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Electronic Supplementary Information

Peptide-Perylene Diimide Functionalized Magentic Nano-platforms for Fluorescence Turn-on Detection and Clearance of Bacterial Lipopolysaccharide

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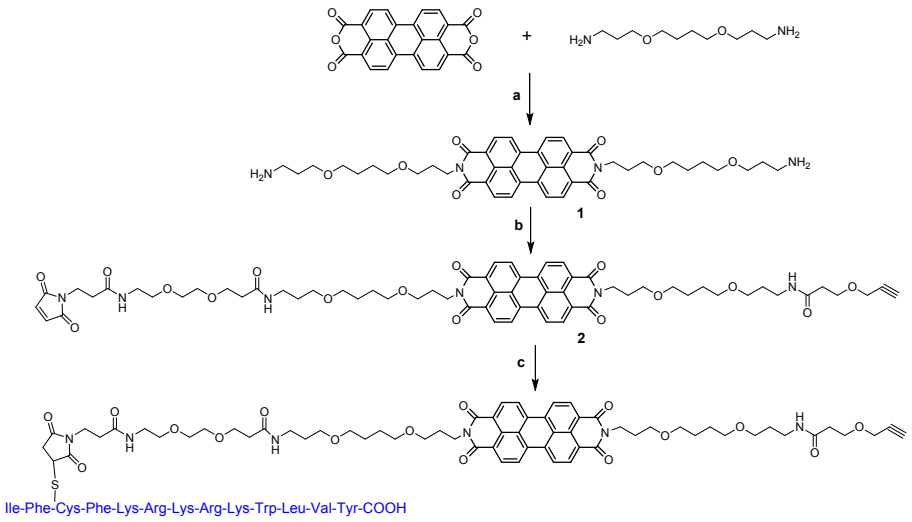
Materials and General methods:

Materials: Anhydrous solvents for organic synthesis were purchased from Aldrich and stored over activated molecular sieves (4 Å). Fmoc-Amino Acids and 2-Chlorotrityl Chloride Resin were purchased from GL Biochem Ltd (Shanghai). Commercially available reagents were used without further purification, unless noted otherwise. Perylene-3,4,9,10-tetracarboxylic dianhydride (Cat No. P1622435), Bovine serum albumin (99 %) were supplied by Sinpharm Chemcial Reagent Co., Ltd. Glutamine were obtained from Life Technologies. L- Ascorbic acid (99 %), Maltohexaose (> 90%), Guanosine diphosphate (> 96%), phosphatidyl ethanol amine (99 %) were purchased from Sigma Aldrich.Thin-layer chromatography (TLC was performed on precoated silica gel 60F-254 glass plates. Lipopolysaccharide (LPS) from *Escherichia coli* 0111 : B4 was purchased from Sigma Aldrich (cat. L3012, the molecular weight varies between 3 and 20 KDa, with an average of 10 KDa). ssDNA (5'-GA TCC TGC GGC CGC TAG TAC TGT CGA CG-3'), dsDNA (5'-GA TCC TGC GGC CGC TAG TAC TGT CGA CG-3') were supplied by First BASE Laboratories.

Instruments: ^{1}H NMR spectra were recorded using 400 MHz spectrometer. Mass spectra (MS) were measured with a Thermo LCQ Deca XP Max for ESI. Analytical reverse-phase HPLC analysis was performed on a Shimadzu HPLC system using an Alltima C-18 (250 × 10 mm) column at a flow rate of 3.0 mL/ min for preparation and a C-18 (250 × 4.6 mm) column at 1.0 mL/ min for analysis. TEM images were recorded on an FEI EM208S Transmission Electron Microscopy (Philips) operated at 100 kV. Fluorescence emission spectra were performed on a Varian Cary eclipse Fluorescence spectrometer. UV absorption spectra were recorded in a 5-mm path quartz cell on a Beckman coulter DU 800 spectrometer. FTIR (infrared spectroscopy) spectrums were recorded on a Shimadzu/Prestige-21 spectrometer.

1. Synthesis and characterization of perylene diimide-LPS recognition peptide conjugate



Perylene diimide-peptide conjugate (3)

Scheme S1: Synthesis of perylene diimide-peptide conjugate. a) imidazole, 125°C, 4h; b) Mal-PEG_2-NHS ester, Propargyl-NHS ester, DIPEA, DMF, r.t, overnight; c) DIPEA, r.t, overnight.

1.1) *Peptide synthesis:*

The peptide synthesis was performed by Fmoc-chemistry based solid phase synthesis using 2-Chlorotrityl Chloride Resin. The coupling reaction was carried out at room temperature using HBTU/DIPEA as coupling reagent. The reaction was allowed to last for 4 hours for coupling with arginine and 2 hours for other amino acids. The deprotection of Fmoc group was performed in 20% piperidine-DMF solution for 20 min. The peptide product was released from resin in the presence of 95% TFA solution (plus 2.5% water and 2.5% triisopropylsilane) followed by stirring for 2 h at rt. The aimed peptide was precipitated by diethyl ether.

