Electronic Supplementary Information

Photoactivated polyprodrug nanoparticles for effective light-controlled Pt(IV) and siRNA codelivery to achieve synergistic cancer therapy

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Synthesis of *trans,trans,trans*-[$Pt(N_3)_2(OH)_2(py)_2$] (Pt(IV))

Pt(IV) was synthesized according to the previous reference.[1] ^{1}H NMR (400 MHz, DMSO-d_6, ppm): 9.08 (a, NCH), 8.26 (c, CH), 7.86 (b, CH). ^{13}C NMR (400 MHz, DMSO-d_6, 298 K) δ (ppm): 149.31 (a, NCH), 142.19 (c, CH), 126.48 (b, CH). IR (cm^{-1}): 3600-3160 (br, NH_3 and OH), 3070 (sh, CH), 2040 (sh, N_3), 550 (sh, Pt-OH). ESI-MS (m/z): calculated for Pt(IV): 471.07; Found for $[Pt(IV) + Na]^+$: 494.2, $[2Pt(IV) + Na]^+$: 965.1. Element analysis calculated for $C_{10}H_{12}N_8O_2Pt$: C 25.46, H 2.54, N 23.76; Found C 25.36, H 2.61, N 23.52. ICP-OES analysis calculated for Pt 41.37; Found 41.30. **Caution!* Although there were no problems ened during this process, heavy metal azides were heat and shock-sensitive detonators. Therefore, any platinum azide compounds must be handled carefully.*

Synthesis of FITC-labeld PP (FITC-PP)

Fluorescein isothiocyanate (FITC) (15 mg, 0.039 mmol) and 100 mg of PP were dissolved in DMSO (5 mL) and stirred for 24 h at room temperature in the dark. Then, the solution was dialyzed (MwCO = 7000) against distilled water for 48 h to remove the unreacted FITC and freeze-dried for further use. The yield of FITC-PP was about 75%.

Cell culture

Ovarian cancer cell line A2780 cell line was purchased from institute of biochemistry and cell biology, Chinese Academy of Sciences, Shanghai, China. The cells were cultured in a humidified atmosphere containing 5% CO_2 at 37 °C using DMEM medium supplemented with 10% fetal bovine serum, 60 mg/mL penicillin and 100 mg/mL streptomycin.

Combination index analysis of Pt and siBcl-2

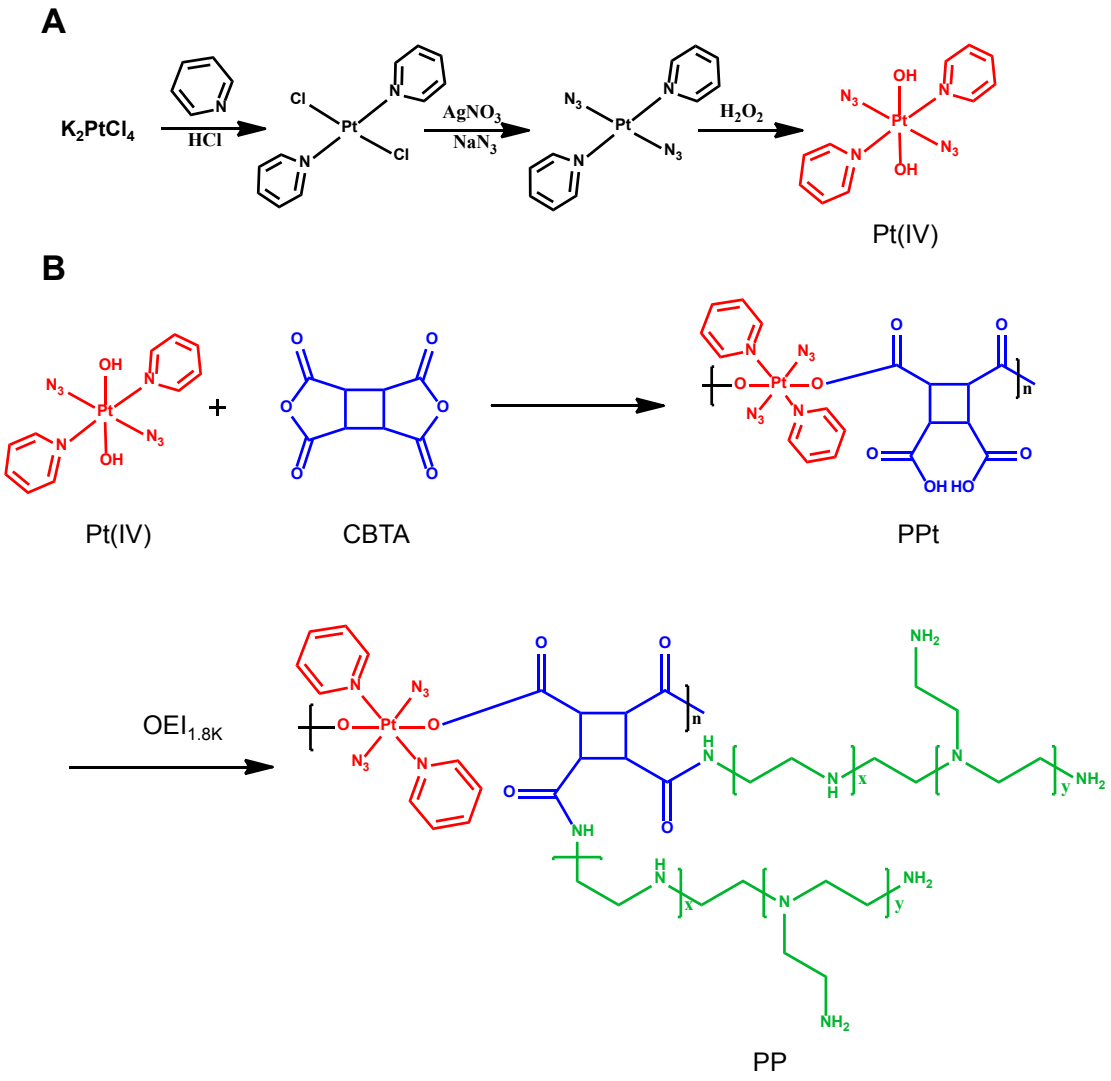
Combination index (CI) was calculated with the equation as below:

