Electronic Supplementary Material (ESI) for Lab on a Chip.
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Single-Shot Multi-Channel Plasmonic Real-Time Polymerase Chain Reaction for Multi-target Point-of-care Testing

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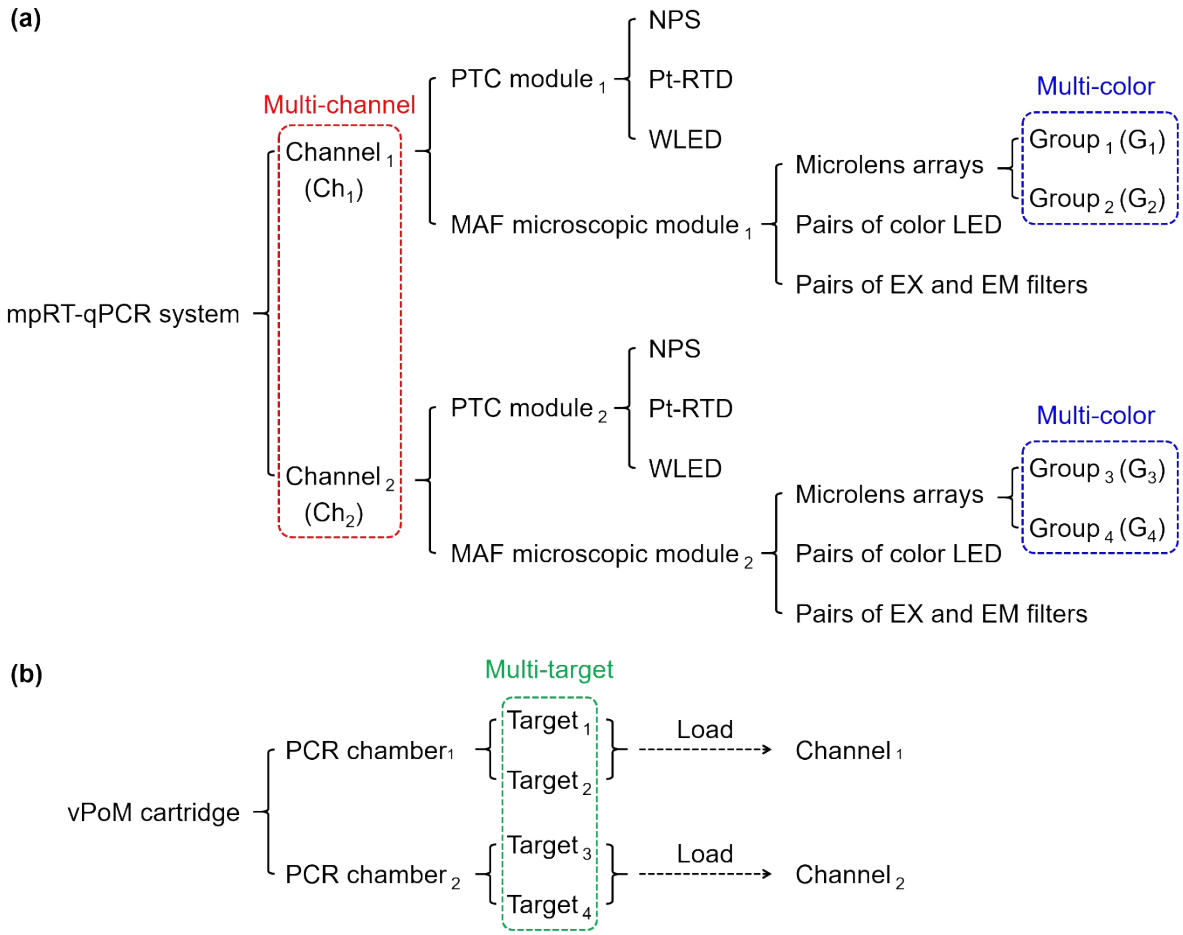

Fig. S1 Structural diagram of multi-channel plasmonic real-time RT-PCR (mpRT-qPCR) assay. Structure maps of **(a)** the mpRT-qPCR system and **(b)** vacuum-assisted polymer-on-metal (vPoM) cartridge.

