

Supplementary Materials

A rapid and specific antimicrobial resistance detection of *Escherichia coli* via magnetic nanoclusters

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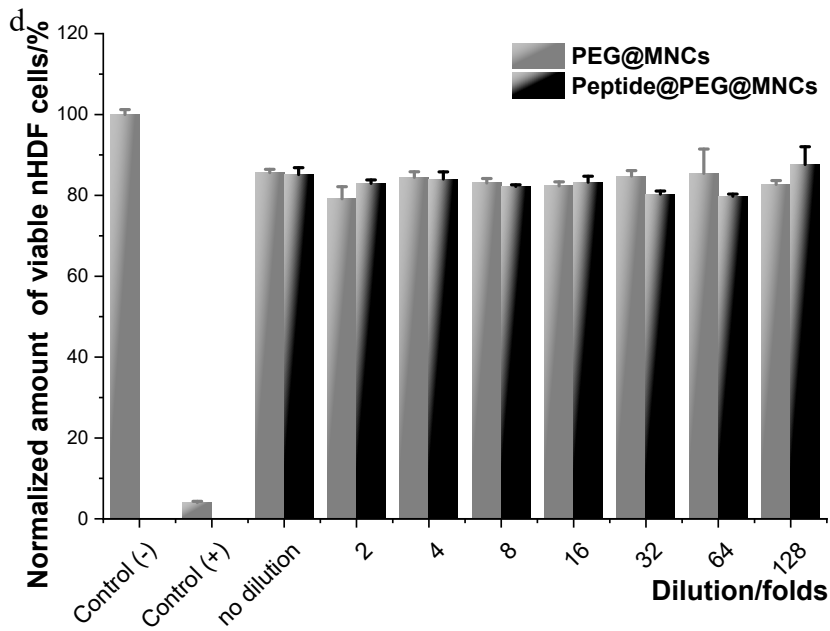
