Electronic Supplementary Information

Daylight-stimulated antibacterial activity for sustainable bacterial detection and inhibition

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Experimental Section

Cytotoxicity Assay：CCK-8 was used to evaluate the cytotoxicity of the nanoparticles. Fibroblasts L929 suspension (100 μL) was added into 96-well microplates, with 5000 cells immersed in the complete growth medium per well, cultivated in a humidified 5% CO_2 atmosphere at 37 °C for 24 h for cell attachment. Subsequently, PEI-CD-Agr-TPEDB (10 μL) at concentrations of 20, 50, 100, 200 and 500 μg/mL, respectively, were added to 96-well plates, followed by incubation for 24 h, respectively. Then, CCK-8 solution was added to 96-well plates at 10 μL per well and incubated for 3 h. The resulting solutions were analyzed at 480 nm on a plate reader (BIO-TEK instruments Inc. EL311S, America). This process was repeated for 8 times in parallel. The results are expressed as the relative cell viability (%) with respect to the blank group only with culture medium. The cell viability in each well was calculated from the obtained values as a percentage of the control wells. The results were presented as a mean and standard deviation for each sample.

Determination of Minimum Inhibitory Concentration: Antimicrobial activity of PEI-CD-Agr and PEI-CD-Agr-TPEDB was assessed by using *E. coli* (Gram-negative) and *S. aureus* (Gram-positive). An overnight culture of *E. coli* and *S. aureus* was inoculated into the Luria-Bertani (LB) broth. Then, bacterial concentrations were determined by measuring optical density (OD) at λ = 600 nm at 0.2 (OD of 0.2 corresponded to a concentration of 10^8 CFU/mL) with a UV-2450 (Shimadzu Co., Japan). Finally, this solution was further diluted four times using sterile nutrient broth. The cell count of bacteria solution was predetermined to be 2×10^7 cells/mL. In order

to test the antimicrobial activity of the nanocomplexes, the diluted bacterial culture was incubated at 37 °C for 16 h in glass culture tubes in the presence of the samples with final concentrations ranging from 8 to 512 μg/mL in multiple of 2, respectively. After incubation, the growth of bacteria was determined by measuring the OD at 600 nm on a Shimadzu UV-2450 instrument. MIC values were defined as the lowest concentration at which more than 90% bacteria was inhibited.

