

iScience, Volume 27

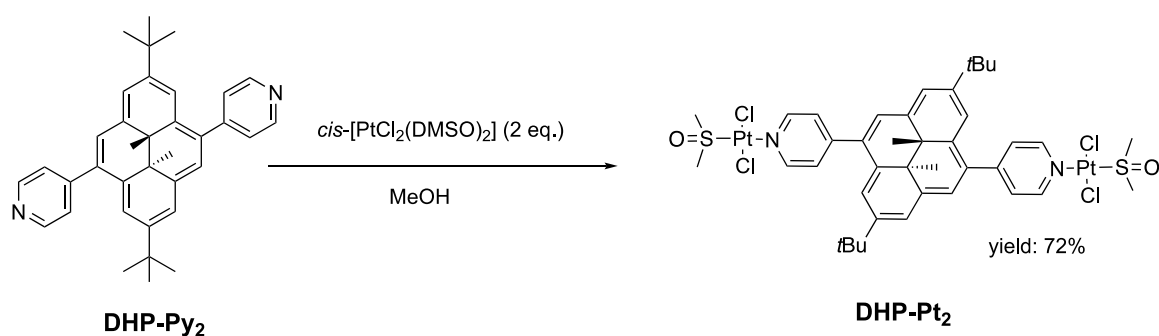
Supplemental information

**Optical modulation of cell nucleus penetration
and singlet oxygen release
of a switchable platinum complex**

Zakaria Ziani, Saioa Cobo, Nathalie Berthet, and Guy Royal

List of Figures, Schemes and Tables

| | |
|--|----|
| Scheme S1. Preparation of DHP-Pt₂ from DHP-Py₂ | 2 |
| Table S1. Crystal Data and Structure Refinement for DHP-Pt₂ | 2 |
| Table S2. IC50 values (μM) for DHP-Pt₂ and CPDO₂-Pt₂ platinum complexes and cisplatin..... | 4 |
| Figure S1. Emission spectrum of DHP-Pt₂ in degassed CD ₂ Cl ₂ | 3 |
| Figure S2. Phosphorescence band of singlet oxygen | 3 |
| Figure S3. Agarose gel electrophoresis images..... | 4 |
| Figure S4. Fluorescence intensity profiles | 5 |
| Figure S5. Thermal back isomerization of CPDO₂-Pt₂ to DHP-Pt₂ and ¹ O ₂ release in tumorous cells..... | 6 |
| Figure S6. ¹ H-NMR spectra of DHP-Pt₂ in CD ₂ Cl ₂ | 7 |
| Figure S7. ¹³ C-NMR spectra of DHP-Pt₂ in CDCl ₃ | 7 |
| Figure S8. Evolution of the ¹ H-NMR spectra of DHP-Pt₂ during illumination | 8 |
| Figure S9. ¹ H-NMR spectra of CPDO₂-Pt₂ | 9 |
| Figure S10. Mass spectra of DHP-Pt₂ i | 10 |
| Figure S11. Mass spectra of CPDO₂-Pt₂ | 10 |



Scheme S1. Preparation of **DHP-Pt₂** from **DHP-Py₂** related to Figure 1.

Table S1. Crystal Data and Structure Refinement for **DHP-Pt₂** related to Figure 1.

| | |
|--|--|
| Formula | C ₄₀ H ₅₀ Cl ₄ N ₂ O ₂ Pt ₂ S ₂ |
| Fw (g mol⁻¹) | 1186.92 |
| Crystal system | Monoclinic |
| Space group | P2 ₁ /c |
| a (Å) | 5.8439(12) |
| b (Å) | 28.182(6) |
| c (Å) | 13.367(3) |
| α (deg.) | 90 |
| β (deg.) | 99.61(3) |
| γ (deg.) | 90 |
| V (Å³) / Z | 2170.6(8) |
| D_x (g cm⁻³) | 1.816 |
| μ (cm⁻¹) | 6.816 |
| Crystal dim. (mm) | 0.10 x 0.27 x 0.38 |
| T (K) | 200 |
| θ range for coll. (deg.) | 3.091-27.000 |
| nb. of rflns. coll. | 24281 |
| Data/restraints/parameters | 3802/0/241 |
| R (I)^a all/R[I > 2σ(I)] | 0.0585/0.0405 |
| Goodness of fit S | 1.122 |
| Δρ_{min}/ Δρ_{max} (e Å⁻³) | -1.047/2.380 |

