Electronic Supplementary Information

S1 Chip Dimensions

Devices are made out of 4 layers of polycarbonate, 1 PDMS layer for valves, and 2 silicone tape layers for bonding. Dimensions are given in Figure S1.1, showing the 4 polycarbonate layers, 1 PDMS layer separating the liquid and gas layer and 2 silicone tape layers to attach two polycarbonate manifold layers for proper external connections.

Liquid inputs are located at the bottom of the device and air inputs are located at the top of the device to prevent punching holes into the PDMS membrane and reduce fabrication complexity.

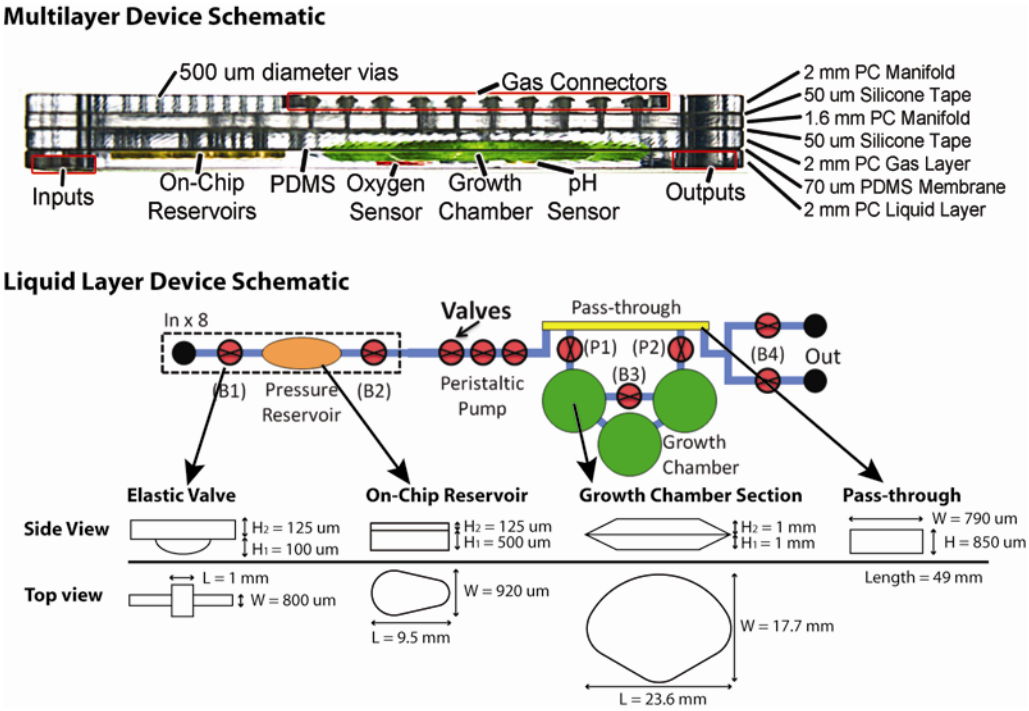

Figure S1.1 Top: actual side view of the fabricated device showing the false color locations of chip components as well as the layer thicknesses for the 7 individual layers.

As shown in Figure S1.1, devices require multiple thickness with dimensions ranging from 100 μm to 1 mm in order to accommodate large volume growth chambers and small volume PDMS valves. Growth chamber wall profiles are also machined with 30 degree angles and rounded corners to enable full membrane deflection to remove growth chamber volume.

A summary of the fabrication process for making the multilayer device is given in Figure S1.2.

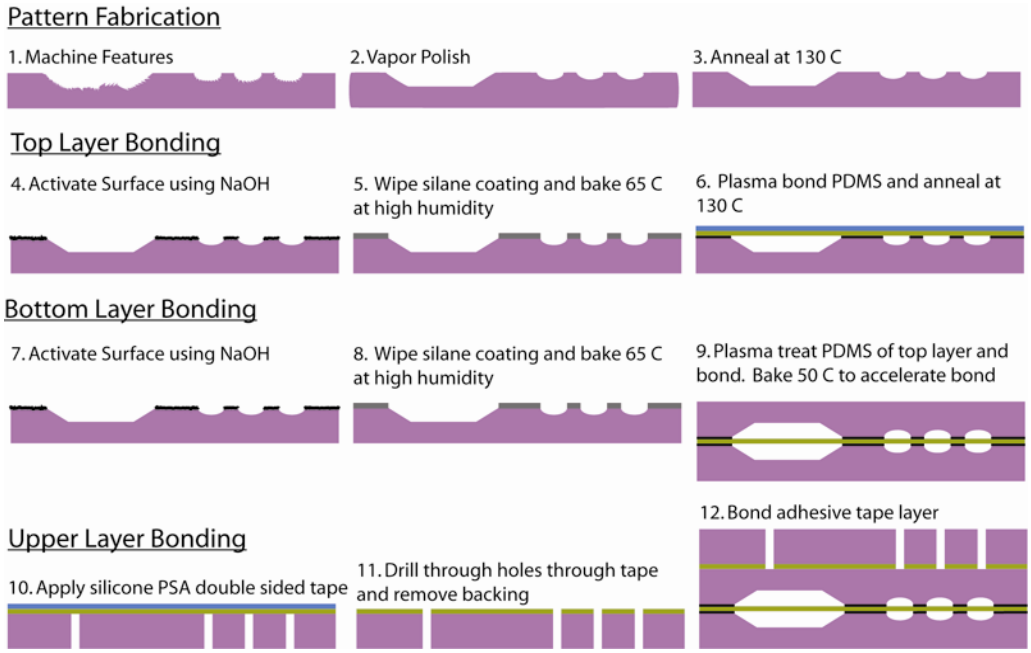

Figure S1.2. Summary of fabrication and assembly steps for making the continuous culture device. Polycarbonate layers are machined and vapor polished. Then PDMS membranes are bound through a silane surface treatment. Finally, upper manifold layers are added using double sided silicone adhesive tape.

