

## Electronic Supporting Information (ESI)

### Hand-held all-in-one (HAO) self-test kit for rapid and on-site detection of SARS-CoV-2 with colorimetric LAMP

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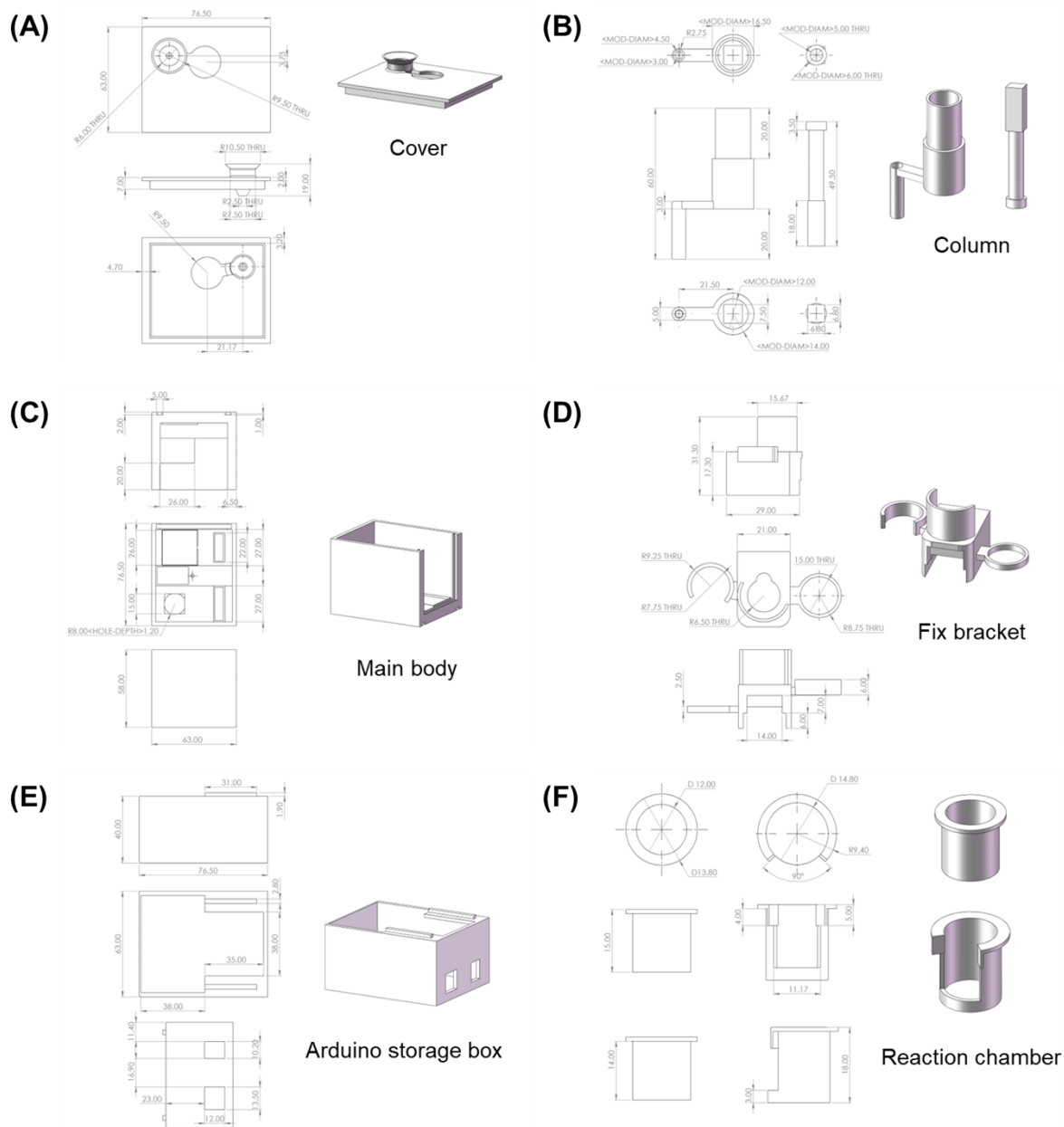
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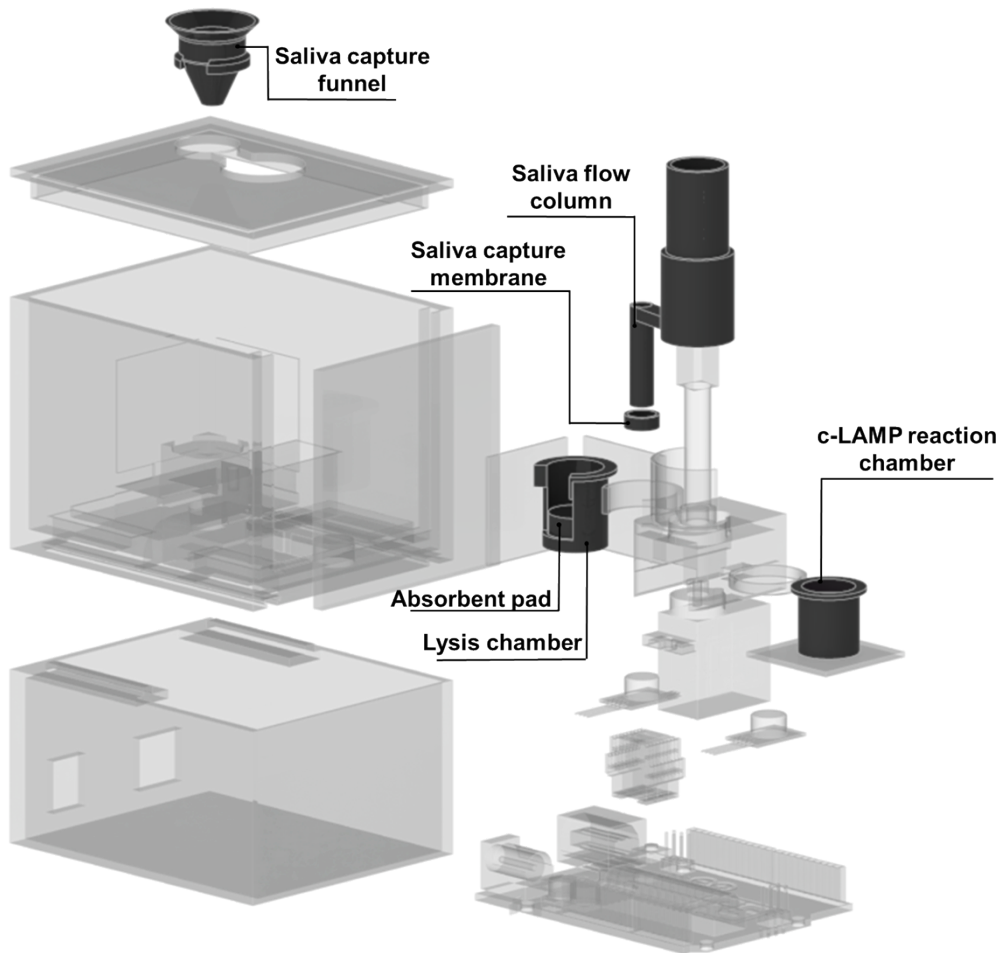
