

Supporting Information

A Cas13a-Microdroplet Platform for Amplification-Free Influenza A virus RNA Diagnostics

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Table S1. DNA and RNA sequences used in this study.

| Name | Sequence (5'-3') |
|---|---|
| Influenza A virus Target RNA | UCUUUCUAUCAUCCCGUCAGGGCCCCCUAAAGCCGAGAUCGCGCAGAGACUGGAAAGUGUCUUUGCAGGAAAGAACACAGACUUGAGGCUCUCAUGGAAUGGCUAAGACAAGACCAAUCUUGUCACCUCUGACUAAGGGAAUUUUAGGAUUUGUGUUCACGCUCACCGUGGCCAGUGAGCGAGGACUGCGUAGACGUUUGUCCAAAUGCCU |
| crRNA | UAAUACGACUCACUAUAGGGAUUUAGACUACCCAAAAACGAAGGGGACUAAAAGCCAUUCCAUGAGAGGCCUCAAGA |
| crRNA IVT Template | TCTTGAGGCTCTCATGGAATGGCTTTAGTCCCCTCGTTTGGGTAGTCTAAATCCCTATA GTGAGTCGTATT |
| RNA Reporter | FAM-UUUUU-BHQ1 |
| Capture Probe | NH2 C12-TTCCCATTTAGGGCATT |
| T7 Promoter | GAAATTAATCGACTCACTATAGGG |
| Influenza A virus - PCR-Forward primer | TAATACGACTCACTATAGGGCTTTCTATCATCCGTCA |
| Influenza A virus - PCR-Forward primer | TTCCCATTTAGGGCATT |

| ACCESSION | Subtype | Gene | Position of forward | | Position of reverse primer | |
|-----------|---------|------|-------------------------|---------------|----------------------------|-----|
| | | | 64 | 84 | 266 | 283 |
| NC_007367 | H3N2 | M2 | ACGT TCTCTCATCGTCCATCA | GGC.....TCC | AAATGCCCTAAATGGGAA | |
| NC_004907 | H9N2 | M2 | ATGT TCTCTCATCCCATCA | AGGC.....TCCA | AAATGCCCTAAATGGGAA | |
| NC_026427 | H7N9 | M2 | ACGT TCTCTCATCCATCA | GGC.....TCCA | AAACGCCCTAAATGGGAA | |
| NC_026431 | H1N1 | M2 | ACGT TCTTCTATCATCCCCTCA | GGC.....TCCA | AAATGCCCTAAATGGGAA | |
| NC_002016 | H1N1 | M2 | ACGT TCTTCTATCATCCCCTCA | GGC.....TCCA | AAATGCCCTAAATGGGAA | |
| NC_007363 | H5N1 | M2 | ACGT TCTCTCATCGTCCCCTCA | GGC.....TCCA | GAATGCCCTAAATGGAAA | |
| NC_007377 | H2N2 | M2 | ACGT TCTCTCATCGTCCCCTCA | GGC.....TCCA | AAATGCCCTAAATGGGAA | |

Figure S1. Alignment of the M2 gene sequences of different subtype of influenza A virus, the gene sequence of strains obtained from National Center for Biotechnology Information is shown in the figure. Highlighted are the primer sequences.

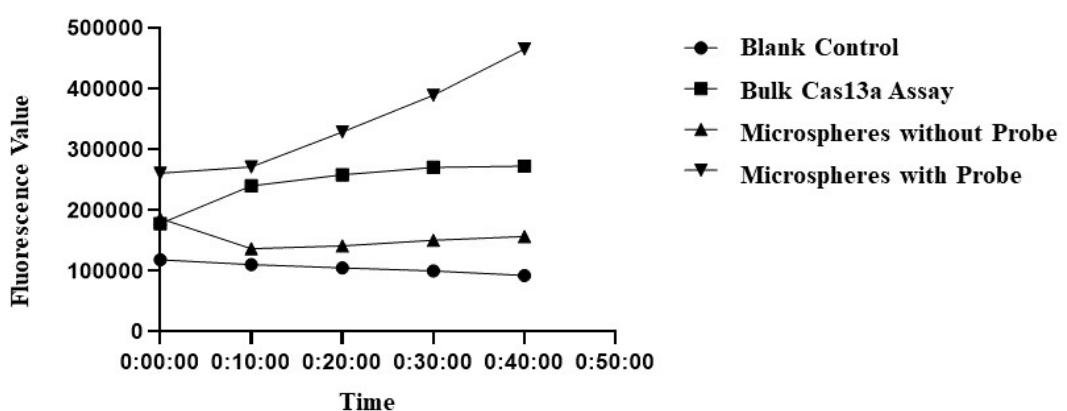


Figure S2. The fluorescence of bulk cas13a assay using the microsphere captured with target RNA.