1.2) *Preparation of Compound **1**:*

Perylene diimide (100 mg, 0.25 mmol), imidazole (3.0 g) and 1,4-Bis (3-aminopropoxy) butane (300 μL, 1.4 mmol) were mixed in a round-bottom flask with a stirring bar, and then heated to 125 °C. After stirring at 125 °C for 4h, the reaction mixture was cooled down to room temperature followed by the addition of 30 mL water. The red precipitate in water was collected by centrifugation at 4000 rpm for 5 min and followed by several times washing with water and diethyl ether. After drying under vacuum, the red solid product (181 mg, 95%) was used directly in the next step without further purification. ^{1}H NMR (400 MHz, DMSO-d6): δ= 8.51 (s, 4H), 8.27 (s, 4H), 7.67 (s, 4H), 4.11 (t, *J*=6.4 Hz, 4H), 3.51 (t, *J*=5.6 Hz, 4H), 3.37-3.40 (m, 12H), 2.83 (t, *J* = 7.2 Hz 4H), 1.92 (t, *J*=5.6 Hz 4H), 1.75 (m, 4H)，1.51 (s, 8H); ESI-MS: m/z 764.38 (calcd) for C44H52N4O8; 765.37 $[M+H]^+$, 1529.05 $[2M+H]^+$ (found).

1.3) *Preparation of Compound **2**:*

Compound **1** (50 mg, 0.065 mmol), Mal-PEG_2-NHS ester (28 mg, 0.075 mmol) and Propargyl-NHS ester (17 mg, 0.075 mmol) were mixed in 4 mL DMF, and then 20 μL triethylamine was added. After stirring at room temperature overnight, the reaction mixture was dropped into 30 mL diethyl ether to give a red precipitate. After 2 times washing, the crude products were purified by column chromatography on silica gel with eluent of methanol/ dichloromethane (1:20) to give 27 mg (35 %) product **2**. ^{1}H NMR (400 MHz, $CDCl_3$): δ= 8.67 (d, *J* = 8 Hz, 4H), 8.61 (d, *J* = 8 Hz, 4H), 6.69 (s, 2H), 4.32 (t, *J*=6 Hz, 4H), 4.16 (d, *J* = 2.4 Hz, 1H), 3.84 (t, *J* = 7.2 Hz, 2H), 3.75-3.80 (m, 4H), 3.30-3.64 (m, 30H), 2.55 (t, *J* = 7.6 Hz, 2H), 2.41-2.47 (m, 4H), 2.05-2.12 (m, 4H), 1.74 (dd, *J* = 12.2 Hz, 6.2 Hz, 4H), 1.55-1.63 (m, 8H); ESI-MS: m/z 1184.53 (calcd) for C64H76N6O16; 1185.31 $[M+H]^+$, 1207.57 $[M+Na]^+$ (found).

1.4) *Preparation of perylene diimide-peptide conjugate (**3**)*

Compound **2** (10 mg, 0.0084 mmol), peptide with sequence (20 mg, 0.011 mmol) were dissolved in 1 mL DMSO, then 5 μL DIPEA was added. The reaction mixture was stirred at room temperature overnight. After precipitation in diethyl ether, the crude products were purified by reverse-phase high performance liquid chromatography (HPLC) with eluting system consisting of A (0.1% TFA water solution) and B (0.1% TFA acetonitrile solution) under a linear gradient, monitored by UV absorbance at 490 nm. The linear gradient stretched over 20 minutes from t = 0 minutes at 20% solution B to t = 20 minutes at 80 % solution B. The reaction yielded the Peptide–perylene diimide conjugate (12 mg, 50 %) after lyophilization. HPLC analysis demonstrated that the purity of the conjugate was over 95%. ^{1}H NMR (400 MHz, DMSO-d6): δ= 10.75 (s), 9.21 (br s), 8.60 (d, *J* = 8.0 Hz), 8.51 (d, *J* = 6.4 Hz), 8.32 (d, *J* = 7.2 Hz), 7.97-8.15 (m), 7.78 (br s), 7.59-7.68 (m), 7.31 (d, *J* = 8.0 Hz), 7.11-7.24 (m), 7.05 (t, *J* = 7.2 Hz), 6.94-7.03 (m)，6.62 (d, *J* = 8.4 Hz), 4.54-4.65 (m); 4.19-4.35 (m); 4.03-4.12 (m); 3.97 (dd, *J* = 8.8 Hz, 4.0 HZ); 3.49-3.62 (m); 2.72-3.27 (m); 2.36-2.43 (m); 2.25-2.32 (m); 1.90 (br s); 1.82 (br s); 1.42-1.62 (m); 1.05-1.30 (m); 0.78-0.89 (m); ESI-MS: m/z 2970.55 (calcd) for C152H211N29O31S; $[M+2H]^{2+}$ 1486.76, $[M+3H]^{3+}$ 991.51 (found).

