Supporting Information

A novel on-demand nanoplatform for enhanced elimination of drugresistant bacteria.

Xiaosong Wei, Jie Li, Haonan Sun, Yufei Zhang, Zhuang Ma, Yayun Bai, Xinge Zhang*

Key Laboratory of Functional Polymer Materials of Ministry Education, Institute of Polymer Chemistry, College of Chemistry, Nankai University, Tianjin 300071, China

* Corresponding author:

E-mail: zhangxinge@nankai.edu.cn

Materials and procedures

Chemicals and strains

 β -D-galactose pentaacetate, Metharcyloylchloride, 1,1,1-Tris(hydroxymethyl)ethane, 2',7'-Dichlorodihydrodihydrofluorescein diacetate (DCFH-DA), Chlorin e6 (Ce6), molecular sieves and 2,4,6-Trimethoxybenzaldehyde were procured from HEOWNS (Tianjin, China). 2-Hydroxyethl methacrylate (HEMA), Ethanolamine (ETA), trifluoroacetic acid (TFA) and N-N-Diisopropylethylamine (DIPEA) were supplied by Aladdin (Shanghai, China). 1,3-Bis(tert-butoxycarbonyl)-2-methyl-2-thiopseudourea and p-Toluenesulfonic acid were purchased from Shanghai Yuanye Bio-Technology Co., Ltd (Shanghai, China) and J&k (Beijing, China), respectively. Cell Counting Kit (CCK-8) was obtained by Zoman Biotechnology Co., Ltd. (Beijing, China). All other reagents of analytical grade were purchased from Tianjin Bohua Chemical Reagent Co., Ltd (Tianjin China) and used directly without further disposition unless additional stated. Aminoglycosides resistant Pseudomonas aeruginosa (P. aeruginosa) and human lung adenocarcinoma cells (A549) were supported by College of Life Sciences (Nankai University) and Tianjin Medical University, respectively. The media for culturing bacteria which consist of Casein tryptone, yeast extract power and agar was procured from Beijing Solarbio Science & Technology Co., Ltd (Beijing, China). The media for culturing cell were composed of trypsin-EDTA solution, fetal bovine serum (FBS), DMEM, penicillin and streptomycin and supplied from Gibco (New York, America). CPADB used in this work was synthesized by our team according to a published literature.

Instruments and characterization

The ¹H NMR spectra of two kinds of block polymers and monomers were monitored by a Bruker AVANCE III 400M instrument. Deuterated solutions such as DMSO, D₂O and CDCl₃ were served according to the solubility of monomers and polymers. Fourier transform infrared spectrometry (FT-IR) acted as another useful analytical mean was recorded on TENSOR II and employed for identifying the structure of polymers. The size and zeta potential of nanoparticles in aqueous phase were identified by dynamic light scatting (DLS) which carried on a Zetasizer Nano ZS. On the other hand, the morphology of self-assembly nanoparticles and particle size in the dry state were performed by Transmission electron microscopy (TEM) on a Tecnai G2 F20 with 200 KV accelerating voltage. The ultraviolet -visible spectroscopy (UVvis) of free Ce6, blank nanoparticles and Ce6-loaded nanoparticles were taken on a UV-2550 in PBS. Additionally, UV-vis also served for the calculation of drug loading capacity (LC) and encapsulation efficiency (EE) of Ce6-loaded nanopartilces. Meanwhile, the generation of reactive oxygen species (ROS), biofilm biomass and CCK-8 results were acquired by a Freedom EVO200 at different excitation wavelength depend on their needs. The photodynamic execution of Ce6-loaded nanoparticles was carried out by Infrared laser with the irradiation of 660 nm. The images of planktonic bacteria and biofilm were collected with confocal laser scanning microscope (CLSM) on a TCS SP8. Additionally, the morphology of biofilm was also analyzed using scanning electron microscope (SEM) on a MERLIN Compact.