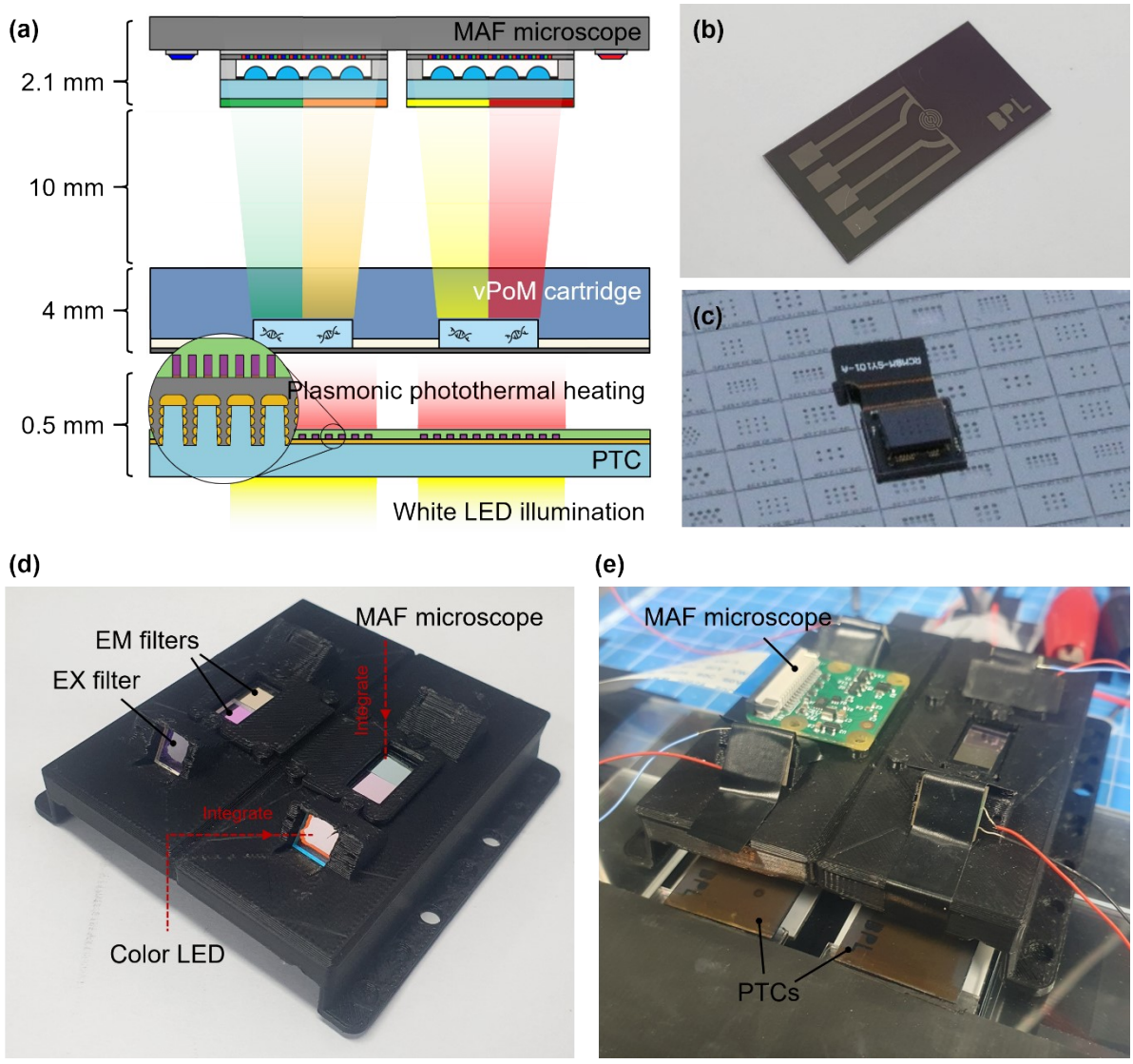

Fig. S2 Detailed configuration of mpRT-qPCR system. (a) Working principle of rapid plasmonic thermocycling and multi-color MAF imaging. Optical photographs of **(b)** the PTC, **(c)** the MAF microscopy, and **(d)** the MAF microscopic modules. **(e)** Detailed experimental setup for the mpRT-PCR assay.

