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Supporting Information

Controlled Release of Singlet Oxygen Using Diphenylanthracene

Functionalized Polymer Nanoparticles

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Experimental section

Materials and Instruments: Butyl methacrylate (BMA, 99%), ethylene glycol dimethacrylate (98%), divinylbenzene (80%) and styrene (99%) were purchased from Sigma Aldrich and distilled before used. All other reactants and spectroscopic solvents were purchase from Sigma Aldrich and used as received. Synthesis grade solvents were distilled and/or dried according standard procedures before used. Bi-distilled water was purified with the Millipore Milli-Q system. Flash column chromatography was performed with silica gel 60 (0.040-0.063 mm) purchased from Merck. NMR spectra were recorded on a Bruker Advanced II (300 or 400 MHz). UV/Vis absorption spectra were recorded on a JASCO V660 spectrophotometer with a Peltier temperature control unit. Steady state fluorescence spectra were recorded in a Spex Fluorolog 1680 Double spectrometer. Dynamic light scattering (DLS) measurements were performed in a Brookhaven instrument (BI-200SM Goniometer and BI-9000 AT correlator using a He-Ne laser (632 nm, 35 mW, model 127, Spectra Physics) and an avalanche photodiode detector. Time-resolved picosecond fluorescence intensity decays were obtained by the single-photon timing method with laser excitation at 350 nm and emission at 420 nm. The set up consists of a diode-pumped solid state Nd:YVO4 laser (Milennia Xs, Spectra Physics) that can synchronously pump a mode-locked Ti:Sapphire laser (Tsunami, Spectra Physics, with tuning range 700-1000 nm, output pulses of 100 fs, and 80 MHz repetition rate that can be reduced down to 4 MHz by a pulse picker) or a cavity-dumped dye laser (701-2, Coherent, delivering 3-4 ps pulses of ca. 40 nJ pulse-1 at 3.4 MHz) working with Rhodamine 6G. Intensity decay measurements were made by alternated collection of impulse and decays with the emission polarizer set at the magic angle position. Impulses were recorded slightly away from the excitation wavelength with a scattering suspension. For the decays, a cut-off filter was used to effectively remove excitation light. Emission light was passed through a depolarizer before reaching the monochromator (Jobin-Yvon HR320 with a 100 lines/mm grating) and detected using a Hamamatsu 2809U-01 microchannel plate photomultiplier. No less than 10 000 counts were accumulated at the maximum channel. Decay data analysis with sums of exponential functions was done with Microsoft Excel. Scanning electron microscopy (SEM) characterization was performed on a JEOL JSM7001F with 15/20 nm Chromium deposition. TEM images were obtained on a Hitachi transmission electron microscope (Model H-8100 with a LaB6 filament) with an accelerator voltage of 100 kV. One drop of water dispersion was placed on a carbon grid and dried in air before observation. AFM images were obtained on a Nanosurf® easyScan 2 FlexAFM atomic force microscope. The images were obtained in tapping mode at room temperature, using Tap190Al-G (budget sensor) rotated monolithic silicon probe tips with symmetric shape, a nominal radius <10 nm, nominal force constant of 48 Nm⁻¹, and nominal resonant frequency of 190 kHz. Nanoparticles Tracking Analysis (NTA) was performed in a Nanosight LM10-HSBF microscope. Irradiations were performed with an Osram HBO- 200 W mercury arc lamp.



Scheme S1. Synthetic routes to dyes 2 and 3.

Compound 5 - 9-(bromo)-10-(4-hydroxyphenyl)anthracene: 4 (2.44 g, 7.25 mmol), 4-hydroxyphenylboronic acid (0.5 g, 3.6 mmol), K₂CO₃ (1.29 g, 9.36 mmol) and Pd(Ph₃)₄ (0.124 g, 0.1 mmol) were added to a mixture of THF:water (9:1, 60 mL) and stirred for 48 h at 70°C under argon atmosphere. The mixture was then cooled to room temperature, diluted with water and extracted with diethyl ether (4×50 mL). The organic phases were dried with MgSO₄, filtered and the solvent evaporated under reduced pressure. The final product was purified by silica flash chromatography using a mixture of hexane/ethyl acetate (9/1) as eluent. After purified by column, the compound was recrystallized from methanol. The final product was obtained as a yellow solid (1.4 g, 4.13 mmol, 57% yield). ¹H NMR (ppm, 400 MHz, DMSO-d₆) δ 9.77 (s, 1H), 8.50 (d, J = 9.2 Hz, 2H), 7.70 (m, 4H), 7.51 (m, 2H), 7.2 (d, J = 8.4 Hz, 2H), 7.02 (d, J = 8 Hz, 2H). ¹³C NMR (ppm, 400 MHz, DMSO-d₆) δ 177.2, 138.2, 131.9, 130.8, 129.6, 127.8, 127.5, 127.3, 127.1, 125.9, 115.5. HRMS (ESI) m/z calculated for [C₂₀H₁₃BrO] 349.0225; found 349.0223 (M+H⁺).

