order to pass most of the fluid over the sensor surface during each half period of operation. This is also important so that most of the fluid has the opportunity to mix as it fills the much larger reservoir volume, which helps homogenize the fluid during each half period.

III. FABRICATION

A. Soft lithography

Soft lithography is used to fabricate the device layers, which involves creating a mold for each layer using photoresist on a blank silicon wafer, pouring or spinning an uncured mixture of PDMS over the mold, baking to cure the PDMS, and then separating the PDMS from the mold.¹⁰

B. Molds

The actuation layer (top) mold is fabricated using Microchem SU8-25 photoresist spun at 1500 rpm for 45 s. To prevent cracking or delamination of the photoresist, the following parameters are used for both a softbake and post-exposure bake (PEB). A wafer is placed on a hotplate at $55\text{ }^{\circ}\text{C}$, ramped to $65\text{ }^{\circ}\text{C}$, and baked for 10 min. It is then ramped to $95\text{ }^{\circ}\text{C}$ and baked for 10 min. After softbaking, the wafer is exposed through a photomask for 30 s in a mask aligner (Karl Suss MA 150) with an intensity of 8.5 mW cm^{-2} . After PEB, the photoresist is developed for 2.5 min in SU-8 developer to create the SU8 mold. The height of the SU8 mold is measured with a profilometer to be $31\text{ }\mu\text{m}$. To ensure that the PDMS separates well from the mold, a hard bake is performed at $200\text{ }^{\circ}\text{C}$ for 5 min.

The control layer mold is also fabricated from SU8-25 using the same process except that the spin speed is 1750 rpm and the exposure time is 25 s. The height of the mold is measured to be $28\text{ }\mu\text{m}$.

The fluid layer mold is fabricated with SPR-220-7.0 photoresist (Rohm and Haas Company, Marlborough, MA) to enable fabrication of rounded fluidic channels, which is necessary to create valves. For improved adhesion of the photoresist, the silicon wafer is primed with hexamethyldisilazane (HMDS) prior to spin coating SPR-220 at 900 rpm for 60 s. The wafer is soft baked at $115\text{ }^{\circ}\text{C}$ for 3.5 min before exposure with the fluid layer mask for 90 s at 8.5

mW cm^{-2} . After developing in MF-24 A for approximately 10 min, the wafer is baked at 120°C for 3 min to reflow the photoresist into a rounded shape. Before reflow, the cross-section of the channel is $200\text{ }\mu\text{m}$ wide by $15.5\text{ }\mu\text{m}$ tall, and after reflow, it is $215\text{ }\mu\text{m}$ wide by $20.5\text{ }\mu\text{m}$ tall. To ensure the PDMS separates from the mold, the mold is treated with oxygen plasma for 1 min at 200 W and is then placed in a sealed container with an open bottle of chlorotrimethylsilane at 80°C for 15 min.

C. PDMS assembly

Sylgard 184 (Dow Corning, Midland, MI) PDMS mixed with different ratios of base and curing agent is used for the various layers. In each case prior to molding and curing, the mixed PDMS is placed in a vacuum chamber to remove bubbles introduced during mixing. The degassed PDMS is then poured or spun onto a mold depending on the layer, following which it is cured in an oven at 80°C . The alignment steps are performed in a custom set up with an x-y-z stage and a stereomicroscope.

For the actuation PDMS layer, a mixture with 4:1 ratio of base to curing agent is poured over the actuation mold. A ring sealed to the wafer confines the PDMS to the mold region. After partially curing for 45 min in an oven at 80°C , the PDMS is removed from the mold and holes are punched for the actuation pressure inlets using a 21 gauge blunt needle (Small Parts, Miramar, FL). The PDMS is then stamped onto a wafer with curing agent spun on it at 4000 rpm, coating the underside of the PDMS with a thin layer of curing agent preparatory to bonding to the control PDMS layer.¹¹ The thickness of the actuation layer is approximately 2.4 mm.

For the control PDMS layer, a mixture with 20:1 base to curing agent is spin coated onto the control mold at 900 rpm to achieve a thickness of approximately $80\text{ }\mu\text{m}$. After baking for 45 min, it is aligned to the actuation layer and the two layers are baked together for 30 min. The resultant bonded piece is removed from the control mold, and holes are punched for the control pressure inlets. The piece is then stamped with curing agent as described previously preparatory to bonding to the fluid PDMS layer.