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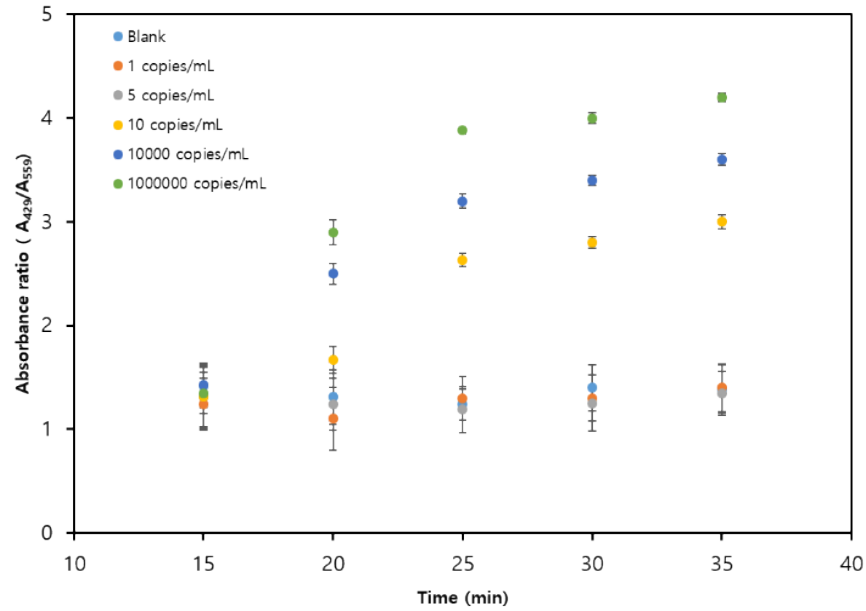
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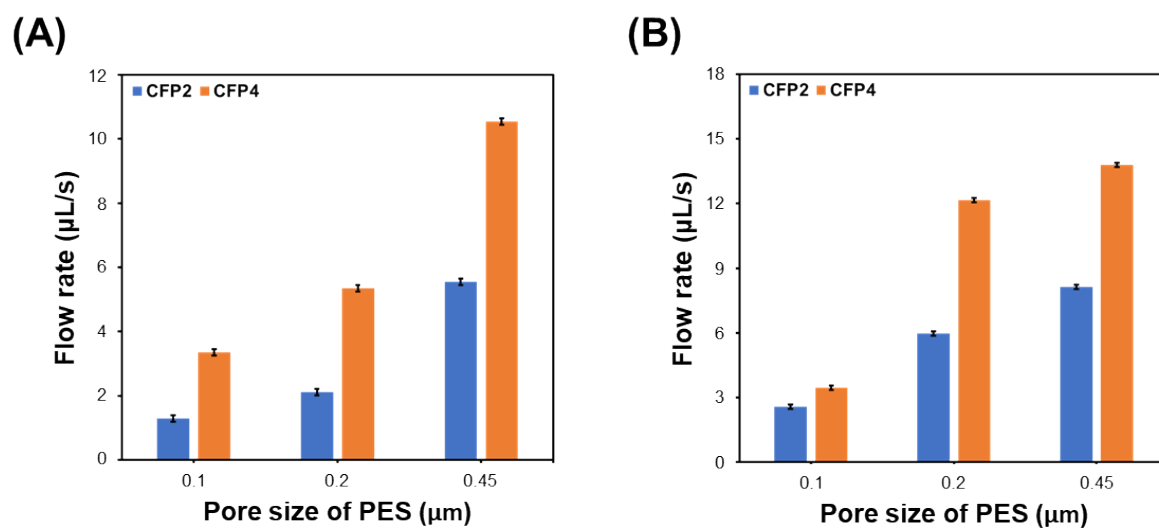
**Fig. S1.** Illustration of the HAO test kit. (A) The cover, (B) column, (C) main body, (D) fix bracket, (E) Arduino storage box, and (F) reaction chamber are shown from the top, side, and front views. Units are in mm.