$$^a R = \sum ||f_o| - |f_c|| / \sum |f_o|.$$

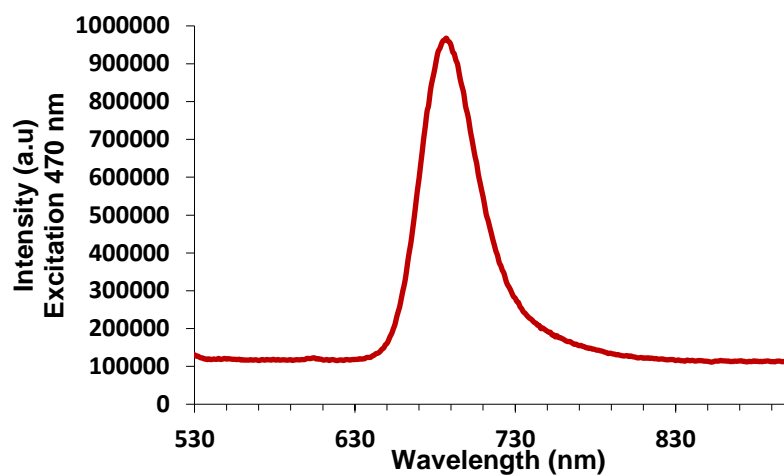


Figure S1. Emission spectrum of **DHP-Pt₂** in degassed CD₂Cl₂ related to Figure 5.

Excitation wavelength: 470 nm.

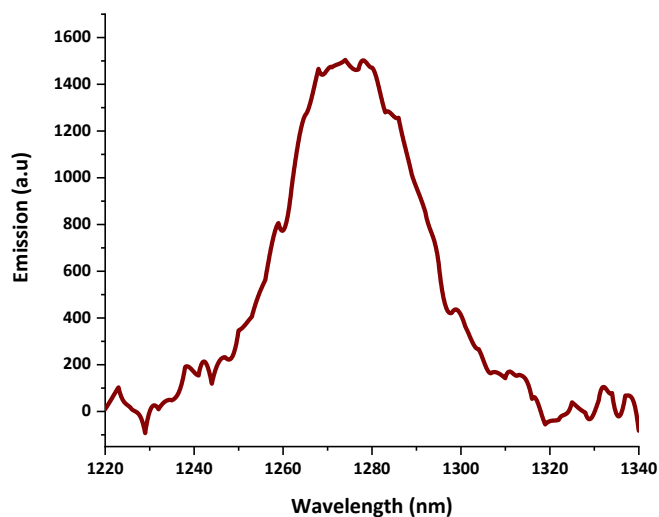


Figure S2. Phosphorescence band of singlet oxygen related to Figure 6.

Phosphorescence band of singlet oxygen characteristic of the deexcitation of the metastable singlet oxygen (¹O₂) to its triplet ground state (³O₂) of a solution of **DHP-Pt₂** in dichloromethane at 25°C. OD = 0.1.

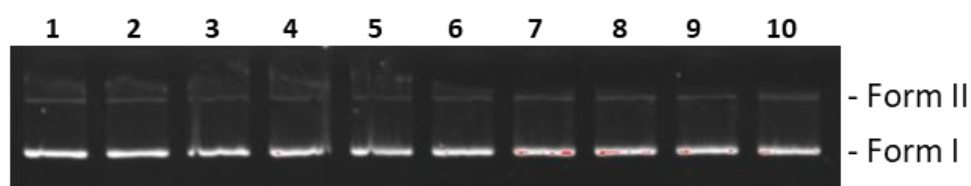


Figure S3. Agarose gel electrophoresis images

Agarose gel electrophoresis images of pBR322 DNA plasmid ($13 \mu\text{g}\cdot\text{mL}^{-1}$) incubated for 16h at 37°C with increasing concentrations of *bis*-pyridine **DHP-Py₂** ligand. Lane 1: pure plasmid DNA; Lanes 2-10: 0.09, 0.19, 0.39, 0.78, 1.56, 3.12, 6.25, 12.5, 25 μM of compound related to Figure 3.