For the fluid PDMS layer, a 20:1 mixture is spin coated onto the fluid mold at 1350 rpm to achieve a thickness of approximately $50\text{ }\mu\text{m}$. After baking for 45 min, the reservoir area is cut out from the fluid layer. The cutting process introduces edge roughness and results in variations of reservoir volume with a tolerance of approximately 10%. The fluid layer is then aligned to the control/actuation bonded piece, and the three layers are baked together for 1 h. The device is then removed from the fluid mold, and holes are punched for the fluid input/outputs.

D. Final device assembly

A fabricated three-layer PDMS device is shown in Fig. 2(a) on a silicon substrate with one pressure source connected to the chip. The PDMS device is designed to be clamped onto a substrate to provide a liquid-tight seal. A glass piece with holes drilled for pressure and fluid inputs and outputs is placed on top of the PDMS device. The final device comprised the glass/PDMS/silicon sandwich placed in a spring-loaded clamp to push the rigid glass piece down onto the PDMS, which in turn creates a seal between the PDMS and the substrate. Fig. 2(b) shows a side view of a clamped final device with all of the pressure and fluid input/outputs connected. Within the flow channels and reservoirs, the silicon surface is hydrophilic while the PDMS surfaces are hydrophobic. The wetting condition in flow channels and reservoirs is, therefore, non-homogeneous. This is typical of many PDMS microfluidic devices on silicon or glass substrates such that in this paper we do not consider the effects of surface modification on volumetric flow rate.

IV. EXPERIMENTAL MEASUREMENT

As seen in Fig. 2(b), fluid and pressure connections to a final device are made with 1 cm long 24 gauge stainless steel tubing (New England Small Tube, Litchfield, NH) bent at $\sim 90^\circ$.

(a)

(b)

FIG. 2. Photographs of fabricated device with coloring matching Fig. 1: (a) a top view with R1 actuated as seen by the orange fluid in the actuation chamber completely displacing the green fluid in the reservoir, R2 is filled with green fluid and the actuation chamber is not actuated. (b) A side view of the PDMS device clamped with the glass piece on top and the appropriate stainless steel and PTFE tubing connections. R1 is again actuated while R2 is not.

One end of the tubing is inserted into the punched holes in the PDMS and the other into PTFE microbore tubing (Cole-Parmer, Vernon Hills IL). PTFE tubing is used to provide the necessary fluid and air pressure to the chip. The tubing connected to the fluid input of the device is also connected to a Harvard Apparatus Pump 33 syringe pump (Harvard Apparatus, Holliston, MA). The fluid output is connected to a segment of tubing used to carry fluid away from the chip. The two control channel ports are connected to solenoid valves (The Lee Company, Westbrook, CT) and a manifold to independently pressurize the control channels to either 0 or 20 psi. The actuation channel ports are likewise connected to solenoid valves but on a different manifold manually set to a pressure between 2.5 and 15 psi. Note that to first order the differential pressure applied across the flow channel that connects the reservoirs is the pneumatic pressure applied to a reservoir through a control channel during actuation.

The control channels to the valves and reservoirs are filled with water (dyed with food coloring for contrast) as shown in Figure 2(a). The water in the reservoir actuation channels provides a method for measuring the flow rate by tracking the water menisci in the tubing.¹² The meniscus movement is a result of the displacement of the actuation membranes when actuation chambers are reciprocally actuated. To ensure that the volume can be accurately derived from the movement of the meniscus, a segment of capillary tubing with an inner diameter of 701 μm

with a tolerance of $10\ \mu\text{m}$ is used (Polymicro, Phoenix AZ). A Photron Ultima APX-RS hi-speed camera (Photron, San Diego CA) is used to image the menisci in the actuation tubing to track their position over time.

Figure 3 shows the complete setup with black and white sample images taken from the high speed video of the menisci. In Fig. 3(a), AV1 and AV2 are the valves controlling the actuation chambers, and CV1 and CV2 are the valves controlling the control channel valves on the chip.