S2 Continuous Culture Sterility

After the continuous culture experiment, growth media is carefully removed from both the growth chamber and from the on-chip reservoirs (labeled the Premixer) while keeping the peristaltic pump closed to prevent contamination. Media is then streaked onto plates containing agar and LB (Luria-Bertani) media. As shown in Figure S2.1, The cells from the growth chamber are the same as the stock culture and the fluid upstream of the peristaltic pump is still sterile.

Figure S2.1 Contamination streaks of the growth chamber and premixer fluids after the continuous culture experiment. The cultures are streaked on the same plate as the initial stock culture demonstrating that there is no contamination of the feed and that the cells have not been overrun by a foreign organism.

S3 Flow rate measurement

To characterize the flow rate of the peristaltic pump for 10 nL volume changes, a measurement system utilizing a triggered CCD camera (Opteon) and a 600 um inner diameter glass capillary (McMaster 8729K57) tube is used as shown in Figure S1.

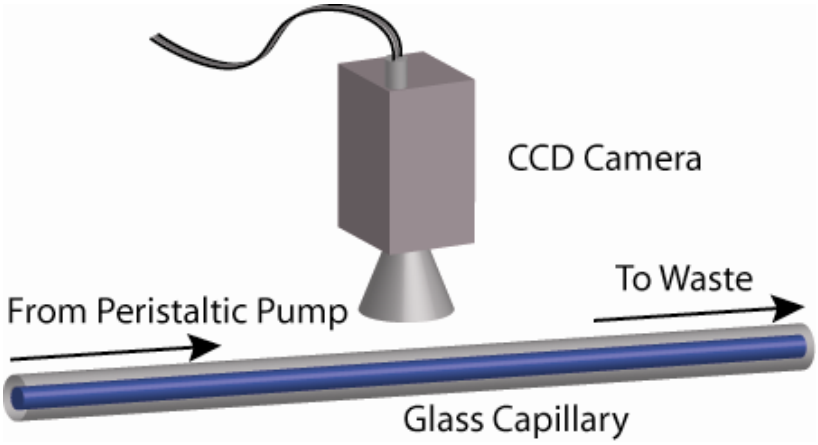

Figure S3.1 Flow rate measurement system. Images are taken every pumping cycle to determine the distance traveled by the fluid plug.

To determine the volume injected per pumping cycle, images were acquired every pumping cycle triggered to step 1 in the cycle, the last step before the output experiences a volume change. Neighboring images were subtracted to obtain a difference image which was then filtered with a threshold into a binary image, removing any differences resulting from spurious noise. The pixel farthest ahead was then stored as the leading edge of the liquid in the capillary. An example of the image processing is shown in Figure S2.

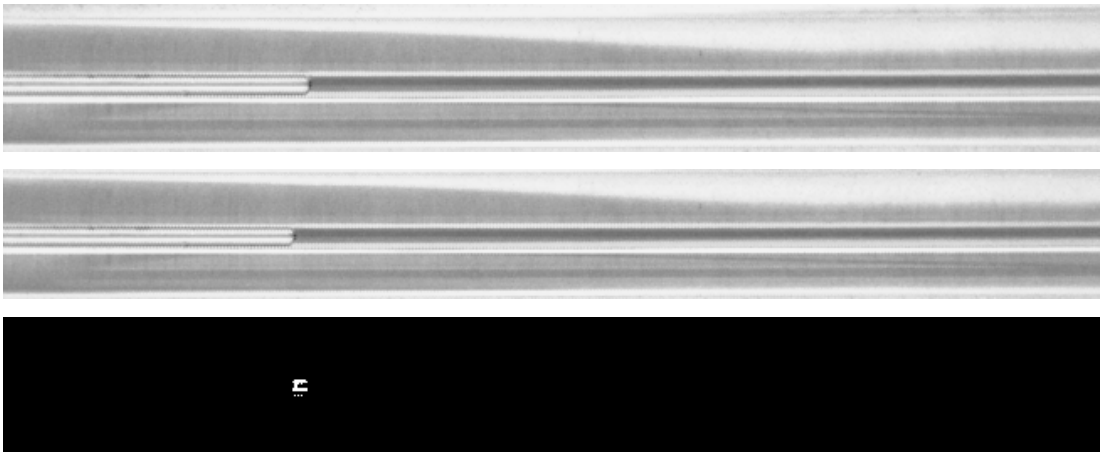

Figure S3.2. Images from two neighboring frames and the processed difference image showing the position and size of the additional fluid plug.

In order to maintain a reasonable field of view to accommodate about 50 injections, the camera was focused on a 40 mm length of the capillary over 626 pixels. For a capillary cross-sectional area of 0.283 mm^2, the volume resolution of the measurement system is approximately 18 nL per pixel. If each injection is 200 nL, then the measurement system can accommodate 55 full injections before the liquid leaves the field of view of the camera.