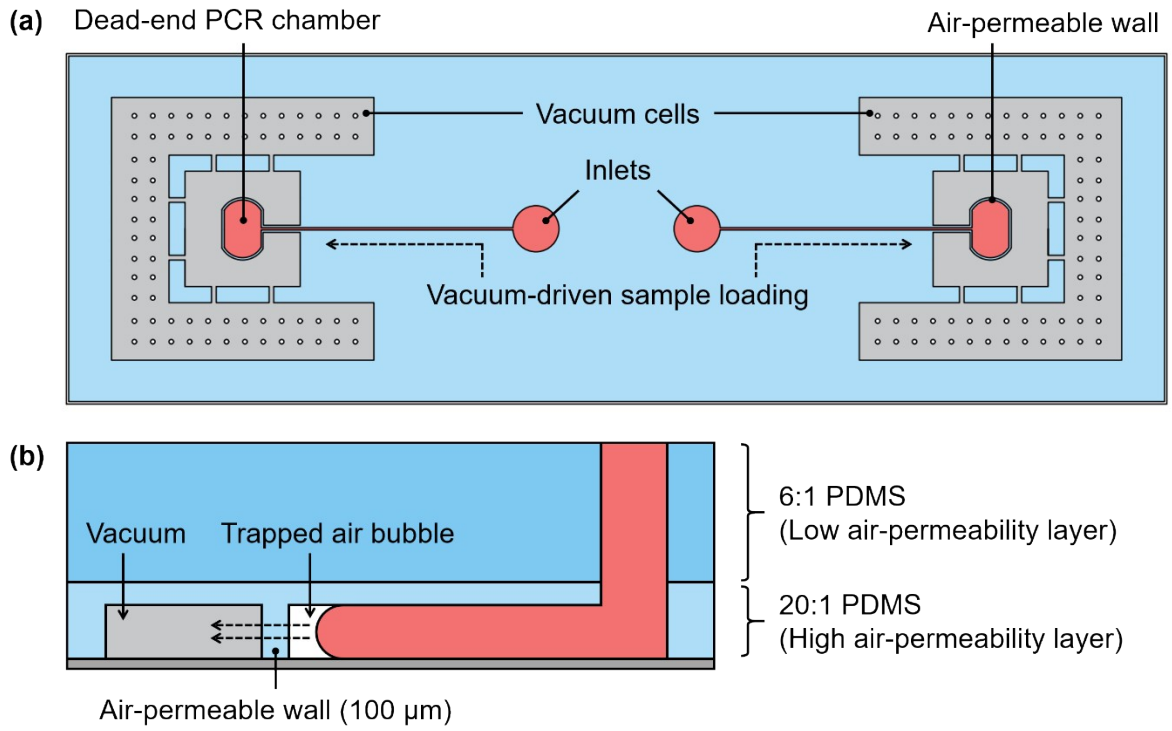

Fig. S3 Layout and working principle of vPoM cartridge. (a) Microfluidic layout of the vPoM cartridge consisting of dead-end PCR chambers, air-permeable walls, and vacuum cells. **(b)** Working principle for the vPoM cartridge. A cross-sectional view shows a bilayer configuration consisting of 20:1 and 6:1 PDMS mixing ratio (base:curing agent) for rapid loading samples and minimal bubble coverage in the vPoM cartridge. The vPoM cartridge was placed in a vacuum desiccator of 80 kPa for one hour. The PCR mixtures were injected into two inlets and spontaneously loaded into the dead-end PCR chambers.