2. The preparation and characterization of peptide functionalized Fe_3O_4@SiO_2 core-shell nanoparticles

2.1) *The preparation and characterization of Fe_3O_4 magnetic nanoparticles*

The synthesis of metal–oleate complex was performed according to the reported process.[1] The monodisperse Fe_3O_4 nanoparticles were prepared according to the following procedure. Briefly, iron oleate (1.0 g, 1.1 mmol) and oleic acid (177.3 μL, 0.56 mmol) were dissolved in 7.1 mL 1-octadecene. The mixture was heated to 100 °C, and kept at that temperature for 1 hour. And then, the reaction mixture was heated to 290 °C with a constant heating rate of 3.3 °C per min, and kept at that temperature for 2 hours. After cooling to room temperature, the particles were precipitated in 50 mL ethanol. The precipitates were separated by centrifugation followed by 3 times washing with the solvent mixture of hexane/ethanol (1:3).[2] A small portion of Fe_3O_4 nanoparticles was suspended in hexane, and a drop of the solution was then dropped onto a copper grid for TEM imaging (Fig. S1). The nanoparticle size distribution was analyzed on 100 individual particles by using Image J.[3]

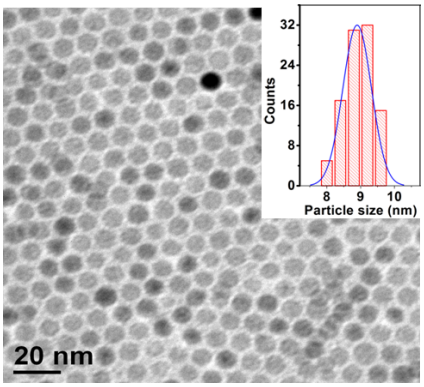


Fig. S1 TEM image of Fe_3O_4 nanoparticles. The inset shows the diameter distribution of 100 individual Fe_3O_4 nanoparticles measured by using Image J.

2.2) *The preparation and characterization of azide group functionalized Fe_3O_4@SiO_2 core-shell magnetic nanoparticles*

3.2 mg of synthesized Fe_3O_4 nanoparticle was dispersed in the solvent mixture containing 30 mL cyclohexane, 4.8 mL Triton-X100, 2 mL 1-hexanol, 0.7 mL distilled water and 300 µL $NH_3.H_2O$. After stirring at 600-700 rpm for half an hour, 26 µL Tetraethyl orthosilicate (TEOS) in 1 mL cyclohexane was dropped into the reaction mixture. After 4 hours, 4 µL of (3-aminopropyl) triethoxysilane (APTS) in 1 mL cyclohexane was dropped into the mixture. After overnight stirring at room temperature, 50 mL pre-cooled ethanol was added into the mixture, and the pH value was adjusted to about 7.0 by 1M HCl at 0 °C. The particle pellets were obtained by centrifugation at 10000 rpm for 8 minutes and washed by water and ethanol.[4-5]

To achieve the surface functionalization with azide group, Fe_3O_4@SiO_2 core-shell nanoparticles (30 mg) were suspended in 2 mL DMF, excess amount of azido-dPEG$_4$·NHS ester (10 mg, 0.026 mmol) and 10 µL DIPEA were then added. The reaction was shaken at room temperature overnight. The nanoparticles were separated by centrifugation at 10000 rpm for eight minutes and washed with DMF and EtOH.

FTIR (infrared spectroscopy) spectra were recorded on a Shimadzu/Prestige-21 spectrometer equipped with IR solution software. The samples were prepared by KBr pellet method before measurements. For organic sample, the compound was mixed with KBr (2 %, m/m). For nanoparticles, the sample was mixed with KBr (20%, m/m) (Fig. S2).

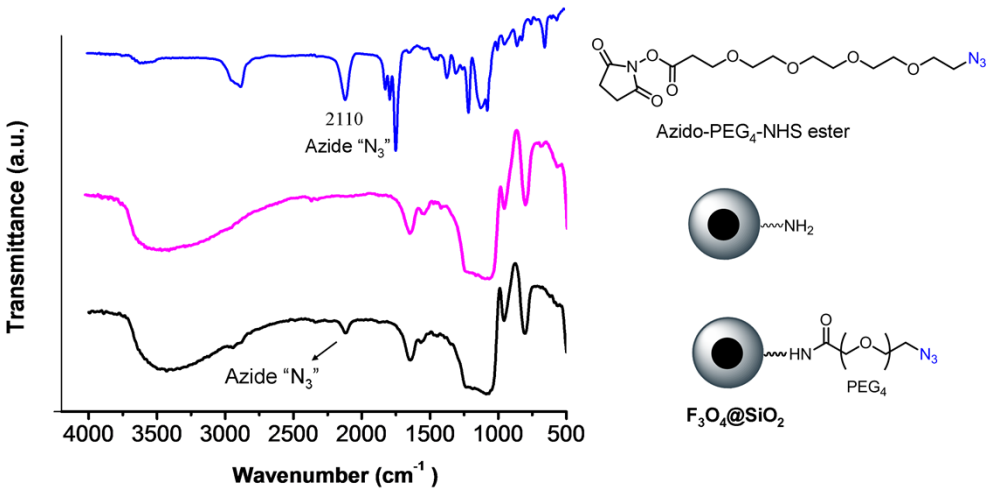


Fig.S2 FTIR spectrum of azide tagged linker, amino group functionalized $Fe_3O_4@SiO_2$, and azide group subsequently functionalized $Fe_3O_4@SiO_2$.

