Electronic Supplementary Material (ESI) for Lab on a Chip.

A microfluidic-integrated lateral flow recombinase polymerase amplification (MI-IF-RPA) assay for rapid COVID-19 detection

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Table S1. Sequences of the SARS-CoV-2 armored RNA particles loaded with the N, ORF1ab, and E genes

Name	Sequence (5'–3')
N	ATTGCCAAAAGGCTTCTACGCAGAAGGGAGCAGAGGCGGCAGTCAAGCCTCTTCTCGTTCCTCATCACGTAGTCGCAACAGTTCAAGA AATTCAACTCCAGGCAGCAGTAGGGGAACTTCTCCTGCTAGAATGGCTGGCAATGGCGGTGATGCTGCTCTTGCTTTGCTGCTGCTTGA CAGATTGAACCAGCTTGAGAGCAAAATGTCTGGTAAAGGCCAACAACAACAAGGCCAAACTGTCACTAAGAAATCTGCTGCTGAGGC TTCTAAGAAGCCTCGGCAAAAACGTACTGCCACTAAAGCATACAATGTAACACAAGCTTTCGGCAGACGTGGTCCAGAACAAACCC
Orf1ab	TGGTGCATCGTGTTGTCTGTACTGCCGTTGCCACATAGATCATCCAAATCCTAAAGGATTTTGTGACTTAAAAGGTAAGTATGTACAAAT ACCTACAACTTGTGCTAATGACCCTGTGGGTTTTACACTTAAAAACACAGTCTGTACCGTCTGCGGTATGTGGAAAGGTTATGGCTGTA GTTGTGATCAACTCCGCGAACCCATGCTTCAGTCAGCTGATGCACAATCGTTTTTAAACGGGTTTGCGGTGTAAGTGCAGCCCGTCTTA CACCGTGCGGCACAGGCACTAGTACTGATGTCGTATACAGGGCTTTTGACATCTACAATGATAAAGTAGCTGGTTTTGCTAA
E	CGGAGTTGTTAATCCAGTAATGGAACCAATTTATGATGAACCGACGACGACTACTAGCGTGCCTTTGTAAGCACAAGCTGATGAGTACG AACTTATGTACTCATTCGTTTCGGAAGAGACAGGTACGTTAATAGTTAATAGCGTACTTCTTTTTCTTGCTTTCGTGGTATTCTTGCTAGT TACACTAGCCATCCTTACTGCGCTTCGATTGTGTGCGTACTGCTGCAATATTGTTAACGTGAGTCTTGTAAAACCTTCTTTTTACGTTTAC TCTCGTGTTAAAAATCTGAATTCTTCTAGAGTTCCTGATCTTCTGGTCTAAACGAACTAAATATTATATTAGTTTTTCT

Table S2. Demographic information for clinically verified cases

Clinical Trial Institution	Sample Type		The Number of Cases	
Fifth Affiliated Hospital of Sun Yat-sen University	Positive	Negative	Positive	Negative
	Patients with severe, intermediate, and mild symptoms	Outpatient screening	37	17

Table S3. Primers designed for off-chip RPA reactions

Name	Sequence (5'–3')	Genome location (N)
COVID-19 RAA F3	GCAACAGTTCAAGAAATTCAACTCCAGGCAGC	28845-28876
COVID-19 RAA R2	TAGTGACAGTTTGGCCTTGTTGTTGTTGGCCT	28911-28842

Table S4. Primers designed for chip-based RPA reactions

Name	Sequence (5'–3')	Genome location (N)
COVID-19 RAA F3	FAM-GCAACAGTTCAAGAAATTCAACTCCAGGCAGC	28845-28876
COVID-19 RAA R2	Biotin-TAGTGACAGTTTGGCCTTGTTGTTGTTGGCCT	28911-28842