Table S2. IC₅₀ values (μM) for **DHP-Pt₂** and **CPDO₂-Pt₂** platinum complexes and cisplatin related to Figure 4.

IC₅₀ values (μM) for **DHP-Pt₂** and **CPDO₂-Pt₂** platinum complexes and cisplatin with A-375, HeLa and HFF-1 cell lines after 24 h, 48 h and 72 h of incubation. Data show means \pm SD of three independent experiments.

| | IC ₅₀ (μM) | | | | | | |
|--|------------------------------------|----------------|----------------|----------------|----------------|----------------|-------|
| | A-375 | | | HeLa | | | HFF-1 |
| | 24h | 48 h | 72 h | 24h | 48 h | 72 h | 48h |
| DHP-Pt₂ | 67.0 ± 6.4 | 52.5 ± 5.4 | 45.5 ± 3.1 | 70.8 ± 7.2 | 59.8 ± 4.9 | 36.3 ± 4.1 | >100 |
| CPDO₂-Pt₂ | 67.6 ± 7.1 | 56.2 ± 4.8 | 46.1 ± 4.2 | 77.6 ± 6.8 | 61.9 ± 5.2 | 40.0 ± 5.1 | >100 |
| Cisplatin | 7.2 ± 1.6 | 2.9 ± 0.5 | 2.5 ± 0.8 | 4.5 ± 0.9 | 2.9 ± 0.6 | 2.7 ± 0.6 | Nd. |

Nd. Stands Not determined

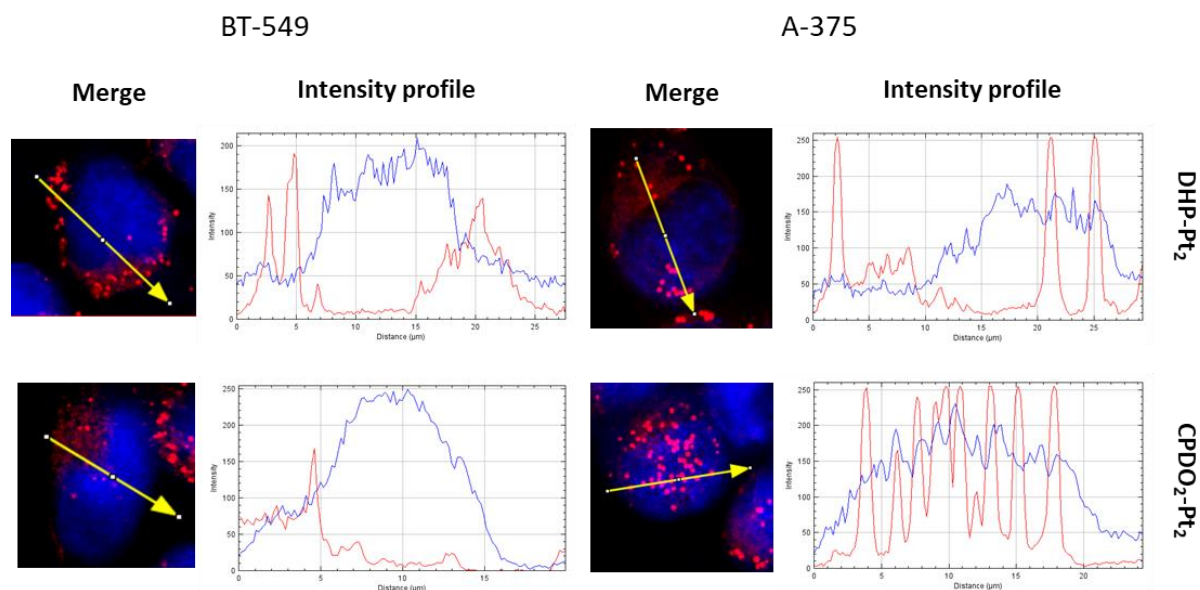


Figure S4. Fluorescence intensity profiles related to Figure 5.