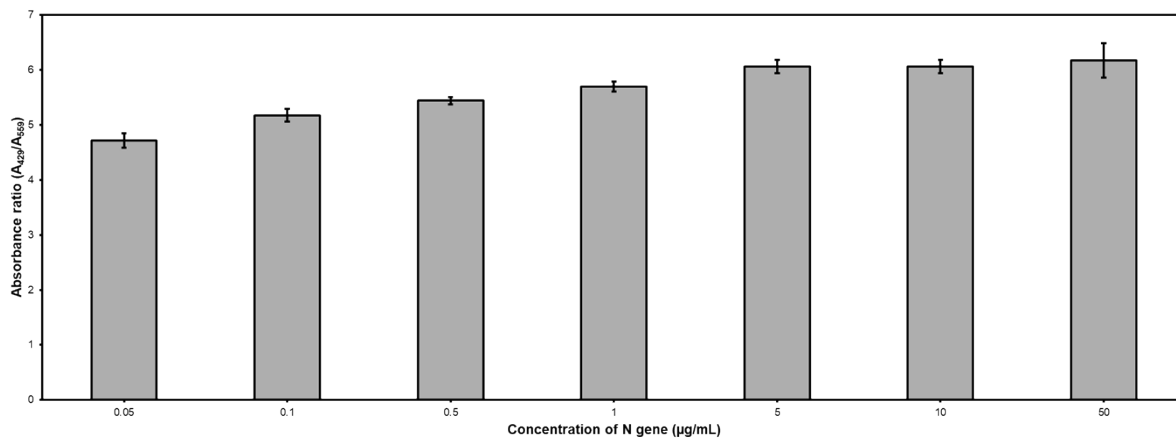
**Fig. S2.** Dark colors exhibited disposable parts, the remaining parts exhibited reusable parts.



