Modularity in the Design and Volume Production of Microfluidic Devices

J. Podczerviensky, and L. Levine

ALine, Inc., 2206 E. Gladwick, Rancho Dominguez, CA 90220; info@alineinc.com

ABSTRACT

Robust and scalable production of functional components integral to microfluidic devices will drive down their cost and improve their performance. We have developed a modular and readily customized approach for routine production of complex microfluidic devices for point of care and sample-to-answer applications. In this paper we present a test design that meters, mixes and pumps fluid reproducibly using a modular fabrication approach.

Keywords: microfluidics, Lab-on-a-chip, sample processing, sample-to-answer, volume production.

1 INTRODUCTION

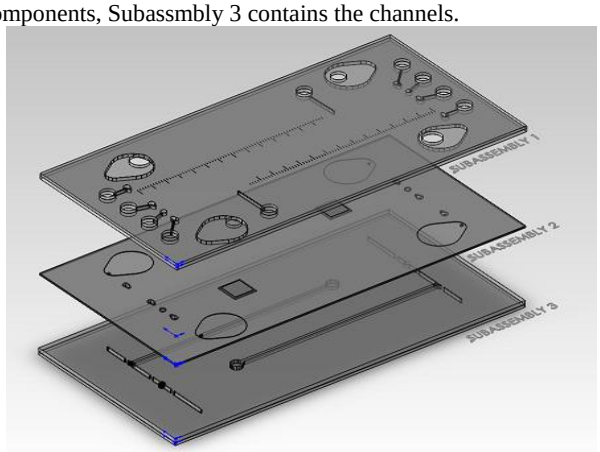
An important requirement for product development is the ability to quickly and iteratively test for functional performance. This is particularly true of microfluidic devices which are complicated by having a high surface area to volume. The behavior of fluids in microfluidic devices is strongly influenced by the surface tension created between the fluid and the plastic it comes in contact with. While modeling of this behavior influences design strategies, the ability to test the performance in real devices drives the product development process. Customers quickly move from 20 or 30 devices, to hundreds and then thousands in support of clinical trials and product launch.

The polymer laminate approach was pioneered in Paul Yager's lab at the University of Washington in the 1980's [1]. It has been used extensively for prototyping microfluidic designs for Lab-on-a-Chip applications. The advantage of the polymer laminate approach is that it is a true rapid prototyping technique that does not require tooling to create a functional device. The process uses designs created in a CAD package which are laser cut. Each laser cut layer is then bonded together using pressure sensitive or thermal bond adhesvies. Polymer Laminate Technology (PLT) offers other advantages not found in traditional rapid prototyping techniques; principally the ability to use a variety of dissimilar materials that are often necessary to create the many functional requirements of microfluidic devices which include pumps, valves, and venting membranes.

Beyond rapid prototyping, PLT is also very compatible with well known roll-to-roll processes for high volume manufacture in which the piece price is driven down to the holy grail of "one dollar".

Our focus has been on the development of the intermediate production capabilties that bridge the gap between rapid prototyping and high volume manufacture (more than 1 million devices per year). To do this we have modularized the production process by designing devices that put the complex components for valves, vents and pumps into one sub assembly. Modularity facilitates design standardization and in process QC, creating a roadmap to high volume production.

Figure 1: Exploded View of Test Device; Subassembly one contains the reservoirs and pneumatic connections to the manifold, Subassemlby 2 contains the vent membrane the valve actuation components, Subassmbly 3 contains the channels.



2 MATERIALS AND METHODS

In this paper we explore the reproducibility of the functions that are incorporated into the device using our modular fabricaton approach. We report here on the dimensional stability of channels of different sizes and the reproducible pump volumes produced by using the PLT process. We also are interested in the inter- and intra-device repeatability of test devices with different widths, depths, and pump area and evaluate the variability of the devices with reference to their geometry, and the optimal geometries to use to get the best performance using the polymer laminate platform.

