Supplementary Information

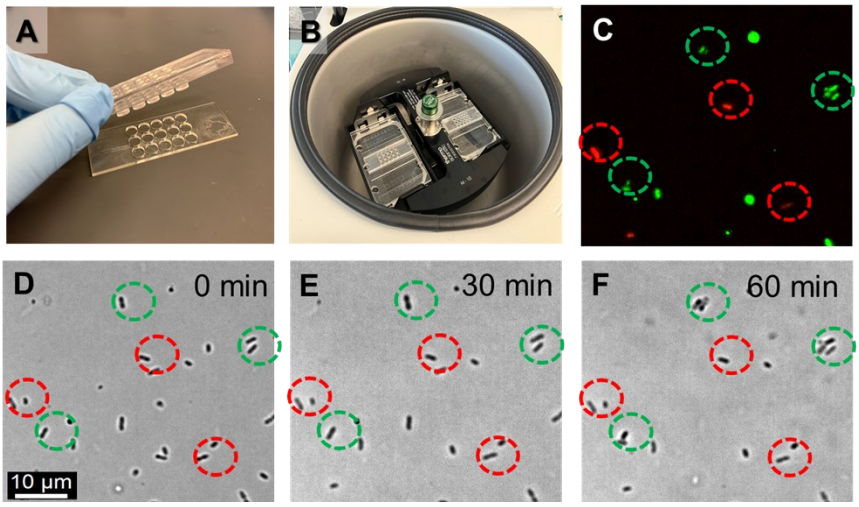

Fig. S1. Loading of gel-encapsulated bacteria into microchamber arrays by centrifugation. (A) A 15-well device with matching PDMS molds for cell encapsulation. (B) Six chips can be centrifuged in parallel with a custom-made chip holder. (C) Single cell killing kinetics measured using cell viability indicators. (D-f) Time-lapse microscopy of individual *E. coli* encapsulated in gel. Green and red circles highlight live and dead bacteria.

