Electronic supplementary information

Integrated microsystems for *in situ* genetic detection of dengue virus in whole blood using direct sample preparation and isothermal amplification

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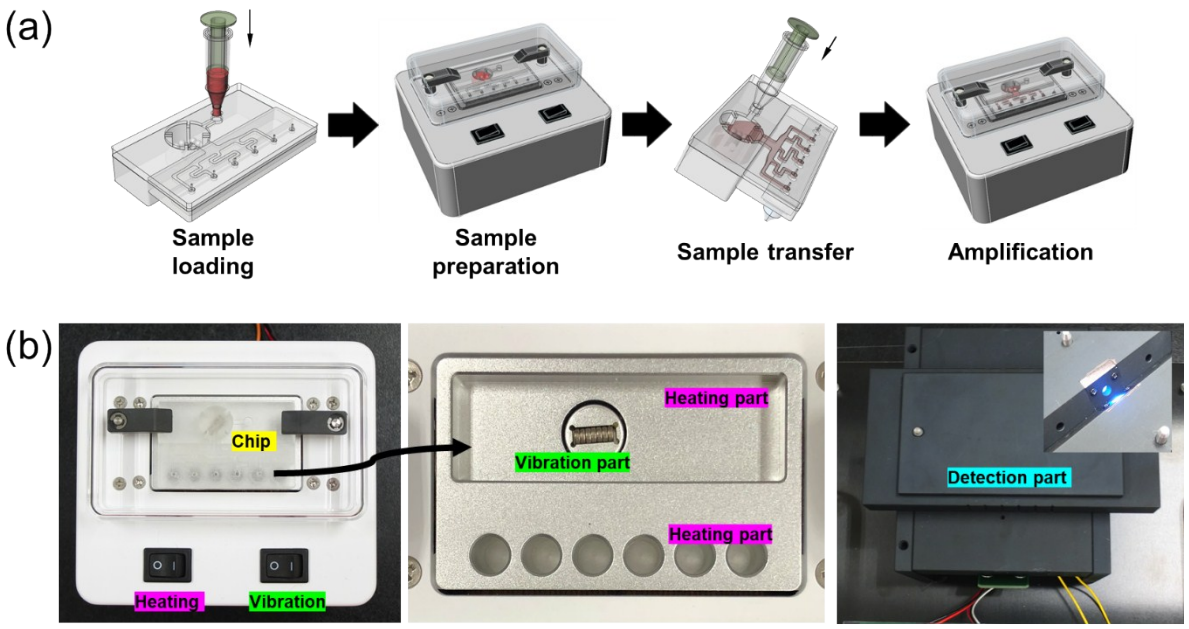
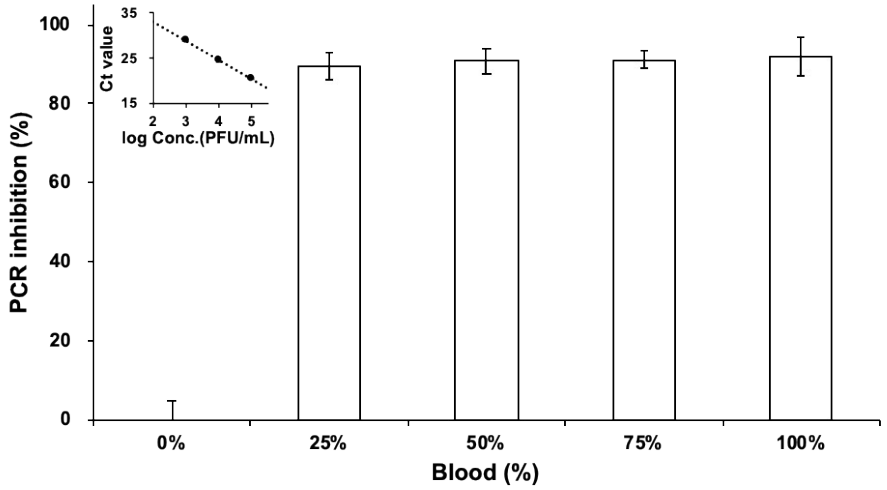
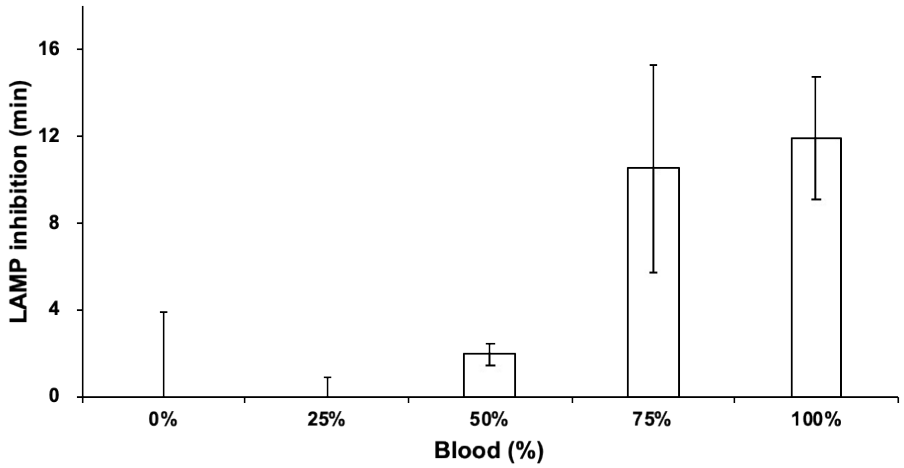