2.3) *The Preparation and Characterization of PDI-peptide conjugated $Fe_3O_4@SiO_2$ core-shell nanoparticles*

The azide group functionalized core-shell nanoparticle (20 mg) was mixed with perylene diimide- peptide conjugate (**3,** 400 μM) in 5 mL water/t-butanol (1:1) mixture containing $CuSO_4$ (10 mmol) and ascorbic acid (10 mmol). The click reaction was keep shaking at room temperature for 48 hours. The nanoparticle was separated by centrifugation at 10000 rpm for 8 min, and followed by washing with H_2O, DMF and then EtOH. The final particles were kept in fridge at 4°C for further use. The amount of PDI-peptide ligand conjugated to particle surface was determined by subtracting the amount of PDI-peptide left in the supernatant from the total amount of ligand added into the reaction mixture. The amount of PDI-peptide in the supernatant was determined by absorbance measurement at 539 nm. The absorbance value was then converted to a concentration value based on a concentration-absorbance standard curve of free PDI-peptide.[6] The amount of PDI-peptide ligand on MNPs was calculated to be 10 nmol/mg. A small portion of final nanoparticles was suspended in ethanol, and a drop of the solution was then dropped onto a copper grid for TEM imaging (Fig. S3). The nanoparticle size distribution was analyzed on 100 individual particles by using Image J.[3]

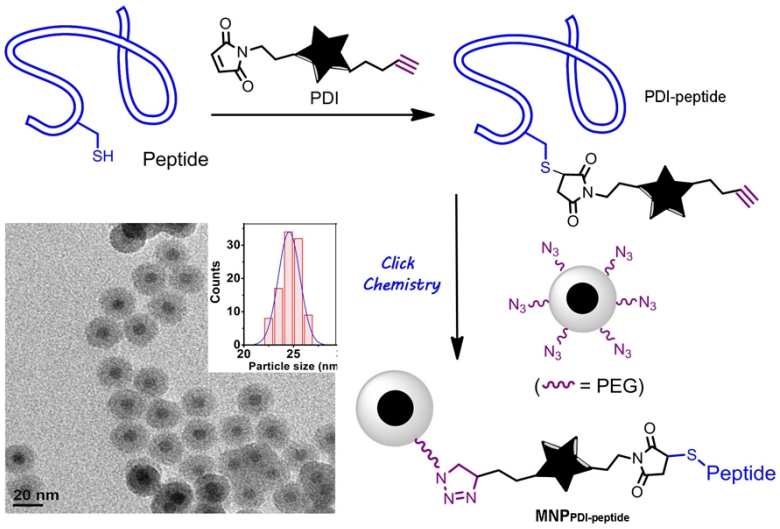


Fig. S3 Schematic illustration of the preparation of $MNP_{PDI\text{-}peptide}$ conjugates, TEM image and particle size distribution analysis of $MNP_{PDI\text{-}peptide}$.

2.4) *The Preparation and characterization of Perylene diimide conjugated $Fe_3O_4@SiO_2$ core-shell nanoparticles without peptide conjugation (MNP_{PDI}).*

As control, the azide group functionalized core-shell nanoparticle (10 mg) was mixed with perylene diimide derivative **2** (400 μM) in 2.5 mL water/t-butanol (1:1) mixture. The click chemistry reaction was performed under the same condition as described above. After 24 hours shaking, the nanoparticle was separated and washed under the processes described above. The presence of perylene diimide on the surface of particle was confirmed by UV-Vis absorption spectra (Fig. S3a).

