Electronic supplementary information for

Microfluidic Filter Device Coupled Mass Spectrometry for Rapid Bacterial Antimicrobial Resistance Analysis

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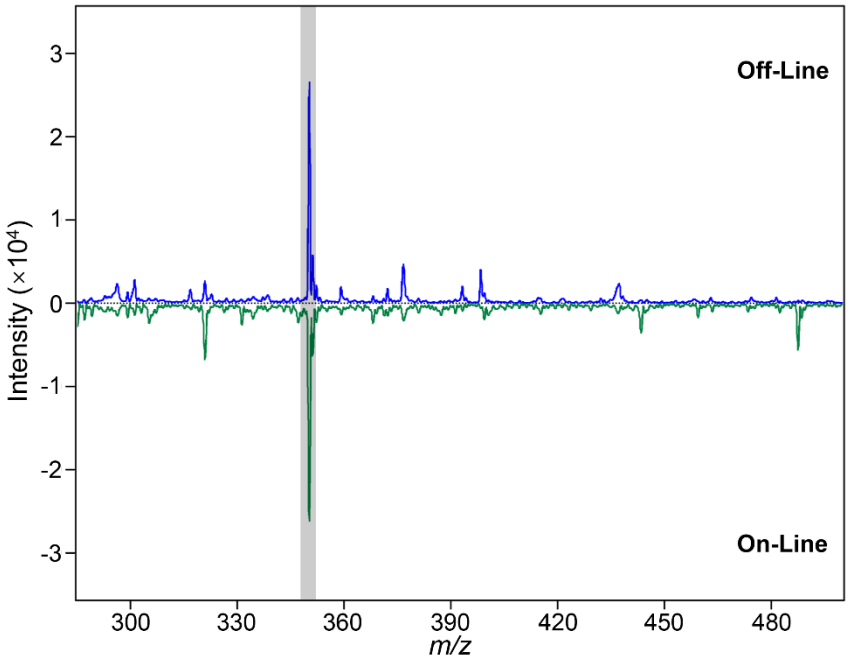

Fig. S1 Mass spectra of AMP by the ESI-MS. Off-Line: 5 μg mL^{-1} AMP aqueous solution was mixed with the ESI buffer (1:1 v/v) in a centrifuge tube and then analyzed by the microchip ESI-MS; On-Line: the ampicillin aqueous solution and the ESI buffer were infused into the microchip via different channels and mixed in the microchip. Peaks corresponding to AMP ([M+H]$^+$ = 350) are highlighted in gray.