$$\mathrm{CI} = \frac{\mathrm{Ca,x}}{\mathrm{ICx,a}} + \frac{\mathrm{Cb,x}}{\mathrm{ICx,b}}$$

$C_{a,x}$ and $C_{b,x}$ are the concentrations of drug A (Pt) and drug B (siBcl-2) used in combination to achieve x% drug effect under irradiation. $IC_{x,a}$ and $IC_{x,b}$ are the concentrations for $PPNP_{siBcl\text{-}2}$ under irradiation and in dark condition to achieve the same effect, respectively. The CI values lower than, equal to, and higher than 1 denote synergism, additivity, and antagonism, respectively.

References

(1) Farrer, N. J.; Woods, J. A.; Salassa, L.; Zhao, Y.; Robinson, K. S.; Clarkson, G.; Mackay, F. S.; Sadler, P. J. A potent trans-diimine platinum anticancer complex photoactivated by visible light. *Angew. Chem. Int. Ed.* **2010**, 49, 8905–8908.



Scheme S1. Synthesis routs and chemical structures of (A) Pt(IV) and (B) PP.

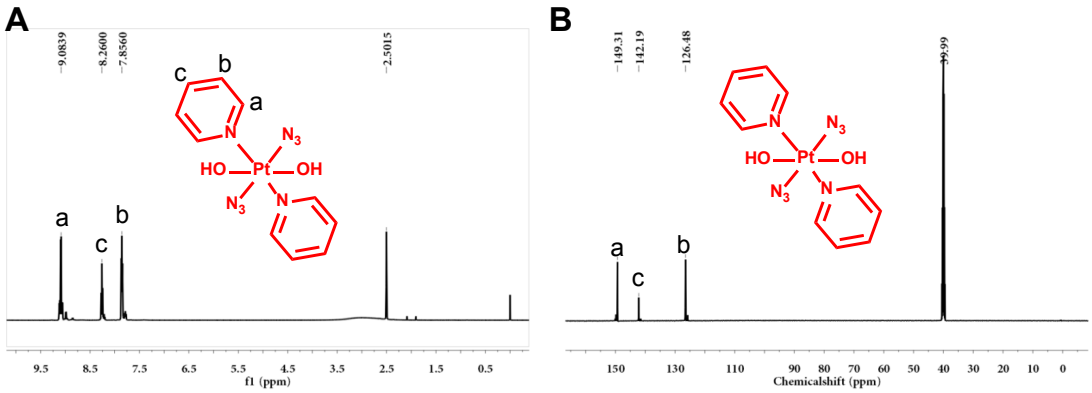


Fig. S1. Characterization of Pt(IV) by NMR. (A) ^{1}H NMR spectra in d_6-DMSO. (B) ^{13}C NMR spectra in d_6-DMSO.