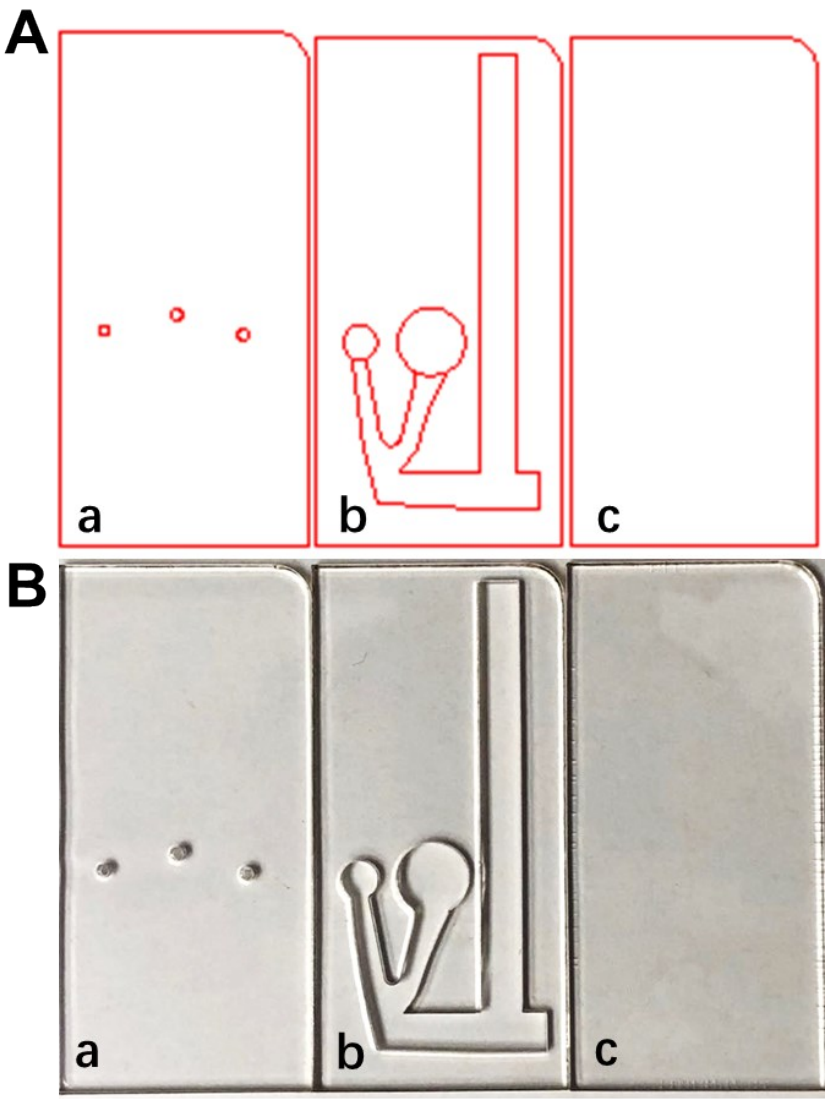

Figure S1. A). Masks used for fabrication of the chip with three layers. B). Photographs of the chip consisting of three layers. (a) Cover PMMA layer, (b) middle PMMA layer, (c) bottom PMMA layer.