Figure 3(b) contains two frames of one video clip showing the movement of the two menisci from one actuation cycle to the next. In frame 1, T1 is connected to the actuation chamber that is pressurized at the point in time the frame was taken. Frame 2 is taken from later in the same video clip when the pressure is released from T1 and applied to T2. From frame 1 to frame 2, the meniscus in T2 moves to the left as a result of pressure applied, and the meniscus in T1 moves to the right as a result of fluid filling the corresponding reservoir, forcing fluid from the actuation chamber into the tubing. It is important to note that the red and green lines are spaced the same distance apart, demonstrating that the volume of fluid displaced in expanding one actuation chamber is the same volume that is displaced in collapsing the opposite actuation chamber. This consistency demonstrates the accuracy of tracking the meniscus to find the displaced volume. To further ensure consistency, the upper tubing was used for all measurements.

The air valves in the actuation manifold are controlled with a custom LABVIEW VI to ensure proper timing of actuation. Twenty different actuation periods are tested from 24 ms to 4 s (one period consists of one actuation of each reservoir) while a duty cycle of 50% was

FIG. 3. (a) Schematic diagram of experimental setup including the menisci being tracked by the hi-speed camera. (b) Two sample frames from a hi-speed video clip demonstrating the movement of the menisci.

maintained for four different pressures: 2.5, 5, 10, and 15 psi. Video clips of the menisci in motion are taken at various frame rates according to the period used. Some of the sample periods and frame rates are listed in Table I. Each video clip is 10 000 frames long in order to image several actuation cycles

To extract information from the hi-speed videos on how far the meniscus moves, a MATLAB script is used to analyze each video frame by frame. A logarithmic edge detection method is employed to find the pixel location of the meniscus. Since the edge is often not a straight line, but distributed across two vertical pixel lines, the average position of the meniscus is determined by averaging the location of each edge pixel. Calibration of the system indicates that each pixel in the image corresponds to $20\text{ }\mu\text{m}$.

V. RESULTS, DISCUSSION, AND DEMONSTRATION

A. Displaced volume

The distance the meniscus moves as a function of time during pump operation is shown in Fig. 4 for three different actuation periods. In each case, increasing meniscus position represents the reservoir filling with fluid, while decreasing meniscus position indicates fluid being expelled from the reservoir. The fluid volume taken in and expelled by the reservoir is equal to the fluid volume moved in the tubing, which can be calculated as the product of the tubing inner diameter and the difference between the maximum and minimum meniscus position. The average maximum and minimum meniscus position for all of the actuations in a video clip is used to calculate the average displaced fluid volume in the tubing, which is plotted in Fig. 5 as a function of actuation period.

From the data in Figs. 4 and 5, it is clear that the higher the actuation pressure for any given actuation period, the further the meniscus moves per actuation, and therefore, the more volume is displaced from the reservoir. Moreover, as the actuation period is increased, the displaced volume asymptotically approaches a limit that is dependent on the actuation pressure [Figs. 4(a) and 5]. The estimated volume of each reservoir is $\sim 300\text{ nl}$. For an actuation pressure of 15 psi, the asymptotic limit of the displaced volume is $\sim 230\text{ nl}$, which is approximately 75% of the reservoir capacity. In this case, the membrane is deflected flush with the bottom of the reservoir in the center of the reservoir. However, around the edges the membrane cannot deflect far enough to conform to the sides and corners of the reservoir, thereby leaving some volume that is not expelled from the reservoir during actuation. On the other hand, for an actuation pressure of 2.5 psi, the pressure is not high enough to deflect the membrane all the way to the floor of the reservoir and a maximum of $\sim 40\text{ nl}$ (13%) is displaced. Furthermore, for a given actuation pressure in Figs. 4 and 5, the displaced volume decreases as the actuation period decreases because the membrane does not have enough time to approach the asymptotic limit for that applied pressure before the reservoir pressures are reversed.

Depending on the application of the pump, it may be desirable to maximize the displaced volume during each actuation, in which case a higher pressure and longer period are needed.

TABLE I. Sample actuation periods and corresponding frame rates of the high speed camera used to measure meniscus motion.

