Supplementary Information

Engineered Polymer Nanoparticles with Unprecedented Antimicrobial Efficacy And Therapeutic Indices Against Multidrug-Resistant Bacteria and Biofilms

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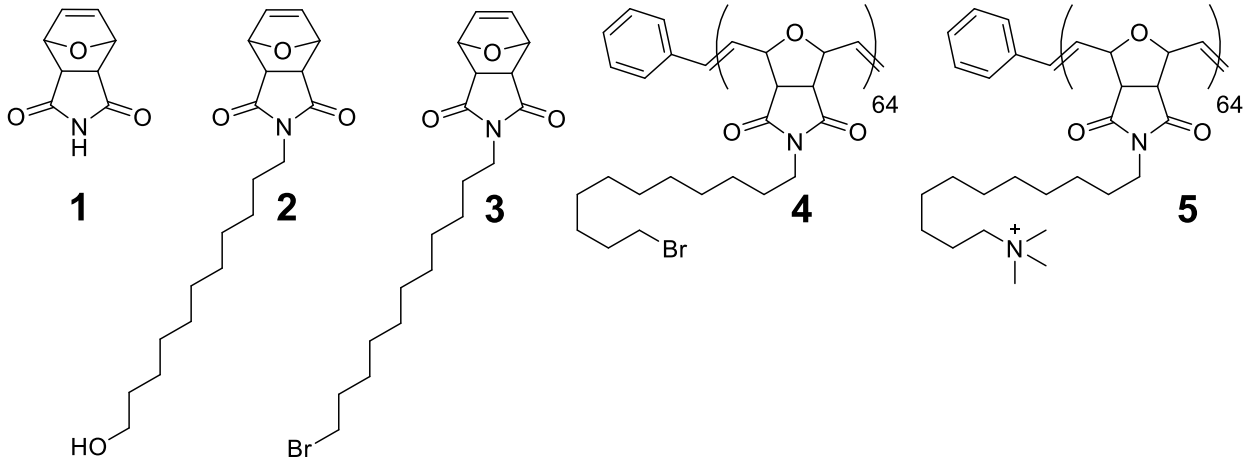
S1. Materials

All chemicals and solvents for syntheses were purchased from Fisher Scientific and Sigma-Aldrich, and used without further purification, unless otherwise stated. The chemicals were used as received. Dichloromethane (DCM) and tetrahydrofuran (THF) used as a solvent for chemical synthesis and dried per standard procedures. All reagents/materials were purchased from Fisher Scientific and used as received. NIH-3T3 cells (ATCC CRL-1658) were purchased from ATCC. Dulbecco's Modified Eagle's Medium (DMEM) (DMEM; ATCC 30-2002) and fetal bovine serum (Fisher Scientific, SH3007103) were used in cell culture. Pierce LDH Cytotoxicity Assay Kit was purchased from Fisher Scientific. The yields of the compounds reported here refer to the yields of spectroscopically pure compounds after purification. ^{1}H NMR spectra were recorded at 400 MHz on a Bruker AVANCE 400 machine.

S2. Synthesis

Oxanorbornene Monomer Synthesis:

Note: Generation of C2 and C6-bridged polyoxanorbornene polymers can be successfully made using the same procedures used to generate C11-bridged polymers however replacing 11-bromoundecanol with bromoethanol or 6-bromohexanol, respectively.



Synthesis of 1: In a pressure tube, furan (4.5ml, 61.7mmol, 1.5eq) and maleimide (4.0g, 41.1mmol, 1.0eq) were added in addition to 5ml of diethyl ether. The tube was sealed and heated at 100^{o}C overnight. Afterwards, the pressure tube was cooled to r.t. and the formed solid was removed, filtered, and washed with copious amounts of diethyl ether to isolate **1** as a white solid (95% yield) and was used without further purification. ^{1}H NMR (400MHz, MeOD) 11.14 (s, 1H), 6.52 (s, 2H), 5.12 (s, 2H), 2.85 (s, 2H).

Synthesis of 2: To a 250ml round bottom flask equipped with a stir bar was added 60ml of DMF. Next, **1** (3.76g, 22.7mmol, 1.0eq) was added along with potassium carbonate (12.59g, 91.1mmol, 4.0eq). The reaction mixture was heated at 50^{o}C for five minutes. Finally, potassium iodide (0.68g, 4.5mmol, 0.2eq) and 11-bromoundecanol (6.00g, 23.90mmol, 1.05eq) were added and stirred at 50^{o}C overnight. Afterwards, the reaction mixture was cooled to room temperature, diluted to 150ml with ethyl acetate and washed with water (7x, 50ml) and brine (1x, 50ml). The organic layer was dried with sodium sulfate, filtered, and rotovaped to yield **2**. **2** was purified by sonication of the rotovaped solid in hexanes and filtered (82% yield). ^{1}H NMR (400MHz, $CDCl_3$) 6.44 (s, 2H), 5.19 (s, 2H), 3.55, (t, 2H), 3.49 (t, 2H), 2.79 (s, 2H), 1.9 (s, 1H), 1.39 (m, 4H), 1.2 (m, 14H).