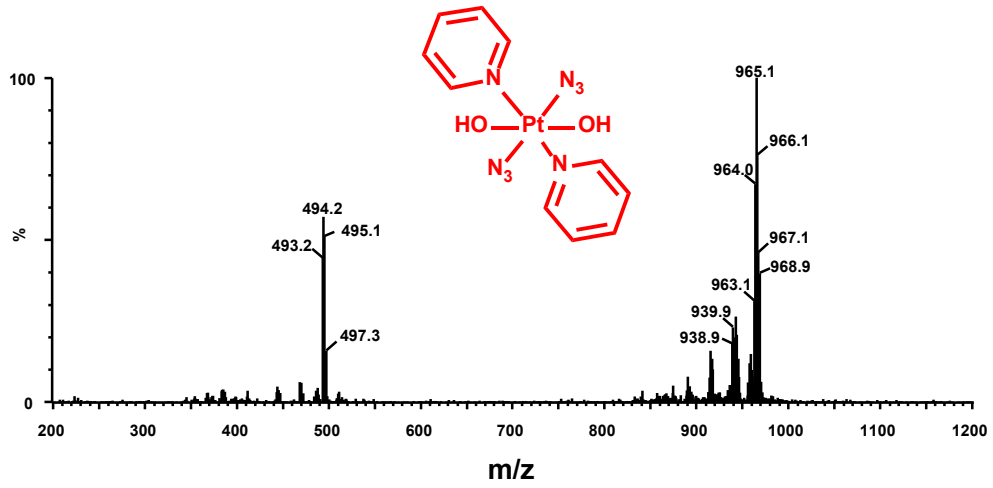


Fig. S2. Characterization of Pt(IV) by ESI-MS.

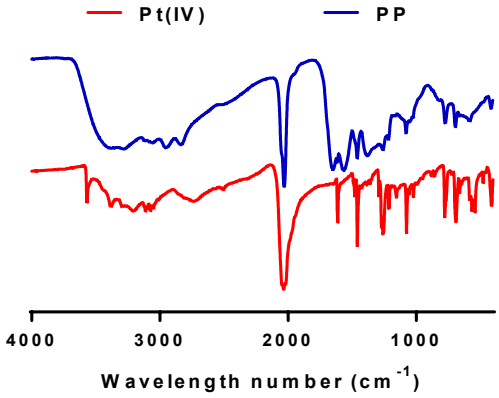


Fig. S3. FT-IR spectra of Pt(IV) and PP.