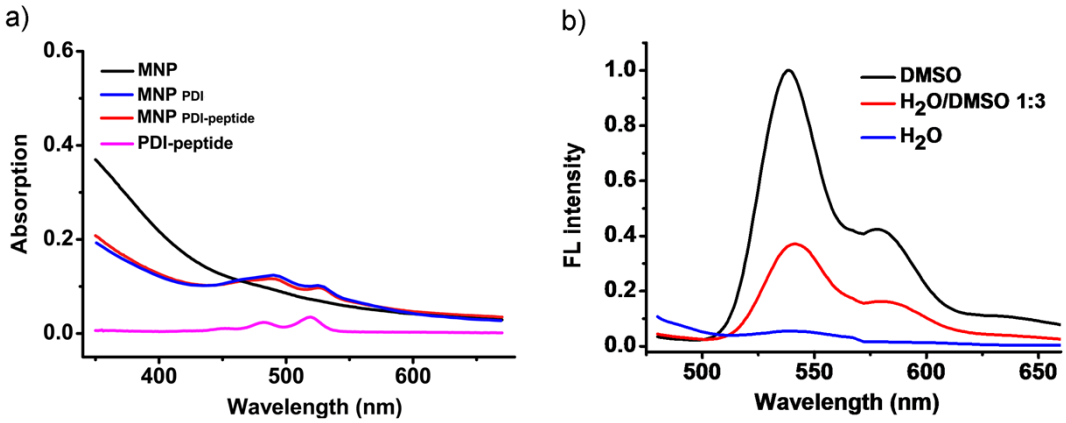


Fig. S4 a) UV-Vis absorption spectra of free PDI-peptide (2.0 μM), MNP (200 μg/mL), MNP_{PDI} (200 μg/mL) and $MNP_{PDI\text{-}peptide}$ (200 μg/mL) in DMSO; b) Fluorescence emission spectra exited at 450 nm of 200 μg/mL $MNP_{PDI\text{-}peptide}$ in different solvents.

3. Selective fluorescence enhancement of $MNP_{PDI\text{-}peptide}$ solutions upon the addition of LPS

3.1) *LPS-triggered fluorescence enhancement* of $MNP_{PDI\text{-}peptide}$ *solution.*

The incubation of $MNP_{PDI\text{-}peptide}$ (100 μg/mL) and LPS was performed in TBS buffer (Tris 50 mmol/L, NaCl 150 mmol/L, pH 7.4). Upon the addition of LPS into $MNP_{PDI\text{-}peptide}$ solution, the emission spectrum $MNP_{PDI\text{-}peptide}$ excited at the wavelength of 450 nm was recorded by a Varian Cary eclipse Fluorescence spectrometer after 15 min incubation at 37 °C. Data normalization was based on the follow concept: sets the fluorescence (FL) peak intensity value $I_{539\ nm}$ at 250 μg/mL (25 μM) LPS as I_{max}, the normalized intensity "*I*" was defined as the measured intensity divided by I_{max}.[7] As the molecular weight of commercial LPS varies between 3 and 20 KDa, we assume an average molecular weight of 10 KDa in our experiment by following the reported method (Ref.4, 5a and 6 in main text, Ref.8 ESI).

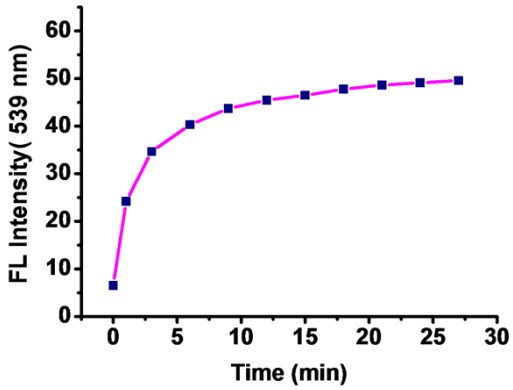


Fig. S5 Increasing fluorescence intensity at 539 nm of $MNP_{PDI\text{-}peptide}$ (100 μg/mL) in TBS upon addition of 100 μg/mL LPS at different time interval.

3.2) *Determination of the detection limit of* $MNP_{PDI\text{-}peptide\ to}$ *toward addition of LPS based on fluorescence turn-on mechanism*

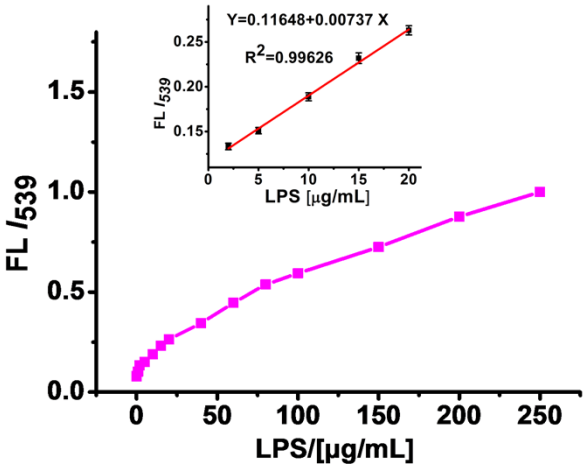


Fig. S6 Plots the normalized FL intensity at 539 nm after 15 min incubation vs the concentrations of LPS (from 0-250 μg/mL). The inset shows the plot and linear fitting of fluorescence intensity at 539 nm vs the concentration of LPS (2 μg/mL -20 μg/mL).

Based on the linear fitting in Fig. S4, the detection limit (C_{min}) is estimated to be 280 ng/mL (28 nmol) by a reported method.[8]

$$C_{min} = \frac{3\sigma}{B}$$

where σ is the standard deviation obtained from three individual fluorescence measurements ($I_{539\ nm}$) of $MNP_{PDI\text{-}peptide}$ (100 μg/mL) without any analyte in TBS buffer (pH 7.4) and B is the slope obtained after linearly fitting the titration curves within certain ranges.