Synthesis of 3: To a 250ml round bottom flask equipped with a stir bar was added **2** (2.64g, 7.87mmol, 1.0eq). Next, DCM (100ml) was added along with tetrabromomethane (3.13g, 9.44mmol, 1.2eq). The reaction was cooled to 0^{o}C using an ice bath. Finally, triphenylphosphine was added in portions (2.47g, 9.44mmol, 1.2eq) and allowed to stir for three hours. Afterwards, the reaction mixture was rotovaped and ethyl ether was added (200ml) and placed in the freezer for 2 hours to precipitate out triphenylphosphine oxide. The reaction mixture was filtered, and the filtrate was rotovaped. Column chromatography was performed to yield **3**, a white solid (79%

yield). ^{1}H NMR (400MHz, $CDCl_3$) 6.51 (s, 2H), 5.27 (s, 2H), 3.45 (t, 2H), 3.41 (t, 2H), 2.83 (s, 2H), 1.85 (q, 2H), 1.55 (q, 2H), 1.41 (q, 2H), 1.29 (m, 12H).

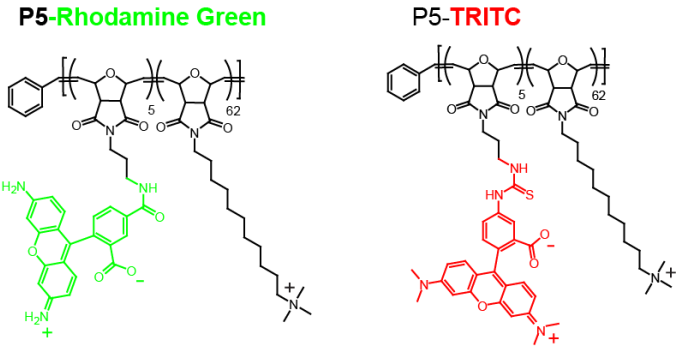
Oxanorbornene Polymer Synthesis:

Synthesis of 4: To a 10ml pear-shaped air-free flask equipped with a stir bar was added **3** (800mg, 2.0mmol, 1.0eq) and 4ml of DCM. In a separate 10ml pear-shaped air-free flask was added Grubbs 3rd generation catalyst[1] (35.4mg, 0.04mmol, 0.02eq) and 1ml DCM. Both flasks were sealed with septa and attached to a schlenk nitrogen/vaccum line. Both flasks were freeze-pump-thawed three times. After thawing, Grubbs 3rd generation catalyst was syringed out and quickly added to the flask containing **3** and allowed to react for 10min. After the allotted time, ethyl vinyl ether (200 μL) was added and allowed to stir for 15 minutes. Afterwards, the reaction was diluted to two times the volume and precipitated into a heavily stirred solution of hexane (300ml). The precipitated polymer was filtered and dissolved into tetrahydrofuran (THF). The polymer was precipitated again into hexane and filtered to yield **4**. MW = 25,698, PDI = 1.04 (determined by THF-GPC using a Polystyrene calibration curve) ^{1}H NMR (400MHz, $CDCl_3$) 6.0 (br, 1H), 5.7 (br, 1H), 4.95 (br, 1H), 4.4 (br, 1H), 3.4 (br, 2H), 3.25 (br, 2H), 1.79 (q, 2H), 1.5 (br, 2H), 1.34 (br, 2H), 1.2 (br, 14H).

Synthesis of 5 Quaternary Ammonium Polymers: To generate the library of quaternary ammonium poly(oxanorborneneimides), **4** (50mg) was added to 20ml vials equipped with a stir bar. Next, excess of the necessary tertiary amines was added (10ml of a 1M trimethylamine solution in THF, all other amines were 200mg) to the vial and purged with nitrogen. First stage of the reactions involved stirring for 30 minutes at 80^{o}C. The polymers precipitated during this time. Half of the THF was evaporated and replaced with methanol which re-dissolved the polymers. The reaction was allowed to proceed overnight at 50^{o}C. Afterwards, the solvent was completely evaporated, washed with hexane 2 times, and dissolved into a minimal amount of water. The polymers were added to 10,000 MWCO dialysis membranes and allowed to stir for 3 days, changing the water periodically. The polymers were filtered through PES syringe filters and freeze-dried to yield all the respective quaternary ammonium polymers **5**. NMR indicated conversion into the desired quaternary ammonium salts.

