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

Flow Cytometry–based Submicron-Sized Bacterial Detection System using Movable Virtual Wall

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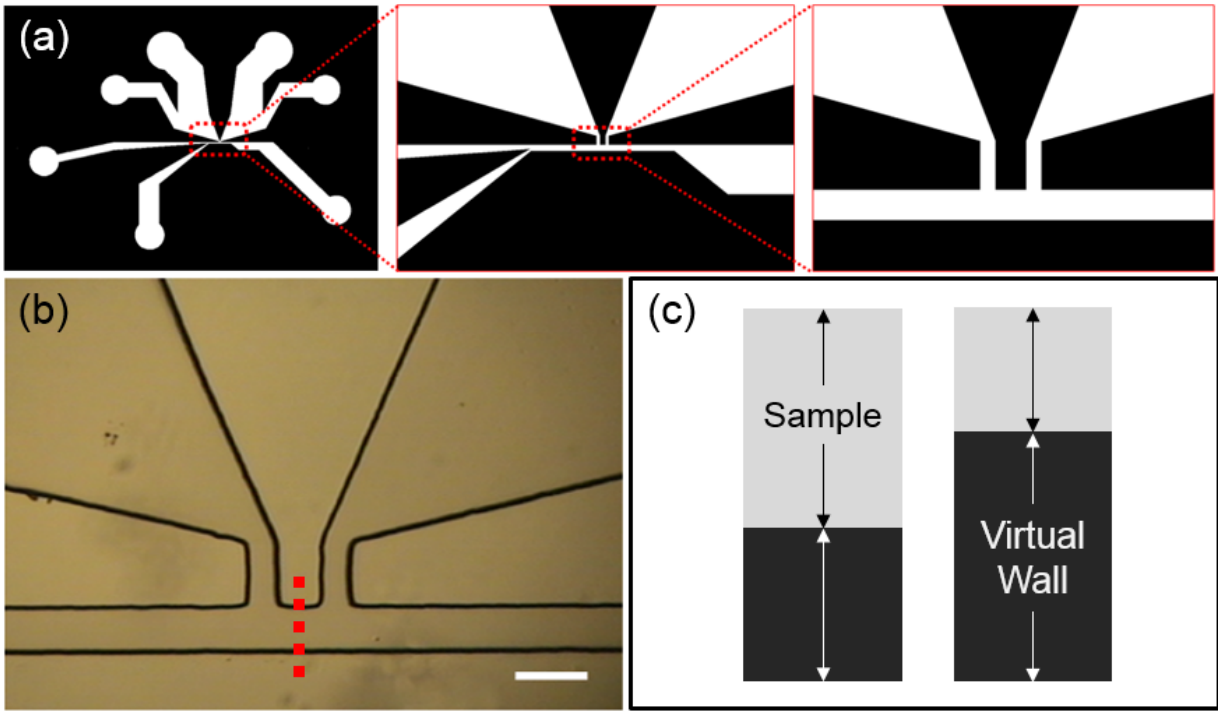

Figure S1. (a) Mask pattern of the microfluidic chip for the flow cytometry–based submicron-sized bacterial detection system. (b) Photographs of the detection region in the fabricated microfluidic glass chip (scale bar = 100 μm). (c) Illustration of varying thicknesses of the detection area (gray) obtained by controlling the flow rate of the virtual wall (black).

