Electronic supplementary information

A subset of new platinum antitumor agents kills cells by a multimodal mechanism of action also involving changes in the organization of the microtubule cytoskeleton

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Table S1. DNA platination in MDA-MB-231 cells treated for 6 h at 2.0 µM concentrations

	pg Pt/µg DNA[a]
1	1.97 ± 0.08
2	4.5 ± 0.5
3	5.9 ± 0.5
4	25 ± 4
5	0.24 ± 0.09
6	1.2 ± 0.1
7	1.67 ± 0.03
8	7 ± 1

[a]All results are expressed as the mean ± SD from three independent experiments.

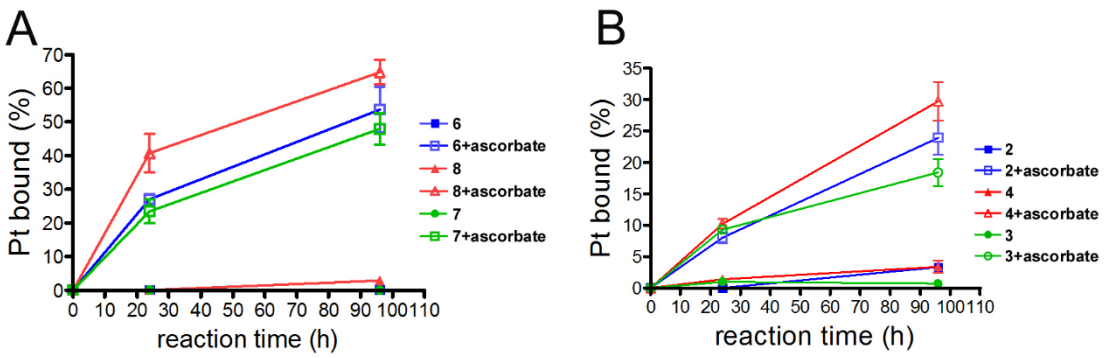

Figure S1. DNA binding of Pt(IV) complexes derived from **5** and **1** in the presence (open symbols) or absence (full symbols) of sodium ascorbate. Calf thymus DNA (64 µgmL^{-1}) was incubated with 2x10^{-5} M Pt(IV) derivatives of 56-MeSS (A) or OX (B) in 10 mM $NaClO_4$ at 37 °C in the presence or absence of 2x10^{-5} M sodium ascorbate.