Fig. S1. (a) 3D modeling image for process and (b) real image of MB-based dengue virus detection modules.

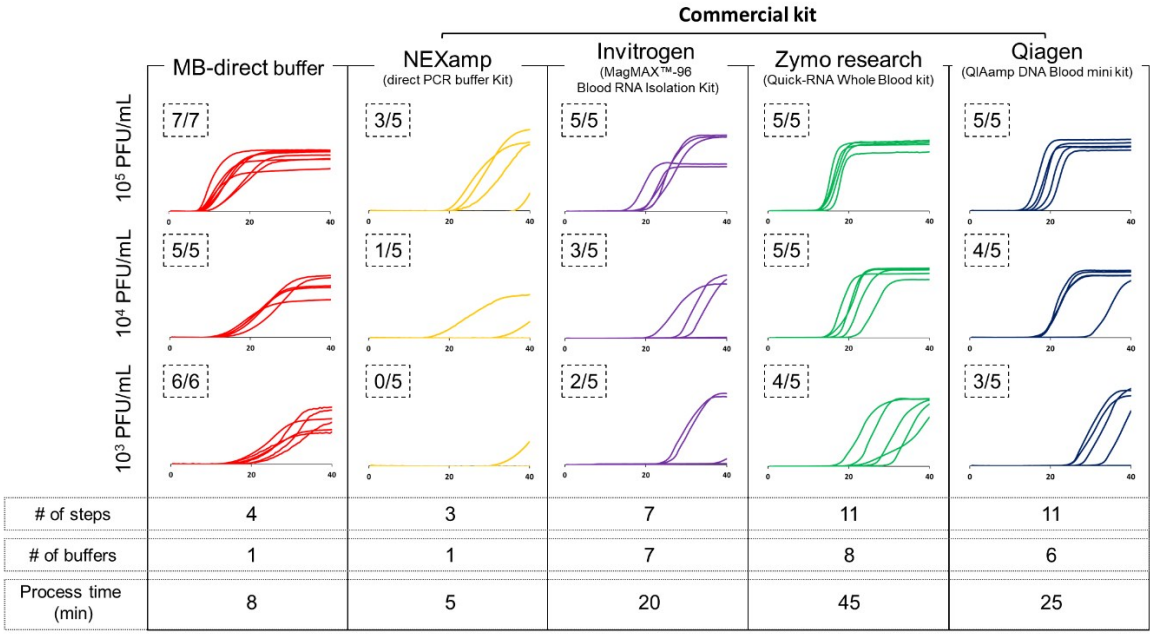


(a)



(b)

Fig. S2. (a) RT-PCR and (b) LAMP reaction inhibition in whole blood; (a) PCR inhibition (%) = 100 - Calculated conc. of sample / Calculated conc. of positive control, (b) LAMP inhibition (min) = Detection time of positive control (min) – Detection time of sample (min)



	MB-direct buffer	NEXamp (direct PCR buffer Kit)	Invitrogen (MagMAX™-96 Blood RNA Isolation Kit)	Zymo research (Quick-RNA Whole Blood kit)	Qiagen (QIAamp DNA Blood mini kit)
# of steps	4	3	7	11	11
# of buffers	1	1	7	8	6
Process time (min)	8	5	20	45	25

Fig. S3. Fluorescence signal for various concentrations of treated dengue virus in whole blood by several kits.