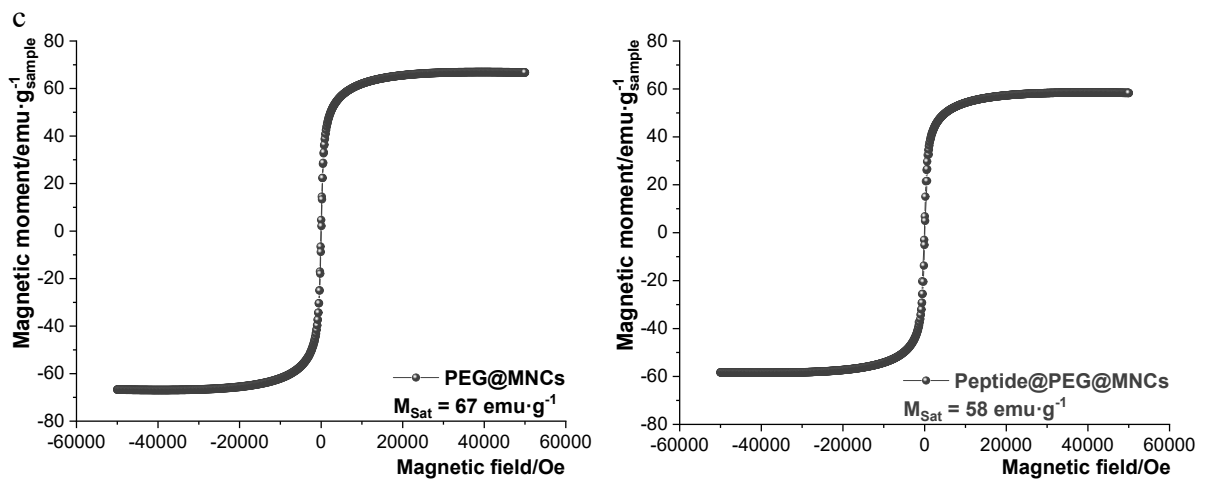
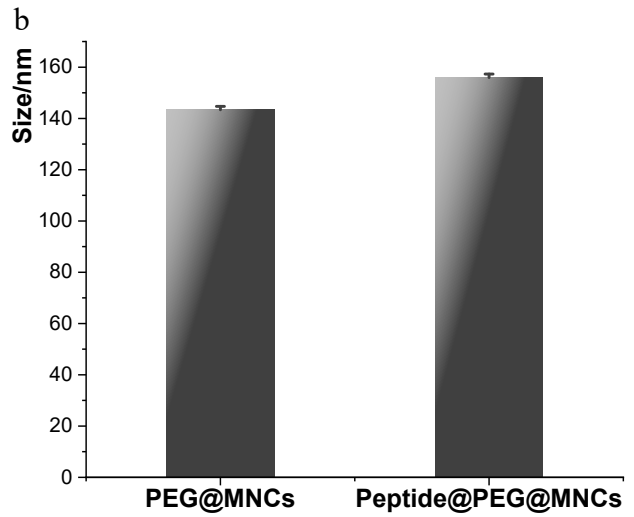
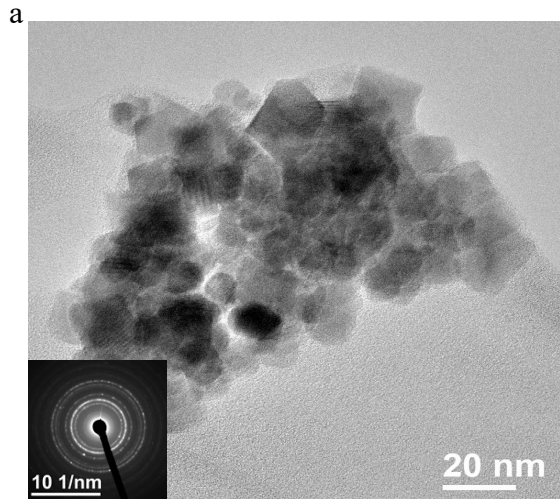
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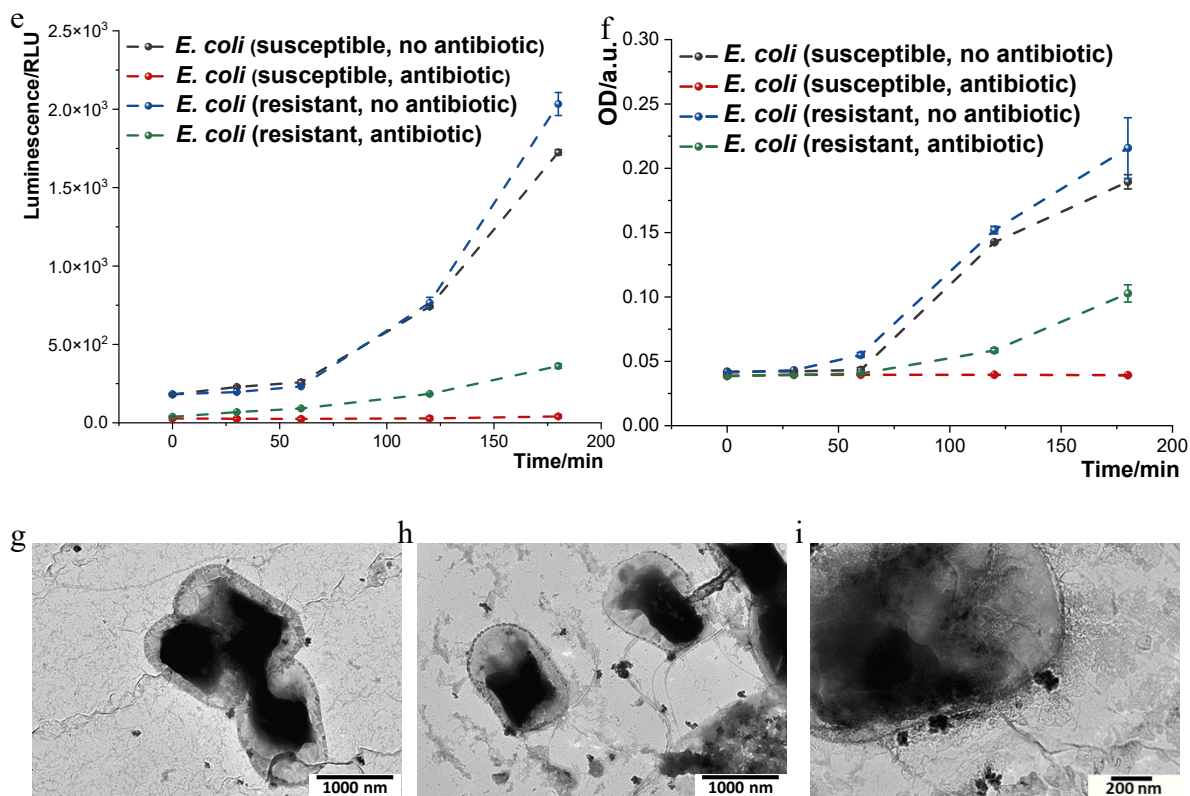