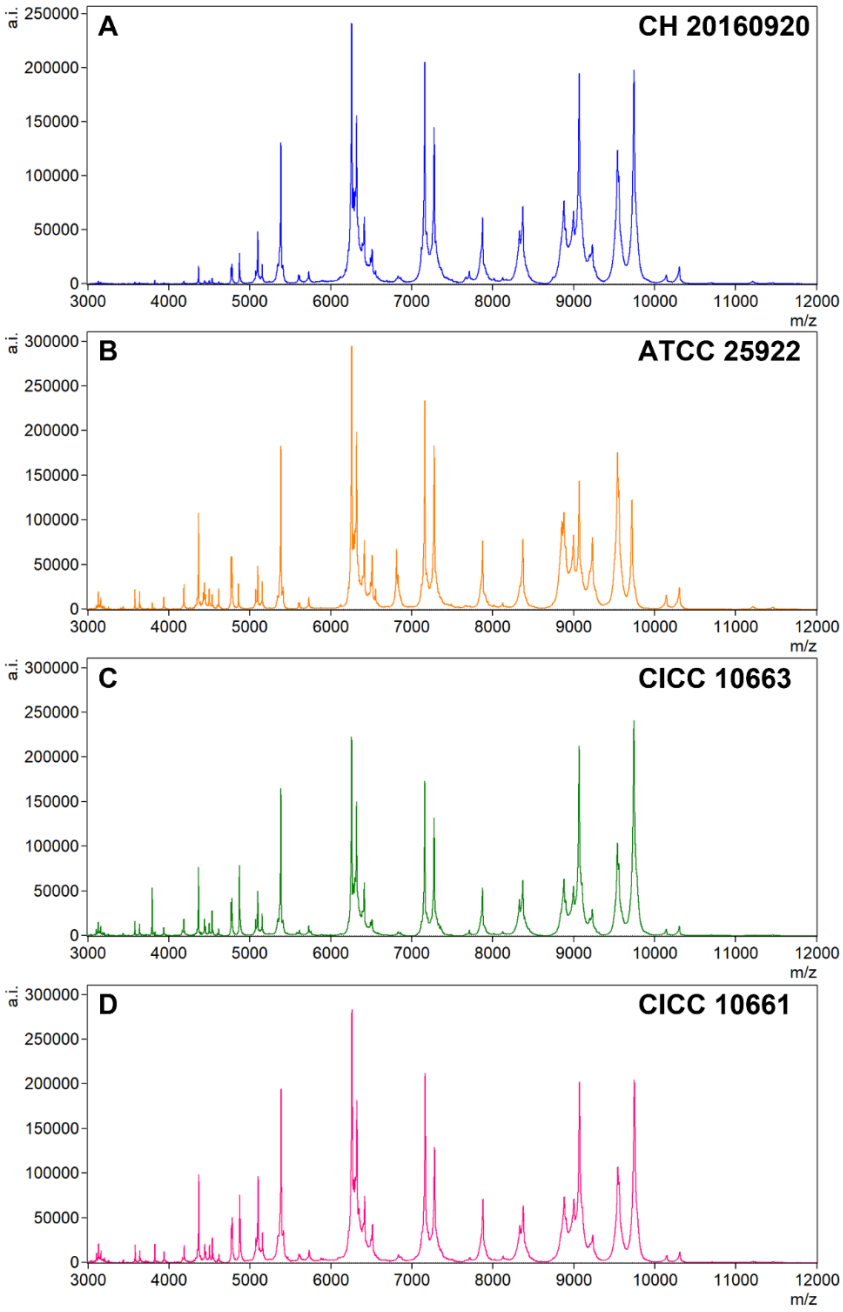

Fig. S2 Mass spectra of four *E. coli* strains including (A) CH 20160920; (B) ATCC 25922; (C) CICC 10663; and (D) CICC 10661 by MALDI-TOF MS. The strains were identified as *Escherichia coli* by searching the mass spectra against a built-in library using the BioExplorer (v3.2, Bioyong Technology Co. Ltd., Beijing, China).