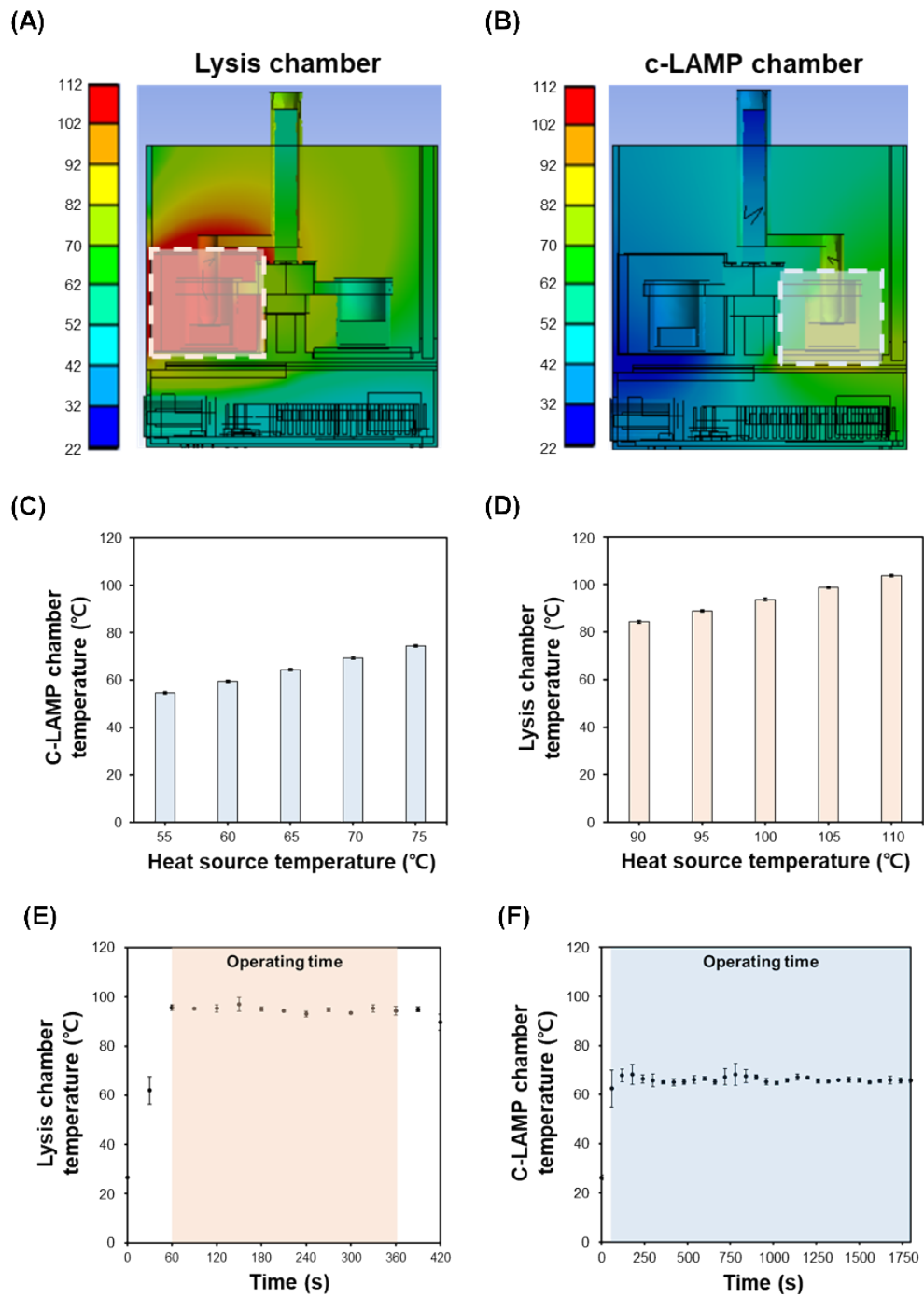
**Fig. S3.** Absorbance ratios were investigated after isolating the different concentrations of the N gene with low, middle, high amounts by using the 3 mm diameters of the PES membrane and 8  $\mu$ m pore size of the CFP across five different time intervals. The results shown that below 10 copies/mL of N genes, all values fell within the margin of error. Simultaneously, the values for 10 copies became distinctly separate from other values after 25 min of LAMP reaction, leading to select 25 minutes as the optimal detection time.