Figure S1. Bacterial capture in PBS towards *E. coli*. (a) PEGylated MNCs before functionalization was imaged by applying a scanning transmission electron microscope (STEM), manifesting a cluster structure. (b) The size of PEG@MNCs before and after functionalization was analyzed by employing a dynamic light scattering (DLS). (c) Analysis of vibrating sample magnetometry (VSM) hysteresis was performed at room temperature towards PEG@MNCs and peptide@PEG@MNCs, including the relative values of saturation magnetization (M_{sat}). (d) The viability of the normal human dermal fibroblasts (nHDFs) was measured after incubation for 24h at 37°C with PEG@MNCs and peptide@PEG@MNCs in DMEM containing 1 % penicillin/streptomycin/neomycin (PSN) at pH 7.4. The cytotoxic cut-off was determined as 70 % viability of the nHDFs measured from the negative control (empty wells). 1 % Triton X-100 in DMEM containing 5 % FCS was regarded as a positive control. No cytotoxicity was observed for all samples aligned with the negative control. Error bars denoted the standard deviations calculated from 8 measurements. The growth analysis of the captured susceptible and resistant *E. coli* without a rinsing process was performed in the presence of 300 $\mu\text{g}\cdot\text{mL}^{-1}$ ampicillin through the utilization of AquaSpark[®] beta-D-glucuronide (e) and optical density (f). (g)-(i) The interaction between susceptible *E. coli* and peptide@PEG@MNCs was analyzed through a transmission electron microscope (TEM). $n = 3$ (biological repeats), mean \pm SD shown.

Table S1. Affinity evaluation was analyzed using single bacterial adhesion force spectroscopies between bacteria and various nanoclusters (MNCs, peptide@MNCs, PEG@MNCs, and peptide@PEG@MNCs). Every type of nanoclusters was deposited on glass petri dishes coated by polydopamine according to published methods(1). Every measurement was carried out at least ten times to derive the average value and standard deviation.

Single bacterial adhesion force between different bacterial pathogens and various surfaces/nN					
	<i>S. aureus</i>	<i>P. aeruginosa</i>	<i>S. epidermidis</i>	Susceptible <i>E. coli</i>	Resistant <i>E. coli</i>
Glass	1.35 ± 0.43	1.39 ± 0.65	0.77 ± 0.57	1.04 ± 0.42	0.66 ± 0.30
polydopamine@Glass	6.25 ± 1.79	8.25 ± 2.39	8.37 ± 1.40	8.17 ± 1.62	2.99 ± 0.59
MNCs	1.47 ± 0.40	1.15 ± 0.68	1.11 ± 0.54	1.81 ± 0.52	1.25 ± 0.44
Peptide@MNCs	1.68 ± 0.86	1.28 ± 0.72	1.23 ± 0.28	1.82 ± 0.57	1.48 ± 0.56
PEG@MNCs	0.61 ± 0.32	0.48 ± 0.24	0.61 ± 0.19	0.42 ± 0.14	0.60 ± 0.32
Peptide@PEG@MNCs	0.77 ± 0.37	1.60 ± 0.44	1.48 ± 0.68	24.44 ± 5.0	23.56 ± 3.84