Compound 6 - **9-(4-hydroxyphenyl)-10-(phenyl)anthracene: 5** (0.8 g, 2.3 mmol), phenylboronic acid (0.36 g, 3 mmol), K₂CO₃ (0.83 g, 5.9 mmol) and Pd(Ph₃)₄ (79 mg, 0.02 mmol) were added to a mixture of THF:water (9:1, 30 mL) and stirred for 48 h at 70°C under argon atmosphere. The mixture was then cooled to room temperature, diluted with water and extracted with diethyl ether (4×50 mL). The organic phases were dried with MgSO₄, filtered and the solvent evaporated under reduced pressure. The final product was purified by silica flash chromatography using hexane/ethyl acetate (8/2) as eluent. After purified by column the compound was recrystallized from methanol (0.74 g, 2.1 mmol, 93% yield). ¹H NMR (ppm, 400 MHz, CDCl₃) δ 7.73 (m, 4H), 7.59 (m, 3H), 7.49 (d, 2H), 7.34 (m, 6H), 7.08 (d, J = 8.4 Hz, 2H). ¹³C NMR (ppm, 400 MHz, CDCl₃) δ 155.1, 139.2, 132.7, 131.5, 130.3, 130.1, 128.5, 127.6, 127.13, 125.1, 125.1, 115.5. HRMS (ESI) m/z calculated for [C₂₆H₁₈O] 346.1353; found 346.1352 (M⁺).

Compound 2 - 9-(phenyl)-10-(4-phenyl methacrylate)anthracene: to a solution of **6** (0.1 g, 0.3 mmol) in dry dichloromethane (15 mL) under argon atmosphere was added Et₃N (87 mg, 0.86 mmol). The mixture was stirred at room temperature for 15 minutes. After this time, methacryloyl chloride (0.33 mg, 0.3 mmol) dissolved in dry dichloromethane (2 mL) was added drop wise. The mixture was stirred for 12 hours at room temperature under argon atmosphere. The solvent was removed under reduced pressure and the crude was purified by silica flash chromatography using hexane/ethyl acetate (9/1) as eluent. The final product was obtained as a yellow solid (78 mg, 0.2 mmol, 62% yield). ¹H NMR (ppm, 400 MHz, CDCl₃) δ 7.71 (m, 4H), 7.55 (m, 7H), 7.35 (m, 6H), 6.46 (s, 1H), 5.84 (s, 1H), 2.15 (s, 3H). ¹³C NMR (ppm, 400 MHz, CDCl₃) δ 166.1, 153.1, 150.6, 139.2, 132.5, 131.5, 130.8, 130.1, 130.0, 128.6, 127.1, 127.2, 121.8, 18.6. HRMS (ESI) m/z calculated for [C₃₀H₂₂O₂] 414.1612, found 414.1614 (M⁺).