**Fig. S4.** Sample flow rate of through PES membrane and CFP. Impact of PES membrane characteristics and CFP pore size on sample flow velocity. Flow velocity of sample absorbed by CFP with pore sizes of 0.1, 0.2, and 0.45  $\mu\text{m}$  after flow through (A) hydrophobic and (B) hydrophilic PES membranes.

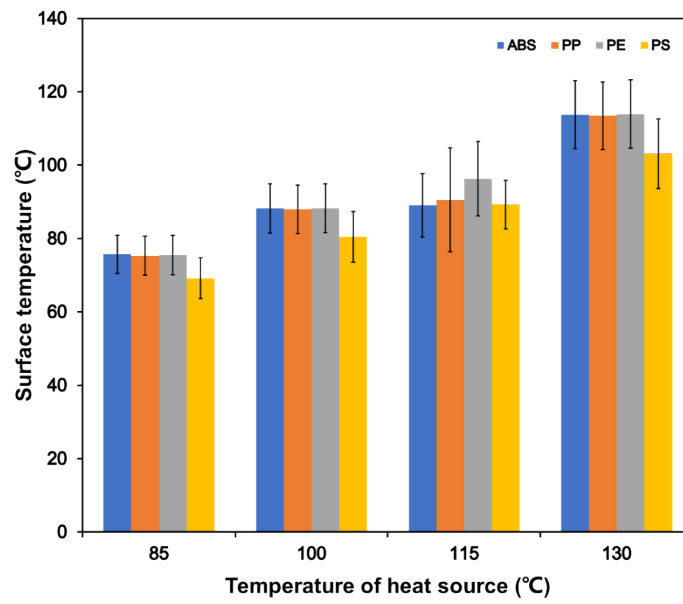


**Fig. S5.** Absorbance ratio at  $A_{429}/A_{559}$  from c-LAMP result by using the PES membrane after passing through the various concentration of N gene.