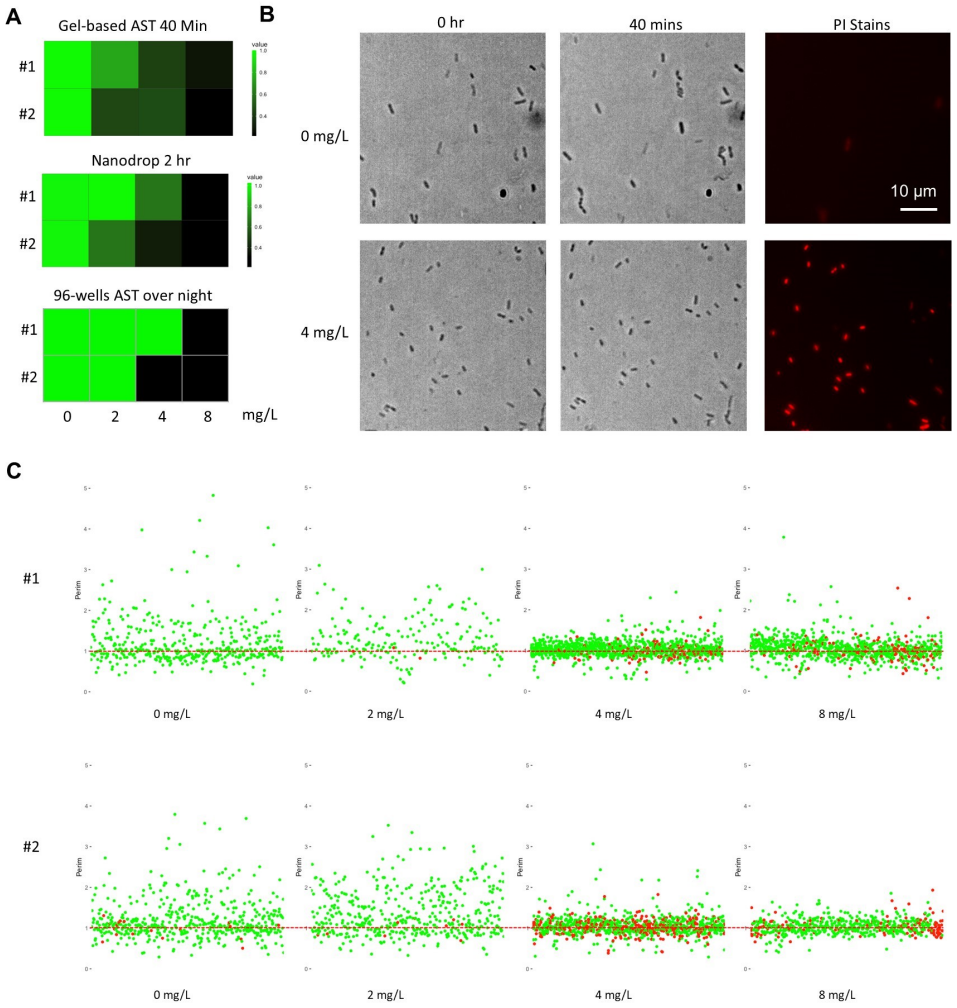

Fig. S2. Single cell response of *K. pneumoniae* to different concentrations of ciprofloxacin. (A) Growth measurements obtained using the microfluidic gel encapsulation platform and 96-well plates. (B) Brightfield and fluorescence images illustrating the response of two *K. pneumoniae* clinical isolates exposed to ciprofloxacin. (C) Changes in the perimeter and area of individual bacteria. Red dots indicate dead cells, as shown by PI staining. Values were normalized to the initial measurement.

Concentration (mg/L)	Percentage of viable cells (%)		
	60 min	90 min	120 min
0	100	100	100
2	96.8	94.4	93.6
20	97.3	96.4	96.2
128	96.3	95.3	95.1

Fig. S3. Percentage of viable cells. The percentage of viable cells was assessed using Propidium Iodide (PI) dye, which evaluates cell viability based on membrane integrity. Some cells were stained with PI, despite ciprofloxacin not directly affecting the bacterial cell membrane integrity.

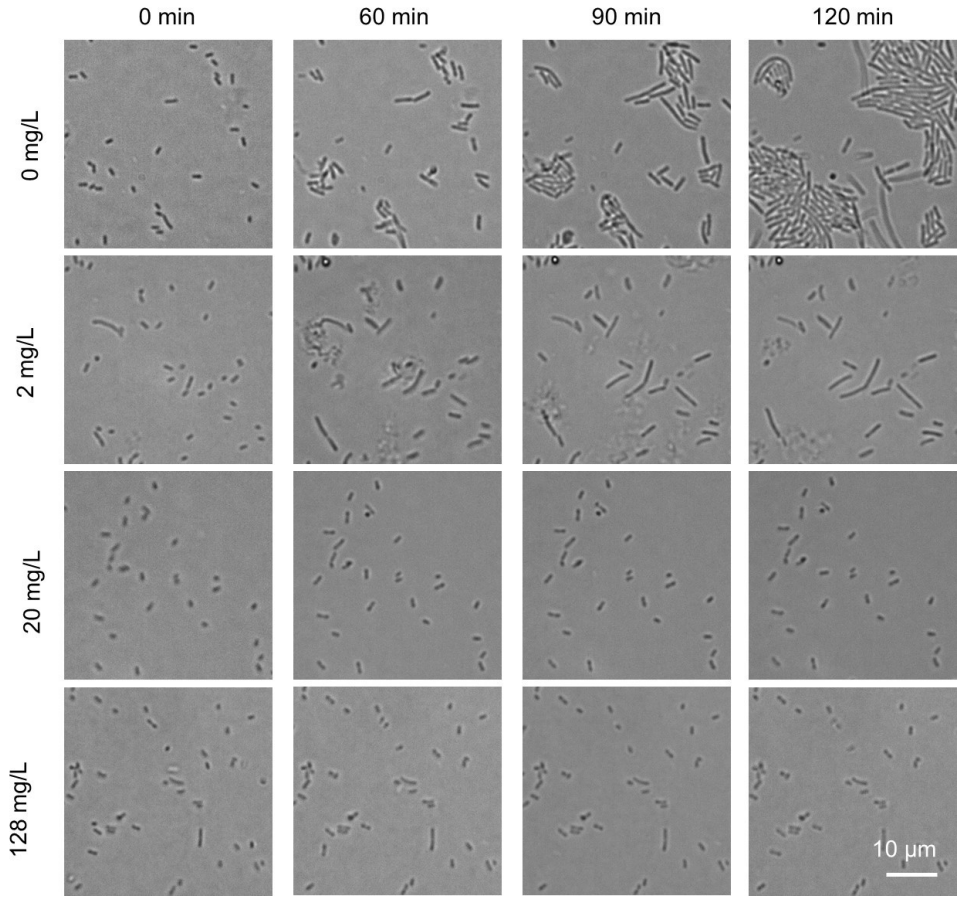

Fig. S4. Time-lapse brightfield images of *E. coli* exposed to ciprofloxacin.