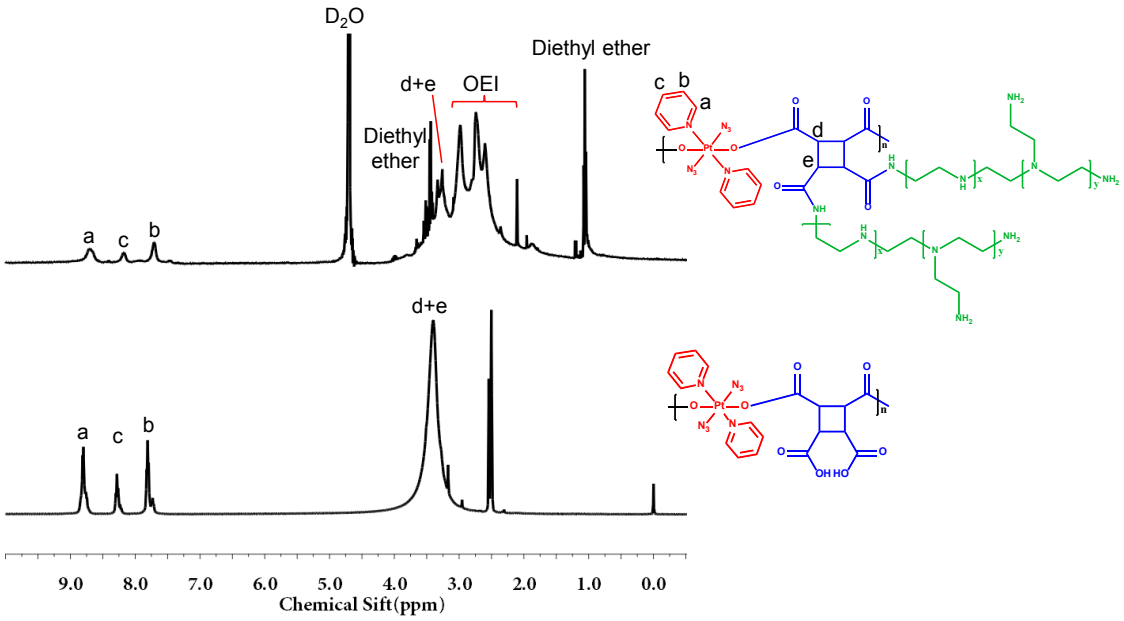


Fig. S4. Characterization of PPt and PP by ^{1}H NMR spectra in d^{6}-DMSO.

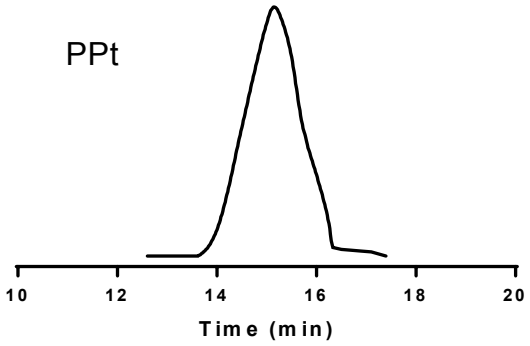


Fig. S5. GPC curve of PPt.

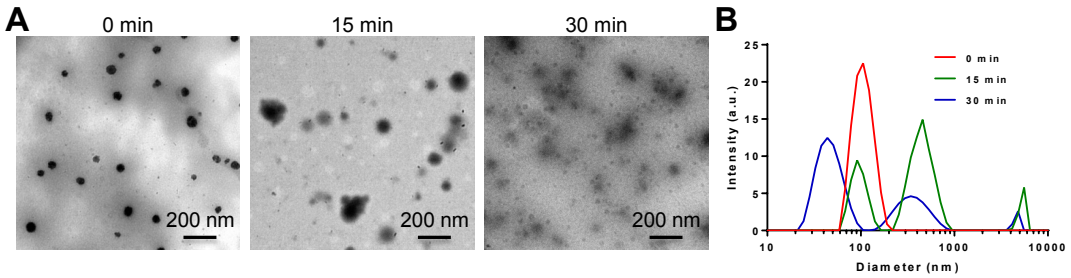


Fig. S6. (A) TEM images of PPNP after irradiation for indicated time intervals: 0 min, 15 min and 30 min. (B) The corresponding size distribution of PPNP after light irradiation. Light source: 530 nm, 20 mW/cm^{2}.

