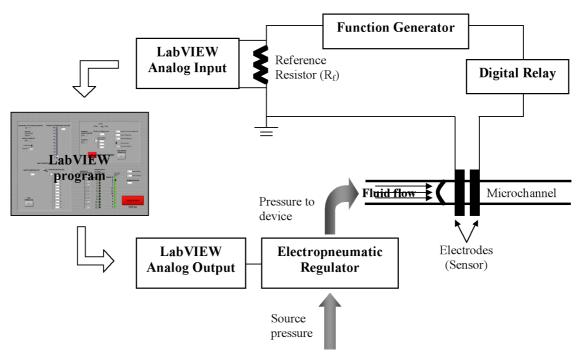
Multiphase Bioreaction Microsystem with Automated On-Chip Droplet Operation

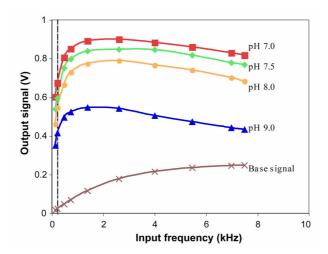
Supplementary Information

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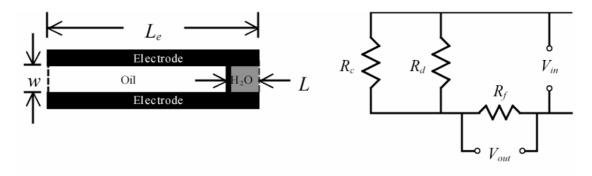
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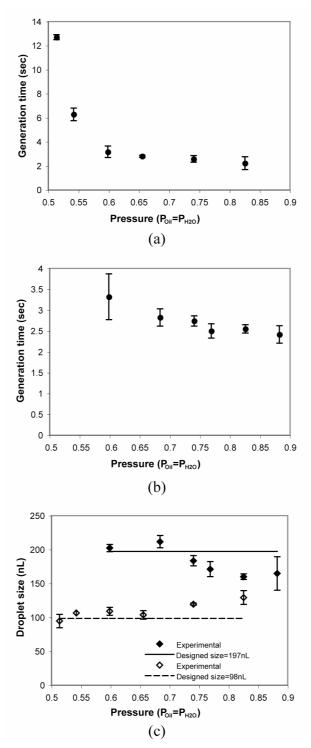
Supplementary Fig. 1 Schematic of the automated droplet-based system setup.



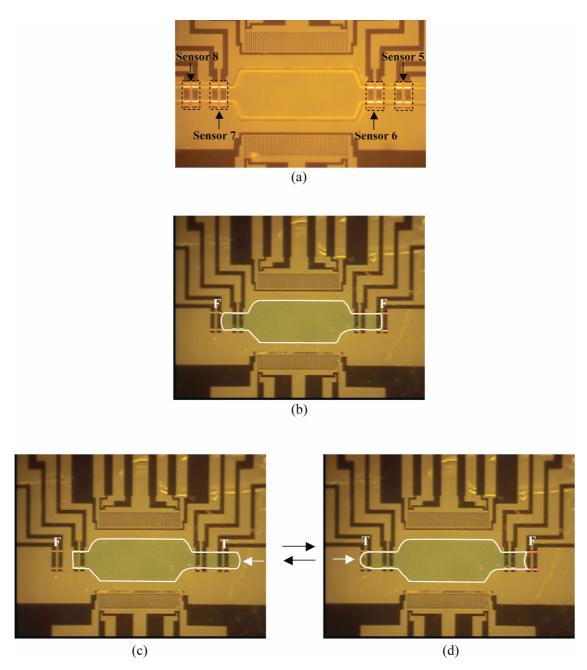
Supplementary Fig. 1Effects of the frequency of input signal and the pH of buffer solution (10mM Tris·HCl). The input signal is a sine-wave AC signal with an amplitude of 1.5V. The ion concentration of buffer solution is 9.19mM, 7.84mM, 5.34mM and 1.03mM for pH equal to 7.0 (■), 7.5 (♦), 8.0 (•) and 9.0 (\blacktriangle), respectively. The dash line shows the frequency (230 Hz) used in the automated device operation.