Fluorescence intensity profiles for the both forms of the bis-platinum complex (**DHP-Pt₂** and **CPDO₂-Pt₂**, red) and the nuclear marker Hoechst 33342 (blue) in the cancer cell lines BT-549 and A-375 (right columns). The regions of the images which were analysed are shown with yellow arrows (left columns).

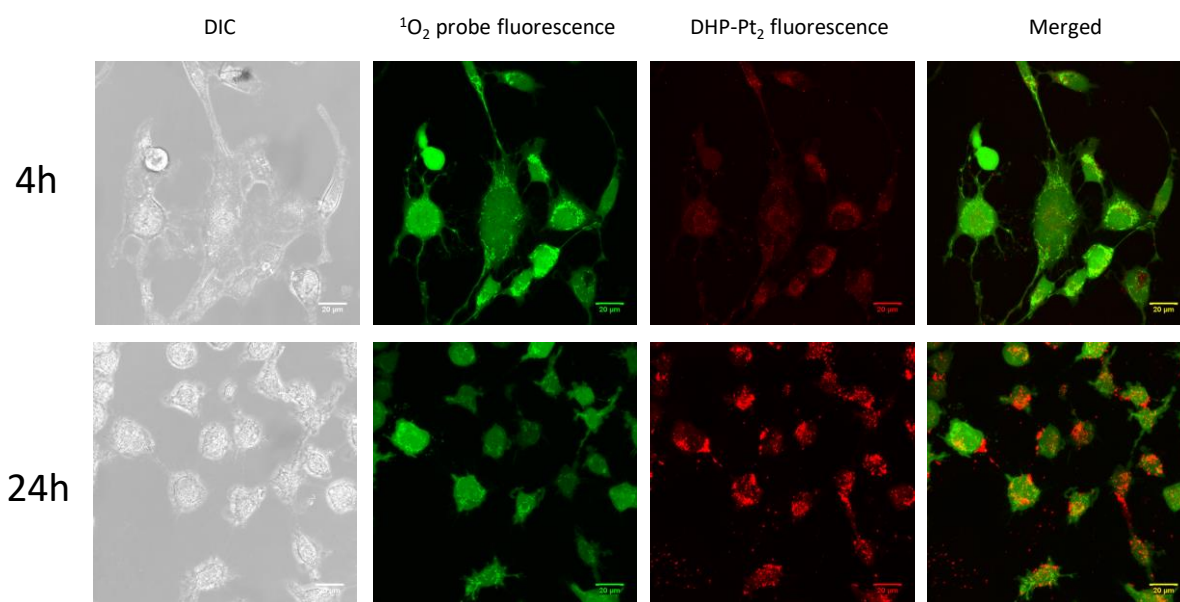


Figure S5. Thermal back isomerization of **CPDO₂-Pt₂** to **DHP-Pt₂** and $^1\text{O}_2$ release in tumorous cells related to Figure 6.

Confocal fluorescence microscope images of A-375 cells treated with 10 μM of **CPDO₂-Pt₂** for 4h and 24h at 37°C. Imaging of $^1\text{O}_2$ release from **CPDO₂-Pt₂** was followed using DCFH-DA as intracellular ROS probe. Scale bars are 20 μm .

At 4h and 24h a significant DCF fluorescence (green) is observed indicating a $^1\text{O}_2$ release from the **CPDO₂-Pt₂** complex in the tumorous cells. The fluorescence of the DCF observed is globally the same at 4h and 24h, indicating that the $^1\text{O}_2$ release step is finished within 4h under our experimental conditions. The increase in **DHP-Pt₂** fluorescence (in red) between microscopy images at 4 h and 24 h illustrates the thermal back isomerization (at $T = 37^\circ\text{C}$) between the initially non-emissive **CPDO₂-Pt₂** form and the fluorescent **DHP-Pt₂** form.