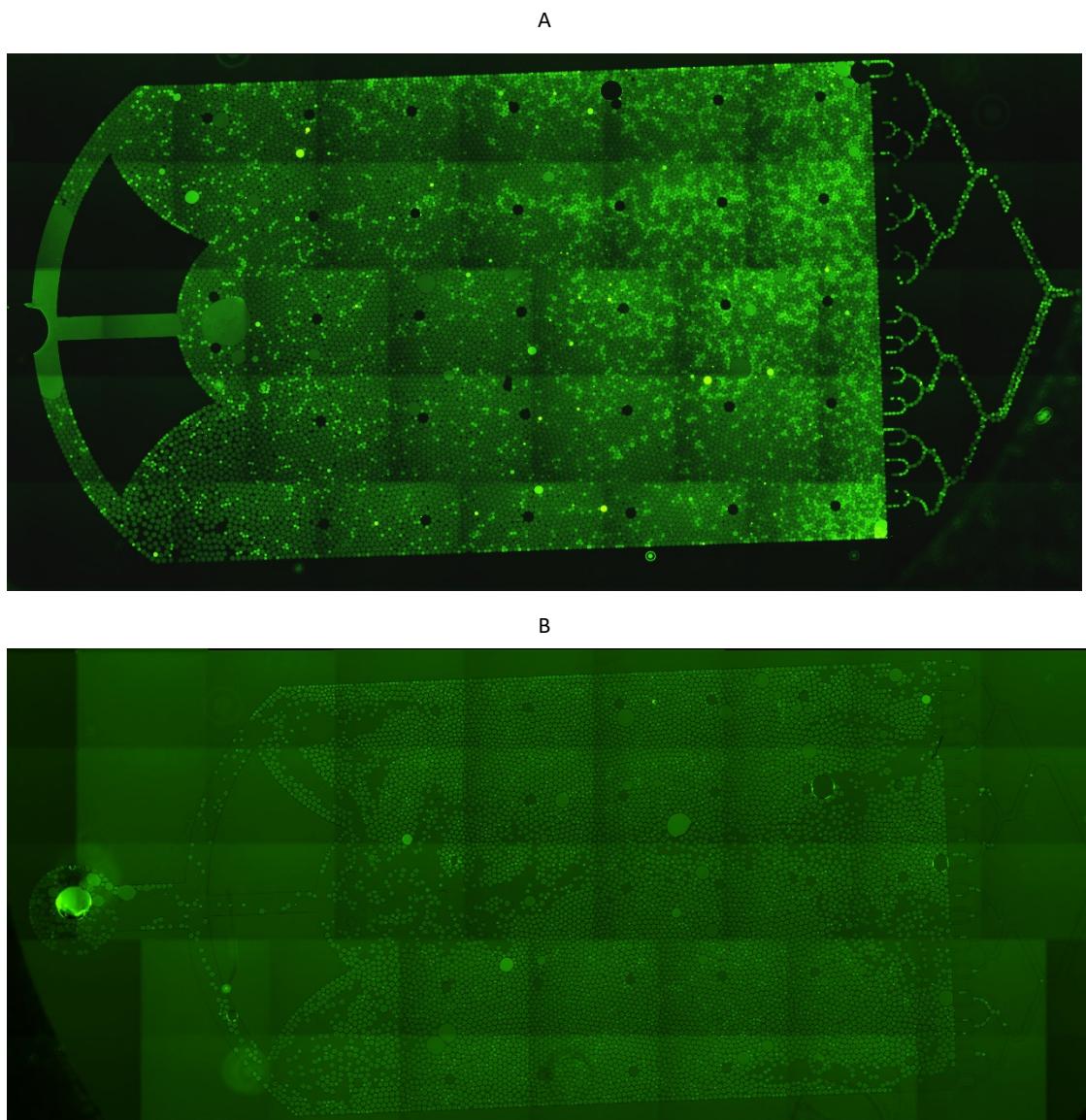


Figure S3. The mosaic fluorescence image of the whole chip with different concentrations of the target RNA. A. The representative fluorescence image for analysis. (10^4 copies/ $20\mu\text{L}$) B. The representative fluorescence image for analysis. (Blank Control)