3.3) *Fluorescence enhancement of $MNP_{PDI\text{-}peptide}$ solution to LPS in different pH conditions*

TBS buffers with different pH values (4.0, 5.0, 5.5, 6.0, 6.5, 7.0, 7.5, 8.0, 8.5, 9.0, 9.5, 10.0, 11.0) were prepared by adjusting the pH with 1.0 M HCl and 1.0 M NaOH water solutions from the mother solution (Tris 50 mmol/L, NaCl 150 mmol/L). All the buffers were sterile filtrated through a 0.2 μm filter before use. The emission spectra (excited at 450 nm) of $MNP_{PDI\text{-}peptide}$ (100 μg/mL) in TBS buffers with different pH values were recorded after 15 min incubation at 37 °C in the absence or in the presence of LPS (100 μg/mL).

3.4) *Selective fluorescence enhancement of $MNP_{PDI\text{-}peptide}$ solution toward LPS and other interferences*

LPS and various small molecular or macromolecular interferences (all at 10 μM) was individually incubated with $MNP_{PDI\text{-}peptide}$ (100 μg/mL) in TBS buffer (pH 7.4). The emission spectrum (excited at 450 nm) was recorded after 15 min incubation at 37 °C (Fig. S5).

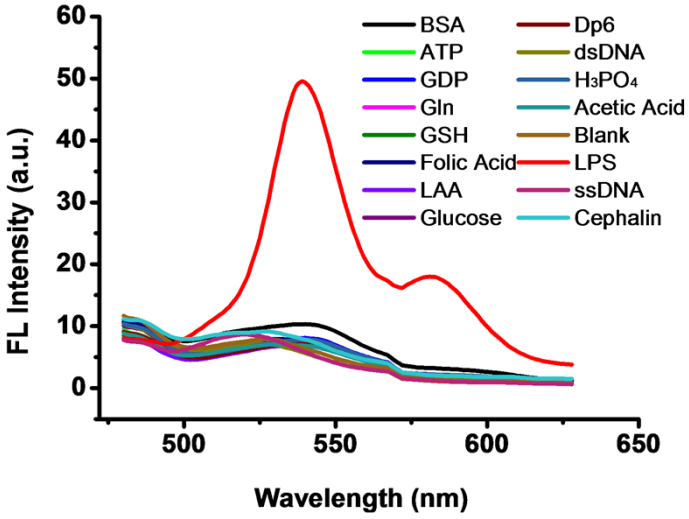


Fig. S7 a) The selective increase of fluorescence of $MNP_{PDI\text{-}peptide}$ (100 μg/mL) upon addition of LPS against various biologically important species (all at 10 μM). BSA: Bovine serum albumin; ATP: Adenosine triphosphate; GDP: Guanosine diphosphate; Gln: Glutamine; GSH: Glutathione; LAA: L-Ascorbic acid; Dp6: Maltohexaose; Cephalin: phosphatidyl ethanol amine. ssDNA: single-stranded DNA; dsDNA: double-stranded DNA; Blank: $MNP_{PDI\text{-}peptide}$ only. The fluorescence spectrum was recorded after 15 min incubation at 37 °C.

3.5) *Competitive binding of $MNP_{PDI\text{-}peptide}$ toward LPS over other important biomolecules*

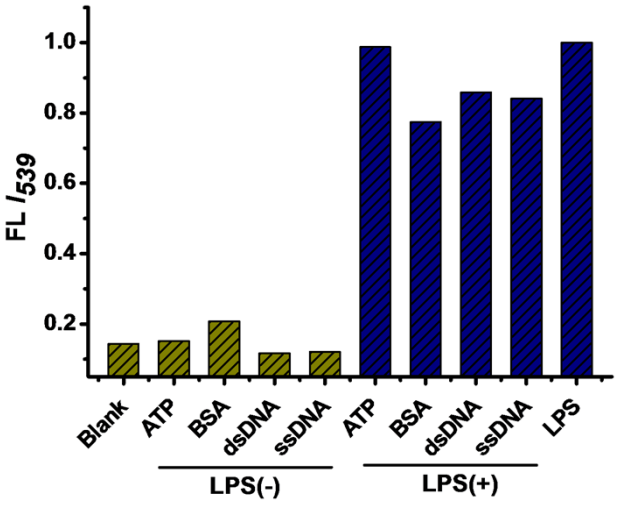


Fig. S8 Competitive binding analysis of $MNP_{PDI\text{-}peptide}$ (100 μg/mL) solution pretreated with ATP, BSA, dsDNA or ssDNA (10 μM) in the absence and presence of LPS (100 μg/mL). The fluorescent intensity was recored at 539 nm.

Basically, $MNP_{PDI\text{-}peptide}$ (100 μg/mL) was first incubated together with 10 μM negatively charged ATP, BSA, ssDNA (5'-GA TCC TGC GGC CGC TAG TAC TGT CGA CG-3'), dsDNA (5'-GA TCC TGC GGC CGC TAG TAC TGT CGA CG-3') in TBS buffer (Tris 50 mmol/L, NaCl 150 mmol/L, pH 7.4) at 37°C, respectively. After 15 min incubation, LPS (100 μg/mL, 10 μM) was then added into the mixtures. The fluorescence spectra excited at 450 nm were recorded at the different time intervals.