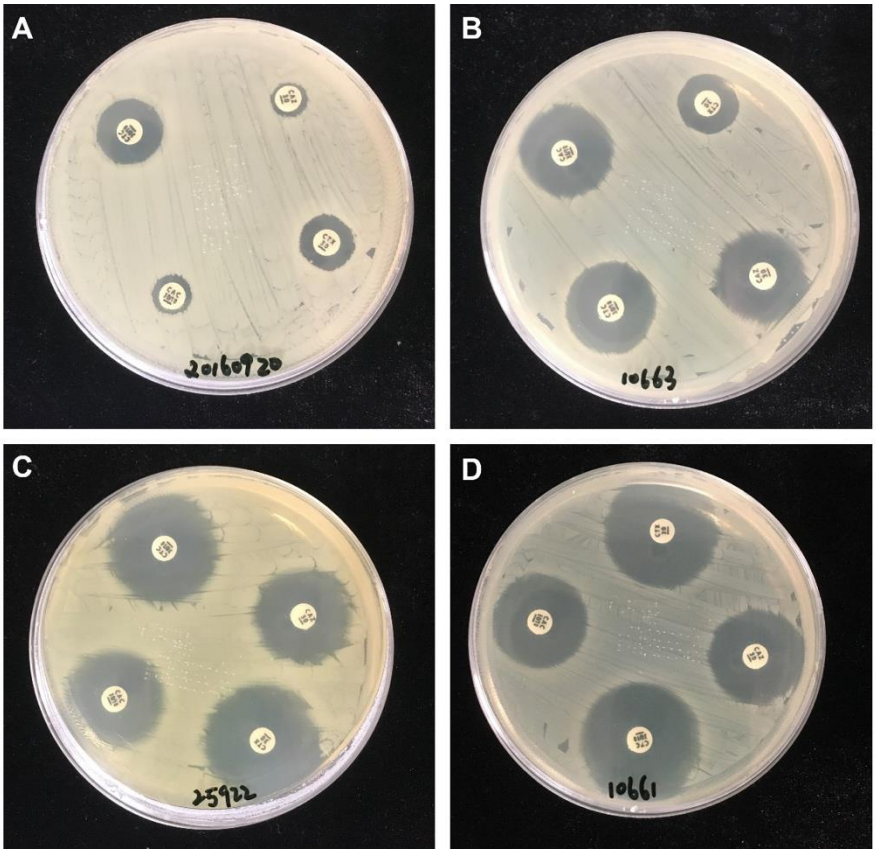

Fig. S3 Double-disk potentiation tests for bacterial antimicrobial resistance analysis: (A) CH 20160920; (B) CICC 10663; (C) ATCC 25922; (D) CICC 10661.

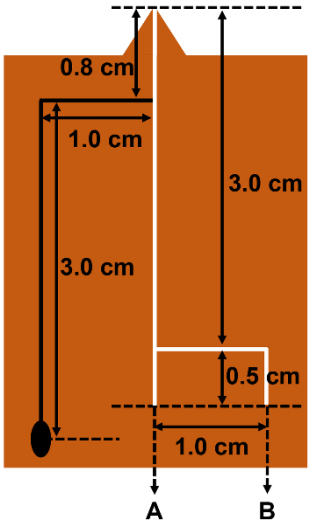

Fig. S4 The PET microchip including two microchannels (White Lines) and one micro-carbon electrode (Black Line). Channel A is for samples from the PMMA chip, and Channel B is for ESI buffer. The depths and widths of all microchannels are 50 and 100 μm, respectively.

Table S1. The result of the double-disk potentiation tests for *E. coli* strains.

Strain No.	CTX (mm)	CTC (mm)	CAZ (mm)	CAC (mm)	ESBL
CH 20160920	11.08	15.04	7.76	8.56	positive
CICC 10663	13.68	20.46	20.36	21.54	positive
ATCC 25922	29.38	30.12	26.00	25.98	negative
CICC 10661	29.46	29.28	24.62	24.20	negative

CTX: cefotaxime; CTC: cefotaxime/clavulanic acid; CAZ: ceftazidime; CAC: ceftazidime/ clavulanic acid. When the difference in diameter of bacterial inhibition ring by CAZ and CAC were $\geqslant$5 mm, or/and by CTX and CTC were $\geqslant$3 mm, the strains could be determined as ESBL-producing bacteria.

Table S2. The RSD of extractive MS signal for single experiments and for three replicates corresponding to **Fig. 3**.

RSD (%)	AMP								CEF							
	m/z 350				m/z 368				m/z 555				m/z 370			
	a			b	a			b	a			b	a			b
No bacteria	2.83	3.51	1.42	1.83	5.39	3.65	3.20	1.44*	4.95	2.43	2.11	4.16	2.53	2.04	2.45	12.8*
CH 20160920	5.07	2.69	5.60	5.03	5.71	6.41	6.57	3.39	5.42	5.31	6.08	3.10	4.19	3.57	4.94	5.18
CICC 10663	6.25	4.07	4.02	1.87	6.42	6.08	3.57	0.95	1.79	3.00	5.00	5.81	2.22	3.07	1.71	3.43
ATCC 25922	5.95	4.59	5.65	4.99	5.09	8.25	9.74	21.1*	1.89	4.12	1.46	6.36	3.16	1.35	8.60	24.5*
CICC 10661	4.93	2.12	8.56	1.71	1.11	1.43	5.79	21.9*	1.54	2.35	1.44	6.32	3.09	2.31	3.15	15.9*

a: RSD of extractive MS signal intensity from single experiment during a period of MS signal recording;

b: RSD of extractive MS signal intensity from three replicates. The mean value of each replicate was used to calculate the RSD of three replicates.

* It should be noted that the signals of the hydrolysis products by the strains ATCC 25922 and CICC 10661, and in the case of no bacteria were from the baseline rather than real MS peaks. Therefore, large RSD was observed.