Synthesis of 2,4,6-trimethoxybenzylidene-1,1,1-tris (hydroxymethyl) ethane

methacrylate (TTMA)

TTMA was synthesized according to a previous protocol.¹ Briefly, the mixture solution composed of 2,4,6-trimethoxylbenzadehyde (550 mg, 2.8 mmol) and 1,1,1tris (hydroxymethyl) ethane (950 mg, 8 mmol) was kept stirring in 40 mL of anhydrous THF. Then, *p*-toluenesulfonic acid (65 mg, 0.375mmol) and 5 Å molecular sieves (20 g) were added. The reaction was executed at 40°C for overnight. Triethylamine (1 mL) was added to neutralize acid, followed by removing molecular sieves and solvent, the crude product was dissolved in 100 mL of DCM and washed with Tris buffer (100 mM, pH 8.0) for three times. The target product was collected by solvent evaporation. In the second stage, the product previously received (500 mg, 1.7 mmol) and TEA (530 µL, 3.8 mmol) were dissolved in 30 mL of DCM, followed by adding 250 µL of methacryloyl chloride dropwise at 0°C. The reaction was transferred into room temperature and kept stirring for overnight. When the reaction accomplished, the obtained mixture was treated with Tris buffer (100 mM, pH 8.0), and the light yellow oily liquid was acquired by removing solvent. Finally, the crude product was transformed into a white solid by purification with silica gel chromatography (EtOAc/hexane, v/v = 1:4). Yield: 51%. ¹H NMR (400 MHz, CDCl₃): δ 0.82 (s, 3H), 1.96 (s, 3H), 3.63 (d, 2H), 3.82 (s, 9H), 4.04 (s, 2H), 4.62 (s, 2H), 5.56 (s, 1H), 5.98 (s, 1H), 6.11 (s, 3H).

Synthesis of 2-O-acryloyloxyethyl-(2,3,4,5-tetra-O-ace-tyl-β–D-galactopyranoside (AcGEA)

AcGEA was synthesized following a previous technique supplied from Murakami et

al.² Raw materials such as β-D-galactose pentaacetate, metharcyloylchloride and boron trifluoride diethyl etherate were added at a certain doses step by step, and it was worth mentioning that the reaction system was controlled in a anhydrous state and nitrogen atmosphere. After the reaction accomplished, yellowish oily liquid was collected as crude product by washing with saturated sodium bicarbonate and saline solution for three times. The ultimate product was acquired by purifying with silica column chromatography (petroleum ether/ethyl acetate = 3:1).Yield: 52%. ¹H NMR (400mHz, DMSO-*d*₆): δ 1.90-2.05 (q, 12H), 2.15 (s, 1H), 3,7-3.8 (m, 2H), 3.9-4.0 (m, 2H), 4.0-4.1 (m, 2H), 4.15-4.25 (d, 1H), 4.75 (d, 1H), 4.95 (t, 1H), 5.15 (t, 1H), 5.25 (d, 1H), 5.7 (s, 1H), 6.05 (s, 1H).

Synthesis of di-(Boc-protected) amino ethyl acrylate

The process for preparing di-(Boc-protected) amino ethyl acrylate was accomplished in two steps. The first step was implemented according to a reference published by Yang et al.³ Firstly, ethanolamine (ETA) (685 μ L, 11.4 mmol) was dissolved in 10 mL of anhydrous DCM, followed by andding N,-N-diisopropylethylamine (DIPEA) (3.0 mL,17.7 mmol). The homogeneous mixed solution was acquired by stirring for 10 mins, followed by adding 1,3-bis (tertbutoxycarbonyl)-2-methyl-2-thiopseudourea (1.1 g, 5.8 mmol). The reaction was performed under stirring for overnight at 25°C. Until the reaction finished, the obtained solution was disposed through nitrogen sparing for 1 h. Consequently, methyl mercaptan (MeSH), a by-product, was eliminated in such a process. The crude product was collected by removing solvent using rotary evaporation. Finally, target product, a white power, was acquired by through a silica column (hexane/ ethyl acetate = 3:1). Yield: 80%. ¹H NMR (400 mHz, CDCl₃): δ 1.5 (s, 18H), 3.57 (s, 2H), 3.78 (s, 2H), 8.68 (s, 1H), 11.45 (s, 1H).