4. Effective clearance of Bacterial lipopolysaccharide (LPS) from bacterial lysates by $MNP_{PDI\text{-}peptide}$

E. coli DH5α (ATCC 53868), or *B. subtilis* (ATCC 33677) cells (10^9 cfu/mL) were suspended in TBS buffer (50 mmol/L Tris, 150 mmol/L NaCl, pH 7.4), and then were lysed under ice bath using an ultrasonic processor (VCX 130, Sonics & Materials, Inc.). The parameters were set at 90 W power and 5 s pulse applied cycles followed by 5 s pauses for a total of 45 min. After centrifugation at 4000 rpm for 10 min, the supernatant was collected and used directly in LPS removal assay.

For LPS clearance, $MNP_{PDI\text{-}peptide}$ was incubated with bacterial lysates in a 2.0 mL centrifugal tube. After 30 min incubation at 37°C, the $MNP_{PDI\text{-}peptide}$ with surface captured by LPS was removed through a simple magnet separation. The supernatant was used for LPS and protein quantification. For a next cycle clearance, the freshly prepared $MNP_{PDI\text{-}peptide}$ (200 μg/mL) was added to the supernatant followed by the same process as described above.

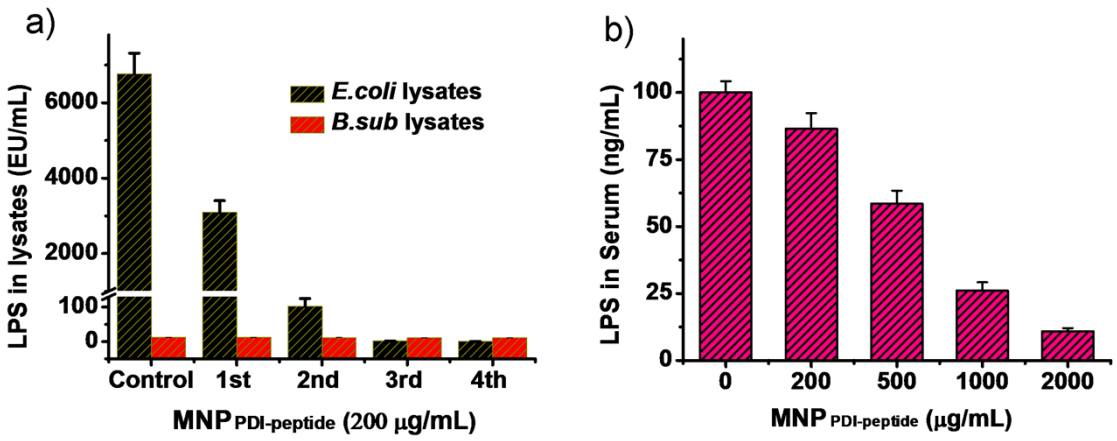


Fig. S9 a) Multi-cycles removal of LPS from *E. coli* and *B. Sub*. cell lysates via. 200 μg/mL $MNP_{PDI\text{-}peptide}$; d) Quantified magnetic LPS removal from contaminated human serum with different amounts of nanoparticles.

4.1) LPS quantification upon the clearance by $MNP_{PDI\text{-}peptide}$ from bacterial lysates

LPS quantification was performed using Limulus amebocyte lysate (LAL) assay (Lonza, Switzerland) according to its standard procedure.[9] To match the range of linearity of LAL assay, ten-fold seral dilutions were prepared from the original cell lysates. Briefly, 25 μL of each tested solution were loaded into 96-well plates and then 25 μL of LAL enzymatic reagent was mixed well with the sample solutions. After 10 minutes incubation at 37°C, 50 μL of substrates solution was added. After further 6 min incubation, 50 μL of stop solution was added. The absorbance of each reaction well at 410 nm was then recorded by a Tecan's Infinite M200 microplate reader. Each experiment contains 3 repeats. The LPS concentration was calculated according to the described methods in the standard protocol of Limulus Amebocyte Lysate (LAL) QCL-1000. According to the reported data, 100 pg of LPS is corresponding to 1.2 endotoxin units [EU].[10]

4.2) Protein quantification upon the efficient clearance of bacterial LPS from bacterial lysates

Total protein concentration in cell lysates before and after LPS clearance with $MNP_{PDI\text{-}peptide}$ was quantified by Bio-Rad Protein Assay (Bio-Rad Laboratories, USA) according to its standard procedure. Briefly, the dye reagent was diluted for 5 times with distilled water before use. 20 μL of the tested sample was mixed with 200 μL of the diluted reagent in 96-well plates. After incubation at room temperature for 5 minutes, the absorbance at 595 nm was recorded by a Tecan's Infinite M200 microplate reader.[11] The protein concentration was determined according to the described methods in the standard protocol (Fig. S6).

