Electronic Supplementary Material (ESI) for Lab on a Chip.

Supplementary Materials

Combinatorial nanodroplet platform for screening antibiotic combinations

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Supplementary Materials

Figure S1. Fabrication of the droplet generation device.

Figure S2. Characterization of the droplet size.

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Movie S1. Demonstration of the combinatorial nanodroplet platform for creating droplets with on-demand reagents, transferring droplets to a tubing for flexible assaying, and delivering droplet to a microchannel for quantitative analysis.

A. Supplementary Figures and Tables

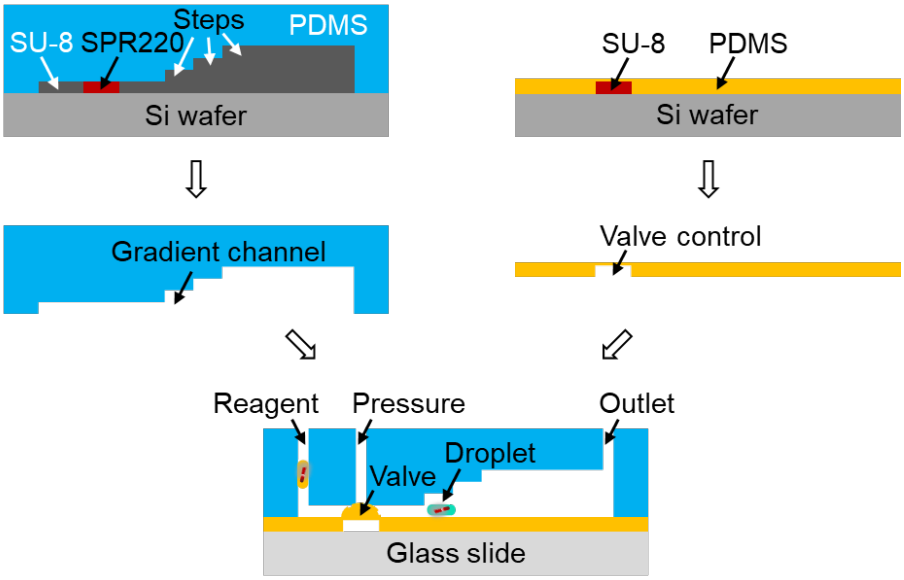

Figure S1. Fabrication of the droplet generation device. The droplet generation device consists of a reagent loading layer, a valve control layer, and a glass substrate. The reagent loading layer is a 3D mold created using one layer of SPR220 photoresistor and four layers of SU-8 and molded using PDMS (left top). The valve control layer is fabricated by casting PDMS on a SU-8 mold (right top). The device was created by bonding the reagent loading layer and the valve control layer on a glass substrate using oxygen plasma treatment for 1 min.