As for the second step, the precursor molecules obtained from the first step (500 mg, 1.65 mmol) was absolutely dispersed in 30 mL of dry DCM. Pyridine (200 μ L, 2.47 mmol), a deacid reagent, was added into the preceding solution. Methacryloyl chloride (190 μ L, 1.98 mmol) was added in a small amount every time at 0°C. Once the operation was completed, the mixed solution was allowed to transfer to room temperature and kept stirring for overnight. The organic solution was washed with saturated sodium bicarbonate solution for three times and concentrated to give crude product until the reaction accomplished. The target product, a white waxy solid, was obtained by silica gel column chromatography (hexane/hexane/ethyl acetate = 5:1). Yield: 55%. ¹H NMR (400 mHz, CDCl₃): δ 1.2-1.7 (t, 18H), 1.96 (s, 3H), 3.75 (s, 2H), 4.28 (s, 2H), 5.59 (s, 1H), 6.16 (s, 1H), 8.65 (s, 1H), 11.48 (s, 1H).

Synthesis of p(GEA-*b*-TTMA)

The galactose-functionalized and guanidine-functionalized block polymers were all synthesized by reversible addition-fragmentation chain transfer (RAFT) equipped with AIBN as the initiator and CPADB as the chain transfer agent, respectively. The detailed procedure was described as follows. Monomer AcGEA (345 mg, 0.75 mmol), AIBN (4.1 mg, 0.025 mmol), and CPADB (13.95 mg, 0.05 mmol) were added into a dry flask comprising 1.5 mL of anhydrous DMF. The air was excluded by bubbling nitrogen for 40 mins, and this device was allowed to transfer to 70°C to initiate the polymerization. The reaction consumed 24 h to accomplish. After quenching in an ice

bath, the macroinitiator, a pink power, was acquired by precipitating in diethyl ether for three times. To confirm the molecular weight of p(AcGEA), the macroinitiator was determined by ¹H NMR and GPC, respectively. To implement the second step reaction, p(AcGEA), another monomer TTMA, and AIBN were added in a certain amount and dispersed in 2.5 mL of anhydrous DMF. The polymerization was achieved, which performed the same operation as described above. p(AcGEA-b-TTMA), a white solid was collected by ether treatment and measured by ¹H NMR to identify a success message. Finally, the galactose-functionalized block polymer could be obtained in the presence of hydrazine hydrate solution (HHA) for taking off OAc groups. Briefly, an amount of 200 mg of p(AcGEA-b-TTMA) was dissolved in 1 mL of DMSO, followed by adding 245 µL of HHA solution dropwise. The mixture solution was kept stirring for overnight at 25°C until the reaction completed. Termination reaction was happened by adding 2 mL of acetone. Lastly, the obtained solution was allowed to transfer to a dialysis bag for removing organic solvent and impurities. Consequently, p(GEA-b-TTMA) like snowflake was collected by lyophilization and measured by ¹H NMR to identify a success message. Other galactose-functionalized polymers were acquired under the same operation condition but by changing the amount of monomer AcGEA. It was worth mentioning that three galactose-functionalized polymers were named as $p(AcGEA_{15}-b-TTMA_{40})$, p(AcGEA₂₅-*b*-TTMA₄₀) and p(AcGEA₃₅-*b*-TTMA₄₀), respectively. The figure in these denominations stands for theoretical molar weight and the following denomination was same as said above.

Synthesis of p(Gua-*b*-TTMA)

The detailed operation was described as follows. Monomer (1.11 g, 3 mmol), CPADB (13.95 mg, 0.05 mmol) and AIBN (4.1 mg, 0.025 mmol) were added to a reaction vessel and dispersed absolutely in 3 mL of dry DMF under stirring. Such a routine as nitrogen sparing and temperature increasing was executed to start polymerization. The reaction was terminated after 24 h through exposing to the air and placing into icy water. The mixture solution was dealt with cold hexane for three times to give a light pink power. The macroinitiator was subsequently detected by ¹H NMR and GPC to supply with composition analysis and molecular weight.