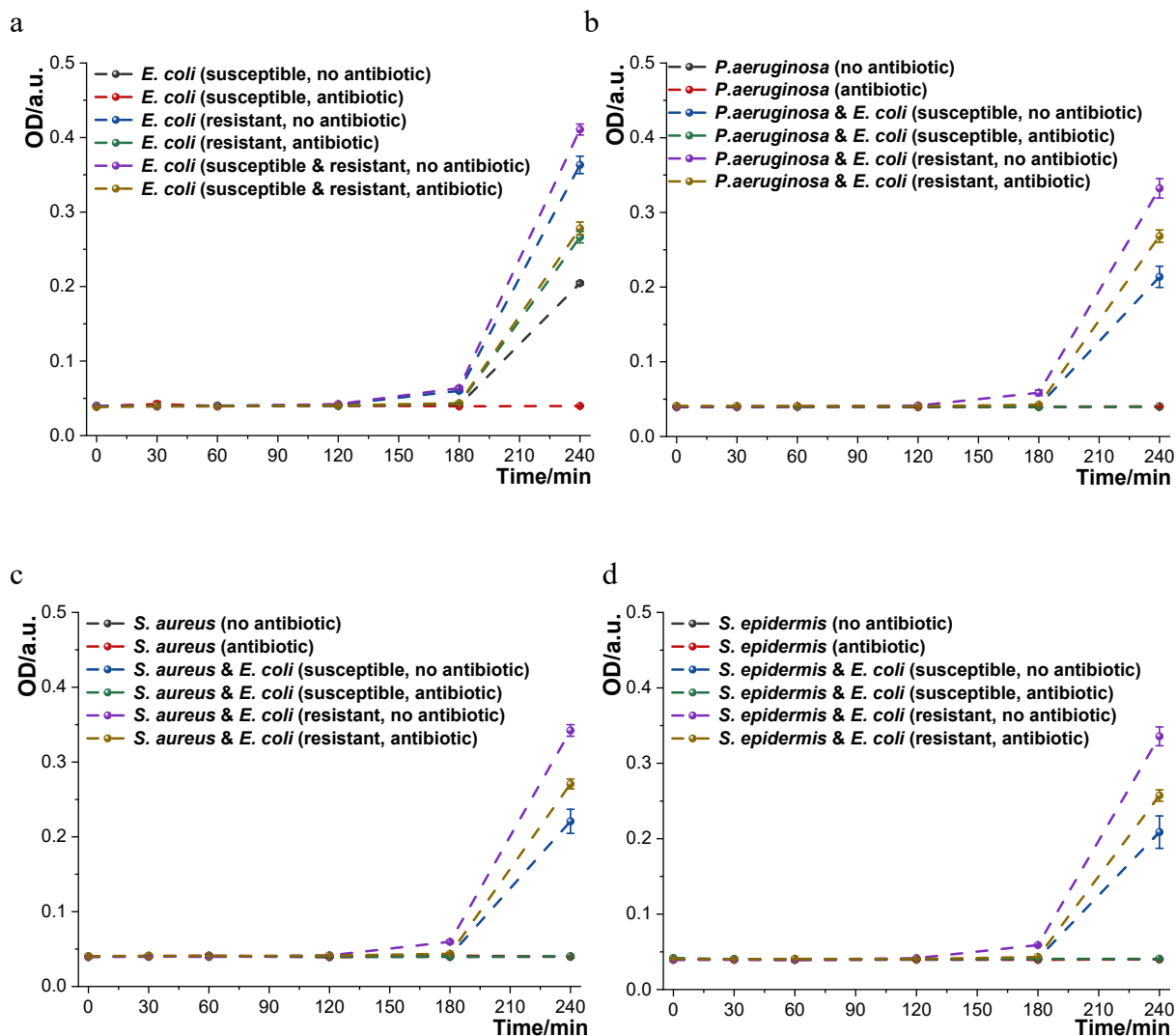
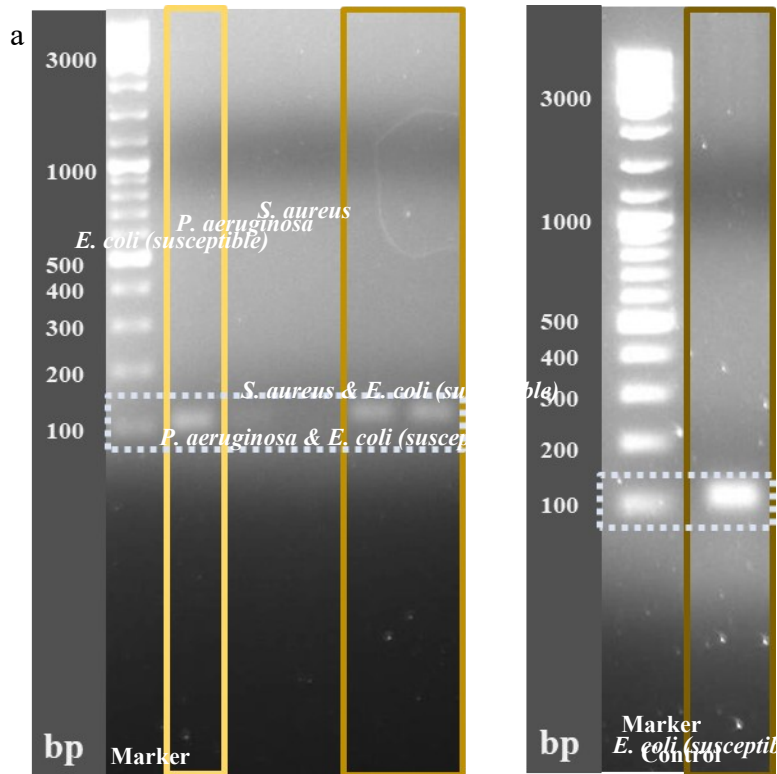