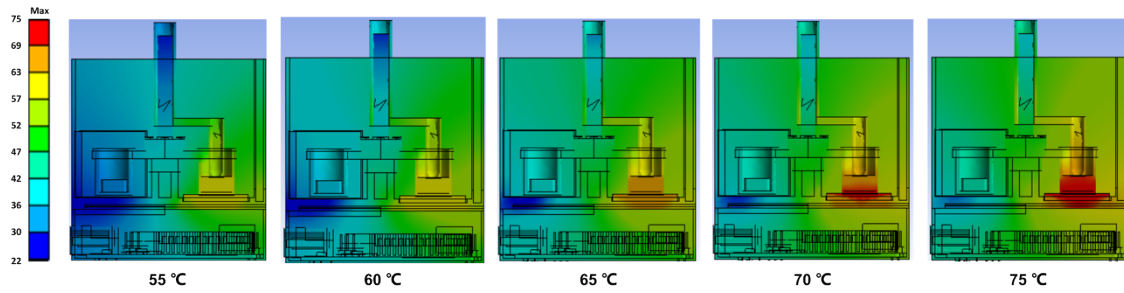


**Fig. S6** Simulation results of temperature distribution of (A) the lysis chamber and (B) the c-LAMP chamber (The region of interests (ROIs) are presented in white dashed box). The temperature provided by the heat source corresponds to the maximum temperatures reached in the target regions of the (C) lysis chamber and (D) c-LAMP chamber. The temperature profile during each period for (E) lysis and (F) c-LAMP corresponding to each chamber.

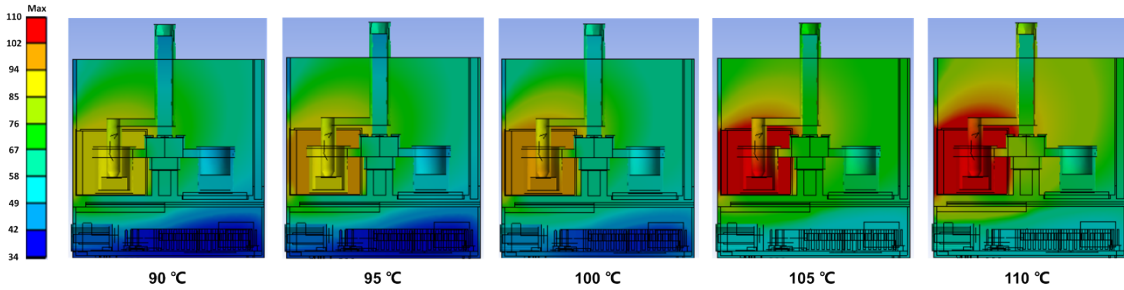
(A)



(B)