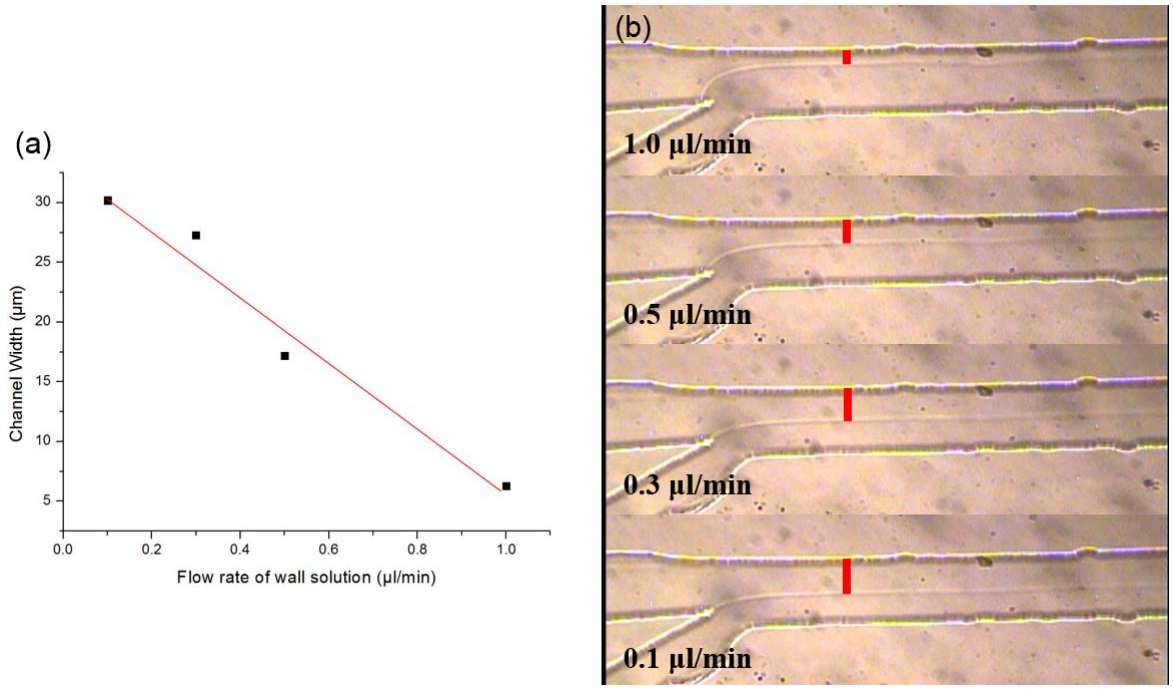

Figure S2. (a) The effective channel width as a function of the flow rate of virtual wall solution. The effective channel has only a high-conductivity solution, excluding the part of a low-conductivity wall solution. (b) Photographs of the main microchannel in which the width of the effective channel (indicated by red lines) varies according to the flow rate of virtual wall solution (0.1, 0.3, 0.5, and 1 μl/min) with a fixed flow rate of a sample solution (1 μl/min).