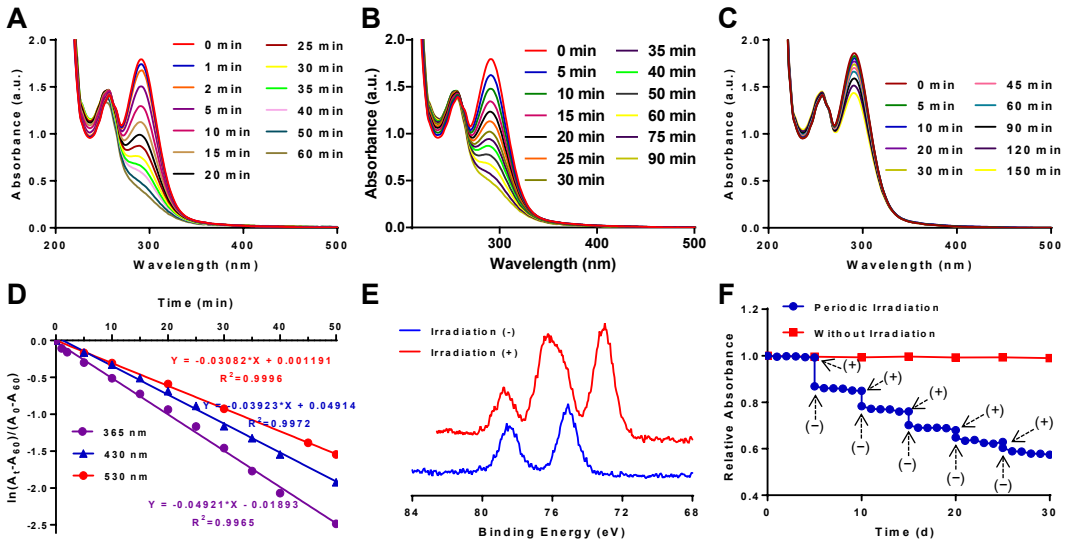


Fig. S7. Photosensitivity of Pt(IV). UV-vis spectra changes of Pt(IV) with different wavelength light irradiation for indicated time intervals: (A) 365 nm, (B) 430 nm and (C) 530 nm. (D) The first order kinetics of Pt(IV) degradation with visible light irradiation. (E) XPS changes of Pt(IV) before and after irradiation. (F) UV-vis absorption of Pt(IV) at 295 nm upon periodic irradiation (irradiation was turned on for 5 min then turned off for 5 days). "–" indicated in the dark, "+" indicated with irradiation. Light source: 530 nm, 20 mW/cm^2.

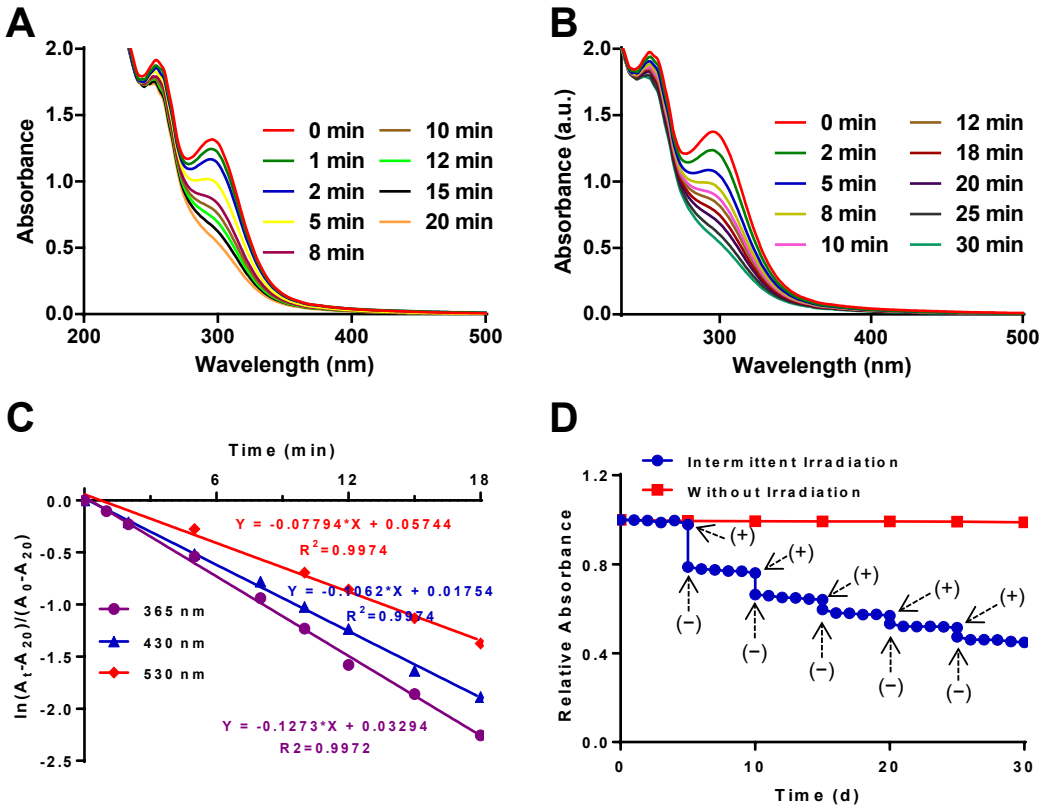


Fig. S8. Photosensitivity of PP. UV-vis spectra changes of PP irradiated with different wavelengths of light for indicated time intervals: (A) 365 nm and (B) 430 nm. (C) The first order kinetics of PP degradation with visible light irradiation. (D) UV-vis absorption of PP at 295 nm upon periodic irradiation (irradiation was turned on for 5 min then turned off for 5 days). "–" indicated in the dark, "+" indicated with irradiation. Light source: 530 nm, 20 mW/cm^2.

