**Figure S1. Real-time quantitative PCR amplification curve.** The designed MGB probes and primers for Escherichia coli and Staphylococcus aureus. A: amplification curve of Escherichia coli; B: amplification curve of Staphylococcus aureus.

**Figure S2. Dynamic range of the ddPCR-based assay.** Detecting the bacterial nucleic acid by ddPCR system, A: The 1D droplet spots of FAM fluorescence for E. coli Clinical strain were shown on the left and middle. B: The 1D droplet spots of VIC fluorescence for Staphylococcus aureus ATCC25913 were shown on the left and middle The estimated copy numbers of E. coli Clinical strain nucleic acid template (copies/20μl) is 1.3×105, 1.3×104, 1.3×103, 1.3×102, 32 and NC. The estimated copy numbers of Staphylococcus aureus ATCC25913 nucleic acid template (copies/20μl) is 1.3×104, 1.3×103, 1.3×102 and NC,. The linear fitting curves of E. coli Clinical strain and Staphylococcus aureus ATCC25913 were shown on the right.