A series of fluidic cards were constructed using commercially available materials including PET (polyethylene ester terphthalate) and cast acrylic (Polymethyl methacrylate). The bonding adhesvies included silicone and solvent acrylic-based pressure sensitive adhesives. PET can be obtained in nominal thicknesses of 12.5 to 250 microns, with a variability of +/- 10%. The

acrylic is typcially 1 to 1.5 mm thick (+/- 10%) The acrylic and silicone pressure sensitive adhesives are transparent and biocompatible with available thicknesses of 25 and 50 microns (+/-5%). The thickness of the channel layer was varied which in turn varied the depth of the cylindrical pump. Within a given pump height, devices were constructed with pump radii varying from 1mm to 4mm.

A pneumatic test station was used to provide regulated pressure and vacuum using a set of Lee Valves with computer control using LabView.

2.1 Device Fabrication

The devices were built in a batch based, modular approach, broken down into subbassemblies for the critical components of the device. Subassemblies are created for the rigid acrylic layer which contains the fluid resevoirs and manifold for the pumps and valves, a second, flexible subassembly for the pumps, valves, vent membranes, and vias to connect the fluid resevoirs to the channels, and a third subassembly for the channel layer, as shown in Figure 1. Having the separtate subassemblies allows for in process QC of the cards critical components before the finished product is made. The devices can be made into fully functional stand-alone devices or can be made to be easily integrated with a plastic injection molded part or to a sensor (electromechanical or optical). In the latter case, the laminate plays an integral role in the device by containing the pumps, valves, and fluidic channels for the funcionality of the device.

The subassemblies are built from plastic substrates (PET, and PMMA) which are patterened on a CO_2 laser cutter and cleaned with isopropyl alcohol. The processed substrates are then aligned using pin fixtures that mate with features cut into the sheets and bonded together using pressure sensitive or thermal bond adhesive films. The surface of the substrates can be treated with an atmospheric argon plasma stream which increases the surface energy of the substrate allowing increased wetting out of the adhesive onto the plastic and higher bonding strength. After the subassemblies are built and tested, they are aligned and laminated together with pressure and heat and then diced to release it from the sheet.

2.2 Diaphragm Pumps and Valves

A schematic of the valves is shown in **Figure. 2 a,b**. There is a gap in the channel layer directly under the valves which is fluidically connected when no pressure is applied to the valve membrane. Liquid in a channel can move up a via layer, into the valve seat and back down the via layer to the channel. The valves are actuated by applying air pressure to a flexible membrane located over the valve seat. When pressure is applied, the flexible membrane blocks the vias, preventing the flow through the valve seat. A detailed analysis of the valve structure as well as the optimal operating conditions can be found in our previous work [2].

A schematic of the diaphragm pump can also be seen in **Figure 2 a,b**. The pump action is created by alternating positive and negative air pressure on an area of a flexible membrane layer located above the fluidic layer and below the manifold layer. It is necessary to have a valve on each side of the pump to allow the pump to pull liquid from the inlet side without pulling from the outlet side and push it to the outlet side without backflow to the inlet. The pump is started with the inlet and outlet valve closed as well as air pressure applied to the pump. The inlet valve is opened and a vacuum is then applied to the pump membrane pulling it up, and drawing in liquid or air from the channels. Once the membrane is drawn up completely, the inlet valve is closed. The outlet valve is then opened and positive air pressure is applied to the pump membrane allowing the liquid to be driven out. The steps are repeated at a rate of 5 sec. per cycle until the channel is nearly empty.

Figure 2a: Filling pump.