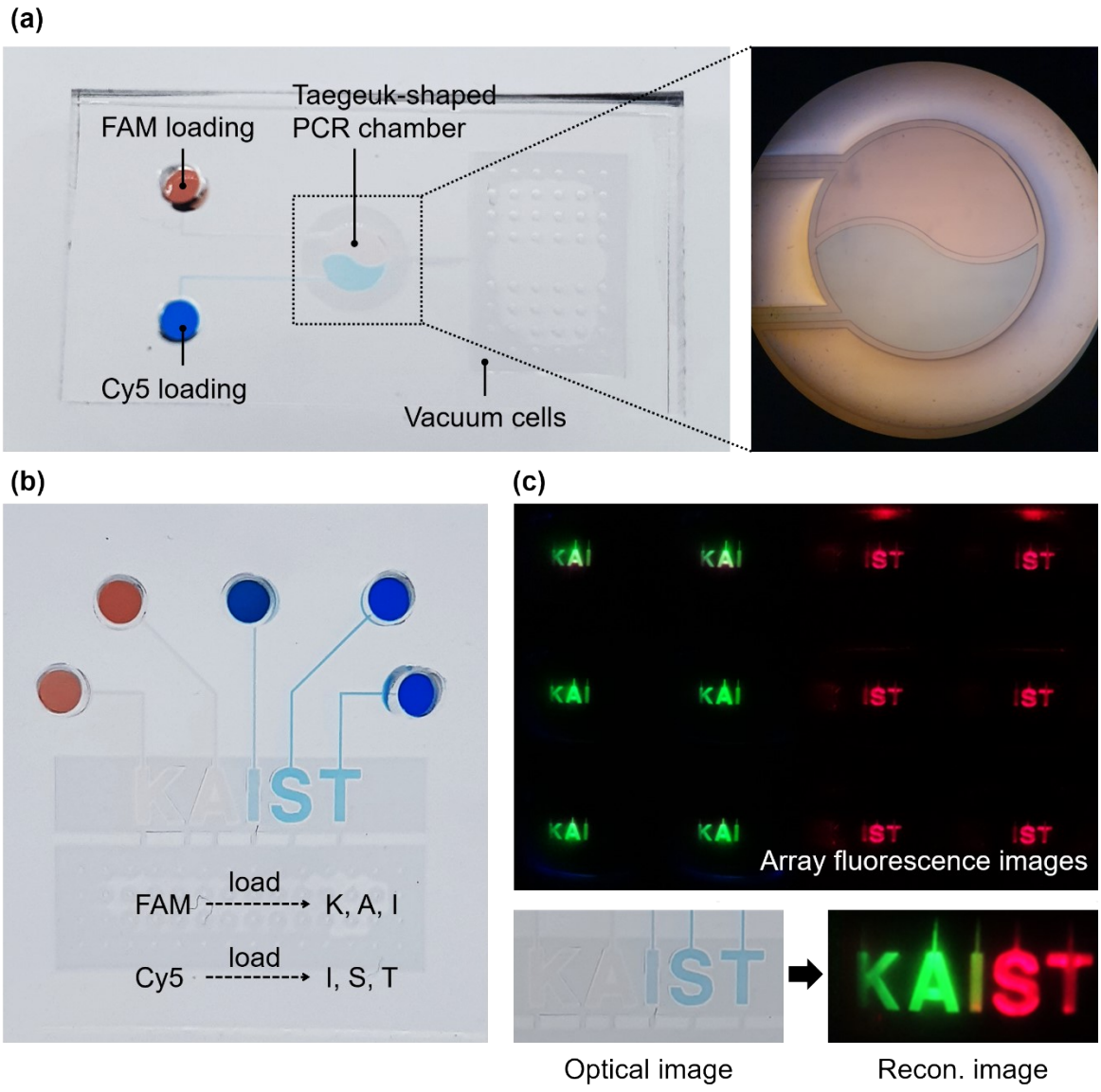

Fig. S4 Two-color array fluorescence images using microlens array fluorescence (MAF) microscope. (a) Optical and microscopic images of PDMS microfluidic chip with taegeuk-shaped PCR chamber. **(b)** Optical photograph of vacuum-assisted PDMS microfluidic chip including letters of 'KAIST'. FAM dyes are loaded into the channels with letters of 'K', 'A', 'I', and Cy5 dyes are loaded into the channels with letters of 'I', 'S', 'T'. **(c)** Captured array fluorescence and reconstructed images for multiplex fluorescence imaging at a single shot.