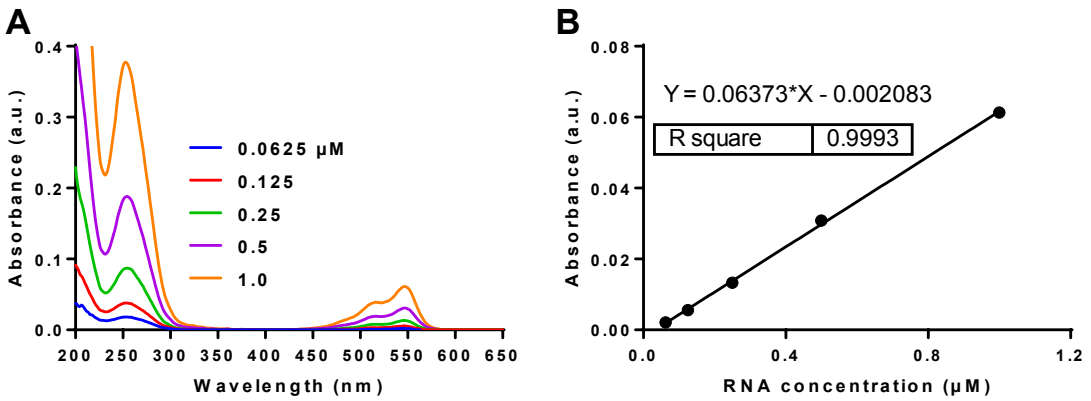


Fig. S9. (A) UV-vis spectrum of the different concentration of Cy3-siRNA. (B) The standard curve of Cy3-siRNA at 547 nm.

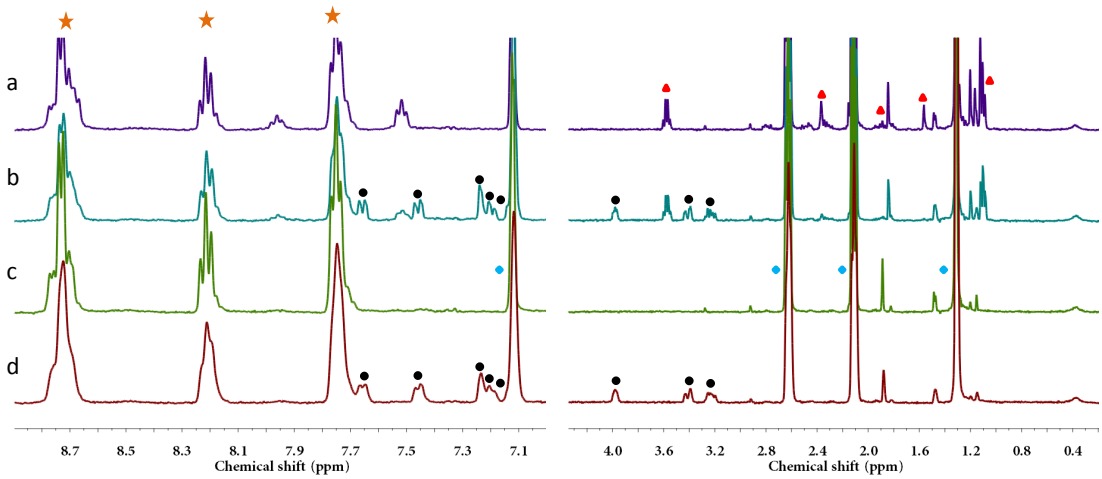


Fig. S10. Detection of azidyl radicals generated from Pt(IV) by ^{1}H NMR spectra. ^{1}H NMR spectra of Pt(IV) and DMPO in D_2O: (a, c) in the absence of Trp, (b, d) in the presence of Trp , (a, b) irradiation for 30 min, (c, d) under dark condition. ★, ^{1}H peaks of Pt(IV); ◆, ^{1}H peaks of DMPO; ●, ^{1}H peaks of Trp; ▲, ^{1}H peaks of photoproducts of DMPO and azidyl radicals. Light source: 530 nm, 20 mW/cm^2.