S3. FRET PNP Formation

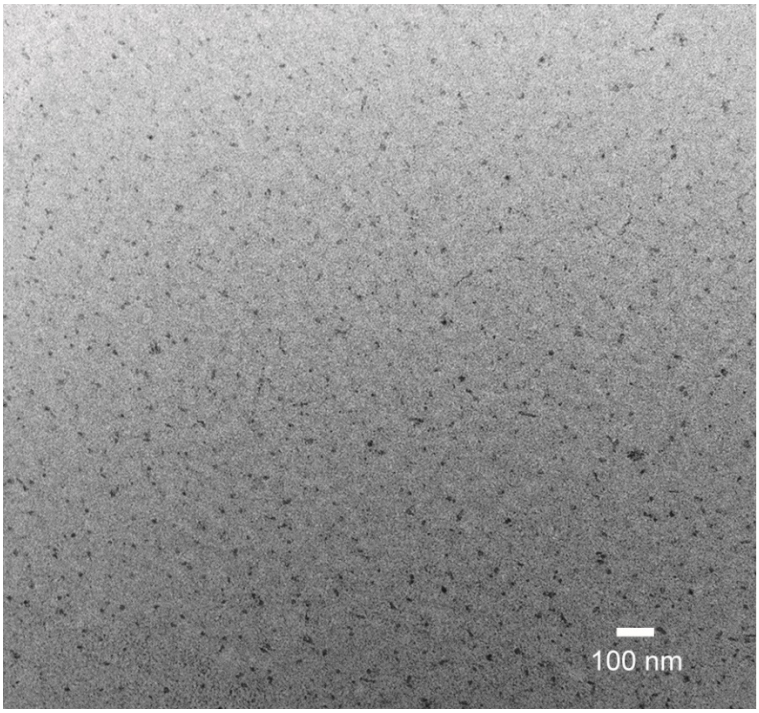
FRET PNPs were generated using the **P5** polymer scaffold, labelled either with donor Rhodamine Green or acceptor TRITC (Functionalized by incorporating a boc-protected amino monomer during the polymerization, followed by purification using a 10,000 MWCO dialysis bag). Keeping **P5-Rhodamine Green**'s concentration constant at ~ 1.6μM in 2ml Eppendorf tubes, increasing concentrations of **P5-TRITC** in MQ water was added and the tubes were sonicated for one minute and allowed to stand for one hour. The solutions were then transferred to a 96-well microplate and the total emission spectrum of both **P5** derivatives were recorded on a SpectroMax M5 microplate reader (Molecular Device) using 480nm as the excitation wavelength (480nm was selected so that only **P5-Rhodamine Green** would be excited).



Supplementary Figure S1: Molecular structures of P5 polymer derivatives used for FRET PNP studies.

S4. TEM characterization of polymeric nanoparticles (PNPs)

TEM samples of polymers were prepared by placing one drop of the desired solution (10 μM) on to a 300-mesh Cu grid-coated with carbon film. These samples were analyzed and photographed using JEOL CX-100 electron microscopy. The average diameter of P5 nanoparticles is 12.7 ± 2.7 nm.



Supplementary Figure S2: TEM image of Polymer nanoparticles.

S5. Determination of MICs, Hemolysis and TNF-alpha secretion

Determination of Antimicrobial Activities of Cationic Polymers: Bacteria were cultured in LB medium at 37 ˚C and 275 rpm until stationary phase. The cultures were then harvested by centrifugation and washed with 0.85% sodium chloride solution for three times.[5] Concentrations of resuspended bacterial solution were determined by optical density measured at 600 nm. M9 medium was used to make dilutions of bacterial solution to a concentration of 1×10^6 cfu/mL. A volume of 50 μL of these solutions was added into a 96-well plate and mixed with 50 μL of polymer solutions in M9, giving a final bacterial concentration of 5×10^5 cfu/mL. Polymer concentration varied in half fold per a standard protocol, ranging from 1024 to 4 nM. A growth control group without polymers and a sterile control group with only growth medium were carried out at the same time. Incubation of the polymers with bacteria was performed for 16 hours. Cultures were performed in triplicates, and at least two independent experiments were repeated on different days. The MIC is defined as the lowest concentration of polymer that inhibits visible growth as observed with the unaided eye.[2]

Determination of Hemolysis of Cationic Polymers: We used the previously established protocol to conduct hemolysis assays on Red Blood Cells.[3] Citrate-stabilized human whole blood (pooled, mixed gender) was purchased from Bioreclamation LLC, NY and processed as soon as received. 10 mL of phosphate buffered saline (PBS) was added to the blood and centrifuged at 5000 r.pm. for 5 minutes. The supernatant was carefully discarded and the red blood cells (RBCs) were dispersed in 10 mL of PBS. This step was repeated at least five times. The purified RBCs were diluted in 10 mL of PBS and kept on ice during the sample preparation. 0.1 mL of RBC solution was added to 0.4 mL of polymer solution in PBS in a 1.5 mL centrifuge tube (Fisher) and mixed gently by pipetting. RBCs incubated with PBS and water was used as negative and positive controls, respectively. All polymer samples as well as controls were prepared in triplicate. The mixture was incubated at 37 ˚C for 30 minutes while shaking at 150 r.p.m. After incubation period, the solution was centrifuged at 4000 r.p.m. for 5 minutes and 100 μL of supernatant was transferred to a 96-well plate. The absorbance value of the supernatant was measured at 570 nm using a microplate reader (SpectraMax M2, Molecular devices) with absorbance at 655 nm as a reference. The percent hemolysis was calculated using the following formula:

Macrophage cell studies and TNF-alpha secretion – RAW 264.7 macrophage cell line was purchased from American Type Culture Collection (ATCC, Manassas, VA). Roswell Park Memorial Institute media (RPMI 1640) supplemented with 10% fetal bovine serum, 1% antibiotics (100 μg/ml penicillin and 100 μg/ml streptomycin) and sodium pyruvate, was used for cell culture. The cells were incubated at 37 °C under a humidified atmosphere of 5% CO_2. The cells were cultured once every four days under the above-mentioned conditions.