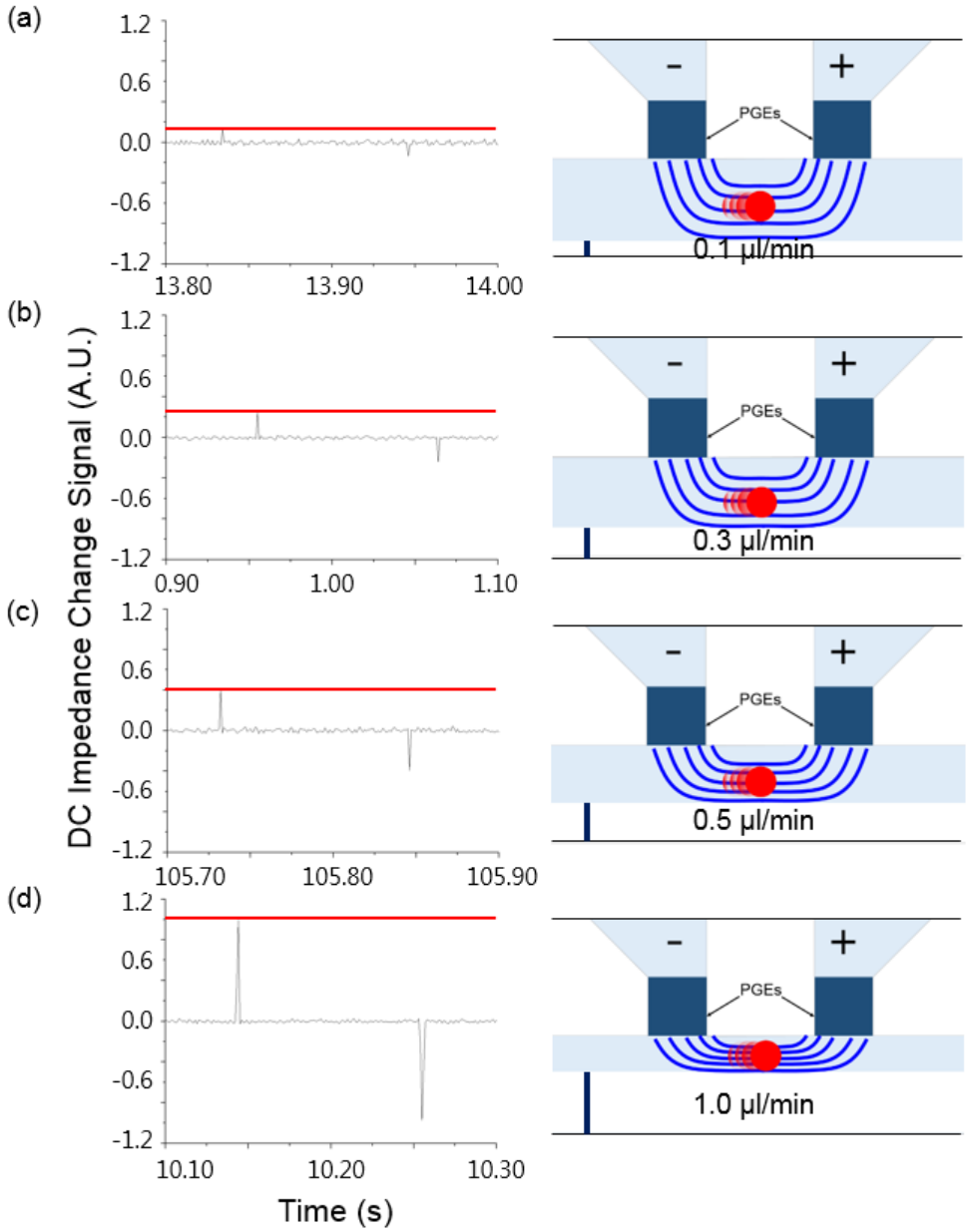

Figure S3. Examples of DC impedance change signals (ΔZ) of 1.69 µm-sized microparticles according to the various flow rates of virtual wall solution: (a) 0.1, (b) 0.3, (c) 0.5, and (d) 1.0 µl/min. With a constant size of microparticle, the amplitude of the ΔZ signals varies as a function of the flow rate of the virtual wall solution. Higher flow rate of the virtual wall solution (indicated by navy blue lines) leads to a smaller detection volume. As the detection volume is reduced, ionic flows are perturbed more by the same-sized microparticles, which gives rise to greater amplitudes of the ΔZ signals (red lines).