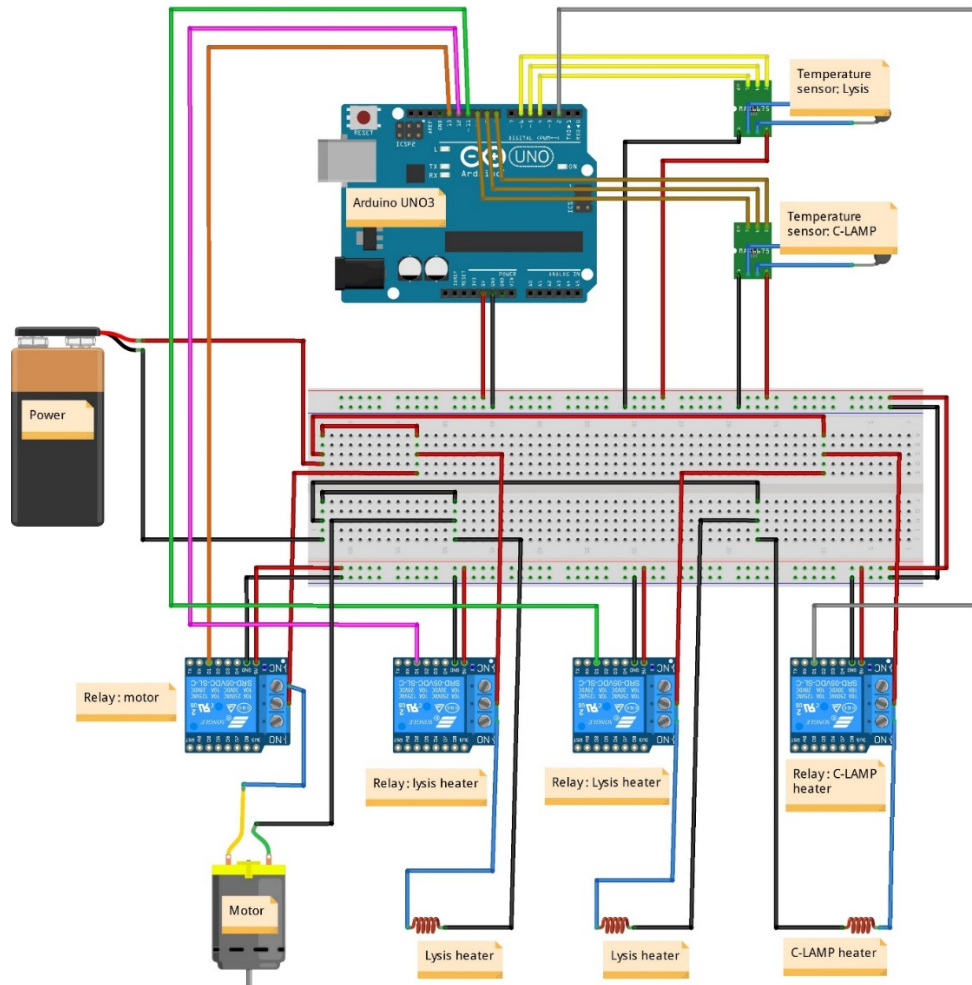


(C)

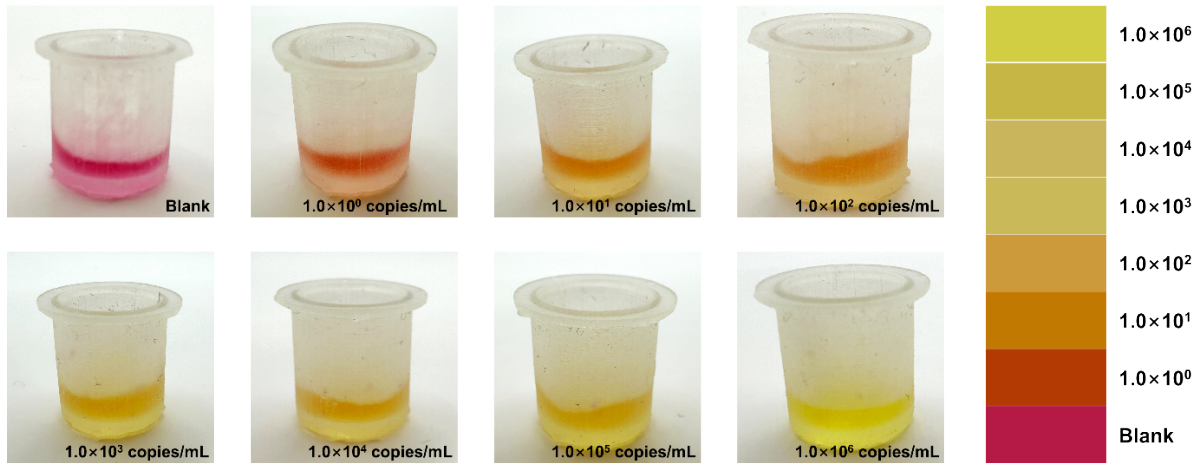


**Fig. S7.** (A) Heat transfer simulation of different heat-resistant materials. The simulation of temperature distribution in (B) lysis and (C) c-LAMP assay for HAO test kit based on ABS material at various heating temperatures. (ABS: acrylonitrile butadiene styrene, PP: polypropylene, PE: polyethylene, PS: polystyrene)





**Fig. S8.** Arduino-based temperature control module and column transfer motor for real-time temperature monitoring and stability maintenance in sample pretreatment and c-LAMP assay in HAO test kit.