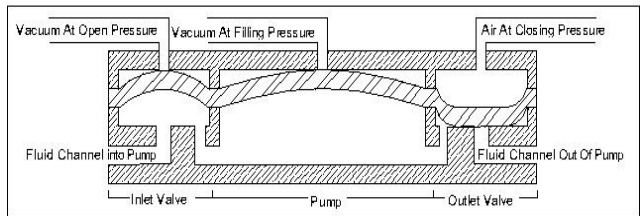
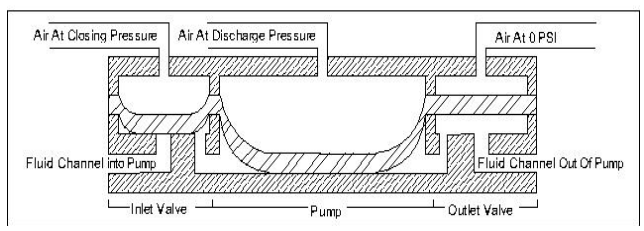


Figure 2b: Discharging pump.



2.3 Test Platform

A test platform was co-developed with Custom Sensor Solutions (Oro Valley, AZ) with computer controlled solenoid valve (Lee Co.) actuation of positive and negative air pressure to the test devices. The platform consists of high (0-60 psi) and low pressure (0-5psi) regulators with digital pressure readout. Air pressure was provided using an air compressor (75 psi). Vacuum was provided by a Tetra aquarium air pump with the outlet reversed, providing 2.5 PSI vacuum. A LabView program was written from routines created in a text document , loaded into LabView, and used to run the experiment. The test devices were connected to the vacuum and pressure sources with hosebarbs and 1/16" ID polyethylene tubing.

Test devices shown in **Figure 1** have a sample and waste resevoir, a cylindrical diaphragm pump with valves on inlet and outlet side of the pump, and a channel of fixed volume for metering of the pump. Tick marks were etched into the top of the acrylic layer along the metering channel at 1 mm increments to measure the volume pumped.

2.4 Test Methods

Pumping Efficiency as a Function of Device Geometry

We evaluated the pressure required to dispense the fluid in the pump by varying the height of the pump below the pump membrane from 0.203 to .406 mm. The volume varied from 0.64 to 20 microliters (uL) as shown in Table 1.

Table 1. Predicted volume based on pump geometry

(uL)	radius (mm)			
height (mm)	1.000	2.000	3.000	4.000
0.203	0.64	2.55	5.74	10.20
0.254	0.80	3.19	7.18	12.76
0.305	0.96	3.83	8.62	15.32
0.406	1.27	5.10	11.47	20.40

The area above the pump membrane was kept at 50 microns and a constant 2.5 psi vacuum was applied to ensure full efficiency and repeatable pumping.

As the diameter and depth of the pump was changed we observed different metered volumes due to the elasticity of the membrane.

The pump radius and channel width were measured for each device using an optical comparator (Sciencescope Model XT-2000). The magnification used for measurements was 45x. The accuracy at this magnification is 6 microns. This data was used for calculating the variability in channel width.

The volume that was pumped was determined by measuring the distance the water traveled in a channel that was downstream of the pump. The channel was long enough to allow a number of pump strokes to be required to fill the channel. This procedure was repeated for several trials for each discharge pressure and pump dimension.

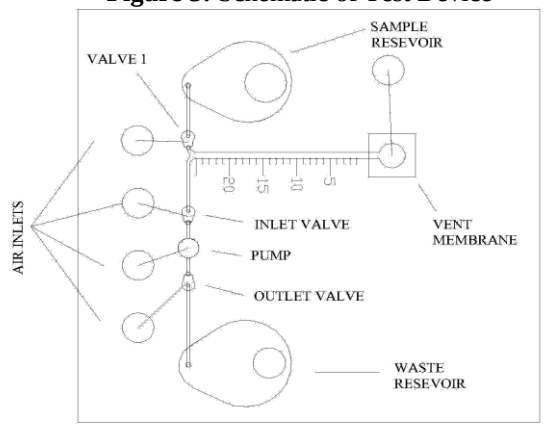
Inter-Device Pump Volume Variability

To measure variability in pump volume from device to device, a series of 7 devices of radii 1.00, 2.00, 3.00 and 4.00 mm were made. The closing pressure for all the valves was maintained at 15 psi. The vacuum to draw the pump and inlet valve diaphragms were maintained at 2.5 psi. The pressure used to discharge the pump varied from 1.00, 5.00, 10.0, and 15.0 psi.