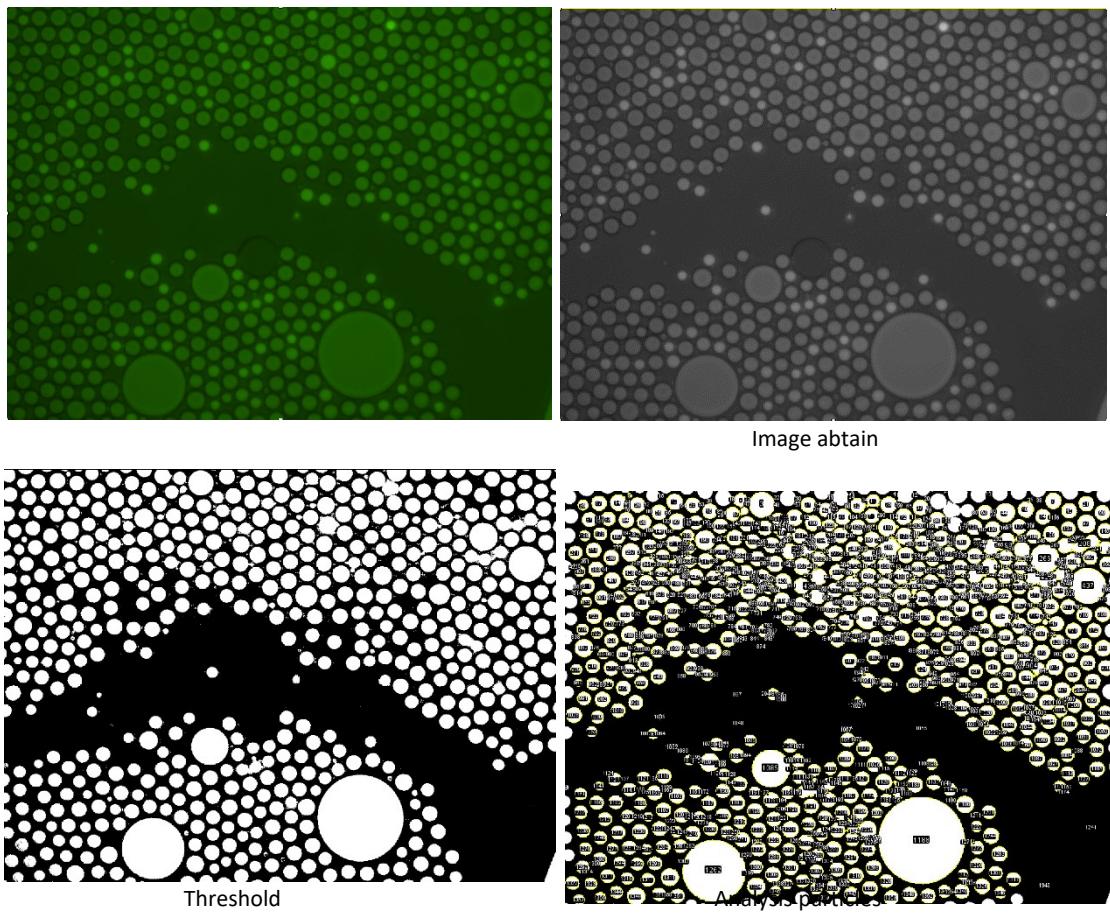


Figure S4. Image analysis and data extraction process of fluorescence image.