Polymer Nanoparticles and LPS treatment – These studies were conducted as per the previously reported protocols.[4] Briefly, to evaluate the effect of polymer on the immune system, 1.0×10^5 of RAW 264.7 cells were cultured in a 24-well plate for 24 h. Then, cells were washed once with cold PBS and treated with different concentration of polymer for 3 h or 24h. The macrophage with 100ng/ml of lipopolysaccharide were the positive control. At the end of incubation, culture media was collected for TNF-α level measurement by ELISA (R&D Systems, MN, USA). Experiments were performed in triplicate.

S6. Propidium Iodide staining and resistance development assay

Propidium Iodide Staining Assay: *E. coli* CD-2, *P. Aeruginosa* ATCC19660 and MRSA CD-489 (1×10^8 cfu/mL) were incubated with 1 μM P5 PNPs in M9 media at 37 ˚C and 275 rpm for 3 h. The bacteria solutions were then mixed with PI (2 μM) and incubated for 30 min in dark. Five microliters of the samples were placed on a glass slide with a glass coverslip and observed with a confocal laser scanning microscopy, Zeiss 510 (Carl Zeiss, Jena, Germany) using a 543-nm excitation wavelength.

Resistance Development: *E. coli* CD-2 was inoculated in M9 medium with 85 nM (2/3 of 128 nM, MIC) of P5 PNPs at 37 ˚C and 275 rpm for 16 h (~ 64 bacterial generations for 1 serial passage). The culture was then harvested and tested for MIC as describe above. *E. coli* CD-2 was cultured without polymer as well every time as a control for comparison of MICs. In the case of P5 PNPs, 20 serial passages were performed giving ~ 1,300 generations.

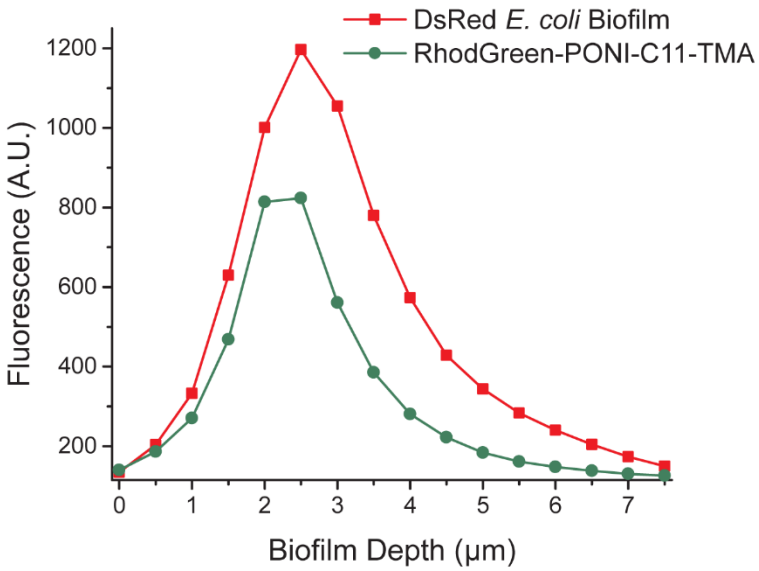
S7. Biofilm formation and penetration studies

Biofilm Formation and treatment: Bacteria were inoculated in lysogeny broth (LB) medium at 37 ˚C until stationary phase. The cultures were then harvested by centrifugation and washed with 0.85% sodium chloride solution three times. Concentrations of resuspended bacterial solution were determined by optical density measured at 600 nm. Seeding solutions were then made in M9 medium to reach an OD_{600} of 0.1. A 100 μL amount of the seeding solutions was added to each well of the 96-well microplate. The plates were covered and incubated at room temperature under static conditions for 1 day. The stock solution of polymers was then diluted to the desired level and incubated with the biofilms for 3 hours at 37˚C. Biofilms were washed with phosphate buffer saline (PBS) three times and viability was determined using an Alamar Blue assay. Minimal M9 medium without bacteria was used as a negative control.[5]