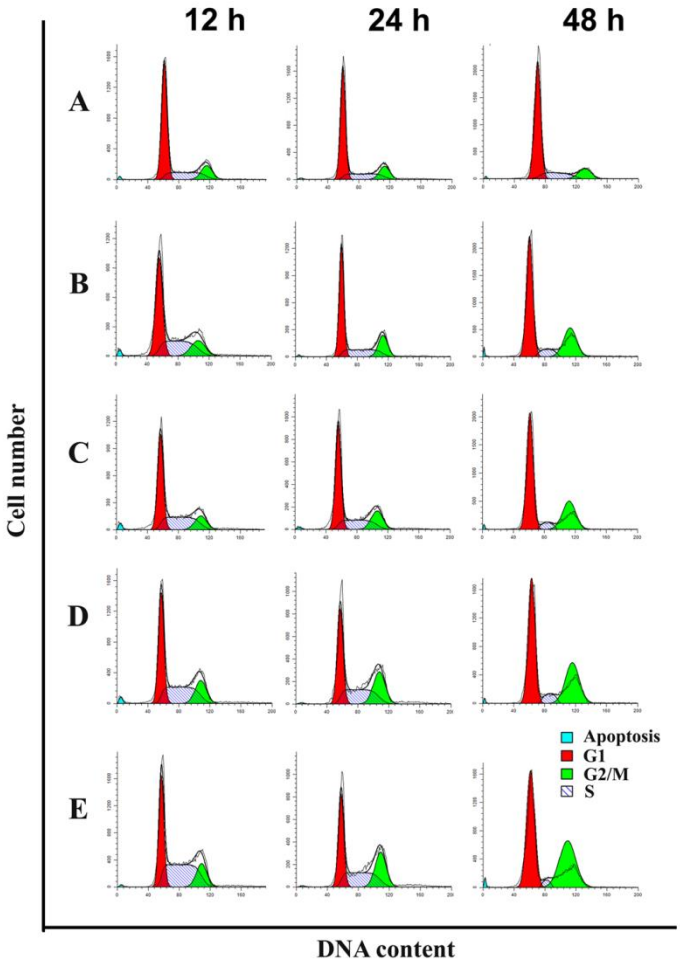

Figure S2. Cell-cycle profiles of MDA-MB-231 cells treated for 12, 24 or 48 h with 5 and its investigated Pt(IV) derivatives at the concentrations corresponding to IC_{30} values (found for these compounds in MDA-MB-231 cells treated for 72 h). The cells were stained with propidium iodide and assessed for cell-cycle distribution by FACS analysis. The colors represent the G1 phase (red), S phase (blue dashed) and G_2/M phase (green) of modeled cell cycle. Cells with the lowest DNA content were considered apoptotic (turquoise). A, control; B, **5**; C, **6**; D, **7**, and E, **8**.

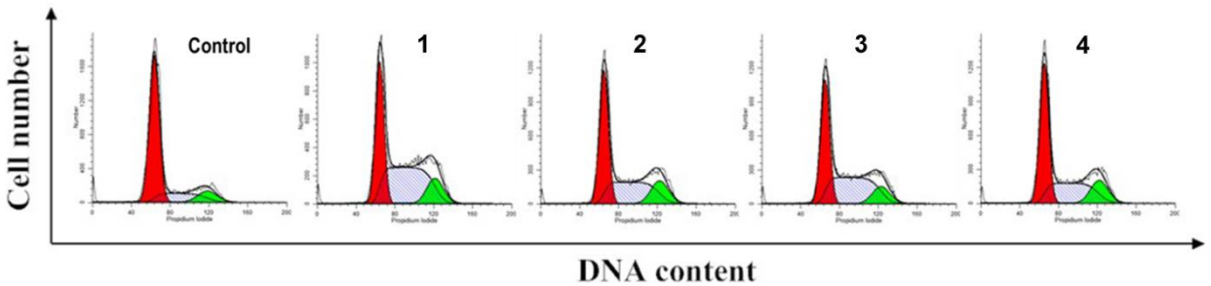

Figure S3. Cell-cycle profiles of MDA-MB-231 cells treated for 24 h with **1** and its investigated Pt(IV) derivatives at the concentrations corresponding to IC_{30} values (found for these compounds in MDA-MB-231 cells treated for 72 h). The other conditions were the same as specified in the legend to Figure S1. The colors represent the G1 phase (red), S phase (blue dashed) and G_2/M phase (green) of modeled cell cycle. Cells with the lowest DNA content were considered apoptotic (turquoise).