Fig. S5 Amplification curves of different starting concentrations in a single channel of the mpRT-qPCR system. FAM-labeled RdRP of (a) 10^4 copies per microliter, (b) 10^3 copies per microliter, and (c) 10^2 copies per microliter with constant concentration of ROX-labeled RNaseP gene.

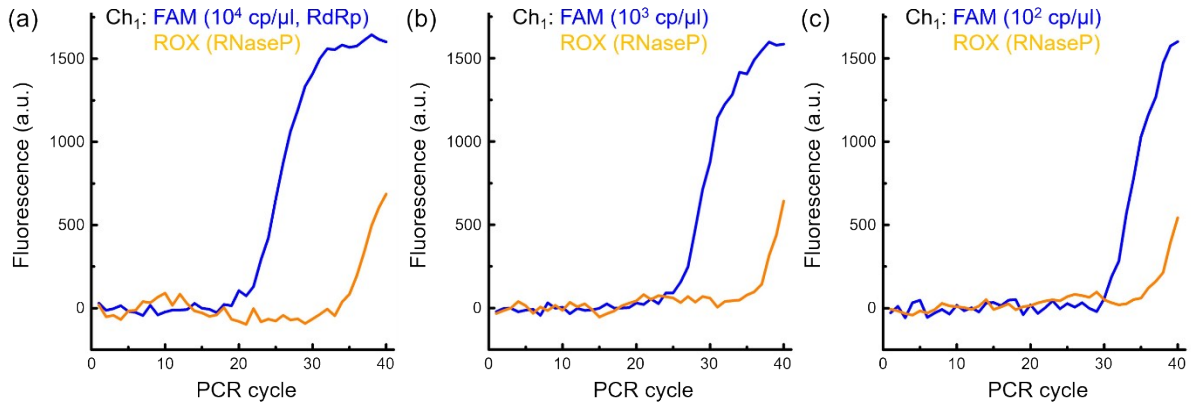

Table S1. Characteristics of color LEDs, excitation & emission bandpass filters, power, luminous/radiant flux depending on fluorescent probes.

TaqMan probe	Color LED	Excitation and emission filter		Power	Flux
FAM	LXML-PB01-0040 (470 nm, blue LED)	480 ± 20 nm	520 ± 10 nm	1.08 W	40 lm
Cy3	LXML-PM01-0100 (530 nm, Green LED)	530 ± 10 nm	565 ± 11 nm	1.12 W	102 lm
ROX	LXML-PL01-0060 (590 nm, Amber LED)	570 ± 10 nm	615 ± 20 nm	1.01 W	61 lm
Cy5	LXM2-PD01-0060 (645 nm, Red LED)	630 ± 10 nm	665 ± 10 nm	0.73 W	50 lm

Table S2. Primer and probe sequence for SARS-CoV-2 and RNaseP gene (W is A/T; R is G/A)

Target	Type	Label	Sequence (5`-3`)
SARS-CoV-2 RdRp target	Forward primer	-	GTGARATGGTCATGTGTGGCGG
	Reverse primer	-	CARATGTTAAASACACTATTAGCATA
	Taqman probe	FAM-BHQ1	CCAGGTGGWACRTCATCMGGTGATGC
SARS-CoV-2 **N target**	Forward primer	-	GGGGAACTTCTCCTGCTAGAAT
	Reverse primer	-	CAGACATTTTGCTCTCAAGCTG
	Taqman probe	Cy5-BHQ3	TTGCTGCTGCTTGACAGATT
RNaseP	Forward primer	-	AGATTTGGACCTGCGAGCG
	Reverse primer	-	GAGCGGCTGTCTCCACAAGT
	Taqman Probe	Cy3-BHQ2 ROX-BHQ2	TTCTGACCTGAAGGCTCTGCGCG