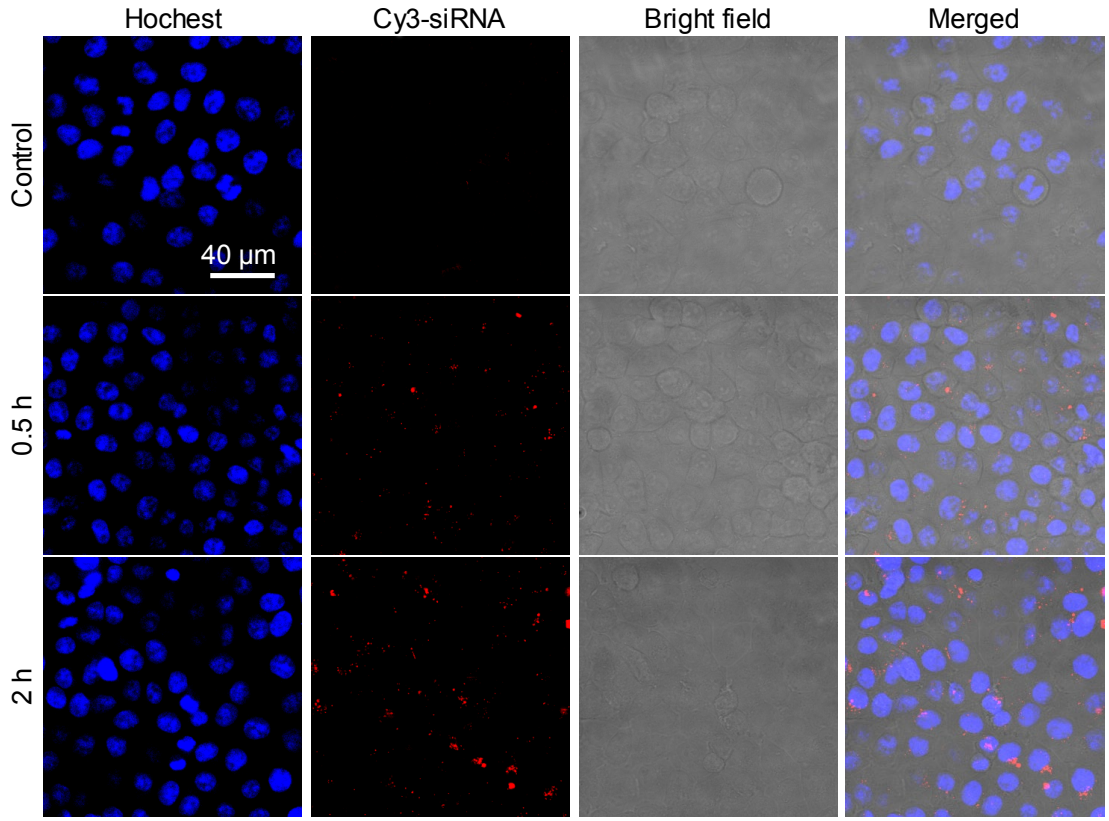


Fig. S11. CLSM images of A2780 cells after incubation with $PPNP_{Cy3\text{-}siRNA}$ for different time intervals.

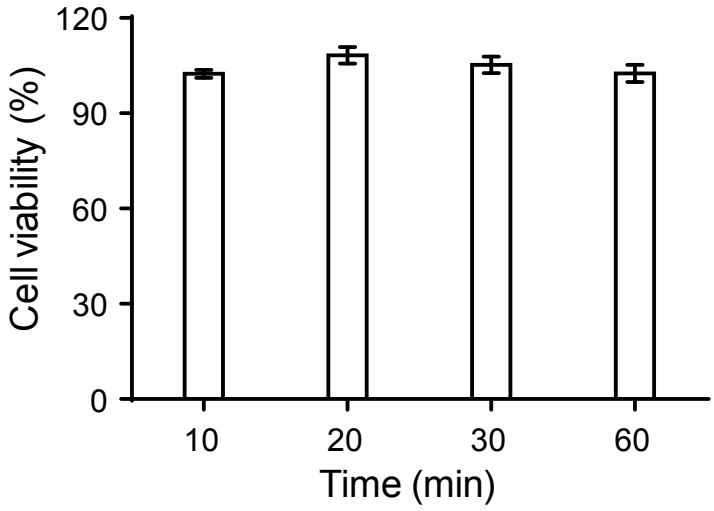


Fig. S12. A2780 cells were irradiated for different time intervals and further incubated for 72 h (530 nm, 20 mW/cm^2).