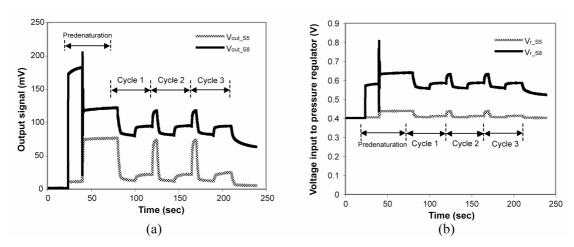
Supplementary Fig. 3 Diagram of simplified circuit model for the analog sensing. w is the gap between the electrodes; L is the length of the aqueous phase covering the electrodes; L_e is the length of the electrode; R_c and R_d are the resistance of the oil and aqueous phase between the electrodes, respectively; R_f is the resistance of the external reference resistor; V_{in} is the voltage input to the analog sensors; V_{out} is the voltage drop across the reference resistor.



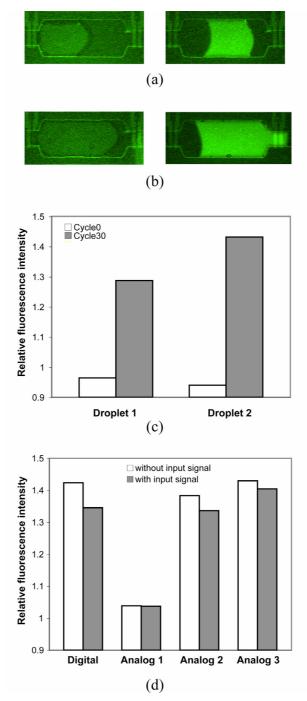
Supplementary Fig. 4 (a)-(b) Effect of pressure on the droplet formation time. (c) Effect of pressure on the droplet size.



Supplementary Fig. 5 (a) Image of the reaction region with four digital confinement sensors. (b) Snapshot of the droplet at room temperature. (c) Oscillation of the droplet during thermal cycling.



Supplementary Fig. 6 (a) Sensor output signal and (b) voltage input to the electropneumatic regulator when Sensor 5 and 8 are monitored during the droplet confinement with thermal cycling. $K_p = 0.0005$ (V_r_S5); $K_p = 0.001$ (V_r_S8); $K_i = 0$.



Supplementary Fig. 7 (a) Images of Droplet 1 (designed size of 98nL) at cycle 0 (left) and cycle 30 (right). (b) Images of Droplet 2 (designed size of 197nL) at cycle 0 (left) and cycle 30 (right). (c) The relative fluorescence intensity of the automated generated droplets at cycle 0 and cycle 30. (d) The relative fluorescence intensity of the droplets with and without input voltage in the digital and analog sensing devices. Digital denotes the reaction product with enzyme concentration of 0.175 U/ μ L in the digital sensing device. Analog 1, Analog 2 and Analog 3 denote the reaction product with enzyme concentration of 0.175 U/ μ L, 0.35U/ μ L, and 0.7 U/ μ L in the analog sensing device.

Supplementary Movie 1 Automated generation of droplet with designed size of 98nL (1x speed).

Supplementary Movie 2 Automated generation of droplet with designed size of 50nL (1x speed).

Supplementary Movie 3 Automated generation of droplet with designed size of 179nL (1x speed).

Supplementary Movie 4 Automated generation of droplet with designed size of 197nL (1x speed).

Supplementary Movie 5 Automated droplet positioning using digital sensing during temperature cycling (1x speed).

Supplementary Movie 6 Automated droplet positioning using analog sensing (1x speed).

Supplementary Movie 7 Automated droplet positioning using analog sensing during temperature cycling (1x speed).