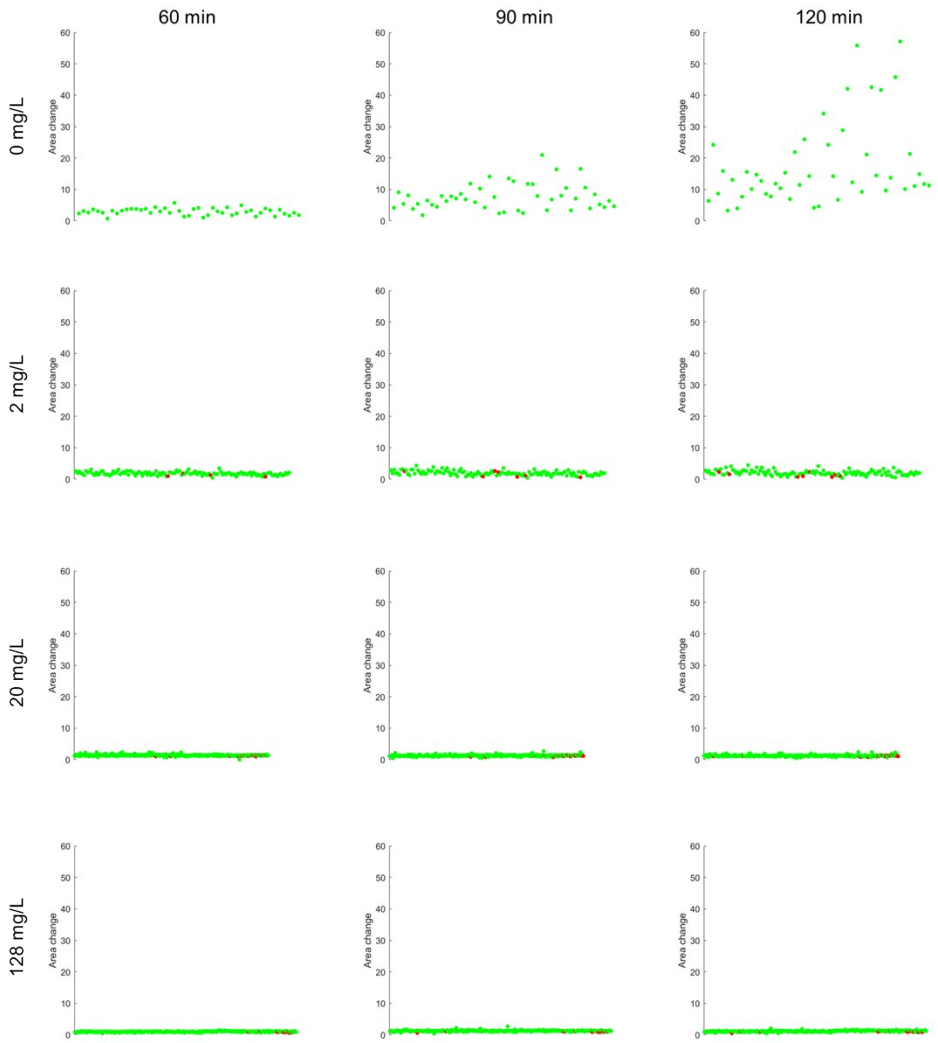

Fig. S5. Kinetics of area of individual *E. coli* exposed to ciprofloxacin. Values were normalized to the initial measurement.