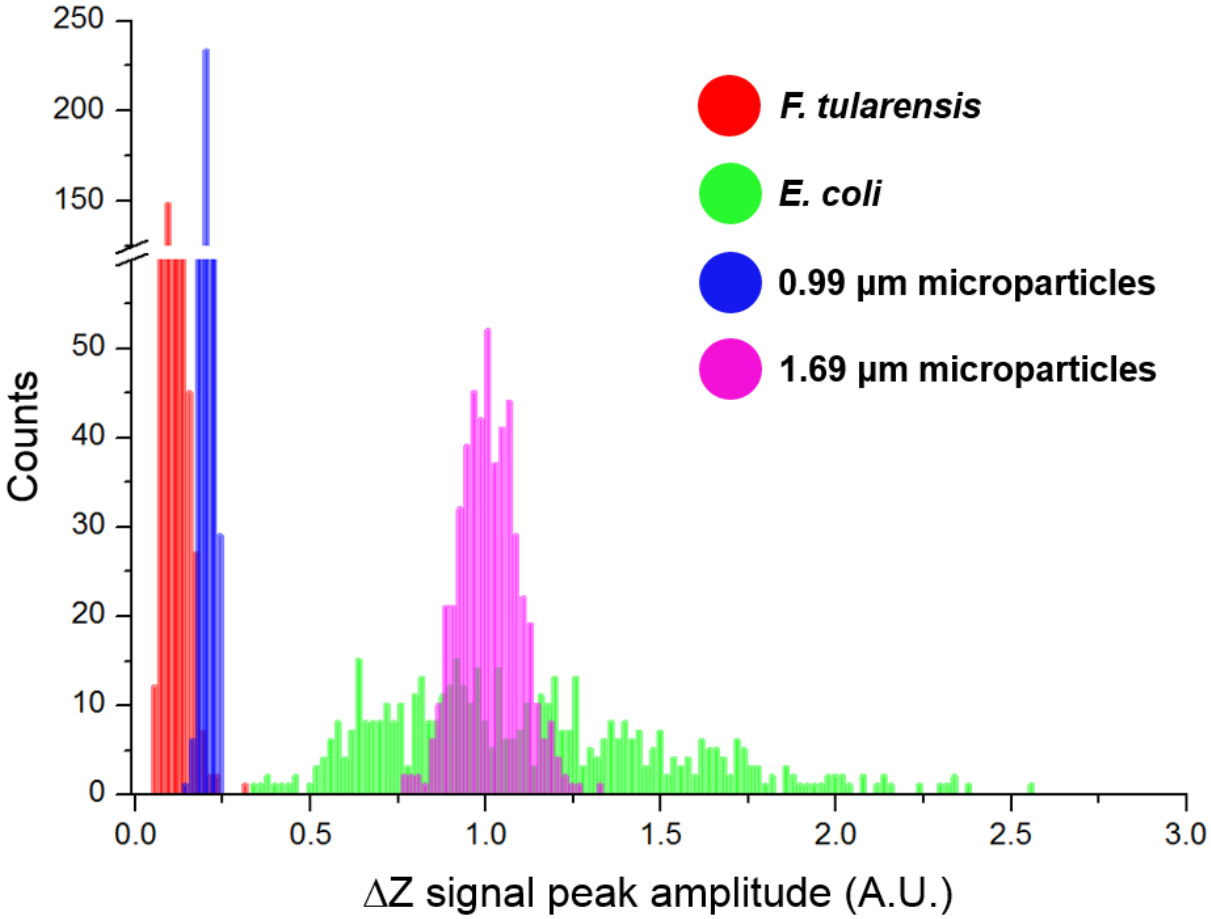

Figure S4. The distribution profile of ΔZ peak amplitudes for target samples (*F. tularensis*, *E. coli*, 0.99, and 1.69 μm diameter microparticles) when the flow rates of the sample solution and virtual wall solution are fixed at 1 μl/min). Since the blue columns indicate 0.99 μm diameter microparticles, *F. tularensis* (red columns) are mostly submicron-sized bacteria, assuming that the shape of the bacteria is spherical (n = 500).

Samples / Peak amplitude (A.U.)	*F. tularensis*	0.99 μm microparticles	*E. coli*	1.69 μm microparticles
Mean (*m*)	0.119	0.202	1.12	1.00
Standard deviation (*σ*)	0.0323	0.0158	0.411	0.0848

Table S1. The mean (*m*) and standard deviation (σ) values of peak amplitudes of the ΔZ signals of the tested samples (*F. tularensis*, *E. coli*, 0.99 and 1.69 μm diameter microparticles) using the flow cytometry–based bacterial detection system where the flow rates of sample solution and virtual wall solution are fixed at 1 μl/min.