Period (s)	Frame rate (fps)
4.022	500
1.022	2000
0.522	4000
0.272	4000
0.123	6000
0.071	12 000
0.043	12 000
0.030	20 000

FIG. 4. Meniscus position as a function of time for the given pressures with three different periods: (a) 1.022 s, (b) 0.123 s, and (c) 0.030 s.

To empty only a fraction of the reservoir, a lower pressure and shorter period may be selected such that the membrane will only be able to move a fraction of its maximum distance before being forced back in the opposite direction.

B. Average volumetric flow rate

The average volumetric flow rate (VFR) can be calculated as the total volume displaced during a half period divided by the duration of the half period. Figure 6(a) shows the average

FIG. 5. Average displaced reservoir volume during one half period.

VFR as a function of the actuation period, while Fig. 6(b) shows the average VFR as a function of the actuation pressure. Note that for 15 psi actuation pressure the maximum observed average VFR is 500 $\mu\text{L}/\text{min}$, which occurs at an actuation period of 24 ms that is the smallest period achievable with our system. As the actuation period is increased, the average VFR decreases. For an actuation period of ~ 120 ms or more, the average VFR falls off nearly exponentially. This is because for a half period beyond ~ 60 ms, very little additional volume is displaced.

It is also important to note that while the maximum average VFR is achieved for the shortest applied actuation period, from Fig. 5 this corresponds to a displaced fluid volume of approximately 100 nL, which is 33% of the fluid volume in a reservoir. Hence, there is a trade-off between the maximum average VFR and the fraction of the fluid in a reservoir that is expelled during an actuation cycle. Maximizing the average VFR mandates selection of shorter actuation periods at the cost of a smaller fraction of fluid in the reservoir being expelled. Likewise, maximizing the fraction of fluid that is pushed through the flow channel during an actuation mandates longer actuation periods with correspondingly lower average VFR.

As a final observation, in Fig. 6(b), we see that the average volumetric flow rate is an essentially linear function of the applied pressure. This is consistent with the theory for pressure-driven laminar flow through a microfluidic channel in which the VFR is linearly dependent upon the pressure across the fluid channel.¹³

C. Improvements: Two-layer design

The reflow pump design can be simplified to use two PDMS layers instead of three. A top view schematic illustration of a two-layer design is shown in Fig. 7(a), in which the reservoirs and flow channels are fabricated in the bottom layer (pink) and the control channels are fabricated in the top layer (light blue—note that overlapping structures in the top and bottom layers are shown in darker blue). In this design, the pump reservoirs are 3.8 mm in diameter and 70 μm tall, and the flow channels are 650 μm wide and also 70 μm tall. The reservoirs have a membrane thickness of only 17 μm , and the reservoir edges are rounded just like the flow channels since the mold for the bottom layer is made with thermally reflowed AZ-50XT photoresist similar to the method described earlier. The rounded reservoir edges are illustrated in the side view shown in Fig. 7(b). In the control layer, all of the control channels, valves, and reservoir actuation chambers are 30 μm tall, and the reservoir actuation chambers are 4 mm in diameter. The two PDMS layers are bonded together with the same process as for the three-layer pump.

Using the method described in Ref. 14, the PDMS device is bonded onto a glass die with Sylgard 184 curing agent as an adhesive and baked at 70 $^{\circ}\text{C}$ for 30 min. The bond strength

FIG. 6. Average volumetric flow rate as a function of (a) actuation period and (b) actuation pressure.

between the PDMS and the glass is strong enough to eliminate the need for a top glass piece and clamping device. Figure 7(c) shows a photo of a completed device with tubing connected to all of the fluid and control ports. Valve V4 can be used to isolate the sensor channel from the reservoirs to permit independent filling and flushing of the reservoirs. During pump operation in which R1 and R2 are reciprocally actuated, valve V4 is open and the other valves are closed.

For a two-layer device, the rounded reservoir edges permit more of the fluid in the reservoir to be expelled when the reservoir is actuated. The situation is exactly analogous to making a PDMS valve with a square-edged flow channel versus a rounded-edged flow channel. The former often will not fully collapse, whereas the latter is easier to fully collapse.¹⁵ In addition, the thinner reservoir membrane permits the reservoir to be collapsed with less pressure than would otherwise be needed. The result is illustrated in Fig. 7(d), in which the measured displaced volume as a percent of the total reservoir volume is plotted as a function of the actuation period for both two-layer and three-layer pump designs. For a 0.5 s actuation period, 93% of the two-layer reservoir volume is expelled compared to 76% for the three-layer case. Hence the two-layer design is not only simpler to fabricate but can also expel a larger fraction of the fluid contained in a reservoir.

