An automatic centrifugal system for rapid detection of bacteria based on immunomagnetic separation and recombinase aided amplification

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Preparation of immune MNPs

The immune MNPs were prepared using EDC method with some modifications¹. Briefly, 1 mg MNPs were washed with 3 mL PB (0.01 M, pH 6.0) twice and added to a 3 mL mixture including 100 µL EDC·HCl (10 mg/mL in PB) and 100 µL NHSS (10 mg/mL in PB), shaking at 220 rpm for 1 h at 37 °C to activate the beads. After washing with PB twice to remove surplus EDC·HCl and NHSS, 100 µg anti-*Salmonella* typhimurium polyclonal antibodies were added to the beads in 3 mL PB (0.01 M, pH 8.0). The mixture was reacted at 220 rpm for 2 h at 37°C and then blocked with 400 µL skimmed milk (100 mg/mL in PB) for 4 h at 37°C with shaking. After washing with PB (0.01 M, pH 8.0) twice, the immune MNPs were resuspended in 1 mL of PB (0.01 M, pH 8.0) containing 15 wt% sucrose and 1 wt% skimmed milk and stored at 4°C.

Method	Time (h)	Equipment Cost (US \$)	Consumable Cost (US \$/sample)	Detection limit (CFU/mL)	Ref
Culture	48	~4,000	~1.5	1	2
PCR	1-3	~40,000	~9	10 ³	3,4
ELISA	2-4	~32,000	~5	104	5,6

 Table S1. Comparison of currently available technologies for bacterial detection.

Primer Type	Nucleotide Sequence		
Probe	CTCTATTGTCACCGTGGTTCAGTTTATCG(FAM- dT)T(THF)T(BHQ1-dT)ACCAAAGGTTCAG[C3-spacer]		
Forward primer	ATTGGCGATAGCCTGGCGGTGGGTTTTGTTGT		
Reverse primer	TACCGGGCATACCATCCAGAGAAAATCGGGCCGC		

Table S2. Nucleotide sequences of RAA primers and prober⁷



Fig. S1 The photo of the sample-in-result-out device.



Fig. S2 The embedded software of the supporting device



Fig. S3 The pictures of the centrifugal disc at different stage. (a) After sample and nucleic acid extraction buffer loading. (b) After the first clockwise centrifugation. (c) After the second clockwise centrifugation.
(d) After the third clockwise centrifugation. (e) After the forth counterclockwise centrifugation.



Fig. S4 The infrared image of the annular heater

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