The method for Boc-deprotection was implemented adapted from a copywriting reported previously.⁴ Firstly, 100 mg of p(Boc-protected Gua) was dissolved in 10 mL of DCM, followed by adding 1 mL of TFA. The mixture was left to stir overnight at room temperature to take off Boc group. Once the reaction was completed, the residual TFA was removed by rotary evaporation. The target product was acquired by dialyzing against distilled water in a dialysis bag (3500 MWCO) for 48 h. The method for preparing p(Gua-*b*-TTMA) was in accord with p(AcGEA-*b*-TTMA) except for solvent. TTMA, p(Gua) and AIBN was dispersed in DMSO at a certain dose, and the reaction was kept on running in a sealed and oxygen-free state for 24 h. After quenching the reaction, the crude product was purifying by dialysis (3500 MWCO) for 2 days. Lastly, p(Gua-*b*-TTMA), a light yellow power, was gained by lyophilization and analyzed by ¹H NMR and GPC.

Preparation and characterization multi-functional nanoparticles and Ce6-loaded

nanoparticles

No matter blank nanoparticles or Ce6-loaded nanoparticles were all established by a nanoprecipitation technique. Typically, 4 mg of copolymer and 0.5 mg Ce6 were added into 4.5 mL of ultrapure water slowly. The obtained Ce6-loaded nanoparticles were dialyzed against distilled water for 12 h. After that, Ce6-loaded nanoparticle suspension was centrifuged with a speed of 1.2×10^4 for 40 min and lyophilization. To test the encapsulation efficiency (EE) and loading capacity (LC) of three nanoparticles, a UV-vis spectrometer was introduced to measure the absorbance at 660 nm and the value of EE and LC were calculated from the following equations:

$$EE\% = \frac{\text{total Ce6 - free Ce6}}{\text{total Ce6}} \times 100$$

$$\text{total Ce6 - free Ce6}$$
(1)

$$LC\% = \frac{\text{total CCO' HCC CCO}}{\text{total mass of NPs}} \times 100$$
(2)

Each sample was tested for three times and all data were obtained in triplicate.

As for other proportional drug loaded nanoparticles, everything stayed the same, except that $p(GEA_{15}-b-TTMA_{40})$ was replaced with $p(GEA_{25}-b-TTMA_{40})$ or $p(GEA_{35}-b-TTMA_{40})$. It's important to note that the drug loaded nanoparticles consisted of $p(GEA_{15}-b-TTMA_{40})$ and $p(Gua_{60}-b-TTMA_{40})$ was denoted as NPs-1@Ce6, $p(GEA_{25}-b-TTMA_{40})$ and $p(Gua_{60}-b-TTMA_{40})$ was denoted as NPs-2@Ce6, $p(GEA_{35}-b-TTMA_{40})$ and $p(Gua_{60}-b-TTMA_{40})$ was denoted as NPs-2@Ce6. The blank nanoparticles consisted of $p(GEA_{15}-b-TTMA_{40})$ and $p(Gua_{60}-b-TTMA_{40})$ was denoted as NPs-1, $p(GEA_{25}-b-TTMA_{40})$ and $p(Gua_{60}-b-TTMA_{40})$ was denoted as NPs-2, $p(GEA_{35}-b-TTMA_{40})$ and $p(Gua_{60}-b-TTMA_{40})$ was denoted as NPs-3. The basic information of nanoparticles such as particle size, size distribution, zeta potential and morphology could be supplied by DLS and TEM instruments. In addition, the confirmation message of drug loading capacity and encapsulation efficiency was able to be acquired by UV-vis with the support of standard curve.

Morphology and size changes of nanoparticles in response to acetal hydrolysis

The hydrolyzed behavior of nanoparticles induced by acid was investigated using DLS and TEM devices. Typically, the nanoparticle solution was prepared freshly and divided in half equally which subsequently treated with equivalent PBS (pH 7.4) and acetate buffer (pH 5.5), respectively. The samples were disposed with stirring mildly at 37°C and extracted at desired time intervals for monitoring the changes. Beyond that, TEM acted another mean was also employed to indicate the change in morphology.