NMR data

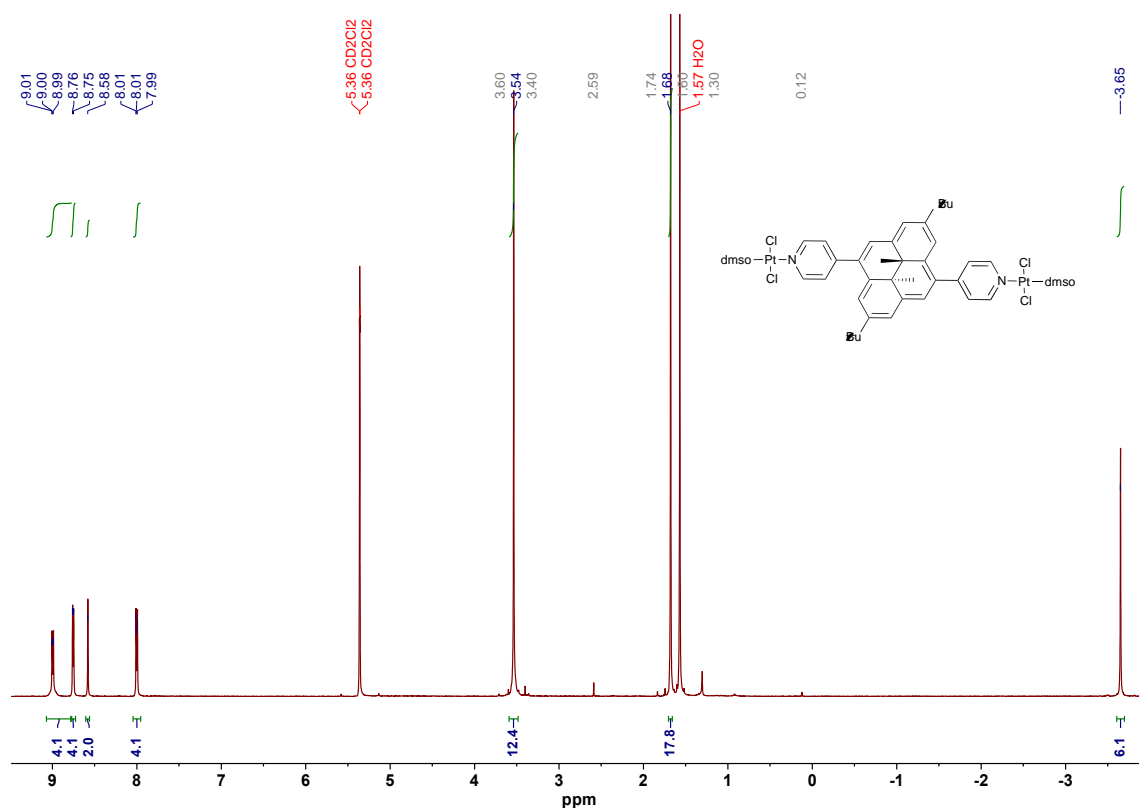


Figure S6. ¹H-NMR spectra of DHP-Pt₂ in CD₂Cl₂ related to Figure 1.