Biofilm-3T3 Fibroblast Cell Coculture: was performed using the previously reported protocol.[5]A total of 20000 NIH 3T3 (ATCC CRL-1658) cells were cultured in Dulbecco's modified Eagle medium (DMEM; ATCC 30-2002) with 10% bovine calf serum and 1% antibiotics at 37 °C in a humidified atmosphere of 5% CO_2. Cells were kept for 24 h to reach a confluent monolayer. Bacteria (*P. aeruginosa*) were inoculated and harvested as mentioned above. Afterward, seeding solutions 10^8 cells/mL were inoculated in buffered DMEM supplemented with glucose. Old medium was removed from 3T3 cells followed by addition of 100 μL of seeding solution. The cocultures were then stored in a box humidified with damp paper towels at 37 °C overnight without shaking. Polymer NPs and other control solutions were diluted in DMEM media prior to use to obtain the desired testing concentrations. Old media was removed from coculture, replaced with freshly prepared testing solutions, and incubated for 3 h at 37 °C. Cocultures were then analyzed using LDH cytotoxicity assay to determine mammalian cell viability using manufacturer's instructions[6]. To determine the bacteria viability in biofilms, the testing solutions were removed and cocultures were washed with PBS. Fresh PBS was then added to disperse remaining bacteria from biofilms in coculture by sonication for 20 min and mixing with pipet. The solutions containing dispersed bacteria were then plated onto agar plates, and colony forming units were counted after incubation at 37 °C overnight.

Biofilm penetration studies using confocal microscopy:10^8 bacterial cells/ml of E2-Crimson (Red Fluorescent Protein) expressing *E. coli,* supplemented with 1 mM of IPTG ((isopropyl β-D-1-thiogalactopyranoside), were seeded (2 ml in M9 media) in a confocal dish and were allowed

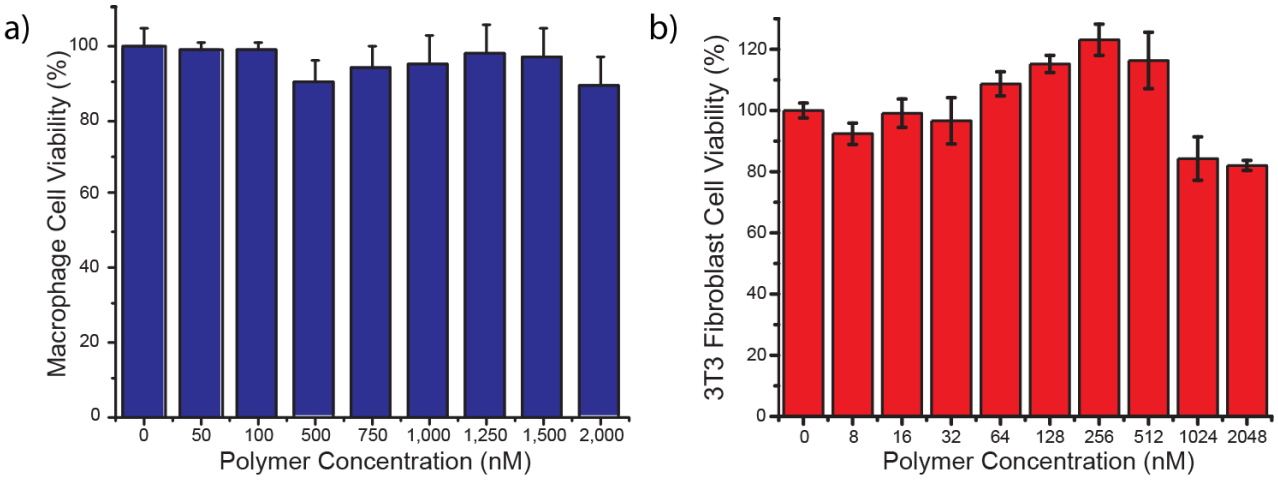
to grow. After 3 days media was replaced by 1000 nM of RhodGreen-P5 PNPs and biofilms were incubated for 1 hour, biofilm samples incubated with only M9 media were used as control. After 1 h, biofilms were washed with PBS three times and were incubated with 100 µM of the substrates for 1 h. The cells were then washed with PBS three times. Confocal microscopy images were obtained on a Zeiss LSM 510 Meta microscope by using a 63× objective. The settings of the confocal microscope were as follows: green channel: λ_{ex}=488 nm and λ_{em}=BP 505-530 nm; red channel: λ_{ex}=543 nm and λ_{em}=LP 650 nm. Emission filters: BP=band pass, LP=high pass.



Supplementary Figure S3: Penetration of Rhodamine Green labelled P5 PNPs (RhodGreen PONI-C11-TMA) into E2-Crimson expressing *E. coli* biofilms. The mean fluorescence of each confocal z-stack image was calculated using ImageJ software.

S8. Mammalian Cell Viability Assay

A total of 20,000 NIH 3T3 (ATCC CRL-1658) cells were cultured in Dulbecco's modified Eagle medium (DMEM; ATCC 30-2002) with 10% bovine calf serum and 1% antibiotics at 37 ºC in a humidified atmosphere of 5% CO_2 for 48 h.[7] Old media was removed, and cells were washed one time with phosphate-buffered saline (PBS) before addition of PNPs in the prewarmed 10% serum containing media. Cells were incubated for 24 h at 37 ºC under a humidified atmosphere of 5% CO_2. Cell viability was determined using Alamar blue assay according to the manufacturer's protocol (Invitrogen Biosource). After a wash step with PBS three times, cells were treated with 220 µL of 10% alamar blue in serum containing media and incubated at 37 ºC under a humidified atmosphere of 5% CO_2 for 3 h. After incubation, 200 µL of solution from each well was transferred in a 96-well black microplate. Red fluorescence, resulting from the reduction of Alamar blue solution, was quantified (excitation/emission: 560 nm/590 nm) on a SpectroMax M5 microplate reader (Molecular Device) to determine the cellular viability. Cells without any PNPs were considered as 100% viable. Each experiment was performed in triplicate.