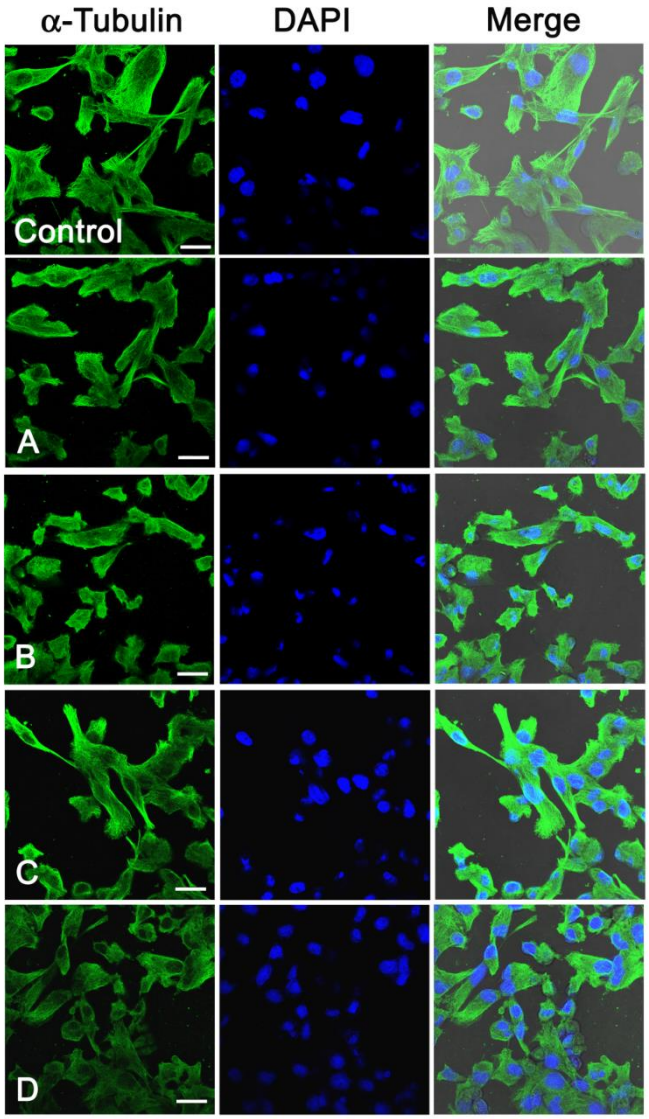

Figure S4. Effects of **5** and its investigated Pt(IV) derivatives on the organization of the structural network (cytoskeleton) within the cell's cytoplasm. MDA-MB-231 cells were incubated with the investigated **5** and its investigated Pt(IV) derivatives at the concentrations corresponding to IC_{50} (72 h; MTT) for 16 h. Images were obtained by confocal microscopy of anti-α-tubulin immunofluorescence (green) preparations; nuclei (DNA) were stained with DAPI (blue). Control cells (top row); cells treated with (A) **5**, (B) **6**, (C) **7,** and (D) **8**. Samples were scanned on confocal laser scanning microscope, and the scale bars are 25 μm.

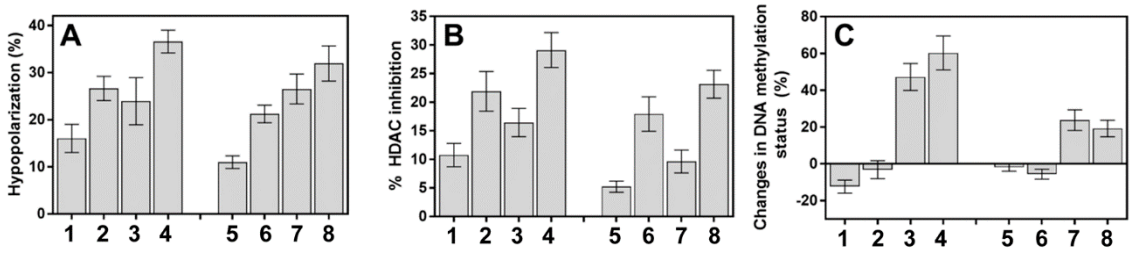

Figure S5. A. Mitochondrial membrane hypopolarization (TMRE fluorescence control – TMRE fluorescence sample) of MDA-MB-231 cells treated for 5 h with the investigated platinum complexes at concentrations corresponding to 3-fold IC_{50} values (determined with MTT; 72 h). After TMRE staining (1 nM; 20 min), the fluorescence (excitation/emission = 560 nm/595 nm) was read on Varian Cary Eclipse. The values are mean ± SD from three independent experiments.

B. HDAC activity (% inhibition) in MDA-MB-231 cells treated for 24 h with the investigated platinum compounds at concentrations corresponding to IC_{30} (determined with MTT assay after 72 h treatment).

C. DNA methylation in MDA-MB-231 cells treated for 24 h with the investigated platinum compounds at concentrations corresponding to IC_{30} (determined with MTT assay after 72 h treatment).