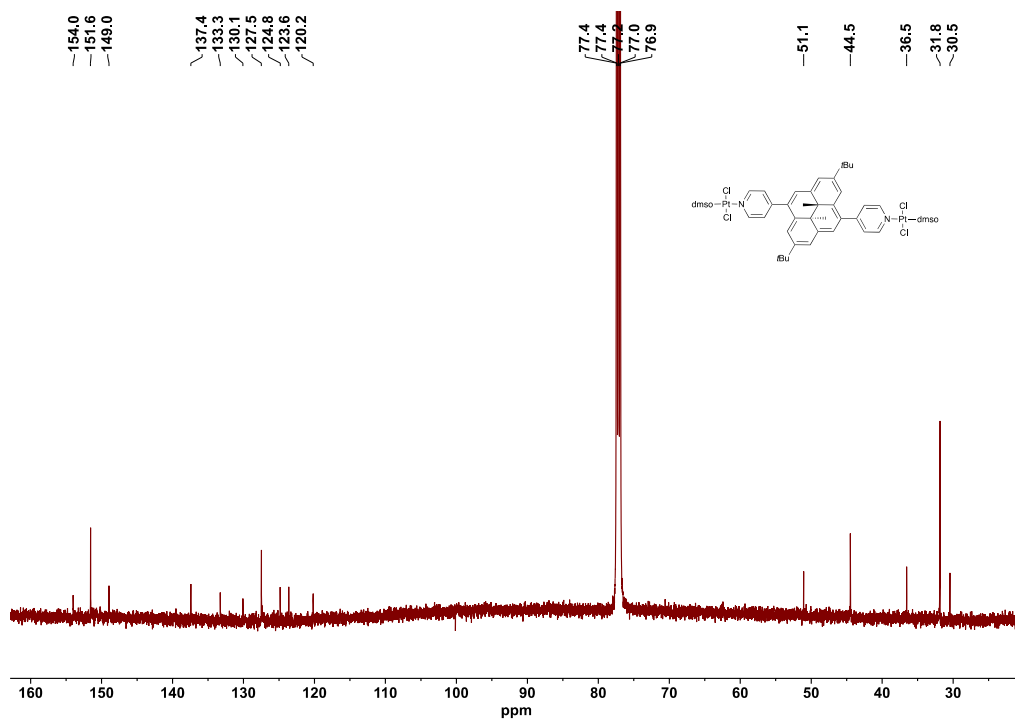


Figure S7. ¹³C-NMR spectra of DHP-Pt₂ in CDCl₃ related to Figure 1.