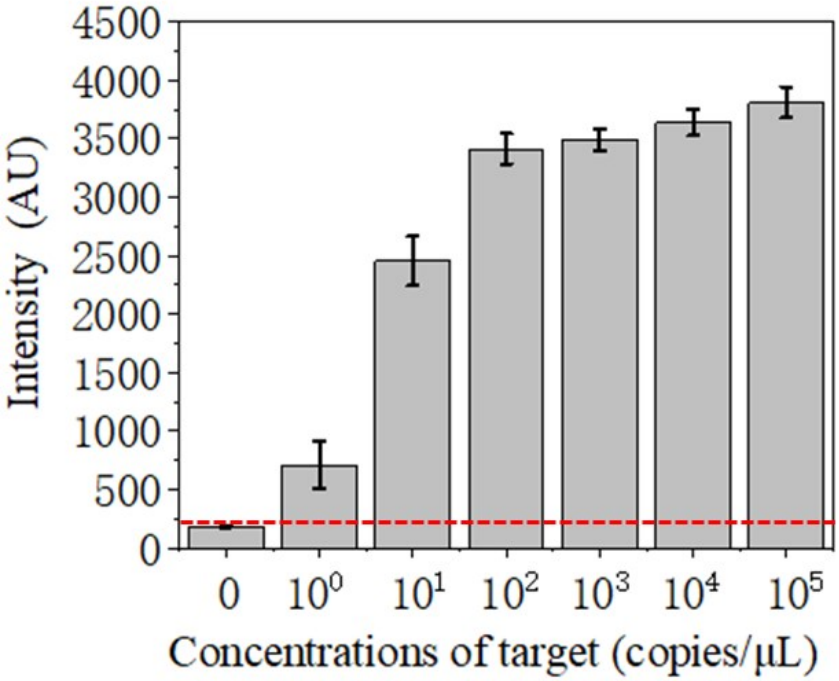

Figure S2. ImageJ grayscale analysis of test strips showing target detection at different concentrations. The red dotted line represents the value of negative samples plus 3 × SD which was used as the cut-off value for a positive signal.

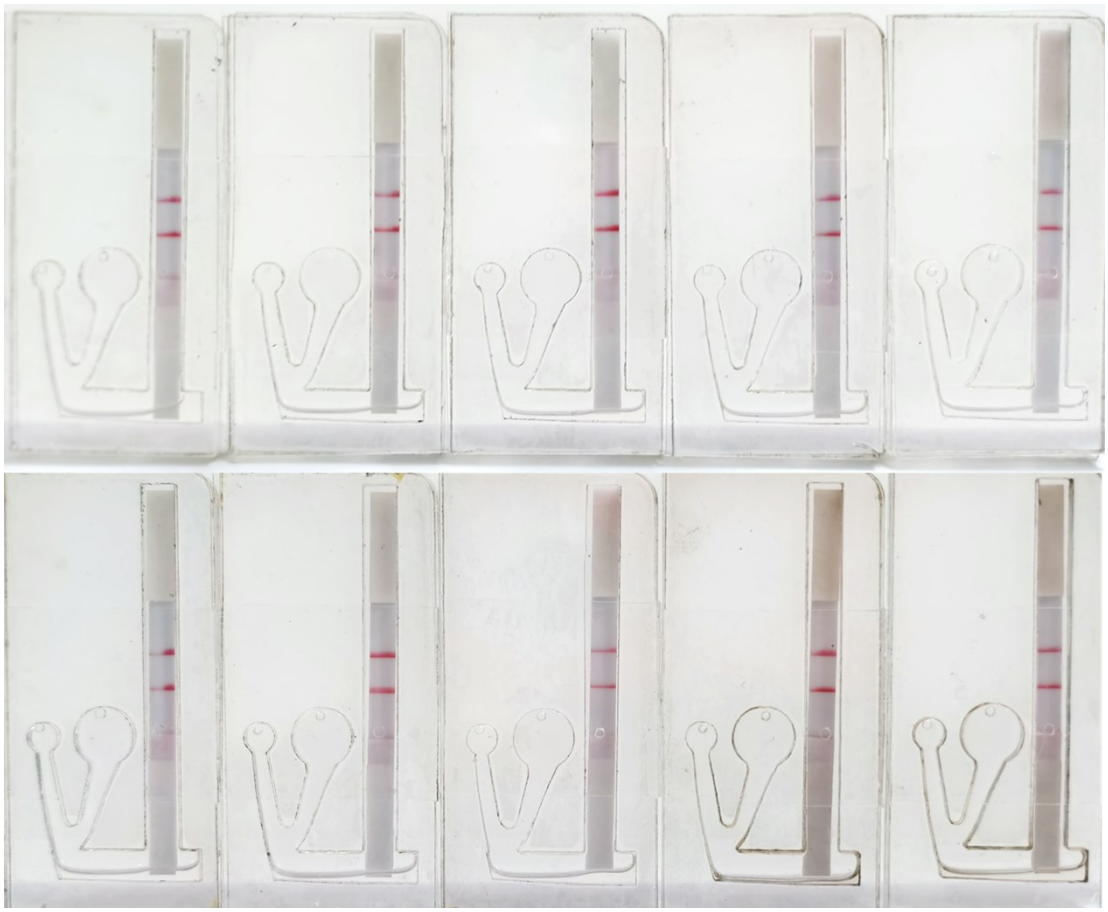

Figure S3. Reproducibility assays were conducted to determine the intra-assay reliability and precision of positive detection using the same N gene extractions.