

Electronic Supplementary Information (ESI)

Capillary force-driven reverse-Tesla valve structure for microfluidic bioassays

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Figure S1. Determination of optimal concentration of Triton X-100

Figure S2. Determination of mixing ratio of the sample and Triton X-100

Figure S3. Absorbance and *p*-NA standard curves

Figure S4. Volume of solution at the outlet according to channel length and flow time

Figure S5. Absorbance after trypsin reaction according to reaction time

Figure S6. Comparison of fluid flows between μ -MixMACS chip, GASI chip, and Tesla chip using computational analysis

Figure S7. Comparison of (a) mixing concentration and (b) cross-sectional view of the outlet after particle motion between μ -MixMACS chip, GASI chip, and Tesla chip using computational analysis

Formula. Calculation formula for reaction efficiency (RE)

Detailed information of nucleic acid amplification:

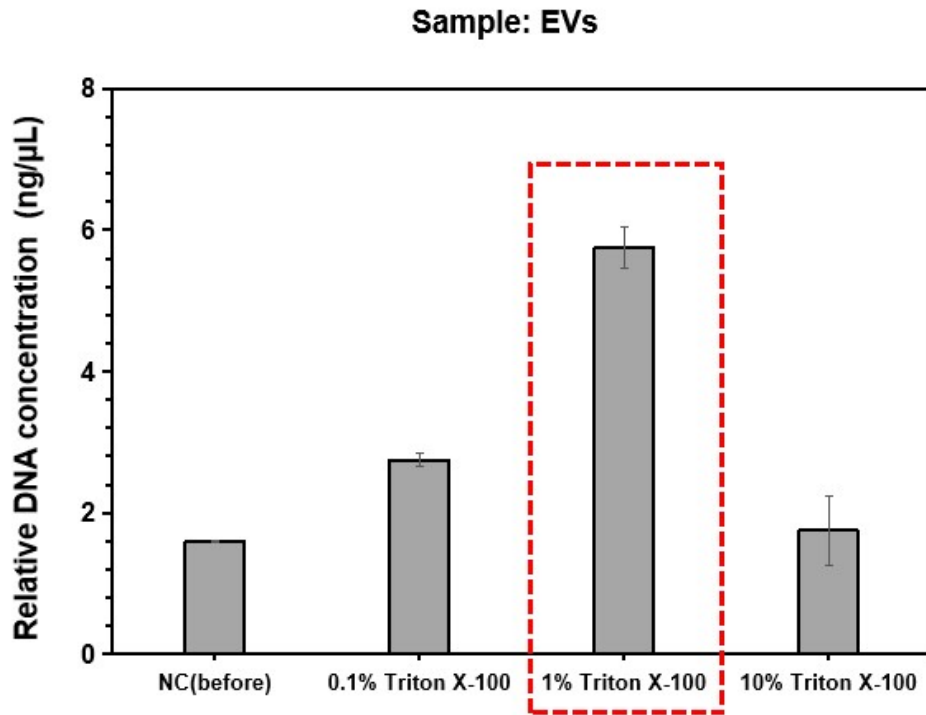


Figure S1. Determination of optimal concentration of Triton X-100

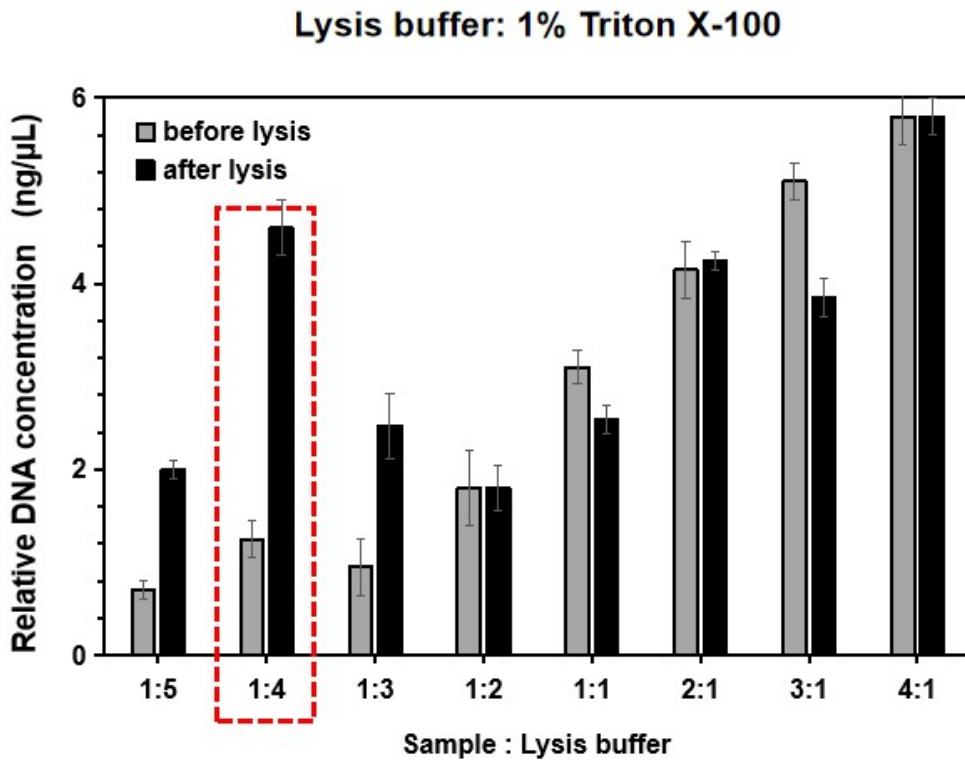


Figure S2. Determination of mixing ratio of the sample and Triton X-100

p-NA Standard Curve

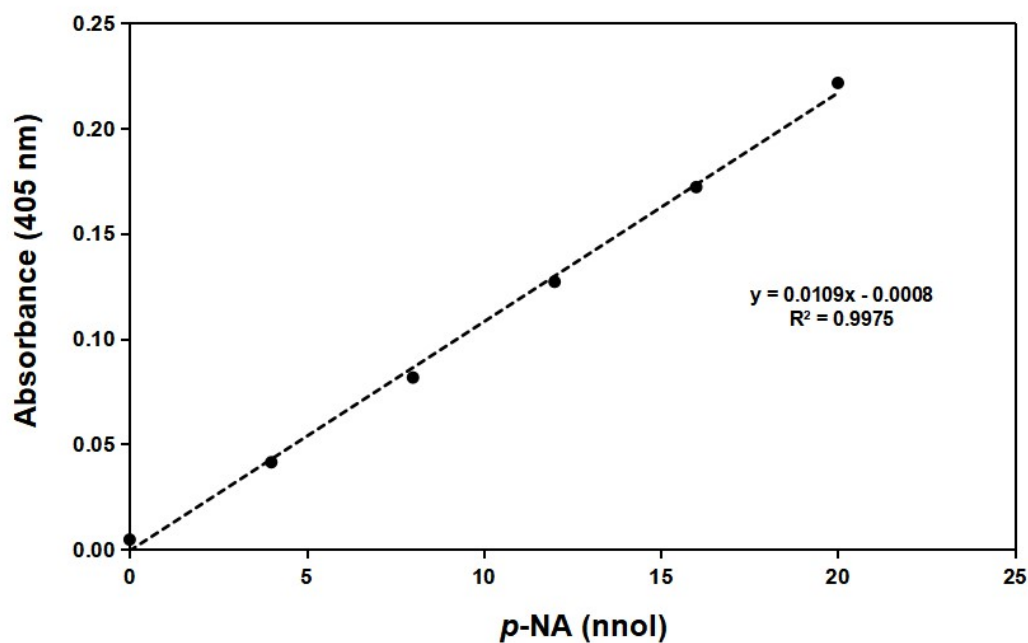


Figure S3. Absorbance and *p*-NA standard curves

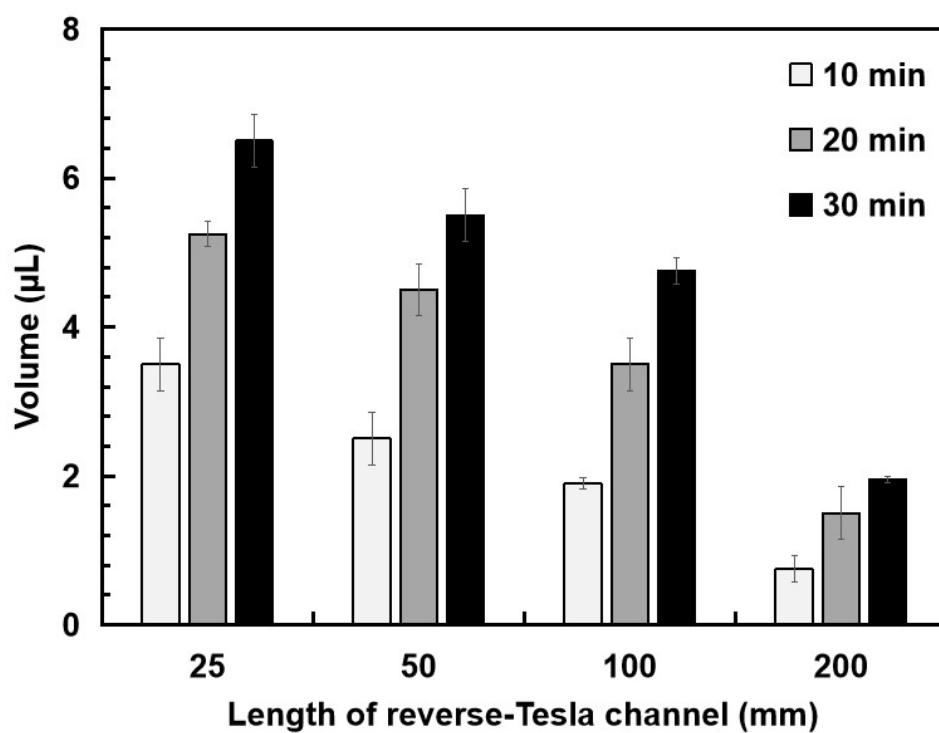


Figure S4. Volume of solution at the outlet according to channel length and flow time