**Fig. S9.** The images of the c-LAMP chamber after terminating the operation for the HAO test at different concentrations of SARS-CoV-2 N gene. The degree of color change could be observed with increasing the amount of the target gene.

**Table. S1.** Comparison of cellulose filter paper for absorb ability of the lysed sample.

<b>Name</b>	<b>Features &amp; applications</b>	<b>Pore size (<math>\mu\text{m}</math>)</b>	<b>Thickness (mm)</b>	<b>Flow rate</b>
<b>Sterlitech™ CFP 2</b>	General filtration, adsorbent conveyance, air/gas contaminant collection	8	0.17	Med-slow
<b>Sterlitech™ CFP 4</b>	Prefilter, organic extractions, biological fluid separations, air monitoring	23	0.21	Fast
<b>Sterlitech™ CFP 40</b>	Soil and air sample analysis, liquid food sedimentation, sample prep for spectrophotometry	23	0.2	Medium
<b>NEWSTAR Cellulose Filter Paper NS 43</b>	Filtration of general fine particles	16	0.22	Med-fast

**Table. S2.** Reproducibility analysis of the color change for SARS-CoV-2 detection based on HAO test kit.

\* SARS-CoV-2 N gene spiked in DI water

Sample number	SARS-CoV-2 concentration (copies/mL)	Intra-assay			Inter-assay		
		Absorbance rate	SD	CV (%)	Absorbance rate	SD	CV (%)
1	$1.0 \times 10^1$	2.68	0.03	1.12	2.69	0.07	2.60
2	$1.0 \times 10^3$	3.56	0.16	4.49	3.62	0.17	4.70
3	$1.0 \times 10^5$	4.36	0.10	2.29	4.39	0.08	1.82

\* SARS-CoV-2 N gene spiked in artificial saliva

Sample number	SARS-CoV-2 concentration (copies/mL)	Intra-assay			Inter-assay		
		Absorbance rate	SD	CV (%)	Absorbance rate	SD	CV (%)
1	$1.0 \times 10^1$	2.39	0.09	3.77	2.46	0.07	2.85
2	$1.0 \times 10^3$	2.95	0.06	2.03	2.92	0.07	2.40
3	$1.0 \times 10^5$	3.46	0.11	3.18	3.58	0.07	1.96

Absorbance rate: value represent the average of absorbance rate after c-LAMP reaction (n = 4)

SD: value represent the standard deviation of parallel result (n = 4)

CV: coefficient of variation= $SD/Absorbance\ rate$

**Table. S3.** (A) Absorbance ratio after c-LAMP reaction with DI water spiked SARS-CoV-2 N gene. (B) Absorbance ratio after c-LAMP reaction with DI water spiked SARS-CoV-2 N gene. LOD calculated based on the sum of the change from the blank sample and three times the standard deviations. Unit of concentration: copies/mL.

(A)

Sample (copies/mL)	Blank	$1.0 \times 10^1$	$1.0 \times 10^2$	$1.0 \times 10^3$	$1.0 \times 10^4$	$1.0 \times 10^5$	$1.0 \times 10^6$
Mean value	1.09	2.71	3.04	3.56	3.95	4.36	4.68
Standard deviation	0.42	0.08	0.17	0.19	0.10	0.11	0.16

(B)

Sample (copies/mL)	Blank	$1.0 \times 10^1$	$1.0 \times 10^2$	$1.0 \times 10^3$	$1.0 \times 10^4$	$1.0 \times 10^5$	$1.0 \times 10^6$
Mean value	1.24	2.42	2.63	2.96	3.20	3.53	3.88
Standard deviation	0.30	0.14	0.09	0.12	0.14	0.15	0.06