Electronic Supplementary Material (ESI) for Dalton Transactions.


# Inducing and monitoring mitochondrial pH changes with a iridium(III) complex via two-photon lifetime imaging

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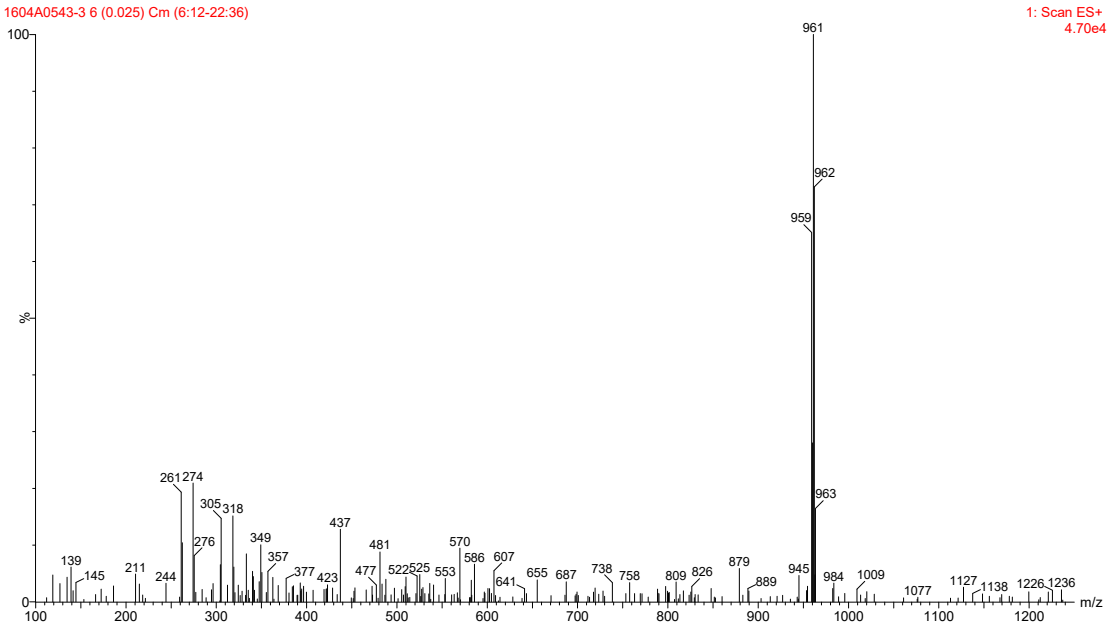