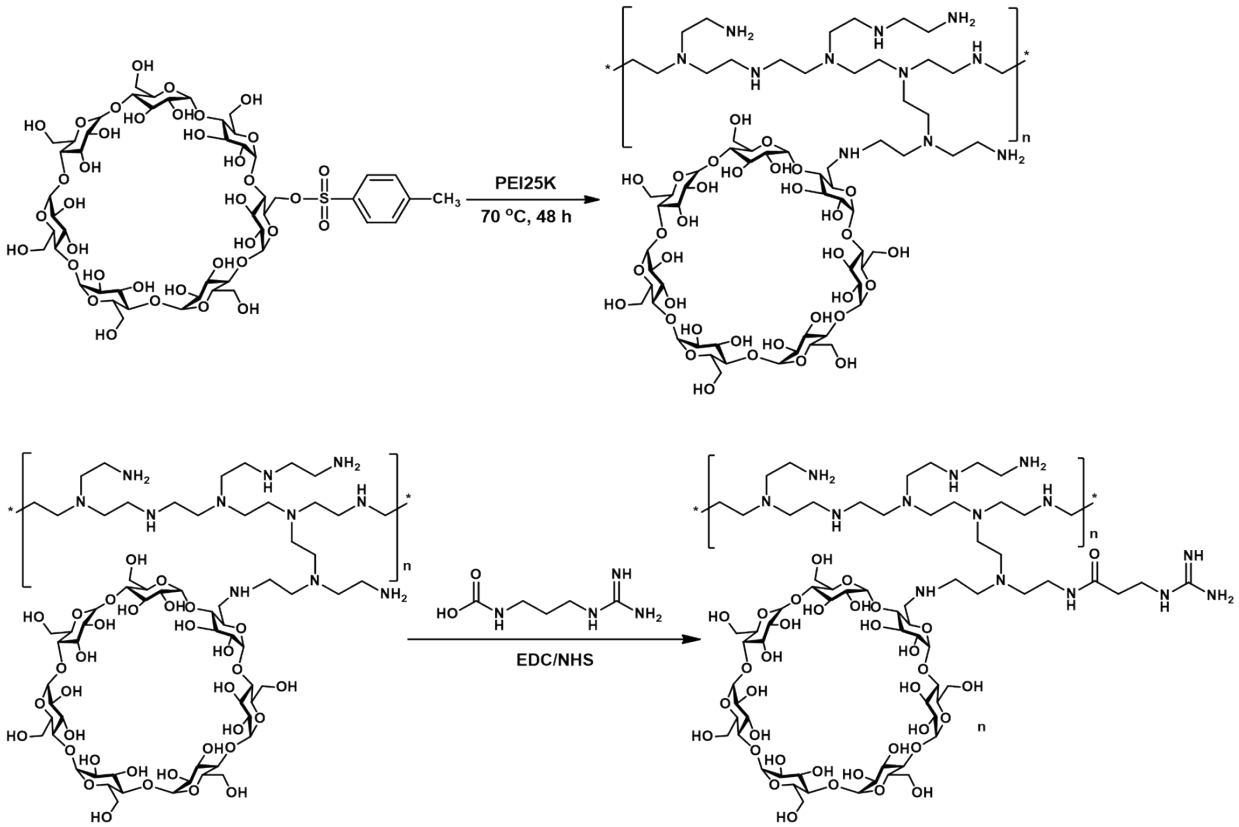
Zone of Inhibition Test: An overnight culture of *E. coli* and *S. aureus* was inoculated into the Luria-Bertani (LB) broth. The bacteria were incubated at 37 °C until a concentration of 2.0×10^9 colony forming units per milliliter (CFU/mL) was reached. The bacterial suspension (50 μL) was inoculated evenly onto an LB agar plate. Then, a sample disk containing the antimicrobial materials was gently placed at the center of the LB agar plate. After incubation overnight at 37 °C, areas of clear media surrounding the disk indicated that the PEI-CD-Agr-TPEDB inhibited bacterial growth. The negative control and positive control were PBS and PEI_{25k}, respectively. The antibacterial activity was evaluated by measuring the diameter of the zone of inhibition around the disk.

Fluorescence Microscopic Observations: A fast-growing bacterial suspension (1.5 mL, OD_{600nm} = 0.5) was centrifuged at 5000 rpm for 5 min and washed with phosphate buffer solution (PBS, 0.01 mol/L, pH 7.4) three times. The supernatant was removed and the remaining bacteria were resuspended in 1.5 mL of PBS. The bacteria were treated with 300 μL of PEI-CD-Agr-TPEDB (32 μg/mL) for 5 min. After interacting with the PEI-CD-Agr-TPEDB, the bacterial cells were rinsed with PBS

three times and imaged using an inverted fluorescence microscope at varying incubation times. The untreated bacterial cells were used as control.

Cytotoxicity of PEI-CD-Arg-TPEDB

The biocompatibility of antibacterial materials is one of the primary concerns limiting wide applications, especially the cytotoxicity to human body cells. In this study, the cell cytotoxicity of PEI-CD-Arg-TPEDB to fibroblasts L929 cell was evaluated by commercial available CCK-8 assay. PEI showed certain toxicity to fibroblasts L929 cell when the concentration increased to 100 μg/mL, but the cytotoxicity of PEI-CD-Arg was confirmed to be markedly reduced. The introduction of multi-hydroxyl groups of CD and essential amino acid (Arg) was confirmed to afford further improvement in the biocompatibility of the PEI-CD-Arg-TPEDB. No cytotoxicity of PEI-CD-Arg-TPEDB was observed to fibroblasts L929 cells, even when the concentration was raised to 500 μg/mL (Figure S8). Thus, the nanocomplex was verified to possess tempting safety profile to fibroblasts L929, thereby portending its safety profile to other human body cells.



Scheme S1. Synthetic route in preparation of PEI-CD-Arg.