Compound 3 - 9,10-bis(4-vinylphenyl)anthracene ^[S1]: a mixture of 4 (0.39 g, 1.13 mmol), 4-vinylphenylboronic acid $(0.5 \text{ g}, 3.38 \text{ mmol}), \text{ K}_2\text{CO}_3$ (0.62 g, 6.76 mmol) and $Pd_2(dba)_3$: $Pd(tBu_3)_2$ (6.8 mg, 0.0075 mmol: 1.9 mg, 0.00375 mmol) in anhydrous toluene (20 mL) was stirred for 48 h at 110°C under argon atmosphere. After this time the mixture was cooled to room temperature, the suspension filtered and the solvent evaporated. The residue was then solubilized in dichloromethane (50 mL) and washed with water (4×50 mL), dried over MgSO₄ and filtered. The solvent was evaporated and the product purified by silica flash chromatography using hexane as eluent. The final product was obtained as a yellow solid (0.38 g, 1 mmol, 90% yield). ¹H NMR (ppm, 400 MHz, CDCl₃) δ 7.73 (dd, J = 3.2 Hz and 6.8 Hz, 4H), 7.66 (d, J = 8 Hz, 4H), 7.45 (d, J = 8 Hz, 4H), 7.34 (dd, J = 3.2 Hz and 6.8 Hz, 4H), 6.9 (dd, J = 11.2 Hz and 17.6 Hz, 2H), 5.92 (d, J = 17.6 Hz, 2H), 5.38 (d, J = 10.8 Hz, 2H). ¹³C NMR (ppm, 400 MHz, CDCl₃) δ 138.7, 136.9, 136.8, 131.7, 129.9, 127.8, 127.1, 126.4, 125.2, 114.3. LRMS (ESI) m/z calc for [C₃₀H₂₂] 382.17, found 383.1 $(M+H^{+}).$

PBMA seed latex preparation: a mixture of butylmethacrylate (BMA, 3.56 g), ethylene glycol dimethacrylate (EGDMA, 3.73 g) was dispersed in a solution of SDS (1.64 g) in water (141.9 g) by magnetic string in a three-necked 250 mL round-bottom reactor equipped with a condenser, mechanical stirrer and gas inlet. KPS (0.11 g) was added and the mixture was

stirred and degassed with argon for 20 min, and allowed to react under constant stirring for 4 h at 80°C in argon atmosphere. After cooling to room temperature, a solid content of 5% of the water-dispersed particles was obtained by gravimetric analysis.

PBMA-2-NP preparation by emulsion polymerization: In a three-necked 500 mL roundbottom reactor equipped with a condenser, mechanical stirrer and gas inlet, PBMA seed latex mixture (0.163 g) was diluted with Millipore water (70 mL). The mixture was stirred and degased with a continuous argon flow for 20 minutes. After this time was heated to 80 °C and two different solutions (solution 1: SDS (0.70 g), KPS (70 mg) and Millipore water (70 g); solution 2: BMA (23 g) EGDMA (3.6 g) and compound **2** (73 mg)) were independently added using a flow controller to the reactor at a constant feed rate over 6 hours. After completing the addition, the mixture was allowed to react for more 2 hours. After this time the reaction was stopped and the mixture cooled to room temperature. After cooling to room temperature, a solid content of 20% of the water-dispersed particles was obtained by gravimetric analysis.

PEGA-2-NP preparation by emulsion polymerization: In a three-necked 500 mL roundbottom reactor equipped with a condenser, mechanical stirrer and gas inlet, PBMA seed latex mixture (16.28 g) was diluted with Millipore water (51.4 mL). The mixture was stirred and degased with a continuous argon flow for 20 minutes. After this time was heated to 80 °C and two different solutions (solution 1: SDS (0.40 g), KPS (40 mg) and Millipore water (15.5 g); solution 2: BMA (13.16 g) EGDMA (1.78 g), compound **2** (75 mg), di(ethylene glycol) methyl ether methacrylate (DEA, 13.55 g) and poly(ethylene glycol) methyl ether methacrylate Mn = 475 (OEGA, 2.97 g) were independently added using a flow controller to the reactor at a constant feed rate over 6 hours. After completing the addition, the mixture was allowed to react for more 2 hours. After this time the reaction was stopped and the mixture cooled to room temperature. After cooling to room temperature, a solid content of 25% of the water-dispersed particles was obtained by gravimetric analysis.

PS-3-NP preparation by miniemulsion polymerization: an aqueous solution of sodium dodecyl sulfate (SDS, 1.16 g), potassium persulfate (KPS, 0.136 g) and NaHCO3 (28 mg) in 53.33 g millipore water and an organic solution of styrene (S, 18 g), compound **3** (36 mg), hexadecane (0.94 g) and divinylbenzene (DVB, 0.22 g) were magnetically stirred for 30 min. The resulting emulsion was then sonicated (Branson 250/450 digital sonifier with micro-tip 1/8, with 50% amplitude and 1 s pulse on/2 s pulse-off) in an ice bath for 120 s. The resulting miniemulsion was added to a three-necked 250 mL round-bottom reactor equipped with a condenser, mechanical stirrer and gas inlet. The mixture was stirred and degassed with argon for 20 min. and allowed to react under constant stirring for 8 h at 70°C in argon atmosphere. After cooling to room temperature, a solid content of 28% of the water-dispersed particles was obtained by gravimetric analysis.