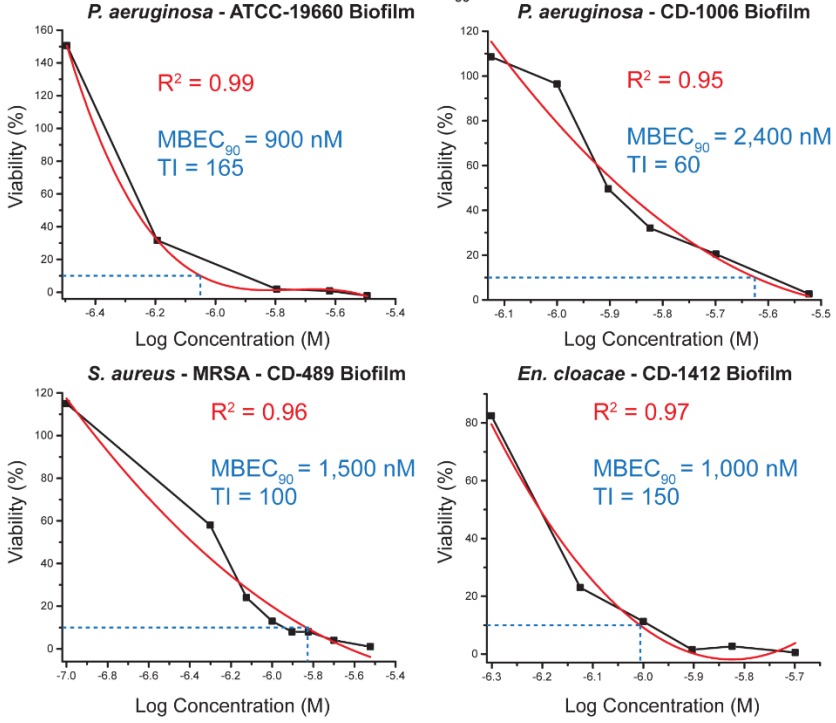


Supplementary Figure S4: Cytotoxicity of PNPs against a) RAW 264.7 cells and b) NIH-3T3 Fibroblast cells.

S9. Therapeutic Indices Against Biofilms

Bacteria were inoculated in lysogeny broth (LB) medium at 37 ˚C until stationary phase. The cultures were then harvested by centrifugation and washed with 0.85% sodium chloride solution three times. Concentrations of resuspended bacterial solution were determined by optical density measured at 600 nm. Seeding solutions were then made in M9 medium to reach an OD_{600} of 0.1. A 100 µL amount of the seeding solutions was added to each well of the 96-well microplate. The plates were covered and incubated at room temperature under static conditions for 1 day. The stock solution of P5 PNPs was then diluted to the desired level and incubated with the biofilms for 3 hours at 37˚C. Biofilms were washed with phosphate buffer saline (PBS) three times and viability was determined using an Alamar Blue assay. Minimal M9 medium without bacteria was used as a negative control. Concentrations were converted to Log, plotted with bacteria viability, and fitted to a curve to determine the minimum biofilm eradication concentration at 90% ($MBEC_{90}$).[8,9] The therapeutic index with respect to red blood cells was calculated by the concentration of P5 PNPs at $MBEC_{90}$ divided by the hemolysis at 50%.