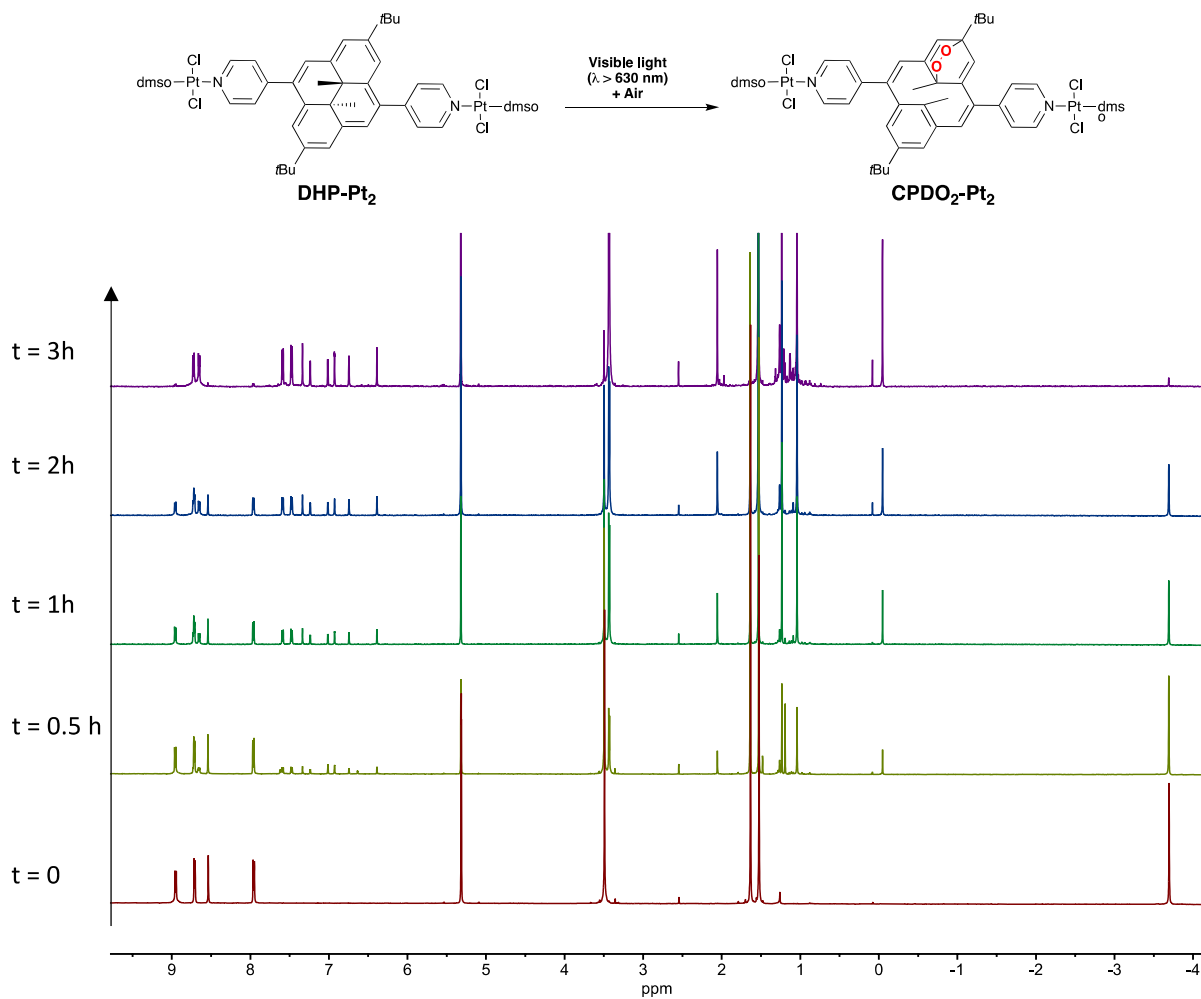


Figure S8. Evolution of the ¹H-NMR spectra of **DHP-Pt₂** during illumination

Evolution of the ¹H-NMR spectra of **DHP-Pt₂** during illumination with visible light ($\lambda > 630$ nm) under aerobic conditions (air, P = 1 atm.). Solvent: CD₂Cl₂ related to Figure 1.

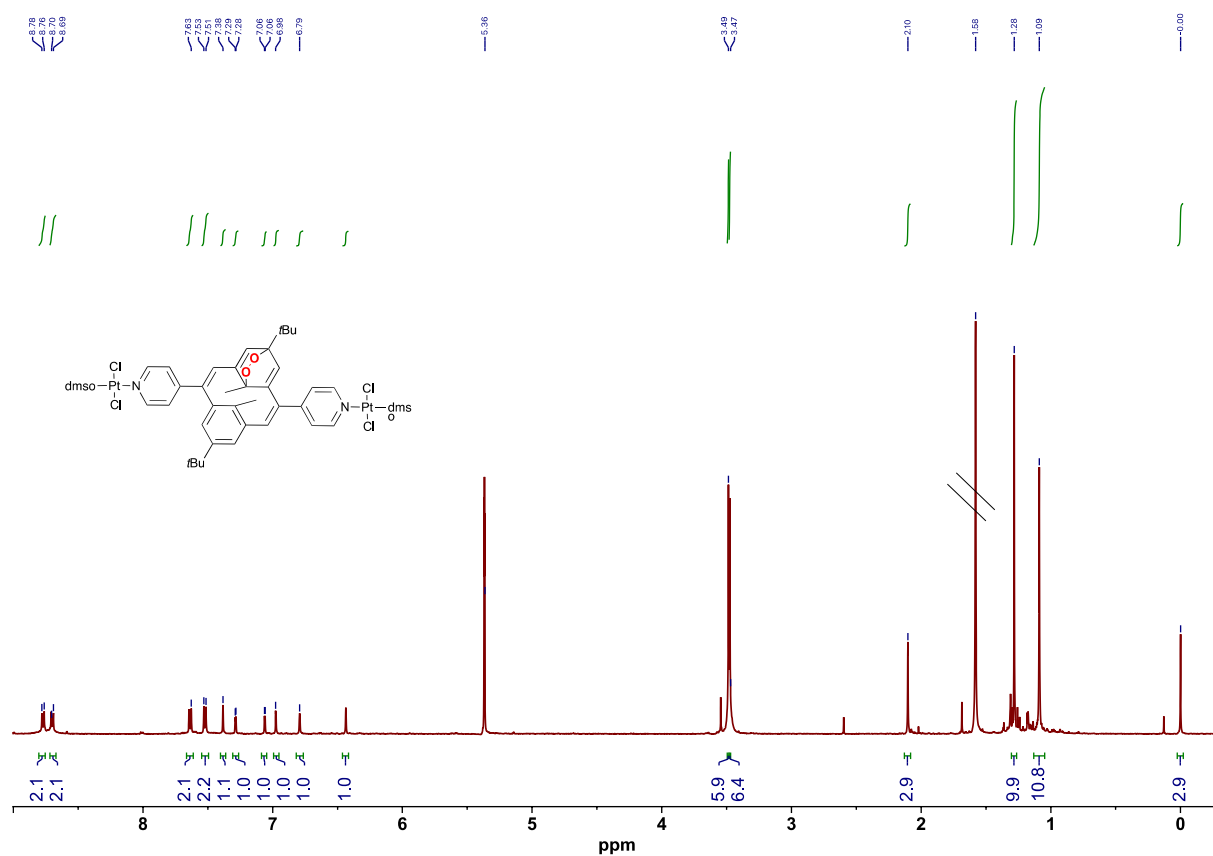


Figure S9. ¹H-NMR spectra of CPDO₂-Pt₂ related to Figure 1.