FIG. 7. Schematic (a) top view and (b) side view of two-layer pump design. The side view is through the dashed line through reservoir R2 in (a). (c) Photo of fabricated two-layer pump design. (d) Displaced volume from a pump reservoir as percent of total reservoir volume for two-layer and three-layer pump designs. In both cases, the actuation pressure is 15 psi. The membrane reservoir membrane thicknesses are 17 and 50 μm , respectively.

FIG. 8. (a) Schematic illustration of two-layer pump integrated on top of 10 mm × 14 mm silicon-on-insulator chip. The flow channel between the reservoirs is positioned over a microcantilever array capable of simultaneous photonic readout of each microcantilever's deflection. (b) Measured deflection for array of microcantilevers during 24 cycles of reflow pump operation. For this particular sample, nine working microcantilevers are monitored; the others in the array suffered from various fabrication defects. (c) Measured deflection during one half period of reflow pump operation.

D. Reflow pump integration with microcantilever array

We now consider a brief example of integrating a reflow pump with a microcantilever sensor array. In this case, the microcantilever array is used to measure the flow rate in the flow channel connecting the reflow pump's reservoirs. A schematic illustration of the two-layer reflow pump is shown in Fig. 8(a) integrated onto a silicon-on-insulator (SOI) microcantilever array chip having the same layout as reported in Ref. 16 and fabricated as described in Ref. 17. The waveguide-based microcantilever differential photonic readout system is discussed in Refs. 18–20. It permits simultaneous readout of all microcantilevers in an array. In Fig. 8(a), the flow channel of the reflow pump is positioned over a 16-microcantilever array. The other microcantilever arrays on the chip are not used. The PDMS device is bonded directly to the SOI microcantilever array chip with the same process as noted above for bonding to glass. Operation of the waveguides on the microcantilever array chip is not affected by the bonded PDMS. The working fluid for deflection measurements is $1\times$ phosphate-buffered saline (PBS) solution (pH 7.4) with 0.2% Triton-X100 surfactant to help prevent bubbles during introduction of fluid to the microcantilever array.

As shown in Fig. 8(b), microcantilever deflection is monitored as a function of time as the reflow pump is actuated with 15 psi and a 5 s period. During reservoir actuation, fluid flow through the channel causes each microcantilever to deflect due to Bernoulli's principle since the microcantilevers are not centered vertically in the flow channels. The magnitude of the deflection response in flowing from reservoir R2 to R1 and vice versa [see Fig. 7(a) for definition of reservoir labels] is somewhat different. This appears to be due to the different bend geometries at the ends of the microcantilever array. A close-up of the microcantilever deflection for a single reservoir actuation (R1 to R2) is shown in Fig. 8(c). As expected from Ref. 17, there is some variation in individual microcantilever response. However, the average deflection across the microcantilevers of an array has been shown to be quite consistent.¹⁷ In this case, the average maximum deflection is 343 nm. Based on calibration with known flow rates using a syringe pump, this corresponds to a flow rate of 180 $\mu\text{l}/\text{min}$.

VI. SUMMARY

We have introduced microfluidic closed-volume reflow pumps in which a small sample volume is rapidly passed back and forth through a fluid channel between two reservoirs. We have demonstrated both two-layer and three-layer pump designs and have used a meniscus tracking system with a high speed camera to measure the volume of fluid displaced between the reservoirs as a function of time, and to characterize the average volumetric flow rate. We find that for the three-layer design with a 300 nl sample volume, the average VFR can be as high as 500 $\mu\text{l}/\text{min}$. Moreover, the two-layer design can pump up to 93% of the sample volume during each half period. We have also demonstrated reflow pump integration with a single-chip SOI microcantilever array. The reflow pump concept is motivated by the need to efficiently use sub- μl sample volumes in conjunction with convection to maximize analyte flux to a surface-based sensor region in order to minimize sensor response time.

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