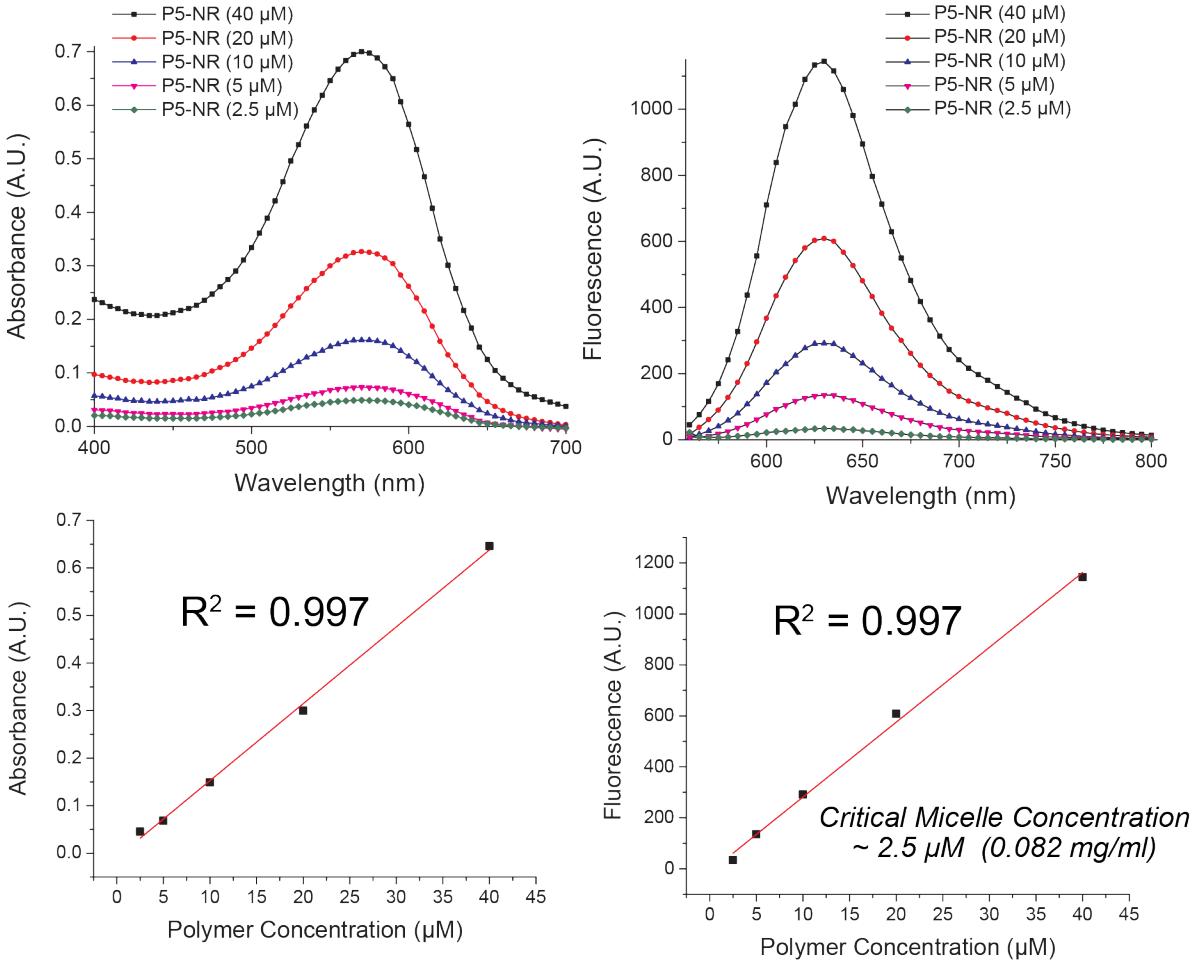
$$TI = \frac{MBEC_{90}}{HC_{50}}$$



Supplementary Figure S5. Therapeutic indices of PNPs against four bacterial biofilms.

S10. Critical Micelle Concentration Study

Critical Micelle Concentration of P5 PNP was determined through dilution of Nile Red encapsulated PNPs. Briefly, 16.0 mg of Polymer P5 and 2.0 mg of Nile Red was dissolved in 2 ml of dimethylsulfoxide in a 7ml scintillation vial. While under vigorous stirring, 3 ml of water was slowly added over the course of 1 hour and allowed to stir overnight. Afterwards, the vial was centrifuged, and the solution decanted to remove precipitated Nile Red that was not encapsulated. Followed by filtration through a PES syringe filter, the solution was transferred to a 3,500 MWCO dialysis bag and allowed to stir in 5L of water for two days, changing the water twice each day. Afterwards, the solution was filtered again through a PES syringe filter yielding Nile Red encapsulated P5 PNPs. Nile Red's fluorescence spectrum was monitored (Excitation = 550nm) as a function of decreasing polymer concentration. it was observed that at 2.5 µM, fluorescence decrease became non-linear. Further dilution was not possible due to limitations in the amount of Nile Red encapsulated. Therefore, the critical micelle concentration was determined to be ~ 2.5 µM and is well within the range of previously reported diblock polymer carriers.



Supplementary Figure S6. Critical micelle concentration of P5 PNPs.

S11. Uropathogenic Strain Information

Table S1 Uropathogenic *E. coli* strain information. All strains were harvested and tested for susceptibility in Cooley Dickinson Hospital Microbiology Laboratory (Northampton, MA). S: Susceptible; I: Intermediate; R: Resistant.

	Riley Strain Name	CD-2	CD-3	CD-19	CD-496	CD-549
	Species	*E. coli*	*E. coli*	*E. coli*	*E. coli*	*E. coli*
	Date Isolated	9/11/2011	9/11/2011	9/12/2011	1/3/12	1/9/12
	Specimen	UCC	UCC	UCC	UCC	UCC
	CFU/mL	>100,000	>100,000	>100,000	>100,000	>100,000
	Note			2 morphologies of *E. coli* with different susceptibilities		
Aminoglycosides	**Amikacin** (Amikin)		S			S
	Gentamicin (Garamycin)	S	R	S	S	R
	Kanamycin High Level					
	Tobramycin (Nebcin)		I			R
Beta-Lactam	**Ampicillin** (Omnipen, Polycillin)	R	R	R	R	R
	Ampicillin/sulbactam (Unasyn)	I	I	R	I	R
	Amoxicillin/CA (Augmentin)					I
	Aztreonam (Zithromax)					R
	Oxacillin (Prostaphlin)					