A schematic of the test device is shown in **Figure 3**. RO filtered water was injected into the sample resevoir via a hand syringe. A hand syringe was also used to pull a vacuum on the vent membrane to draw water from the reservoir into the metering channel. Valve 1 in the test device is closed to shut off connection of the sample port with the rest of the device..

The water was pumped out along a distance of the metering channel using the pump routine described in Section 2.2. This routine was created in a text document as a series of binary numbers functioning as on-off commands for the solenoid valves in the test platform which in turn governed the delivery of air and vacuum to the pneumatic

Figure 3: Schematic of Test Device



components of the test device. As the pump was being cycled, liquid would travel down the metering channel into the pump. Once the pump cycling was complete, the distance the water traveled down the metering channel as well as the number of pump strokes were recorded. Each device and pressure required different number of pump cycles to drain the channel, which ranged from 1 cycle to 15 cycles.

Intra-Device Variabiltiy

Using 5 psi, the pump volume was recorded for five repetitions on each device using the same method as the previous experiment for measuring interdevice variability.

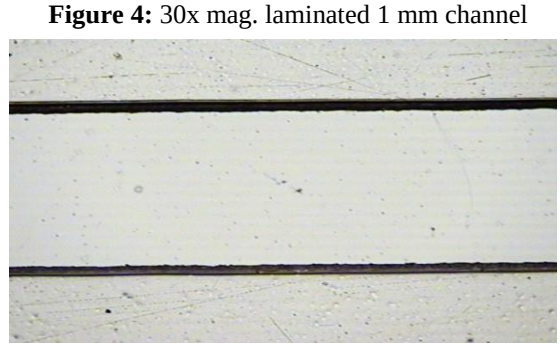
Pumping Efficeincy

By taking into account all the area above and below the membrane that could contribute the pump volume, the efficeincy is calculate as the ratio of the measured pump volume divided by the calculated pump volume multiplied by 100. The efficiency was determined for each device and for the different radii, pump heights, and discharge pressure as well as the inter- and intra- device variability in pump volume.

Channel Dimension Repeatability

Devices with channels ranging from 125 microns wide to 2 mm wide, and from 125 microns to 1 mm tall and up to 5 inches long were fabricated using a variety of lamination strategies. After fabrication and capping the

Figure 4: 30x mag. laminated 1 mm channel



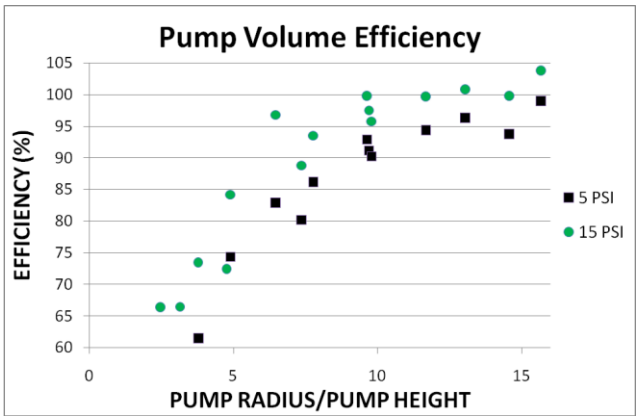
channel, the average width was measured using the optical comparator along the length in several locations, depending on the total length, and the average channel width and standard deviation was calculated.

3 RESULTS

Pumping Efficiency

When the ratio of pump radius to the height of the pump is at least 6, 15 psi of air pressure was sufficient for greater than 95% pumping efficiency as shown in Figure 5 below.