**Fig. S1** ESI-MS characterization of complex **MitoIr-NH**, 961$[M\text{-}PF_6]^+$.

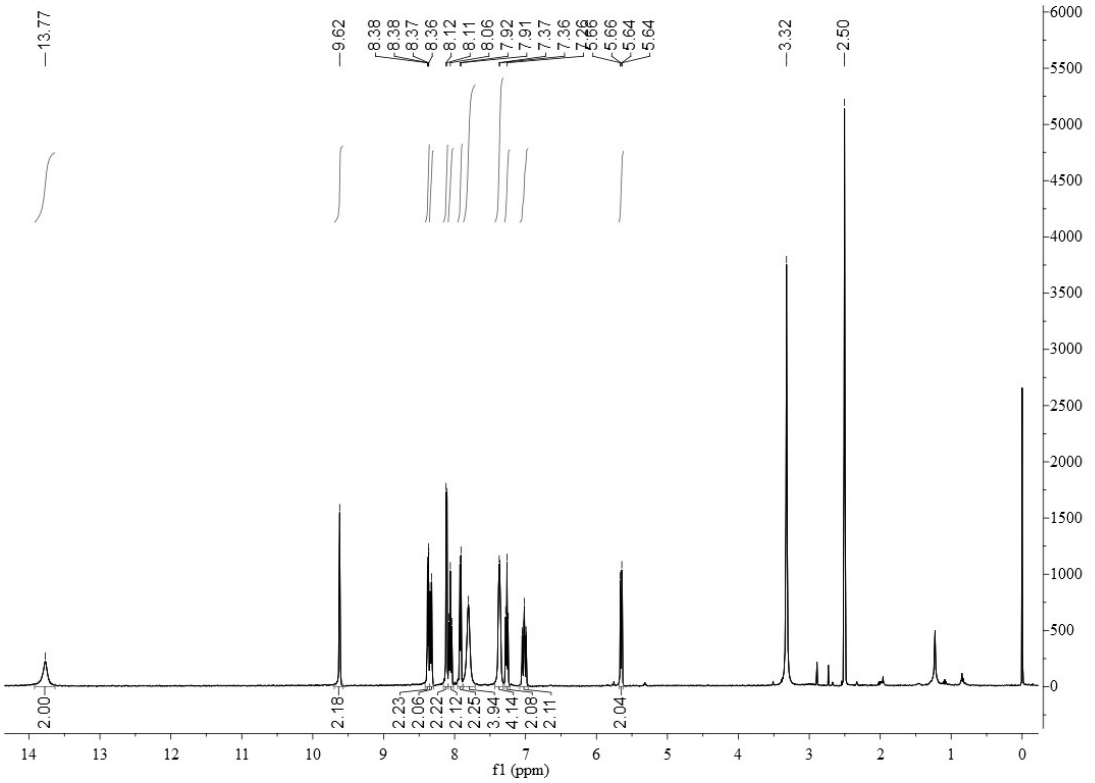


**Fig. S2** $^1$H NMR characterization of complex **MitoIr-NH**.

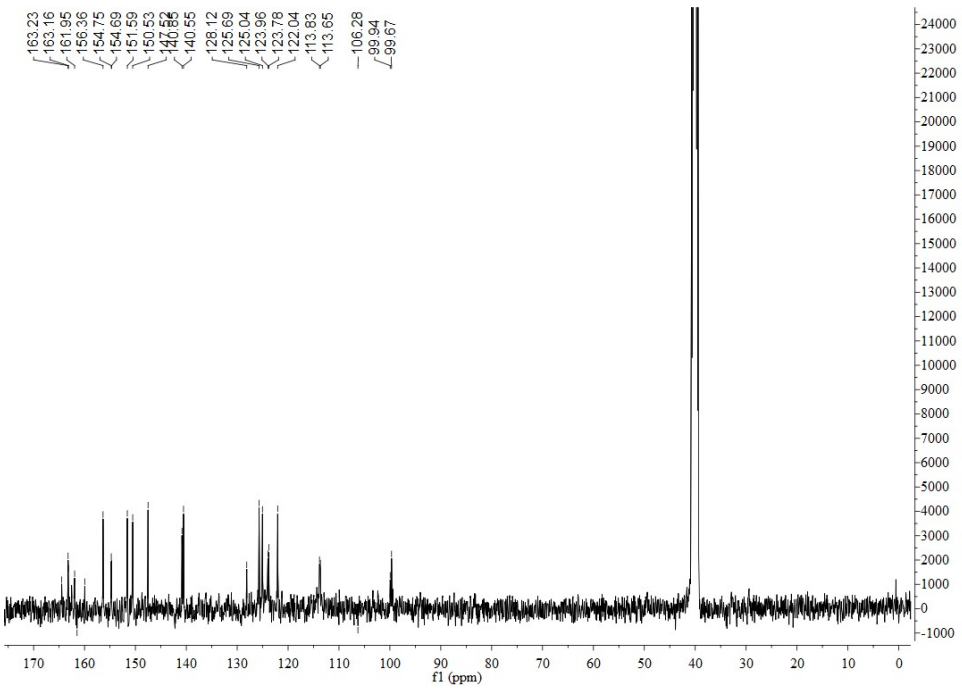


**Fig. S3** $^{13}$C NMR characterization of complex **MitoIr-NH**.

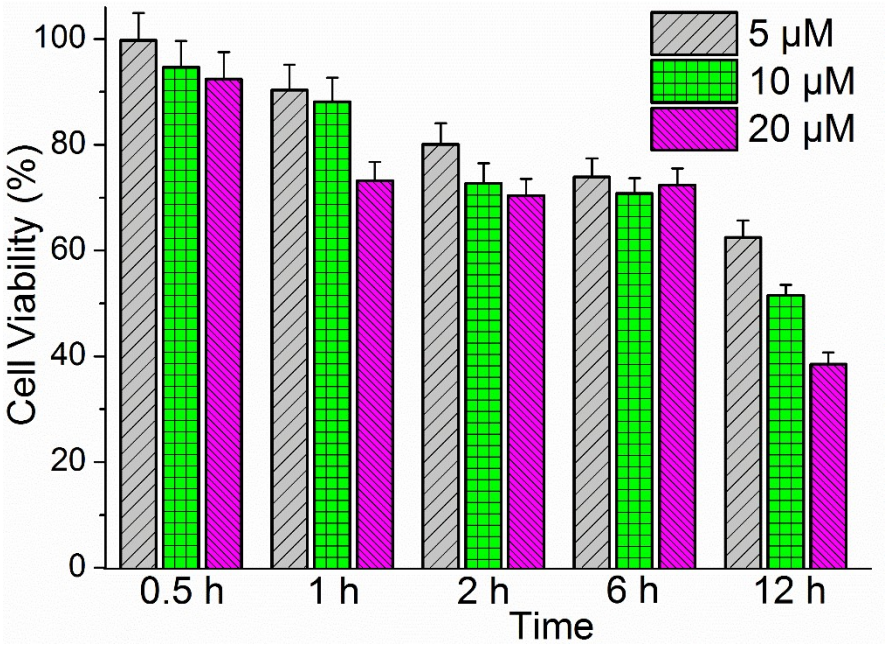