	Penicillin					
	Piperacillin (Pipracil)					R
Cephalosporin	**Ceftazidime** (Fortaz, Taxicef)					
	Cefaclor (Ceclor, Ceclor CD)					R
	Ceftriaxone (Rocephin)	S	S	S	S	R
	Cefotaxime					R
	Cefazolin (Ancef, Kefzol)	S	S	I	R	R
	Ceftizoxime					R
	Cefepime (Maxipime)	S	S	S	S	R
	Cefoxitin (Mefoxin)	S	S	S	S	R
	Cefuroxime-Sodium					
	Cefuroxime-Axetil (Ceftin)					R
Carbapenem	**Ertapenem**					
	Imipenem (Primaxin)				S	S
	Meropenem (Merrem)					
Macrolides	**Azithromycin** (Azactam)					
	Erythromycin					
Fluoroquinolone	**Ciprofloxacin** (Cipro)	S	S	R	S	R
	Levofloxacin (Levaquin)	S	S	R	S	R
	Ofloxacin (Floxin)					
Lincosamides	**Clindamycin** (Cleocin)					
Oxazolidinones Folate pathway inhibitors	**Linezolid** (Zyvox)					

Antimycobacterial	**Rifampin** (Rifadin, Rimactane)					
Folate pathway inhibitors	**Trimethoprim /Sulfa** (Gantanol)	S	R	S	S	R
Tetracycline	**Tetracycline**					R
Glycylcyclines	**Tigecycline**					S
Glycopeptides	**Vancomycin** (Vancocin)					

Table S2. Uropathogenic *E. cloacae complex, P. aeruginosa,* and *S. aureus* strain information. All strains were harvested and tested for susceptibility in Cooley Dickinson Hospital Microbiology Laboratory (Northampton, MA). S: Susceptible; I: Intermediate; R: Resistant.

	Riley Strain Name	CD-866	CD-1412	CD-1545	CD-1006	CD-23	CD-23	CD-489
	Species	*E. cloacae complex*	*E. cloacae complex*	*E. cloacae complex*	P. aeruginosa	*P. aeruginosa*	*S. aureus*	*S. aureus - MRSA*
	Date Isolated	3/29/12	6/7/12	6/19/12	4/23/12	9/12/11	6/21/11	1/3/12
	Specimen	UCC	UCC	UCC	UCC	UCI	UCC	UCS
	CFU/mL	10-100,000	>100,000	>100,000	>100,000	>100,000	MODERATE	>100,000
	Note		Urine from nephrostomy tube			Extended panel of susceptibilities for the P. aeruginosa		
Aminoglycosides	**Amikacin** (Amikin)		S	S				
	Gentamicin (Garamycin)	S	I	I	S	I	S	S
	Kanamycin High Level							

	Tobramycin (Nebcin)		R	R		S		
Beta-Lactam	**Ampicillin** (Omnipen, Polycillin)				S	R		
	Ampicillin/sulbactam (Unasyn)				S	R	S	R
	Amoxicillin/CA (Augmentin)					R	S	R
	Aztreonam (Zithromax)							
	Oxacillin (Prostaphlin)						S	R
	Penicillin						R	R
	Piperacillin (Pipracil)					S		
Cephalosporin	**Ceftazidime** (Fortaz, Tazicef)							
	Cefaclor (Ceclor, Ceclor CD)						S	R
	Ceftriaxone (Rocephin)	S	S	S	S	R	S	R
	Cefotaxime						S	R
	Cefazolin (Ancef, Kefzol)	R	R	R	S	R		
	Ceftizoxime					R		
	Cefepime (Maxipime)	S	S	S	S	I		
	Cefoxitin (Mefoxin)	R	R	R	S	R		
	Cefuroxime-Sodium						S	R
	Cefuroxime-Axetil (Ceftin)					R		
Carbapenem	**Ertapenem**							
	Imipenem (Primaxin)	S			S	S	S	R

	Meropenem (Merrem)							
Macrolides	**Azithromycin** (Azactam)						R	
	Erythromycin						R	
Fluoroquinolone	**Ciprofloxacin** (Cipro)	S	S	R	S	R		
	Levofloxacin (Levaquin)	S	S	R	S	R	S	R
	Ofloxacin (Floxin)							
Lincosamides	**Clindamycin** (Cleocin)						R	
Oxazolidinones Folate pathway inhibitors	**Linezolid** (Zyvox)		S	S			S	S
Antimycobacterial	**Rifampin** (Rifadin, Rimactane)						S	S
Folate pathway inhibitors	**Trimethoprim /Sulfa** (Gantanol)	S	R	R	S	R	S	S
Tetracycline	**Tetracycline**					R	S	S
Glycylcyclines	**Tigecycline**							
Glycopeptides	**Vancomycin** (Vancocin)						S	S

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