Figure S2. Bacterial capture in artificial urine. (a)-(d) The growth analysis of the captured susceptible and resistant *E. coli* with a rinsing process was performed in the presence of 300 $\mu\text{g}\cdot\text{mL}^{-1}$ ampicillin through applying AquaSpark[®] beta-D-glucuronide and the relative optical density, from a suspension of every single bacterial strain or a mixture of bacterial strains. Single bacterial strain: susceptible and resistant *E. coli*, *P. aeruginosa*, *S. aureus*, and *S. epidermidis*; mixed bacterial strains: susceptible and resistant *E. coli* respectively with *P. aeruginosa*, *S. aureus*, and *S. epidermidis*, and a mixture of susceptible and resistant *E. coli*. n = 3 (biological repeats), mean \pm SD shown.



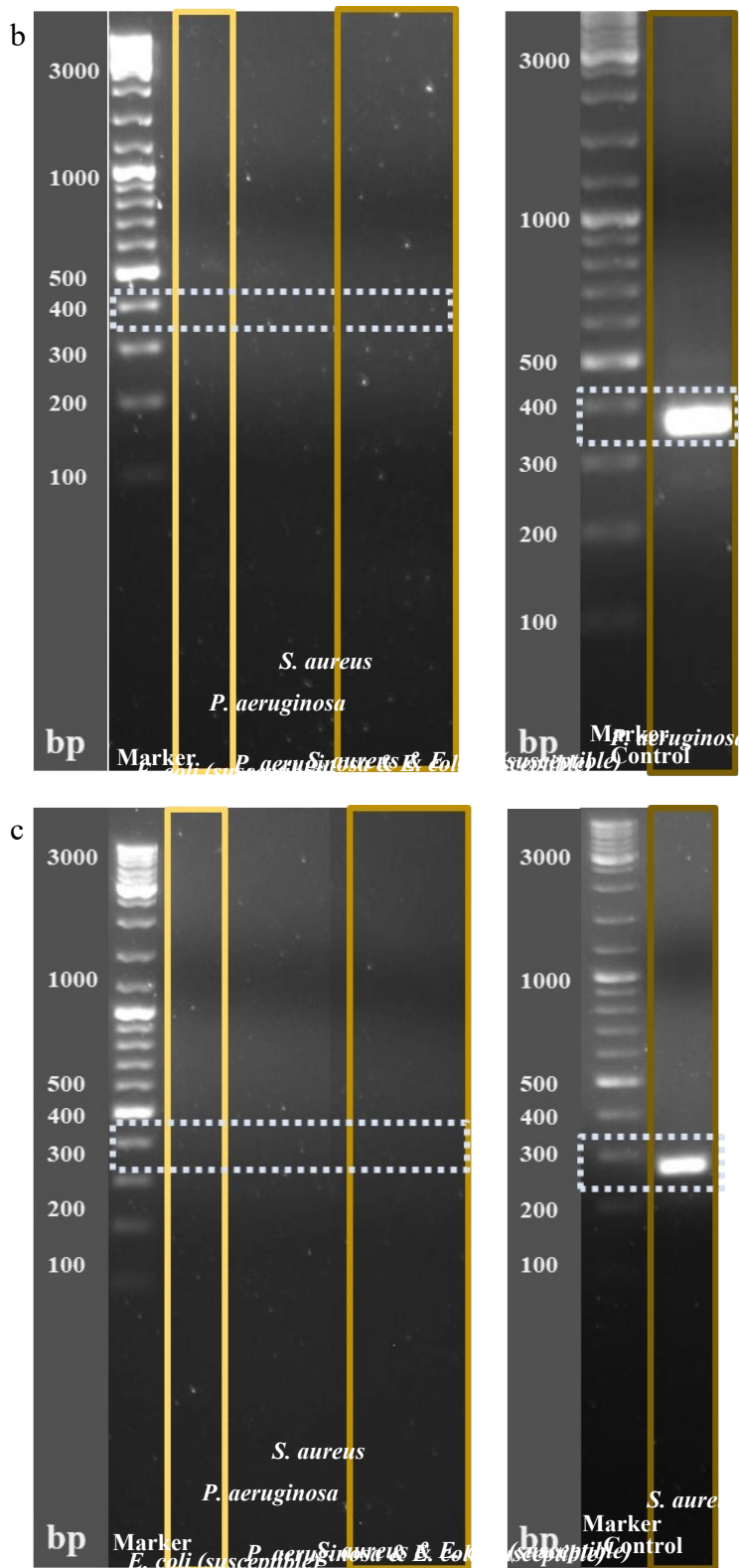


Figure S3. Capture specificity to *E. coli*. Polymerase chain reaction (PCR) analysis was conducted to understand the specificity of the fabricated peptide@PEG@MNCs to *E. coli*. Thereby, the *lacY* gene of *E. coli* (a), the *toxA* gene of *P. aeruginosa* (b), and the 16 rRNA gene of *S. aureus* (c) were respectively targeted. Moreover, we respectively applied susceptible *E. coli*, *P. aeruginosa*, and *S. aureus* as the positive controls. The analyzed peptide@PEG@MNCs were separated after the interaction and rinsing process from the

artificial urine spiked by susceptible *E. coli*, *P. aeruginosa*, *S. aureus*, and susceptible *E. coli*, mixed with *P. aeruginosa*, and *S. aureus*, respectively. Only the samples isolated from artificial urine spiked by susceptible *E. coli* and the bacterial mixtures containing susceptible *E. coli* lead to distinctive bands, revealing the existence of *E. coli*. Hence, the fabricated peptide@PEG@MNCs have the specificity to capture *E. coli* from the infected media.

Reference

1. M. Mittelviehhaus, D. B. Müller, T. Zambelli, J. A. Vorholt, A modular atomic force microscopy approach reveals a large range of hydrophobic adhesion forces among bacterial members of the leaf microbiota. *The ISME journal* **13**, 1878-1882 (2019).