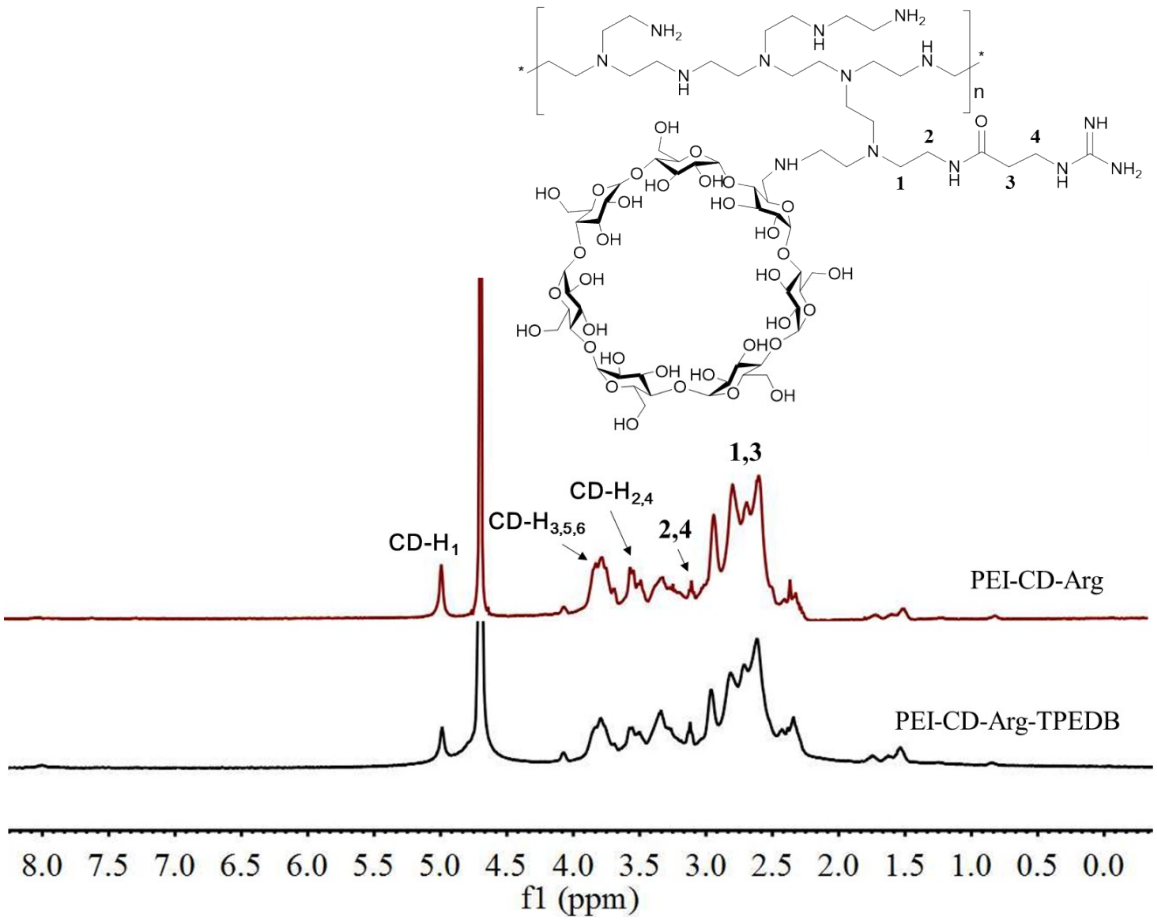


Figure S1. ^{1}H NMR spectrum of PEI-CD-Arg and PEI-CD-Arg-TPEDB in D_2O.