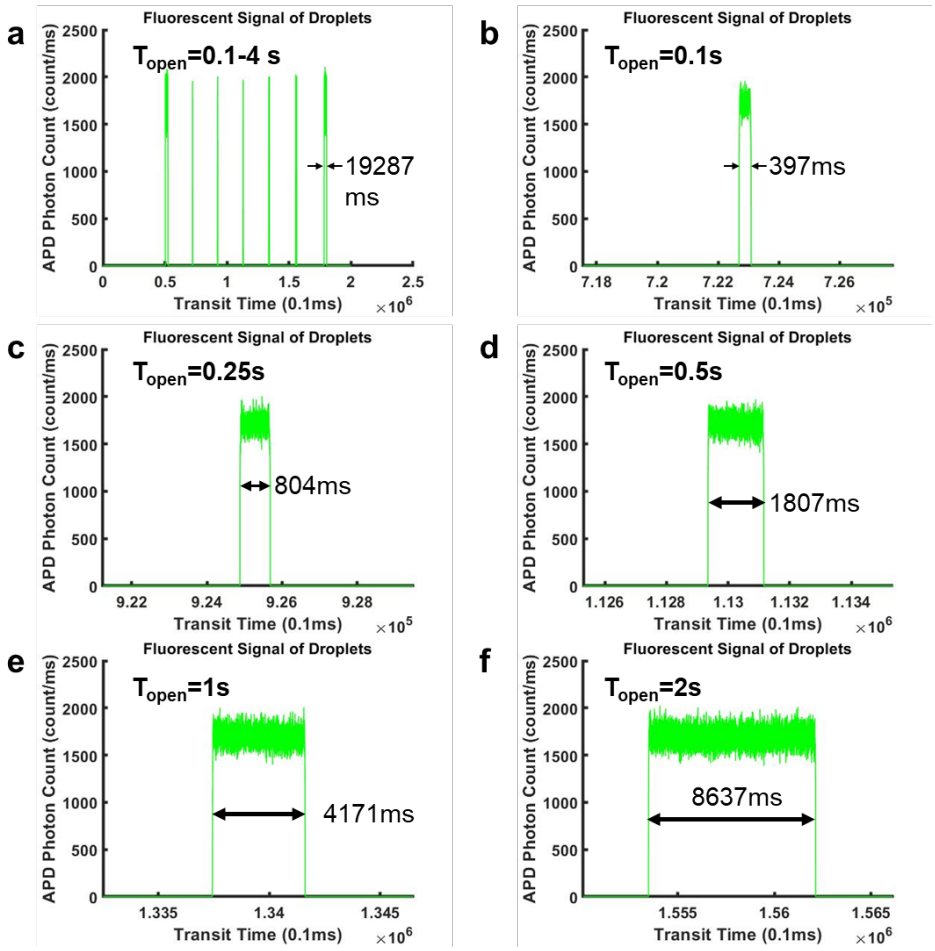

Figure S2. Characterization of the droplet size. (**a**) A set of droplets was generation with variable valve opening duration and delivered into the downstream channel to measure the size. (**b-f**) The close-up views of the droplets with valve opening time from 0.1s to 2s.

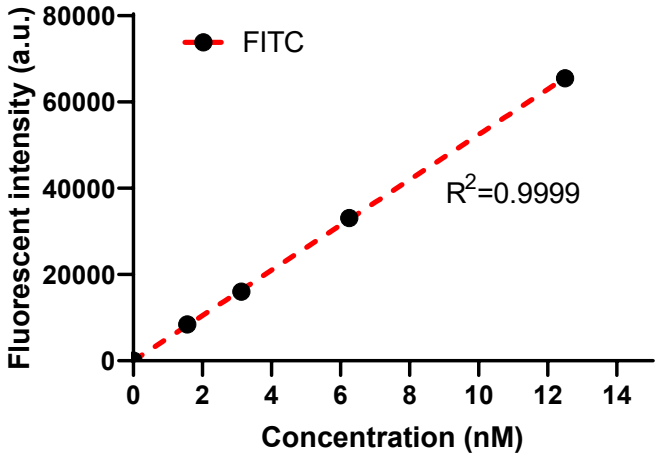

Figure S3. Linearity between the fluorescence intensity and the concentration of FITC dye. Linear regression was performed using GraphPad Prism 8. Data represent mean ± SEM (n = 3).

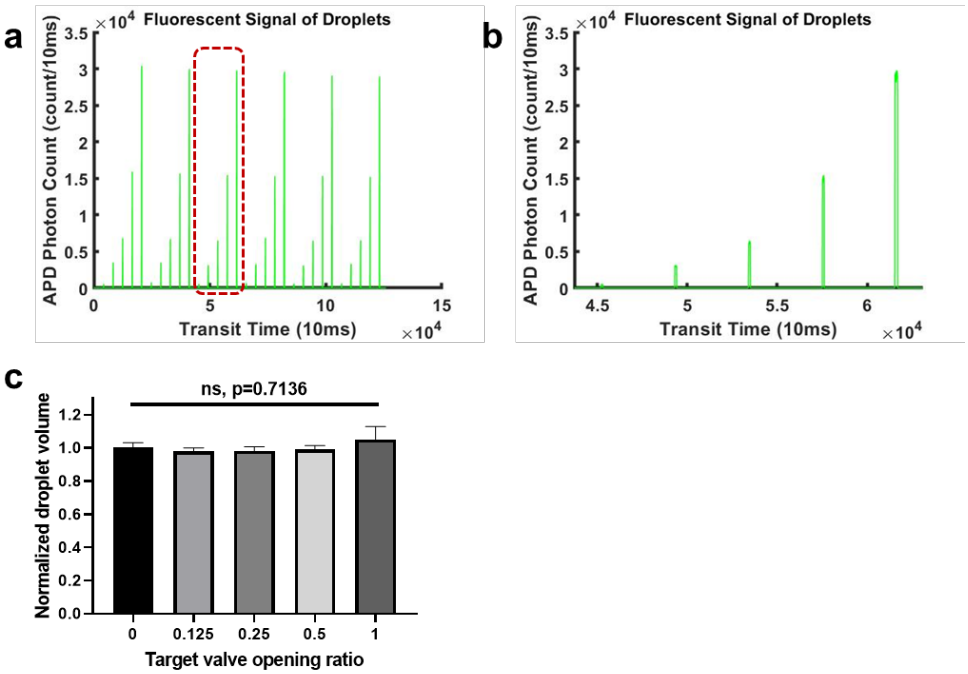

Figure S4. Characterization of the relationship between the target concentration in droplets and the opening time ratio of respective valve. (**a-b**) Series of droplets were created with different valve opening time of the target fluorescence dye. Close-up view of a set of droplets was shown. (**c**) The droplet size remains when the total valve opening time is constant though the opening time/ratio of the target sample varies. One-way ANOVA testing was performed to determine the difference among groups. Data represent mean ± SEM (n = 5).