In vitro ROS generation and detection of various samples by PDT

In this trial, DCFH-DA was introduced as a probe to reflect the generation of ROS. Firstly, 1 mL of nanoparticle solution (NPs-1@Ce6, NPs-2@Ce6, NPs-3@Ce6 or free Ce6) was mixed with 1 mL of DCFH-DA (50 µmol/L). The mixture solution was transferred into a 24-well plate and incubated for 10 mins in the dark and then exposed to a 660 nm light. Finally, the supernatants came from the certain point (1 min, 2 min, 3 min, 4 min and 5 min) were taken out and then measured by a microplate reader with excitation at 488 nm and emission at 525 nm. It was important to mention that all data was given in triplicate. Interestingly, the color images that correspond to test data were achieved at the same time.

Interaction between P. aeruginosa and nanoparticles

The interaction between *P. aeruginosa* and nanoparticles was determined by ultraviolet spectrophotometer and confocal laser scanning microscope (CLSM). At

first, the bacteria was revived in LB liquid and cultivated in a shaker with a constant temperature. After 12 h cultivation, the obtained bacterial suspension was diluted with fresh fluid medium until the concentration reached 10⁹ CFU/mL. To explore the effect of time on bacterial capture, the bacterial suspension was centrifuged for 5 mins and the obtained sediments were subsequently dispersed with nanoparticles again. Finally, equivalent supernatants supplied from different samples were collected at preconceived time to measure the OD₆₀₀. On the other hand, the effect of concentration of nanoparticles on bacterial capture was similar as the procedure described above. In briefly, the nanoparticle solution was diluted into varied concentration gradient ranging from 1000 µg/mL to 62.5 µg/mL and co-cultivated with bacterial precipitation for the same period. After cultivating silently on the shaker, the equal volume of supernatants was taken out for the bacterial capture assay. In case of visual analysis, CLSM was introduced as a praised technique in this experiment for providing additional support information. The fresh bacterial sediments were resuspended with nanoparticles (1 mg/mL) and then incubated in a shaker at 37°C for appropriate time. Subsequently, each sample was stained with acridine orange (AO) for 15 mins in the dark. Until the bacteria was labeled, sterile PBS was added to wash the dye. Furthermore, a control group with only PBS was executed during this process.

Antimicrobial properties of Ce6-loaded nanoparticles by PDT

To evaluate the antibacterial activity of Ce6-loaded nanoparticles, drug-resistant *P. aeruginosa* was selected as the participant for this assessment test. Briefly, the

bacterial suspension cultured for overnight was diluted with acetate buffer to a concentration of 1×10^9 CFU/mL and managed with centrifugation for providing the pure bacteria. Subsequently, the pure bacteria was redispersed with Ce6-loaded nanoparticle solutions and co-incubated for 4 h in the dark. At last, each sample was exposed under a 660 nm laser for different time (0 min, 1 min, 3 min and 5 min). 30 μ L of the diluted solution (10000 \times) from each sample was seeded on an LB agar plate, followed by growing at 37°C for 12 h to form bacterial colonies.

The above was the contents of the colony count method, however, the bacterial Live/Dead staining method was also performed which could present the intuitionistic images for antibacterial assay. Activities in the early stage of this experiment were similar as the above described so that it was no need to say more. Consequently, after treatment, each sample was stained with AO and EB for 15 mins in the dark, followed by washing with sterile PBS for three times. At the end of this operation, each sample was observed using a Leica TCS SP8 CLSM (excitation at 488 nm). By the way, green fluorescence was the representation of lived bacteria and the dead bacteria present the red fluorescence.