**Fig. S4** Cytotoxicity of **MitoIr-NH** determined by MTT assay. A549 cells were treated with complex **MitoIr-NH** at the indicated concentrations and time intervals.

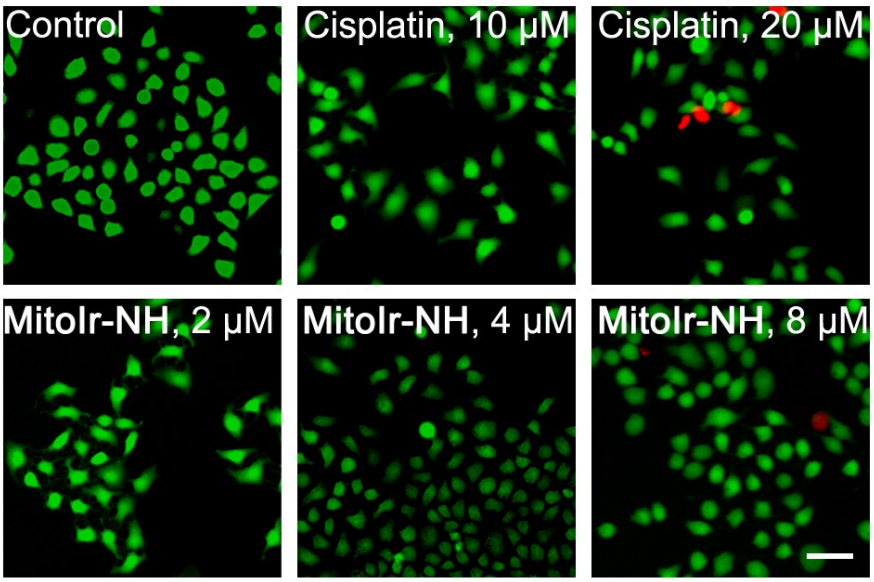


**Fig. S5** Fluorescence images of A549 cells costained with calcein AM (staining live cells, green color) and propidium iodide (staining dead cells, red color) after treated with complexes at indicated concentrations for 4 h. Scale bar: 50 μm.