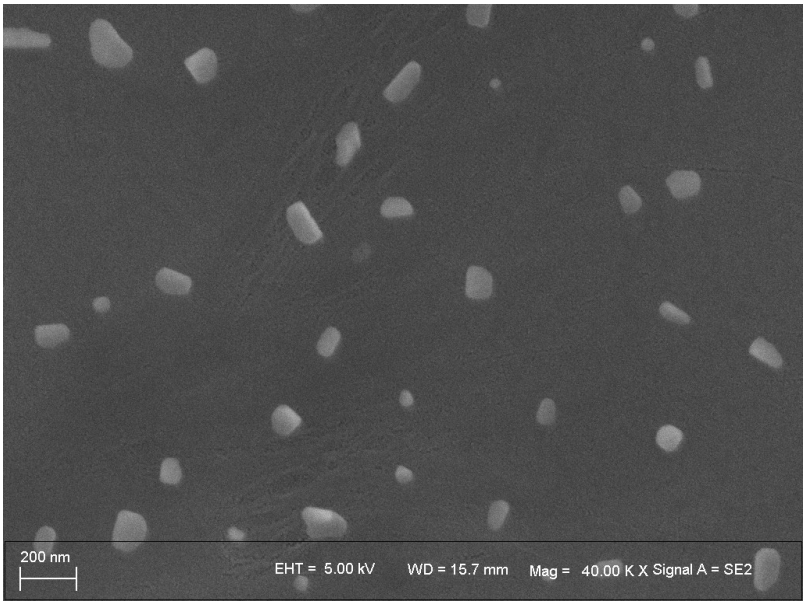


Figure S2. SEM images of PEI-CD-Arg-TPEDB.

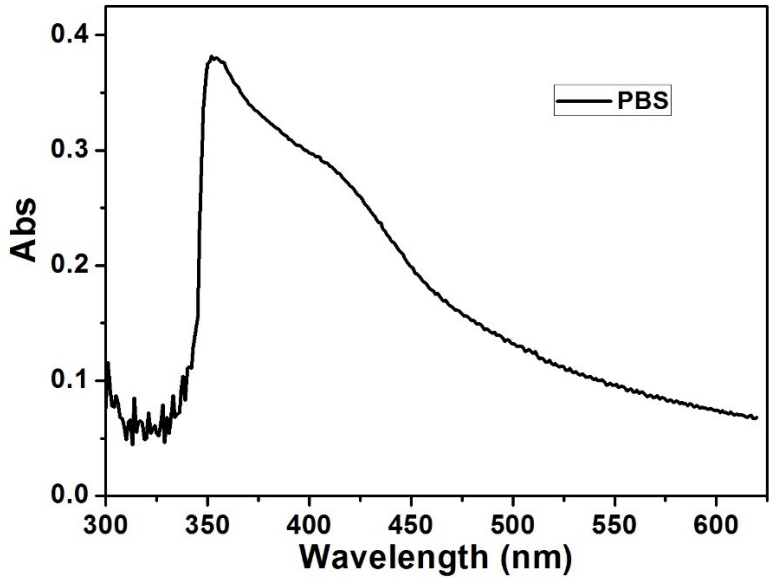


Figure S3. UV-vis absorption spectra of PEI-CD-Arg-TPEDB in PBS.

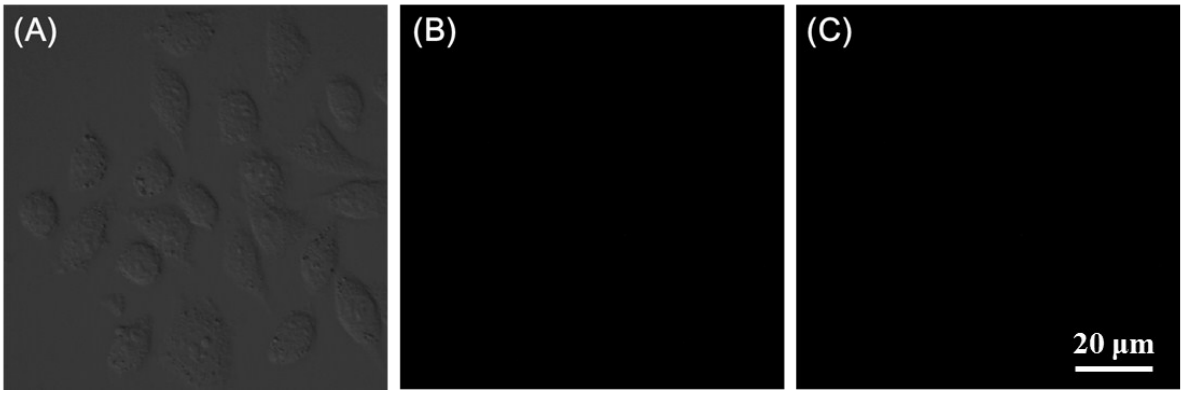


Figure S4. CLSM images of L929 cells. Excitation wavelength: 405 nm for TPEDB and 488 nm for DCFH. Emission detecting ranges: (A) Bright-field images of the L929 cells; (B) 450-488 nm; (C) 505-525 nm.