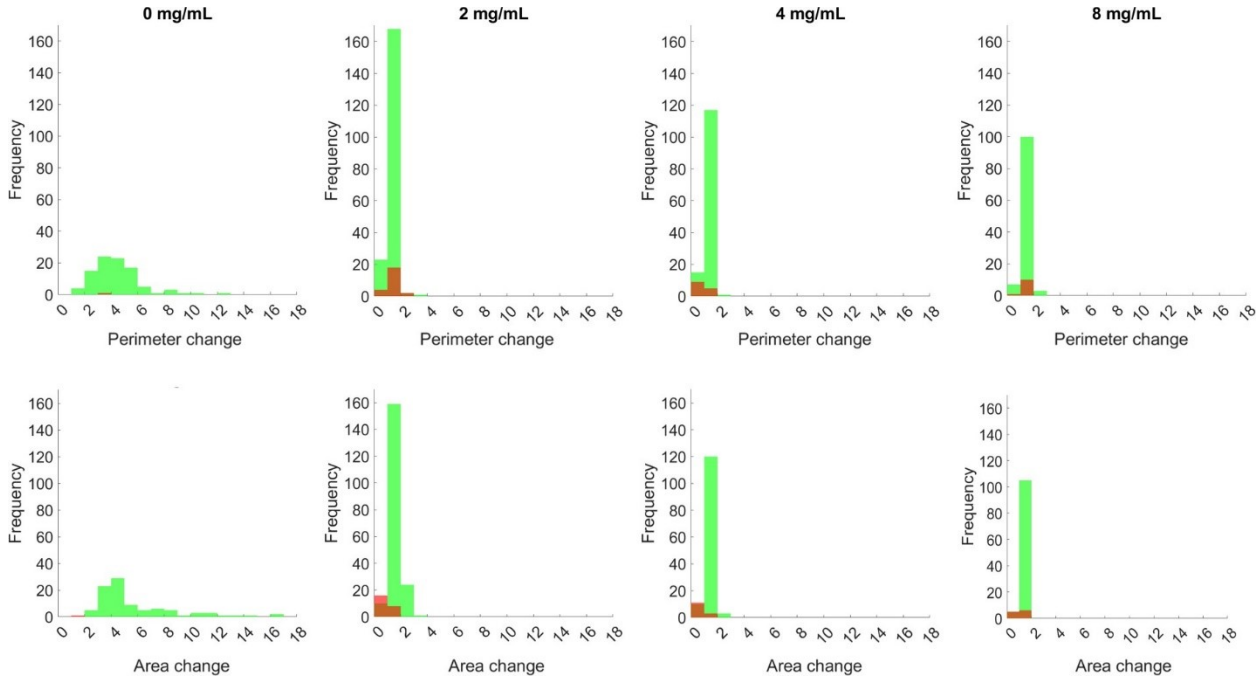

Fig. S6. Single cell response of *E. coli* to different concentrations of ciprofloxacin after 90 minutes. Histograms of bacterial response of uropathogenic *E. coli* (EC137) in culture broth. Red bins indicate dead cells, as shown by PI staining. Values were normalized to the initial measurement.

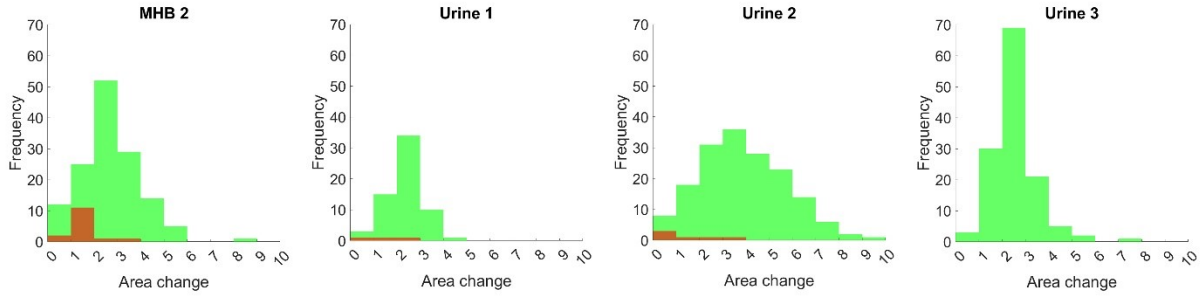

Fig. S7. Single cell response of *E. coli* to ciprofloxacin across different media. Histograms of bacterial response of uropathogenic *E. coli* (EC137) to ciprofloxacin. Red bins indicate dead cells, as shown by PI staining. Values were normalized to the initial measurement.