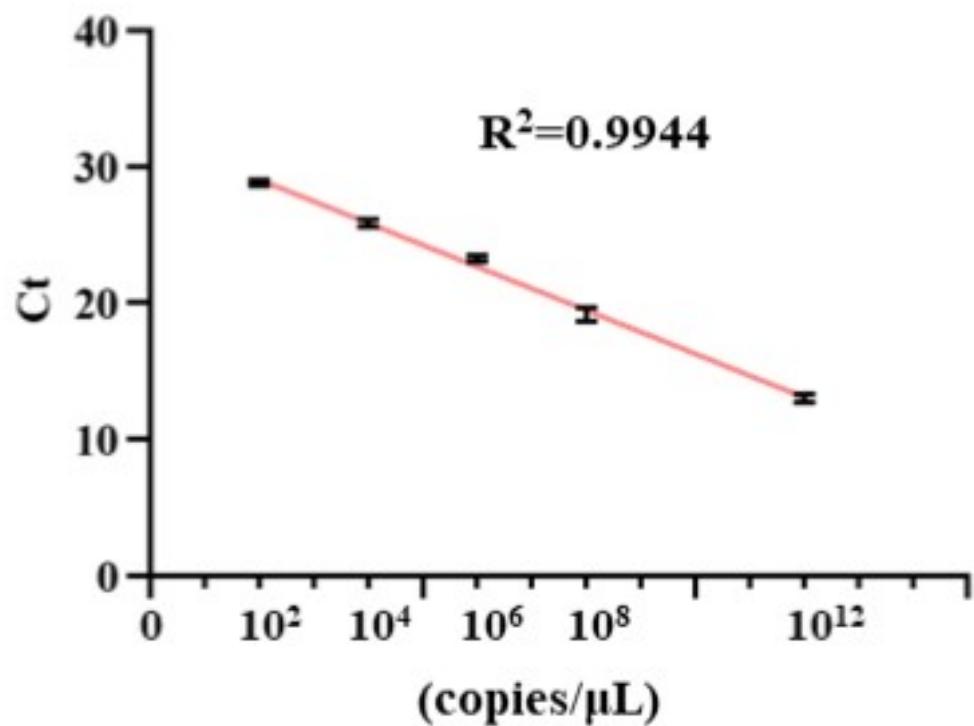


Figure S5: Quantification range of RT-qPCR

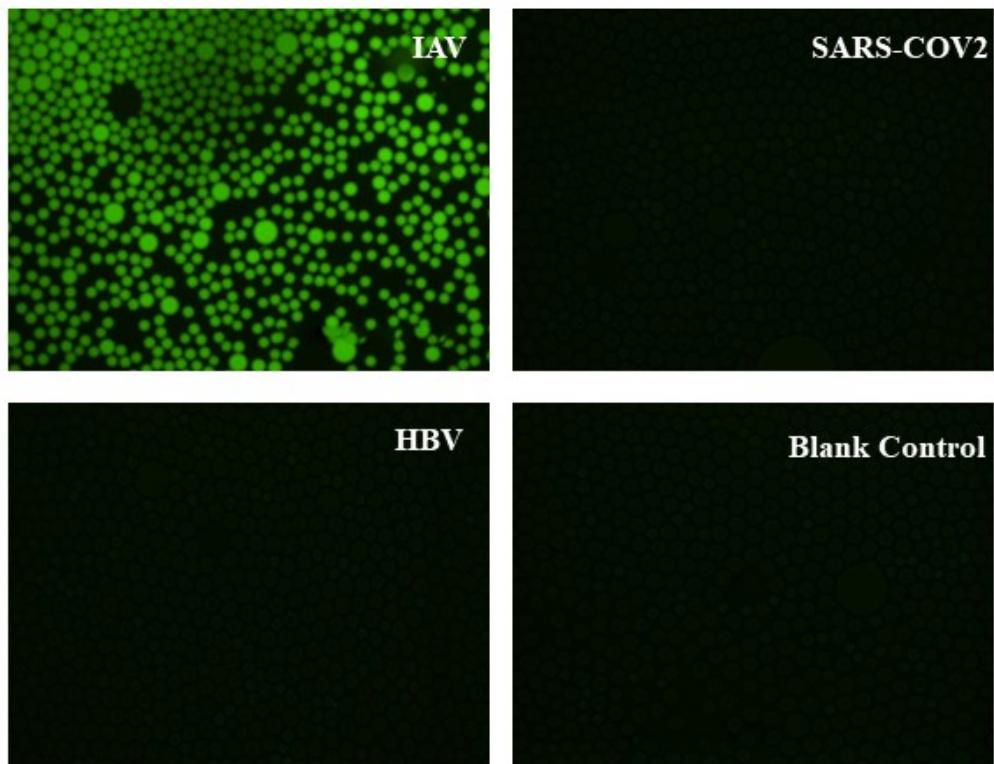


Figure S6. The specificity of this assay.