Endoperoxide formation and thermolysis: The functionalized nanoparticles were irradiated in a quartz cuvette (1×1 cm). In a typical procedure, nanoparticles were diluted to a 0.6 wt% with water and methylene blue was added to obtain a concentration of 3.5×10^{-6} M. The mixture was irradiated with a mercury lamp with continuous oxygen bubbling. The 546 nm peak (region where only methylene blue absorbs) was isolated with optical filters. The photo-

oxidation was monitored by fluorescence spectroscopy. After irradiation, methylene blue was removed from the solution by adding 5% (v/v) of an ionic change resin (Bio-Rad AG 501- X8 20-50 mesh). The mixture was gently stirred for 2 hours and the resin removed by filtration. The nanoparticles were stable in water dispersion for at least several weeks. For oxygen release, the dispersion were transferred to capped vials and placed in a ventilated heating oven at 90 °C. The thermolysis was monitored by recording the fluorescence emission spectra with the sample at room temperature.

Photophysics data of compounds 2 and 3



Figure S1. Normalized absorption and fluorescence emission spectra of 1 (solid lines), 2 (dashed lines) and 3 (dotted lines) in toluene.

Sample	λ_{abs}^{max} (nm)	λ_{em}^{max} (nm)	$ au_F$ (ns) ^a	
			non-degassed	degassed
DPA 1	339; 355; 374; 394	407; 429	5.8	7.9
Compound 2	339; 356; 374; 395	410; 430	5.8	7.9
Compound 3	338; 353; 372; 392	425; 441	2.9	

Table S1. Photophysical properties of compounds 2, 3 and pristine DPA (1) in toluene.

^a single exponential.

Microscopy characterization of PEGA-2-NP



Figure S2. TEM (A, 100 kV) and SEM (B, 15 kV) images of PEGA-2-NP nanoparticles.



Figure S3. AFM image of an isolated PEGA-2-NP nanoparticle.



Figure S4. NTA results of a **PEGA-2-NP** dispersion, showing a representative video frame of the measurements (A), the particle diameter distribution (B) and a representation of number of particles (vertical axis) as a function of particle size and relative particle scattering intensity (C), showing that there is only one particle population, with no significant aggregation.



Figure S5. Normalized fluorescence spectra in water of **PS-3-NP** (solid line), **PBMA-2-NP** (dotted line) and **PEGA-2-NP** (dashed line) nanoparticles.

Table	S2.	Photophysical	properties	of	nanoparticles	PBMA-2-NP,	PEGA-2-NP	and	PS-3-
NP in	wate	er dispersion.							

Sample	DPA derivative	λ_{em}^{max} (nm)	$ au_F$ (ns)		
			non-degassed	degassed	
PBMA-2-NP	2	409; 427	8.3 ^a	8.2 ^a	
PEGA-2-NP	2	410; 428	9.1 ^a	8.8 ^a	
PS-3-NP	3	418	6.5 ^b	6.6 ^b	

^a single exponential; ^b average from a biexponential fitting: 7.6 ns (73%) and 3.5 ns (27%).



Figure S6. Time dependence of the fluorescence intensity (407 nm) at 90°C of **PBMA-2-NP** (circles), **PEGA-2-NP** (squares) and **PS-3-NP** (triangles).



Figure S7. Variation of the diameter of **PEGA-2-NP** with temperature, measured by dynamic light scattering. The sequence was: 20°C, 40°C, 60°C, 75°C, 60°C, 20°C.