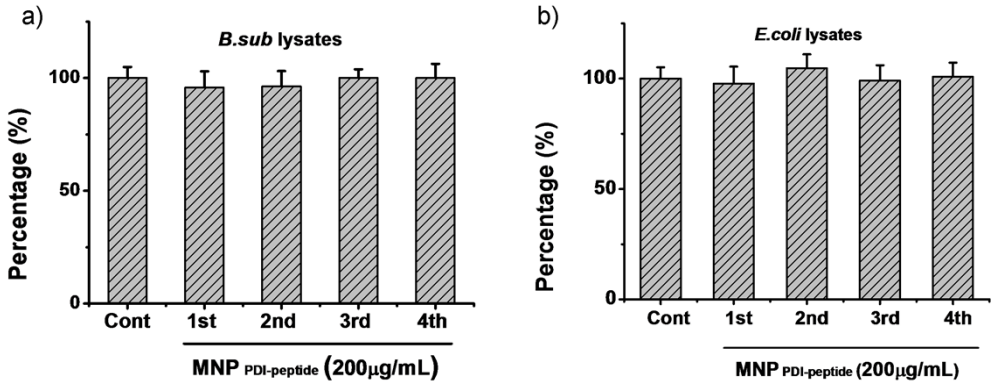


Fig. S10 Protein recovery after 1-4 cycles treatments with 200 μg/mL $MNP_{PDI\text{-}peptide}$. The protein quantification was performed using Bio-Rad Protein Assay.

5. Efficient clearance of bacterial LPS from contaminated human serum sample by using $MNP_{PDI\text{-}peptide}$

Human serum (from human male AB plasma, Sigma Aldrich) without further dilution was sterile filtrated through a 0.2 μm filter. Pure LPS water solution (1.0 μL, 500 μg/mL) was added into human serum (5.0 mL) to reach 100 ng/mL to prepare the contaminated samples.[12-13] Then the LPS containing serum was incubated with different concentrations of $MNP_{PDI\text{-}peptide}$ (0, 200, 500, 1000, 2000 μg/mL) at 37°C for 30 min. After removal of the LPS captured $MNP_{PDI\text{-}peptide}$ by magnetic separation, LPS content in serum was quantified by LAL assay as described before.

6. Detoxification evaluation of contaminated human serum samples by quantifying NO generation in macrophage cultures.

The serum samples after LPS clearance with different concentrations of $MNP_{PDI\text{-}peptide}$ (0, 200, 500, 1000, 2000 μg/mL) were mixed with basic RPMI 1640 respectively (serum to medium 1:3) to form the cell culture medium. The mouse macrophage RAW 264.7 cells were plated at a density of 1×10^6 cells/well in 24- well plates for 12 h, followed by treatment with the as prepared cell culture medium for a further 24 h. The amount of NO production in the medium was detected with the Griess reaction (Life Technologies, USA). 75 μL of each supernatant was mixed with 65 μL water and 10 μL Griess reagent in a 96-well plate. The absorbance at 548 nm was recorded after 30 min incubation at rt. NO quantification was processed according to the standard procedure supplied with Griess reagent (Invitrogen).[14]

7. Cell viability test of $MNP_{PDI\text{-}peptide}$

Briefly, RAW 264.7 cells were seeded in a 24-wells plate with a density of 1.0×10^6 cells per well in RPMI 1640 with 10% FBS. After 12 hours' incubation at 5% CO_2 and 37°C, the medium was changed with fresh medium in the presence of $MNP_{PDI\text{-}peptide}$ at different concentrations. The cell viability

measurements were determined after 24 hours' incubation. The particle containing medium was then replaced with fresh RPMI-1640 (phenol red free) containing 10% TOX-8 reagent. After 2 hr incubation, the fluorescence intensity at 590 nm was measured at excitation wavelength of 560 nm by a Tecan's Infinite M200 microplate reader. The cell viability rate (VR) was calculated according the reported methods (Fig. S7).[15] RAW 264.7 cells were purchased from Sigma-Aldrich.

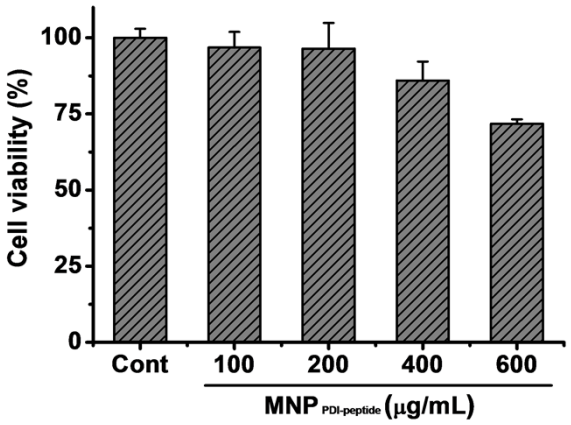


Fig. S11 Cell viability evaluation based on TOX-8 assay after 24 h incubation in the presence of 100, 200, 400, 600 μg/mL $MNP_{PDI\text{-}peptide}$.

References

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