P. aeruginosa biofilm formation and dispersion assay

To investigate whether or not Ce6-loaded nanoparticles have impact on the dispersing of biofilms by PDT, this experimental study was performed in two aspects Firstly, the seed from monocolony of drug-resistant *P. aeruginosa* was transferred to fresh liquid medium and kept in an incubator for overnight. Subsequently, 200 μ L of the diluted suspension with an OD₆₀₀ of 0.01 was introduced to a 96-well plate and kept in a

stationary state for forming mature biofilms. To study the influence of time on biofilm dispersion, three sets of Ce6-loaded nanoparticles was prepared in advance. The liquid medium was replaced with nanoparticle solutions, followed by co-incubating in the dark for 4 h. Next, three experimental groups were treated with a 660 nm light source for the appointed intervals and the PBS group was taken as a control. Finally, the various samples were washed with PBS, fixed with methyl alcohol and stained with crystal violet within the required time. The message of quantitative analysis was provided by a microplate reader and the images correspond to quantitative value was produced at the same time. On the other hand, the effect of concentration on biofilm dispersion was performed as the same steps except that Ce6-loaded nanoparticles were diluted into various concentrations ahead of schedule and exposed under a 660 nm laser for the equal time period. At last, the results including data and image were also acquired by reading a plate.

Morphological analysis of biofilms by CLSM and SEM

To intuitively observe the morphology of biofilms after treating with various groups, CLSM and SEM were chosen for recording the image information. For CLSM images, the cover slips treated with high pressure were selected as a surface for biofilm formation. Briefly, the bacterial dilutions with an OD_{600} of 0.01 and cover slips were co-incubated in a 12-well plate at 37°C without moving. Until the mature biofilms formed, the residual medium was removed and then substituted with nanoparticle solutions and sterile PBS, respectively. After irradiation with a 660 nm light for a certain period, the residual biofilm were washed with PBS mildly for three times and

then fixed with 4%wt glutaraldehyde solution. Subsequently, the fluorescent dyes such as EB and FITC-ConA were introduced for labeling bacteria and polysaccharide, respectively. After staining for 15 mins, the samples were washed with PBS for removing the residual dye. Eventually, the cover slips were put on the microscope slides and photographed in three dimensions. As for SEM analysis, the microscope slides were cut into many small pieces which could be put into a 12-well plate. Afterward, no matter growth conditions of biofilms or experimental treatment for biofilms was executed in the same as shown above. However, the fluorescent dyes were absent in the post processing. Until the biofilms were fixed by glutaraldehyde solution, the samples were dehydrated with graded ethanol (30%wt, 50%wt, 70%wt, 90%wt and absolute ethanol). It was worth mentioning that the samples need to be treated with spray-gold before imaging in order that the bacteria could be endowed with electrical conductivity.

Biofilm permeation assay

To investigate biofilm permeation behavior of nanoparticles, Nile red was introduced with the purpose of path tracking. In brief, the mother liquors of Nile red and polymer with a concentration of 10 mg/mL were prepared in advance. Afterward, Nile red and polymer were mixed evenly with the weight ratio of 1:2, followed by injecting ultrapure water. Until the nanoparticles formed entirely, the resulting solution was transferred to a dialysis bag (3500 MWCO) and dialyzed against ultrapure water for 48 h. The detailed steps for biofilms formation was performed as mentioned earlier. Once the mature biofilms were obtained, cleaning with PBS was carried out for

removing bacterial excretion. Subsequently, the biofilms were immersed in Nile redloaded nanoparticle solutions for 4 h, followed by washing with PBS again. Ultimately, the biofilms stained with FITC-ConA were examined under the CLSM with the excitation wavelength at 488 nm (FITC) and 561 nm (Nile red), respectively.

Assessment of cytotoxicity of Ce6-loaded nanoparticles

The A549 cells were used to evaluate the cytotoxicity of nanoparticles. Primarily, the A549 cells stored in liquid nitrogen were resuscitated and cultured in DMEM supported with 10% FBS and 1% mixture solution of penicillin and streptomycin. Afterward, the cells were inoculated to a 96-well plate and maintained in an incubator filled with 5% CO₂ at 37°C to grow adhering to the wall. Diluted nanoparticles in grace concentrations (62.5 μ g/mL to 1000 μ g/mL) were added into the cells and cultured for further 24 h. Finally, CCK-8 was added to all the samples for another 2 h and monitored whenever necessary. The result was given by recording at 450 nm and cells treated with nothing were accepted as the control group. Average values of three data sets were calculated for giving cytotoxicity analysis.