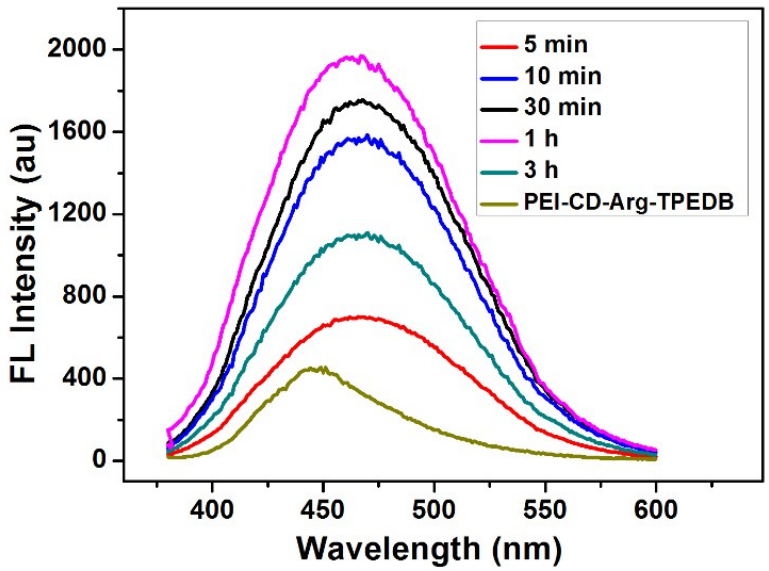


Figure S5. Fluorescence spectra of PEI-CD-Arg-TPEDB incubated with *S. aureus* as a function of incubation time.

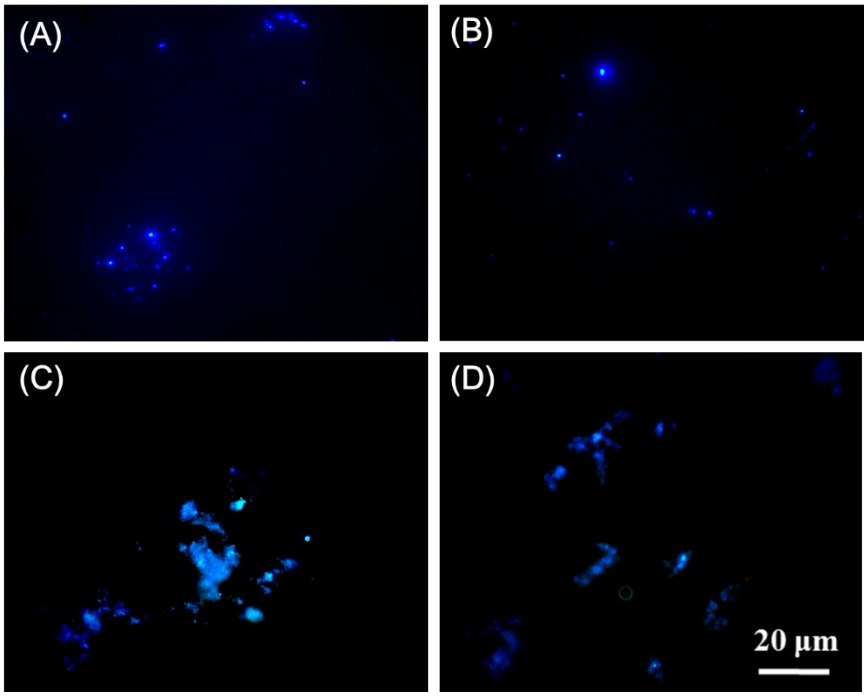


Figure S6. Fluorescence observation of (A, B) *S. aureus* and (C, D) *E. coil* treated with PEI-CD-Arg-TPEDB for (A, C) 1 d, and (B, D) 3 d.