Endoperoxide formation and thermolysis

The functionalized nanoparticles were irradiated in a quartz cuvette $(1 \times 1 \text{ cm})$. In a typical procedure, nanoparticles were diluted to a 0.6 wt% with water and methylene blue was added to obtain a concentration of 3.5×10^{-6} M. The mixture was irradiated with a mercury lamp with continuous oxygen bubbling. The 546 nm peak (region where only methylene blue absorbs) was isolated with optical filters. The photo-oxidation was monitored by fluorescence spectroscopy. After irradiation, methylene blue was removed from the solution by adding 5% (v/v) of an ionic change resin (Bio-Rad AG 501- X8 20-50 mesh). The mixture was gently stirred for 2 hours and the resin removed by filtration. The nanoparticles were stable in water dispersion for at least several weeks. For oxygen release, the dispersion were transferred to capped vials and placed in a ventilated heating oven at 90 °C. The thermolysis was monitored by recording the fluorescence emission spectra with the sample at room temperature.



Figure S8. Time dependence at room temperature of the fluorescence intensity (407 nm) of **PBMA-2-NP** after irradiation.



Figure S9. Time dependence of the fluorescence intensity (407 nm) of **PBMA-2-NP** during two irradiation (green arrows) / thermolysis at 90 °C (red arrows) cycles.

Estimation of the maximum singlet oxygen concentration delivered upon thermolysis of 9,10-diphenylanthracene endoperoxides

Considering the following kinetic scheme

$$DPA - O_2 \xrightarrow{k} DPA + {}^{1}O_2$$
$${}^{1}O_2 \xrightarrow{1/\tau} {}^{3}O_2$$

Application of the quasi-steady-state approximation to singlet oxygen gives

$$[^{1}\mathbf{O}_{2}]_{max} = k \tau [DPA - O_{2}]_{0}$$

Studies performed with 9,10-diphenylanthracene in dimethylformamide at 90
^oC

 $k = 9 \times 10^{-5} \text{ s}^{-1}$, rate constant for the thermolysis of 9,10-diphenylanthracene endoperoxides in dimethylformamide at 90 °C.

 $\tau = 17.9 \ \mu s$, singlet oxygen lifetime in dimethylformamide, obtained from ref. [S2].

 $[DPA] = 4 \times 10^{-5} \text{ M of } 9,10$ -diphenylanthracene in dimethylformamide.

$$[{}^{1}\mathbf{0}_{2}]_{max} = 9 \times 10^{-5} \, s^{-1} \times 17.9 \times 10^{-6} \, s \times 4 \times 10^{-5} \, M$$
$$[{}^{1}\mathbf{0}_{2}]_{max} = 6.4 \, \times 10^{-14} \, M$$

• Studies performed with polymer nanoparticles (PBMA-2-NP) functionalized with 9,10-diphenylanthracene endoperoxides at 90 °C in water

$$[{}^{1}\mathbf{O}_{2}]_{max} = k \tau [np] n E P$$

 $k = 1 \times 10^{-4} \text{ s}^{-1}$, rate constant for the thermolysis of 9,10-diphenylanthracene endoperoxides in the polymer nanoparticles at 90 °C.

 $\tau = 4.4 \ \mu s$, singlet oxygen lifetime in water, obtained from ref. [S3].

Nanoparticles concentration - $[np] = 6.9 \times 10^{11}$ nanoparticles per mL of emulsion = 6.9×10^{14} nanoparticles per L of emulsion.

 $nEP = 5.8 \times 10^{-20}$ mol of endoperoxide per nanoparticle or 35 000 endoperoxide molecules per nanoparticle.

$$\begin{bmatrix} {}^{1}\text{O}_{2} \end{bmatrix}_{max} = 1 \times 10^{-4} \, s^{-1} \times 4.4 \times 10^{-6} \, s \times 6.9 \times 10^{14} \frac{np}{L} \times 5.8 \times 10^{-20} \, mol/np$$
$$\begin{bmatrix} {}^{1}\text{O}_{2} \end{bmatrix}_{max} = 1.8 \, \times 10^{-14} \, M$$

References

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Figure S1-a: ¹H NMR spectrum of compound 5.





Figure S1-b: ¹³C NMR spectrum of compound **5**.



Figure S1-c: HRMS spectrum of compound 5.



Figure S2-a: ¹H NMR spectrum of compound **6**.



Figure S2-b: ¹³C NMR spectrum of compound **6**.



Figure S2-c: HRMS spectrum of compound 6.



Figure S3-a: ¹H NMR spectrum of compound 2.



Figure S3-b: ¹³C NMR spectrum of compound **2**.



Figure S3-c: HRMS spectrum of compound 2.



Figure S4-b: ¹³C NMR spectrum of compound 3