Figure 5: Pump Efficiency vs Ratio of Pump Radius to Height Below Pump Membrane



Inter-Device Variability

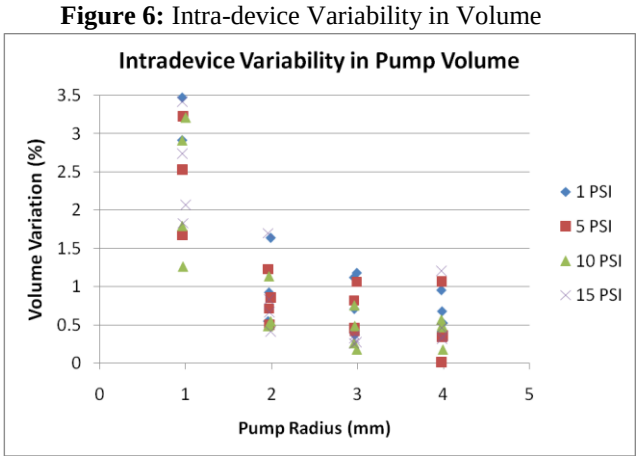
Variability in pump volume decreased nearly linearly as the pump radius increased. Interdevice variability was 6.5% for 1 mm pumps, 4.7% for 2 mm pumps, 4.5% for 3 mm pumps, and 2% for 4 mm pumps as shown in Table 2.

Table 2: Summary of Interdevice Variation				
Radius	1 mm	2 mm	3 mm	4 mm
Avg. Volume Pumped (uL)	0.75	4.10	9.16	16.65
Standard Deviation (uL)	0.05	0.19	0.41	0.34
Percent Variation	6.47	4.71	4.50	2.05

Intra-Device Variability

For pump radii of 2 mm, 3 mm, and 4 mm, the average intradevice variability in pump volume was under 1% and stayed under 1.5% for nearly all of these devices under all pressures. For pump radius of 1mm, the average intradevice variability was 3% and was as high as 8% when the pump pressure was 1psi and the efficiency below 10%.

Figure 6: Intra-device Variability in Volume



Channel Width and Height Variability

Channels shorter than 5 inches can be laminated to other substrates with about 20-25 microns of variation along the length. With typical channel widths of 250-1000 microns, this gives at most 2-8% variability.

Table 3: Summary of Channel Measurements			
Average channel width (mm)	0.55	1.00	1.63
Avg. intrachannel std. dev. (mm)	0.023	0.012	0.010
Avg. intrachannel % variation	4.31	1.21	0.62
Interchannel std. dev. (mm)	0.032	0.021	0.020
Interchannel % variation	5.81	2.06	1.23

4 DISCUSSION

Optimal pump efficiency was acheived at 15 psi for a ratio of pump radius to pump height at or greater than 6. This includes cylinder heights between 0.2 mm to 0.4 mm, and radii greater than 2 mm, with pump volumes between 0.64 and 20.4 microliters. With an alignment tolerance of +/- 75 microns our alignment procedure has little effect on the variability of the volume pumped. In these devices the cylinder is composed of 3 stacked layers.

Initially, the experiment was set up without a vacuum being pulled on the inlet valves membrane to assist the opening of the valve. As the pump would draw liquid in, it would also partially close the inlet valve; restricting flow to the pump. As the pump volume increased, the effect on the inlet valve was greater resulting in a slower and more variable fill rate. With constant vacuum above the inlet valve, the pump would fill in a matter of seconds.

The pump volume is dependent linearly on height and on the square of the radius, therefore variablity in the thickness of material would have little influence on the pump volume compared to the effect of mis-alignment on the radius. With these alignment tolerances, pump performance above a 1 mm radius is acceptable.

5 CONCLUSION

The fabrication process is critically important for performance of microfluidic devices. By chosing instrument parameters and device geometries that minimize both inter and intra-device variability, laser cut and laminated devices are suitable for commerical products. Scalability of the production process has been demonstrated using a modular fabrication process for complex on board processes including valves, pumps and vents.

REFERENCES

[1] P. Yager, "Bioengineering Tutorials"

[2] J. McDowell, J. Goldstein, W. Penrose, L. Levine, "On-Board Pneumatic Valves for Multiplexed Applications in Microfluidic Devices," info@alineinc.com, 2008.