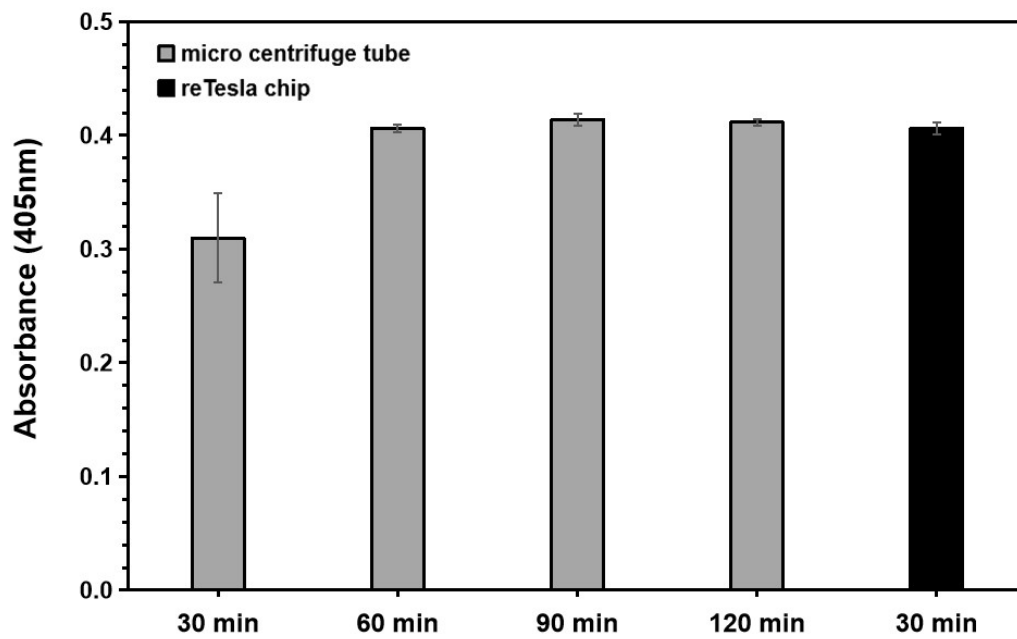


Figure S5. Absorbance after trypsin reaction according to reaction time

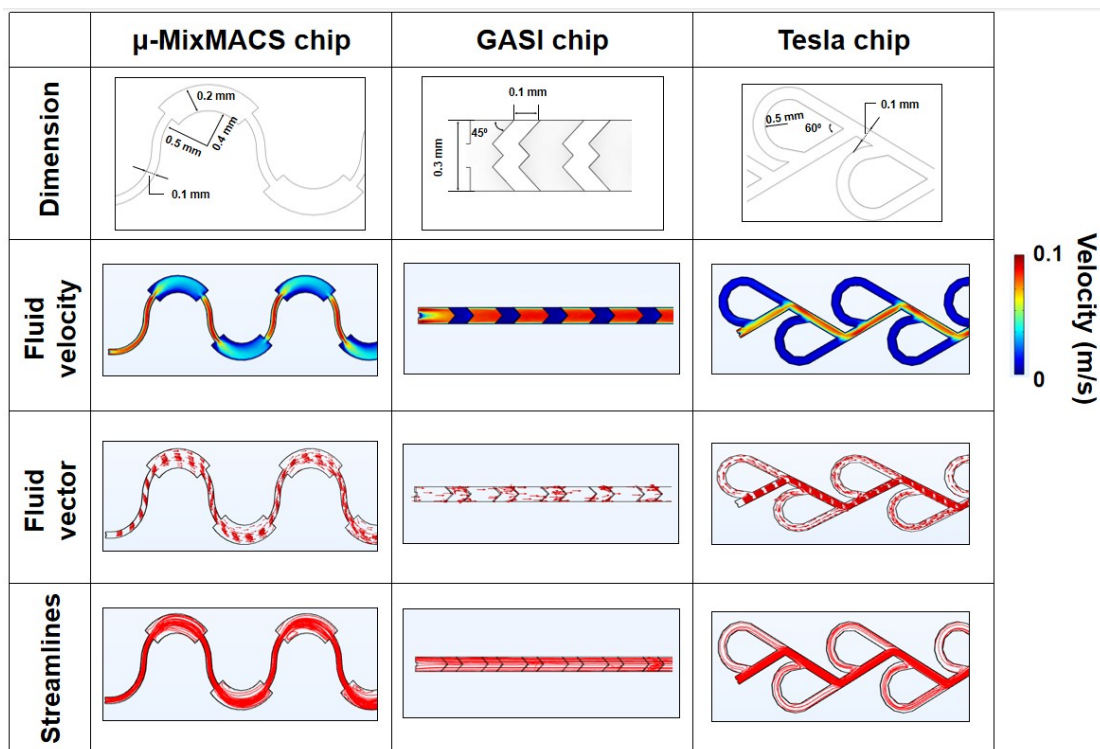


Figure S6. Comparison of fluid flows between μ-MixMACS chip, GASI chip, and Tesla chip using computational analysis

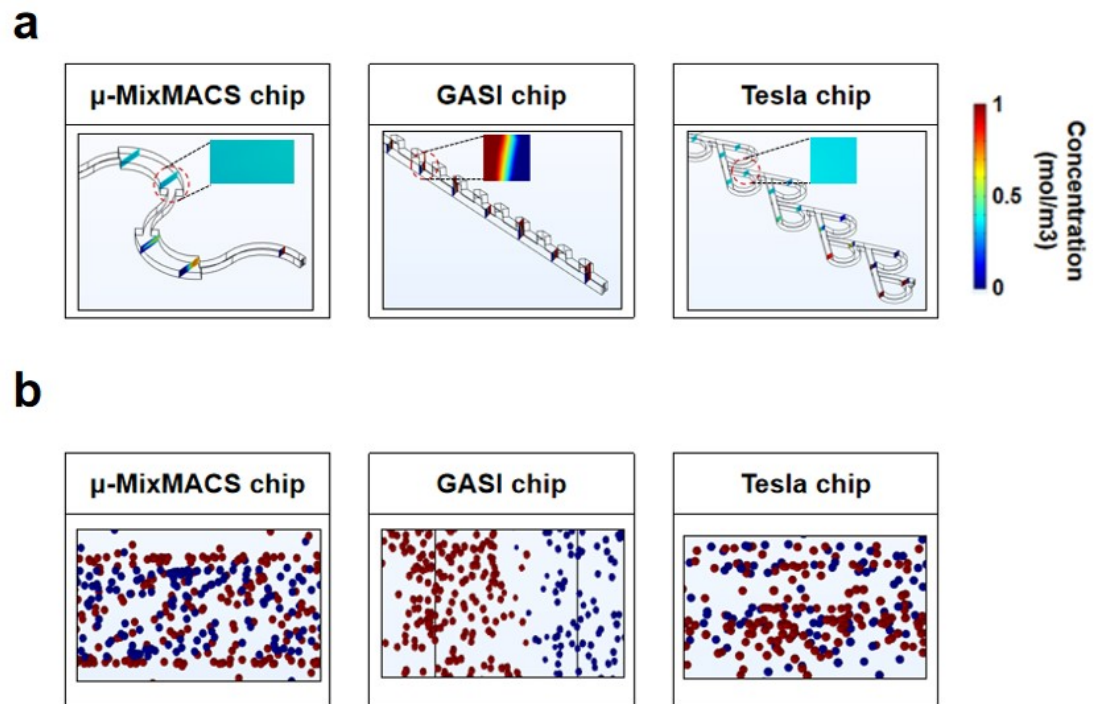


Figure S7. Comparison of (a) mixing concentration and (b) cross-sectional view of the outlet after particle motion between μ -MixMACS chip, GASI chip, and Tesla chip using computational analysis

Formula. Calculation formula for reaction efficiency (RE)

1. The formula for calculating the RE for cell lysis is expressed as follows:

$$RE_{cell\ lysis} = \left(1 - \frac{C_{after}}{C_{before}}\right) \times 100\%$$

where C_{before} is the sample concentration before cell lysis, and C_{after} is the sample concentration after cell lysis.

