

Electronic supplementary information for

Microfluidic chip with movable layers for the manipulation of biochemicals

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Pressure generated at moment of contact:

To measure pressure variations in the chambers, we connected a pressure sensor at the bottom side of the bottom chamber (Figure S1). The pressure sensor was connected to a data acquisition unit (NI USB-6008, National Instruments) and then linked to a PC running LabVIEW software. We set the reference position ($z = 0$ mm) of the top chamber as the bottom and top chambers contact with a mild contact force. Then we demounted the top and bottom layers, loaded solutions into the chambers, and mounted the layers in the instrument.

For the experiment shown in Figure 4c of the main text, the bottom and top chambers did not merge initially at $z = 0.25$ mm, and the pressure in the bottom chamber was 0 Pa. Then, at $z = -0.1$ mm, the top chamber moved down and mechanically pressed the bottom chamber, increasing the pressure. Afterwards, stepping the motors up by 0.25 mm every 5 s created a stepwise change of pressure, and a liquid bridge was formed between the chambers. Pressure

became negative and finally returned to zero at the moment of liquid bridge rupture. This result shows that, in the normal case ($z \geq 0$), the pressure inside the chambers is attributed to Laplace pressure generated by the concave shape of the liquid bridge.

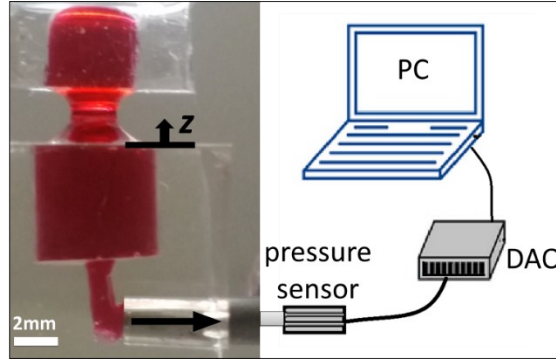


Figure S1. Experimental set up for measuring the pressure inside the chambers.

Selecting tip geometry:

To fully collect beads, we examined trapezoid tips (sharp, moderate, and wide tip size) and round-vertex-cone tips (Figure S2). Polydimethylsiloxane (PDMS) was used to fabricate the tips using 3D printed molds. All PDMS tips were treated with oxygen plasma to make the surface of the tips hydrophilic. In the experiment, we used a bead with 2 μm diameter and a 150 mT external magnetic field. The tips were dipped to the same depth in the bottom chamber to collect beads. Figure S2 illustrates the bead collection process. States 1 and 2 show the photos just before and after rupture of liquid bridges, respectively, under the application of the magnetic field. State 3 shows the photos after the magnetic field was removed. For the trapezoidal tips, the liquid bridge profile at state 1 shows that a considerable number of beads existed below the neck of the liquid bridge. At state 2, the number below the neck did not remain at the tip after the liquid bridge ruptured. Rather, they had moved down to the bottom chambers. This reduces the bead-collection efficiency. When we removed the magnetic field at state 3, the beads at the top

of the bottom chamber sunk. On the other hand, a round-vertex-cone tip shows that, at state 1, all beads were located above the neck of the liquid bridge, thereby achieving high bead collection efficiency at state 2. Thus, the round-vertex cone shape was used in the remainder of the study due to its high collection efficiency.

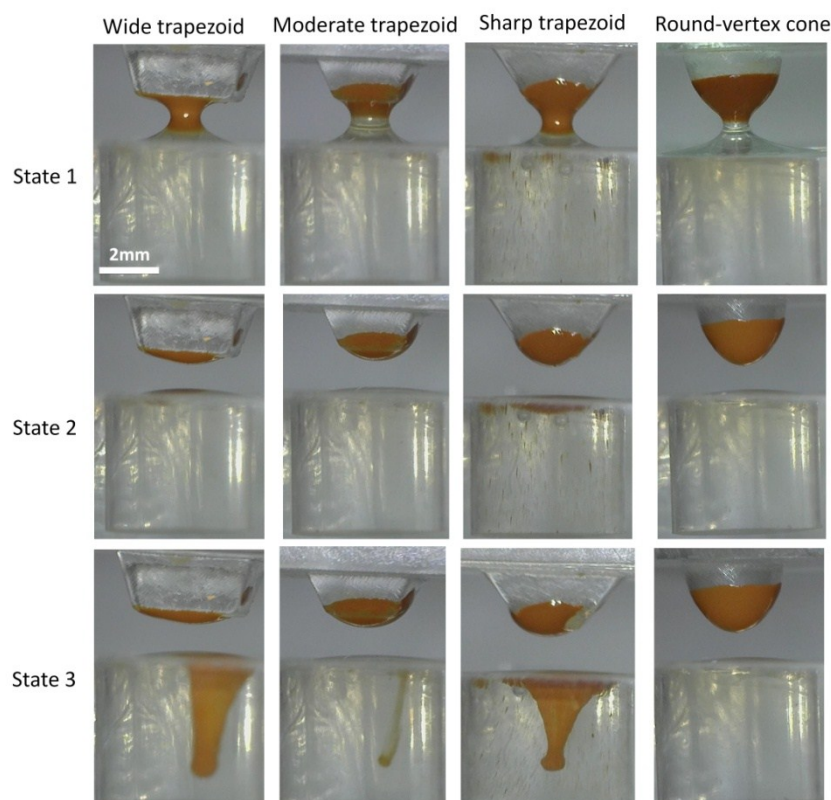


Figure S2. Effect of tip geometry on bead collection.

