## **Supplementary Material**

Sub-7-second genotyping of single-nucleotide polymorphism by high-resolution melting curve analysis on a thermal digital microfluidic device

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## ESI Text

Device fabrication and assembly. The fabrication procedure is shown in Fig. S1.

Step a: The bottom plate (60 mm × 30 mm) of the device was a glass slide patterned with two arrays of film chromium electrodes (130 nm thick, 2 mm x 2 mm size) by standard lithographical and wet-etch methods (Newway Photomask Making Ltd.)

Step b: The patterned bottom plate was then coated with 25  $\mu$ m SU-8 2025 (MicroChem Corp.) by soft lithography processes as the dielectric layer. The contact pads and ground pad were left uncoated.

Step c: Then 100 nm Teflon AF was spin coated on top of the dielectric layer (1% w/w Telfon AF in FC-40 at 1000 r.p.m. for 60 s) and then baked on a hot plate at 90 °C for 5 min and followed by 4 hours of backing at 160 °C in an oven for gasification of Teflon.

Step d: The top plate (54 mm × 20 mm) was an Indium Tin Oxide-coated (ITO) glass (Kaivo, China).

Step e: Inlet and outlet holes were drilled on the top plate by a laser cutting machine (ZLGEM-AZ1, Shanghai Zhikaijie Laser Technology) at different places for oil and sample introduction.

Step f: Next, 100 nm of Teflon AF was coated over on the ITO layer following the same protocol as on the bottom plate (step c).

Step g: Finally, the bottom plate (electrodes side faced up) and the top plate (ITO side face down) was

bonded together using UV glue (T305, TG) with a 200  $\mu$ m thick glass frame (laser cutted) spacer. Liquid metal (Coollaboratory Liquid Ultra, Coollaboratory) was deposited between the ground pad and the top plate for grounding the top plate when applying electric field across the droplets.



Fig. S1. Fabrication details of T-DMF device.



Fig. S2. Schematic of the proposed T-DMF device.



**Fig. S3.** 3D mode for finite element model (FEM) simulation of the thermal electrode's characteristic. (a) 3D mesh building of the FEM of T-DMF device, displaying its surface temperature. The inset shown the 3D mesh building of the FEM of droplet and thermal electrode, displaying its lower surface temperature. (b) Side view schematic of the model.



**Fig. S4.** Measuring system for thermal digital microfluidic device (T-DMF). (a) The system is comprised of a T-DMF device holder for electrical connection, control electronics for task execution and signal acquisition, a fluorescence microscope for images capture, and a computer software for data hub and overall system coordination; they together handle the entire operating procedure. (b) Experimental preparation of the T-DMF device for ultrafast MCA.



**Fig. S5.** Circuit diagram and schematic of the thermal modulator with on-chip thermal electrode. (a) Transport mode: one side of the thermal electrode is switched to the AC signal supplier by the control software. Thermal mode: the control software records the temperature signal from the ADC (Analog to Digital Convertor) and feeds it back to the temperature control system, for modulating the power inputted to the heater. (b) Schematic diagram shows how the controller switches between the two modes of operations of the thermal electrode. A resistance meter for temperature sensing was kept screening all the time. A capacitance meter was connected initially when the electrode was idle. For the transport mode, an AC signal power supply was connected to one side of the thermal electrode with the other side flouting. For the thermal mode, a tunable power source was connected to one side of the thermal electrode with the other side grounded.



**Fig. S6.** Sample droplet loading procedure. (a) Droplet loading activated. The coverage of the sample on the first electrode is a reliable trigger signal to activate the procedure. (b-c) Droplet loading in process. The electrodes are actuated cooperatively for droplet generation. (d) Droplet loading is completed.