2. The formula for calculating the RE for the proteinase assay is expressed as follows:

$$RE_{proteinase\ assay} = \left(1 - \frac{C_{before}}{C_{after}}\right) \times 100\%$$

where C_{before} is the *p*-NA content before trypsin cleaves the substrate, and C_{after} is the *p*-NA content after trypsin cleaves the substrate.

3. The formula for calculating the RE for nucleic acid amplification is expressed as follows:

$$RE_{NAA} = \left(1 - \frac{C_{before}}{C_{after}}\right) \times 100\%$$

where C_{before} is the DNA content before amplification, and C_{after} represents the DNA content after amplification.

Detailed information of nucleic acid amplification:

Target: SARS-CoV-2 N gene. The sequences are shown in Table S1 [1].

The nucleic acid amplification reaction uses LAMP. LAMP is commonly used isothermal amplification technologies that rapidly detect target nucleic acids using LAMP-specific primers and strand-displacement DNA polymerase.

Reagents:

1. WarmStart Multipurpose LAMP/RT-LAMP 2X Master Mix with UDG (NEB #M1708) (consisting of Bst 2.0 WarmStart DNA Polymerase, Isothermal Amplification Buffer, MgSO₄, and dNTP mix) (New England Biolab USA)

2. 50X LAMP fluorescent dye (B1700SVIAL) for fluorescence measurements. (New England Biolab USA)

Fluorescent probe (P): For the detection of SARS-CoV-2 N gene, FAM-labeled probe was used. FAM (Carboxyfluorescein) is a fluorescent compound with an excitation peak at 493 nm and an emission peak at 517 nm.

3. LAMP Primer: forward inner primer (FIP), backward inner primer (BIP), forward loop primer (LF), backward loop primer (LB), forward outer primer (F3), and backward outer primer (B3) (IDT, USA) [2].

Table S1. Target (SARS-CoV-2 N gene) fragment sequences [1]:

SARS- CoV-2	<i>N</i> <i>gene</i>	AUGUCUGAUAUAUGGACCCCAAAAUCAGCGAAAUGCACCCCGCA UUACGUUUGGUGGACCCUCAGAUUCAACUGGCAGUAACCAGAA UGGAGAACGCAGUGGGGCGCGAUAACAACAACGUCGGCCCCAA GGUUUACCCAAUAUAUCUGCGUCUUGGUUCACCGCUCUCACUC AACAUUGGCAGAUACCAAAUUGGCUACUACCGAAGAGCUACCA GACGAAUUCGUGGUGGUGACGGUAAAUGAAAGAUCUCAGUCC AAGAUGGUAUUUCUACUACCUAGGAACUGGGCCAGAAGCUGGA CUUCCCUAUGGUGCUAACAAGACGGCAUCAUAUGGGUUGCAA CUGAGGGAGCCUUGAAUACACCAAAAGAUCACAUUGGCACCCG CAAUCCUGCUAACA AUGCUGCAAUCGUGCUACAACUCCUCA GGAACAACAUUGCCAAAAGGCUUCUACGCAGAAGGGAGCAGAG GCGGCAGUCAAGCCUCUUCUCGUUCCUCAUCACGUAGUCGCAAC AGUUCAAGAAAUUCAACUCCAGGCAGCAGUAGGGGAACUUCUC CUGCUAGAAUGGCUGGCAAUGGCGGUGAUGCUGCUCUUGCUUU GCUGCUGCUUGACAGAUUGAACCAGCUUGAGAGCAAAAUGUCU GGUAAAAGGCCAACAACAACAAGGCCAACUGUCACUAAGAAAU CUGCUGCUGAGGCUUCUAAGAAGCCUCGGCAAAAACGUACUGC CACUAAAAGCAUACAAUGUAACACAAGCUUUCGGCAGACGUGGU CCAGAACAACCCAAGGAAAUUUUGGGGACCAGGAACUAAUCA
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CAACAAUCCAUGAGCAGUGCUGACUCAACUCAGGCCUAA

References

- [1] J. Talap, M. Shen, L. Yu, S. Zeng, and S. Cai, *Talanta.*, 2022, 248 , 123644.
- [2] J. P. Broughton , X. Deng , G. Yu , C. L. Fasching , V. Servellita , J. Singh , X. Miao , J. A. Streithorst , A. Granados and A. Sotomayor-Gonzalez , *Nat. Biotechnol.*, 2020, 38 , 870-874.