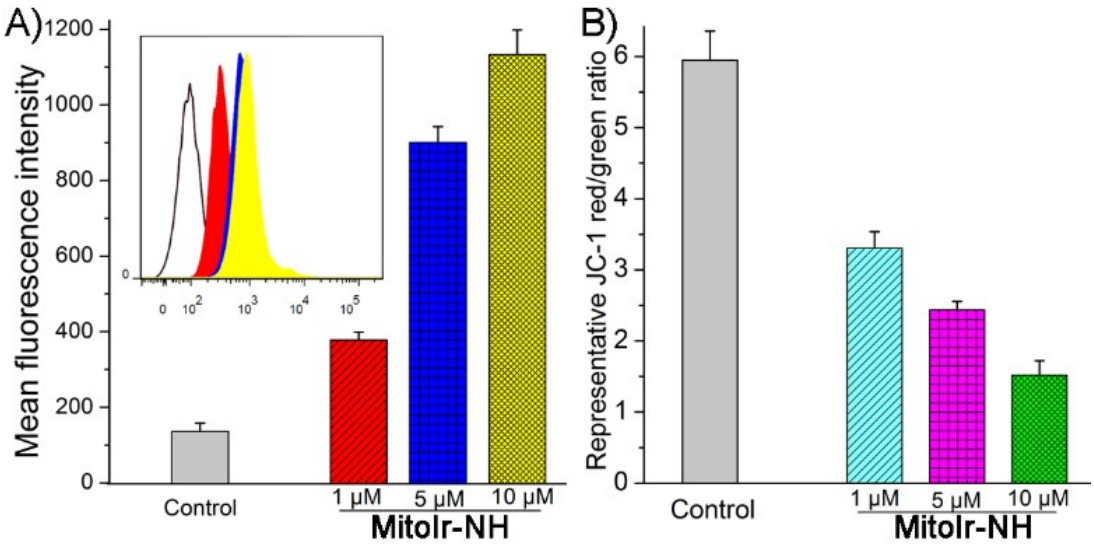


**Fig. S6** A) Intracellular ROS production measured by DCF fluorescence in A549 cells after treatment with complex **MitoIr-NH** at indicated concentrations for 6 h, insert: flow cytometry. B) Effects of complex **MitoIr-NH** on MMP analyzed by JC-1 staining and flow cytometry. A549 cells were treated with complex **MitoIr-NH** at the indicated concentrations for 6 h. JC-1 was excited at 488 nm and monitored simultaneously at 530 ± 15 nm and 590 ± 15 nm.

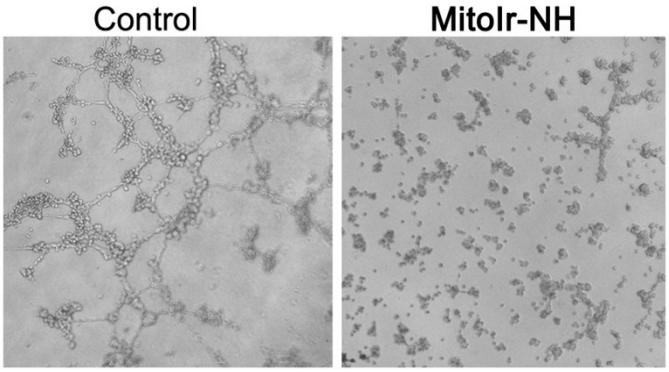


**Fig. S7** Effects of **MItoIr-NH** (0.5 μM) against tube formation of EA.hy926 on matrigel. EA.hy926 cells were seeded on matrigel in medium were treated with **Ir1-Ir3** for 5 h.