Figure S1. The synthetic route of two amphipathic block copolymer by reversible

addition-fragmentation chain transfer (RAFT) polymerization.

Samples	feeding ^a	product ^b	Mn ^b	Mn ^c	Mw/Mn ^c
p(AcGEA ₁₅ - <i>b</i> -TTMA ₄₀)	15/40	10/14	9.7	7.7	1.10
p(AcGEA ₂₅ - <i>b</i> -TTMA ₄₀)	25/40	20/15	14.3	12.9	1.35
p(AcGEA ₃₅ -b-TTMA ₄₀)	35/40	27/15	21.9	13.4	1.32
$p(GEA_{15}-b-TTMA_{40})$	15/40	8/14	8.3		
p(GEA ₂₅ -b-TTMA ₄₀)	25/40	15/15	11.8		
$p(GEA_{35}-b-TTMA_{40})$	35/40	18/15	14.5		
$_$ p(Gua ₆₀ - <i>b</i> -TTMA ₄₀)	60/40	45/20	14.4	22.6	1.52

Table 1. Characteristics of two sets of block polymers.

^aThe theoretical values. ^bCalculated from ¹H NMR. ^cDetermined by GPC.

Table 2. Characteristics of Constructed Nanoparticles.

Samples	Ce6/Polymer	Dh	PDI^b	Zeta	EE	LC
	а	$(nm)^b$		$(mv)^b$	(%) ^c	(%) ^c
Blank NPs-1	0/1	144.9 ± 1.415	0.104	12.0 ± 0.781		
Blank NPs-2	0/1	143.8 ± 0.416	0.156	11.1 ± 0.153		
Blank NPs-3	0/1	143.4 ± 2.066	0.141	8.29 ± 0.459		
NPs-1@Ce6	0.125/1	175.8 ± 1.514	0.108	-8.6 ± 0.146	68.9	7.9
NPs-2@Ce6	0.125/1	164.1 ± 2.425	0.111	-11.5 ± 0.896	71.7	8.2
NPs-3@Ce6	0.125/1	159.2 ± 3.301	0.077	-12.5 ± 0.977	75.5	8.6



Figure S2. ¹H NMR spectrum of AcGEA in DMSO- $d_{6.}$



Figure S3. ¹H NMR spectrum of TTMA in CDCl₃.



Figure S4. ¹H NMR spectrum of Boc-Gua in CDCl₃.



Figure S5. ¹H NMR spectrum of p(GEA-*b*-TTMA) in DMSO-*d*₆.



Figure S6. ¹H NMR spectrum of p(Gua-*b*-TTMA) in DMSO-*d*₆.



Figure S7. FT-IR spectra of p(Gua-*b*-TTMA) and p(GEA-*b*-TTMA).



Figure S8. UV-vis spectra of blank nanoparticles, free Ce6 and Ce6-loaded nanoparticles in ultrapure water.



Figure S9. Photographs of colonies and responding amount of *P. aeruginosa* on the LB agar plates treated by acetate buffer and blank nanoparticles, respectively.



Figure S10. 3D CLSM photographs of biofilms severally managed with free Ce6 (A). SEM images of *P* .*aeruginosa* biofilm managed with free Ce6 (B). Quantitative analysis of biofilm eradication treated with free Ce6 during 10 min (C). Quantitative analysis of biofilm eradication treated with free Ce6 with a serial concentration (5-80 μ g/mL) for 10 mins using crystal violet staining (D).



Figure S11. Corresponding quantity images of biofilm eradication from free Ce6 treated with serial concentrations or different time.



Figure S12. Cell viability of A549 after co-cultivated with different doses of nanoparticles for 24 h.

References

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