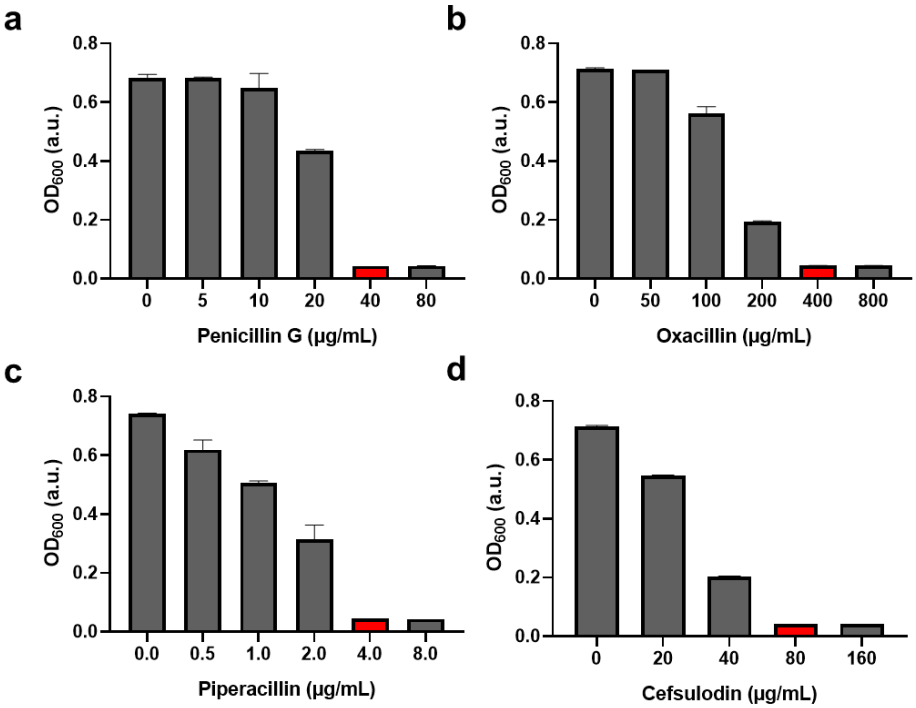

Figure S5. MICs of antibiotics involved in this study. The MICs of antibiotics, including Penicillin G (PEN), Oxacillin (OXA), Piperacillin (PIP), and Cefsulodin (CEF), were determined using standard microdilution methods. Measuring bacterial growth with antibiotics at various concentration using optical density at 600 nm. The antibiotic concentration increased in a 2-time order and the minimal inhibitory concentration (MIC) of each antibiotic was highlighted in red. Data represent mean ± SEM ($n \geq 3$).

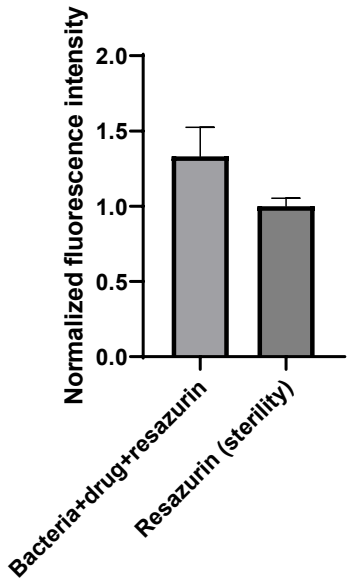

Figure S6. No bacterial contamination was detected during the incubation in the droplet system. The fluorescence intensity of the droplets consisting of bacteria, drug, and resazurin and the droplets consisting of resazurin was normalized to evaluate the bacterial contamination. We found that the fluorescence signal in the sterility droplets was lower than that in the droplets containing bacteria, drug, and resazurin, suggesting that no bacterial contamination was detected in the droplet system. Data represent mean ± SEM (n = 5).

Supplementary Movies

Movie S1. Demonstration of the combinatorial nanodroplet platform for creating droplets with on-demand reagents, transferring droplets to a tubing for flexible assaying, and delivering droplet to a microchannel for quantitative analysis.