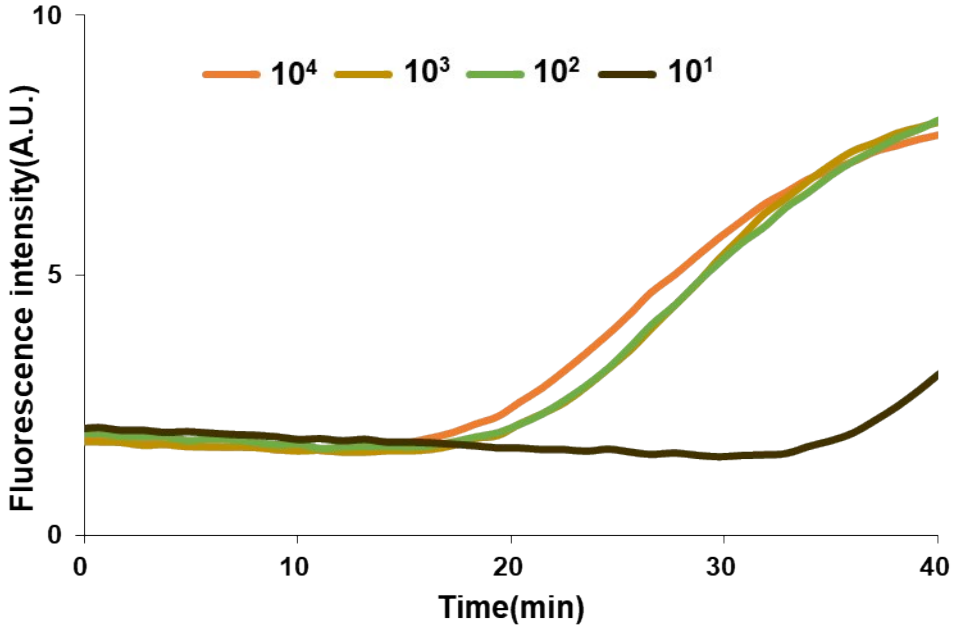


Fig. S4. Limit of detection for dengue virus spiked in whole blood by MB-direct buffer system

Table S1. The experimental details of real time RT PCR and LAMP

	PCR and LAMP reagents				Amplification conditions		
	Components		Amount (∝L)	Total (∝L)	Process		Temperature - Time
One-step RT-PCR (One-step RT-PCR set)	Reaction buffer		2.0	**10.0**	Reverse transcriptase		50 □ (20 min)
	Enzyme mix		0.5		Pre-incubation		95 □ (10 min)
	Primer set		2.0		Denaturation	45 cycles	95 □ (20 sec)
	Sample		1.0		Amplification		60 □ (30 sec)
	dH_2O		4.5				
Two-step RT-PCR	RT (Superscript® II Reverse transcriptase)	Enzyme	1.0	**20.0**	Reverse transcriptase		65 □ - 42 □ - 42 □ - 72 □ (5 min-2 min - 50 min – 15 min)
		Oligo dT	1.0				
		Reaction buffer	7.0				
		Sample	11.0				
		dH_2O					
	PCR (TB Green Premix Ex Taq II)	Enzyme Mix	10.0	**20.0**	Pre-incubation		95 □ (5 min)
		Primer set	1.0		Denaturation	45 cycles	95 □ (10 sec)
		RT Sample	2.0		Amplification		60 □ - 72 □ (20 sec – 30 sec)
		dH_2O	7.0				
PCR (TB Green Premix Ex Taq II)	Enzyme Mix		10.0	**20.0**	Pre-incubation		95 □ (5 min)
	Primer set		1.0		Denaturation	45 cycles	95 □ (10 sec)
	Sample		2.0		Amplification		60 □ - 72 □ (20 sec – 30 sec)
	dH_2O		7.0				
LAMP (Mmio® dengue detection kit)	Reaction buffer		12.5	**25.0**	Isothermal Amplification		58 □ (40 min)
	Enzyme mix		1.0				
	Primer set		2.0		Termination		80 □ (5 min)
	Sample		2.0				
	dH_2O		7.5				