Automated DNA extraction on the chip:

We coded the motions of top and bottom layers in open-source software (IDE, Arduino). Then we uploaded the code on microcontroller, which in turn controls the stepper motor for the DNA extraction process. Figure S3 illustrates the automations extraction process where lysis, bead binding, washing1&2 and elution are performed. For DNA extractions, initially, the A549 crude cells and reagents were pipetted on the chambers. The place of chamber that contains crude cells

was on the top chip. It was aligned with lysis chamber on the bottom layer, then brought in contact to perform lysis process. Twenty sequential contacts were executed to assure mixing of cells with the lysis solution. Cell Lysis took 20 min at room temperature. In the second step, the bottom layer rotated 45° to allow the chamber with binding buffer that contains magnetic silica bead to align with the lysed cell mixture, then the top layer made another 20 times sequential contacts between the binding buffer chamber and the chamber with lysed cells, so magnetic silica beads were added to the lysed mixture. Binding process took 1 min and positively charged magnetic beads were bounded with negatively charged DNA fragments. At this stage, DNA was ready to be collected so the tip was brought to mixture chamber while magnetic field was applied to collect the beads at the tip. After the bead collection at the tip, the bottom layer rotated 45° to align the tip, which has the beads, above the washing chamber. The tip released the beads on washing buffer for 1 minute then collected them. Washing process was executed twice to assure protein-free DNA. For elution, the beads were dropped in the elution chamber to break up the bonding between the silica beads and DNA fragments. Finally, the beads were collected leaving the elution chamber with pure DNA.

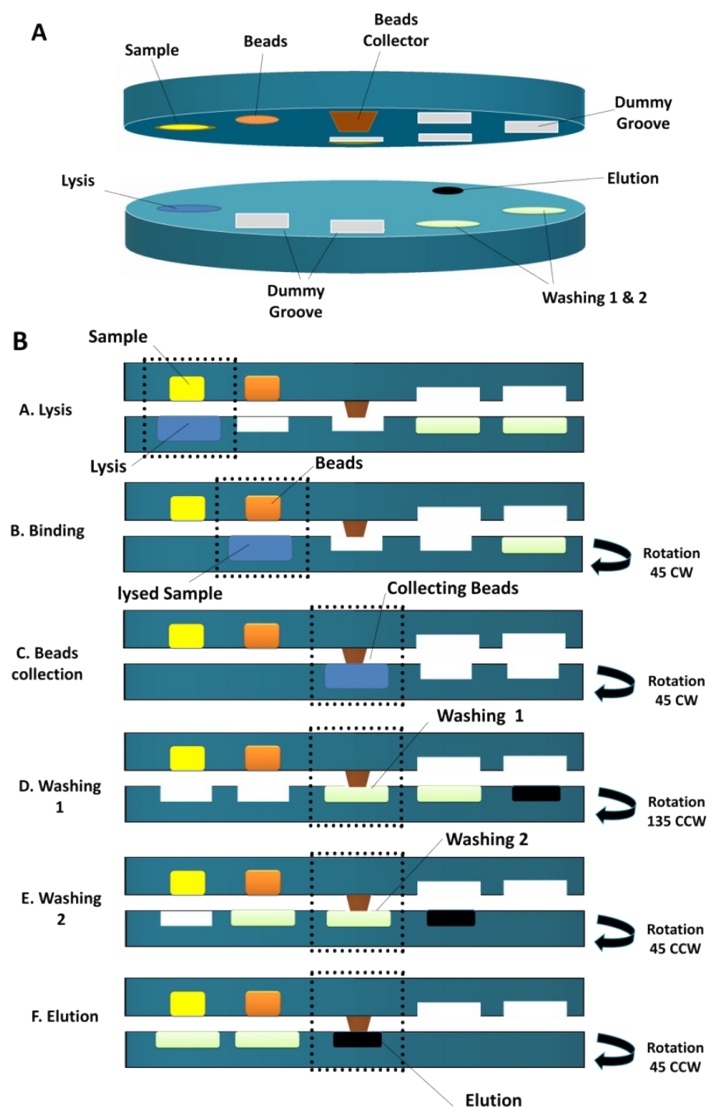


Figure S3. Automated DNA extraction.

GAPDH gene primer information:

The PCR primers were designed according to nucleotide sequences in the GenBank database (Homo sapiens GAPDH gene sequence, GenBank No.NG007073.2).

Supplementary Table S1

Primer name	orientation	Primer sequences 5' to 3' orientation	PCR product size in bp
GAPDH	Forward	ACCACAGTCCATGCCATCAC	363
	Reverse	GTCAAAGGTGGAGGAGTGGG	