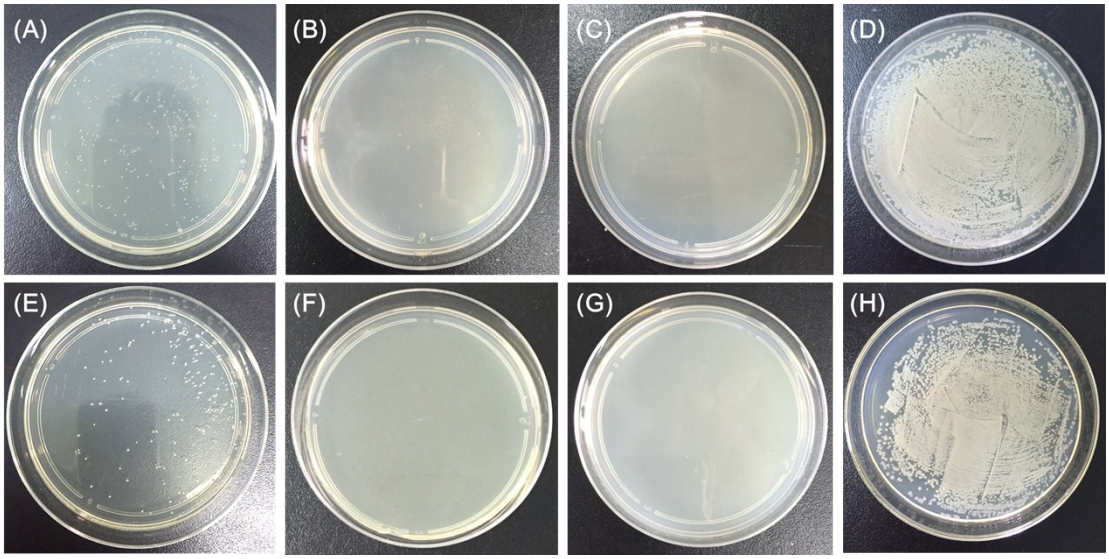


Figure S7. Inhibition of *E. coli* (A-D) and *S. aureus* (E-H) by PBS (D, H) and PEI-CD-Arg-TPEDB (A-C, E-G) (10 μM) under dark (D, H) and irradiated for 30 min (A, E), 1h (B, F) and 2h (C, G).

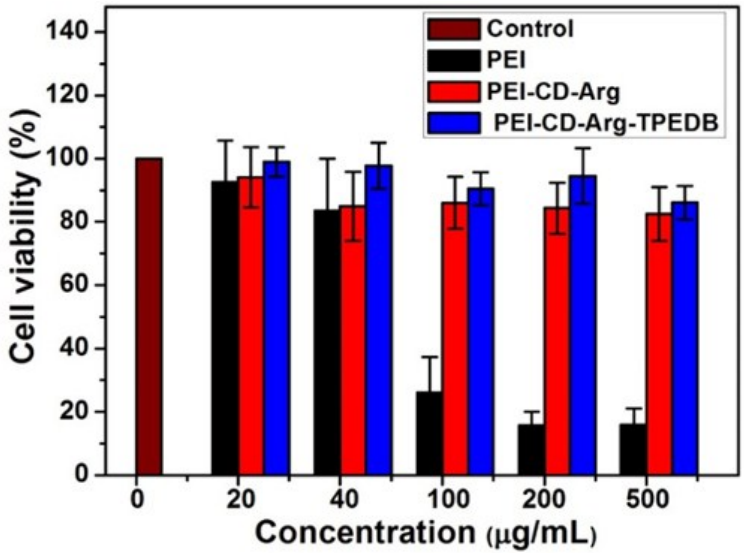


Figure S8. Cell viability of fibroblasts L929 treated with PEI, PEI-CD-Arg and PEI-CD-Arg-TPEDB (0−500 μg/mL).