¹H-NMR spectra of CPDO₂-Pt₂ formed by illumination with red light of a solution of DHP-Pt₂ in CD₂Cl₂ (<5% of DHP-Pt₂ are remaining).

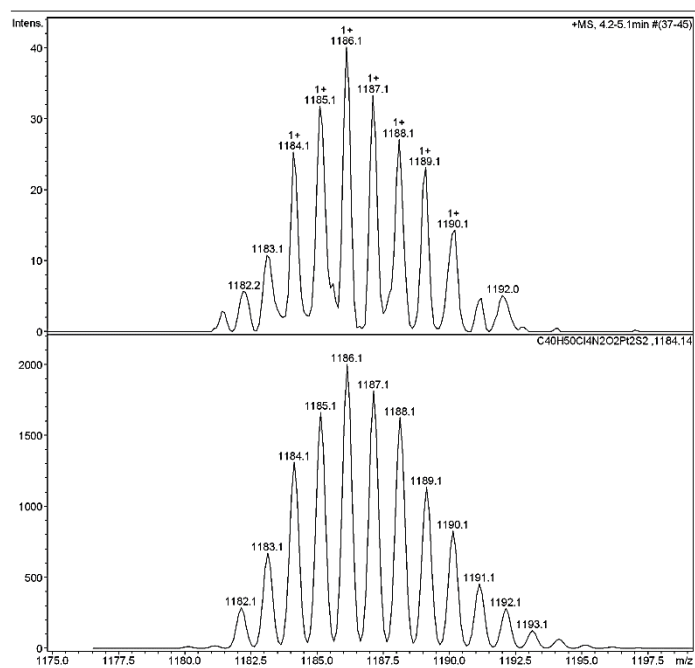


Figure S10. Mass spectra of **DHP-Pt₂** related to Figure 1.

Experimental (top) and calculated (down) electrospray ionization mass spectra of **DHP-Pt₂** in CH₂Cl₂.

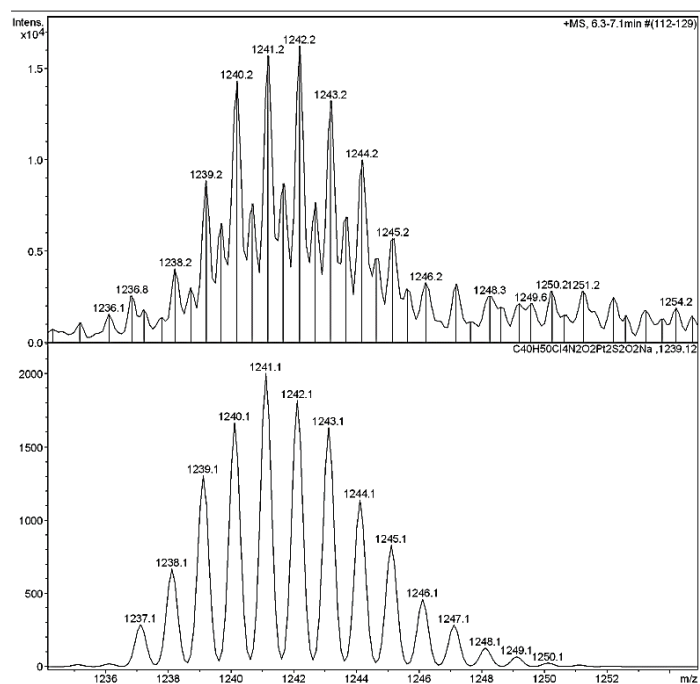


Figure S11. Mass spectra of **CPDO₂-Pt₂** related to Figure 1.

Experimental (top) and calculated (down) electrospray ionization mass spectra of **CPDO₂-Pt₂** in CH₂Cl₂.