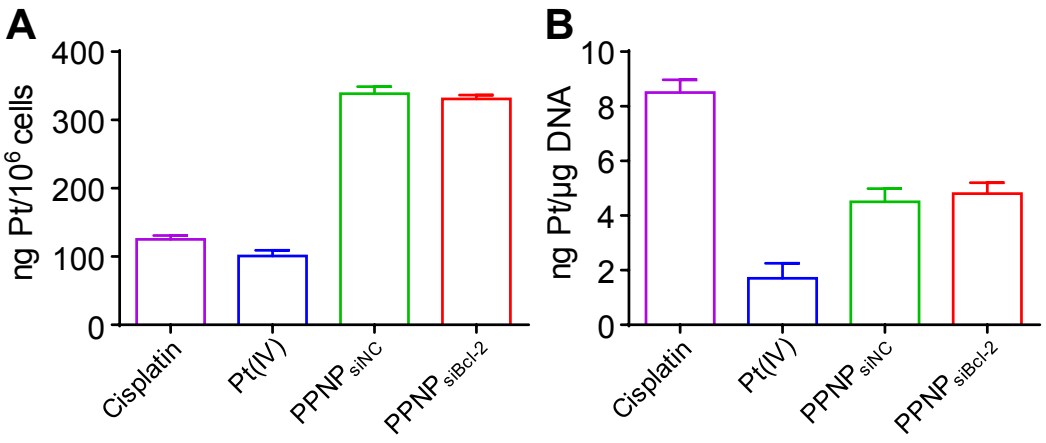


Fig. S13. (A) Pt accumulation and (B) Pt-DNA adducts in A2780 cells after treatment with different drugs for 4 h in dark.

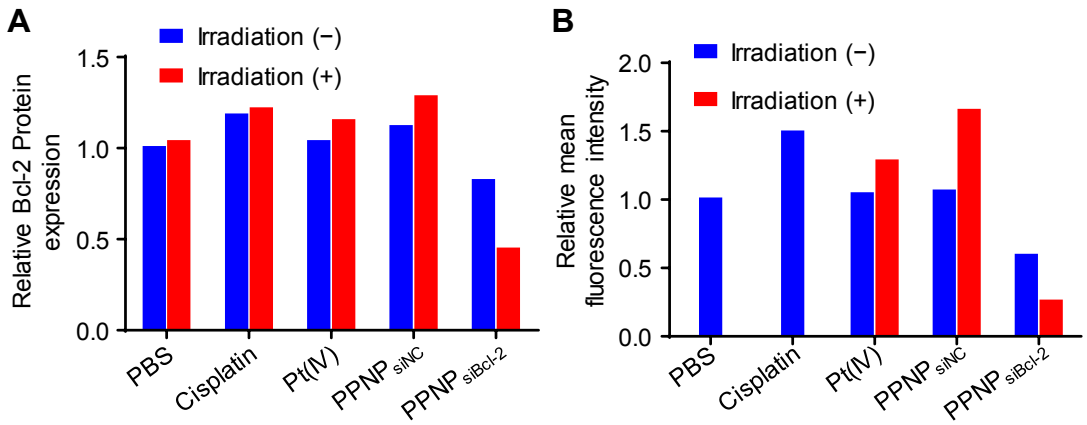


Fig. S14. (A) Relative Bcl-2 protein expression and (B) relative mean fluorescence intensity of A2780 cells after treatment with different drugs. "−" indicated in the dark, "+" indicated with irradiation.

Table S1. Elemental analysis of Pt(IV)

	Pt(IV)
C	25.36[a]/25.46[b]
H	2.61[a]/2.54[b]
N	23.52[a]/23.76[b]

[a]Found Value;[b]Calculated Value

Table S2. ICP analysis of Pt content in Pt(IV), PPt and PP.

	Pt(IV)	PPt	PP
Pt/wt%	41.30[a]/41.37[b]	28.82[a]/29.21[b]	10.76[a]/12.44[b]

[a]Found Value;[b]Calculated Value

Table S3. GPC characterization of PPt and PP.

Polymer	$M_n/10^4$	$M_w/10^4$	PDI	DP	$OEI_{1.8K}$ (Number)
PPt	1.38	1.67	1.21	21	/
PP	3.24	4.34	1.34	/	10

[a]Found Value;[b]Calculated Value