

Accepted Article

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This manuscript has been accepted after peer review and appears as an Accepted Article online prior to editing, proofing, and formal publication of the final Version of Record (VoR). This work is currently citable by using the Digital Object Identifier (DOI) given below. The VoR will be published online in Early View as soon as possible and may be different to this Accepted Article as a result of editing. Readers should obtain the VoR from the journal website shown below when it is published to ensure accuracy of information. The authors are responsible for the content of this Accepted Article.

To be cited as: *Angew. Chem. Int. Ed.* 10.1002/anie.201704828
Angew. Chem. 10.1002/ange.201704828

Link to VoR: <http://dx.doi.org/10.1002/anie.201704828>
<http://dx.doi.org/10.1002/ange.201704828>

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Nanoscale Metal-Organic Layers for Deeply Penetrating X-ray Induced Photodynamic Therapy

Guangxu Lan^{‡a}, Kaiyuan Ni^{‡a}, Ruoyu Xu^a, Kuangda Lu^a, Zekai Lin^a, Christina Chan^a, and Wenbin Lin^{*a}

Abstract: We report the rational design of metal-organic layers (MOLs) that are built from $[\text{Hf}_6\text{O}_4(\text{OH})_4(\text{HCO}_2)_6]$ secondary building units (SBUs) and $\text{Ir}[\text{bpy}(\text{ppy})_2]^+$ or $\text{Ru}(\text{bpy})_3^{2+}$ -derived tricarboxylate ligands (Hf-BPY-Ir or Hf-BPY-Ru ; bpy = 2,2'-bipyridine, ppy = 2-phenylpyridine) and their applications in X-ray induced photodynamic therapy (X-PDT) of colon cancer. Heavy Hf atoms in the SBUs efficiently absorb X-rays and transfer energy to $\text{Ir}[\text{bpy}(\text{ppy})_2]^+$ or $\text{Ru}(\text{bpy})_3^{2+}$ moieties to induce PDT by generating reactive oxygen species (ROS). The ability of X-rays to penetrate deeply into tissue and efficient ROS diffusion through ultrathin 2-D MOLs (~ 1.2 nm) enable highly effective X-PDT to afford superb anticancer efficacy.

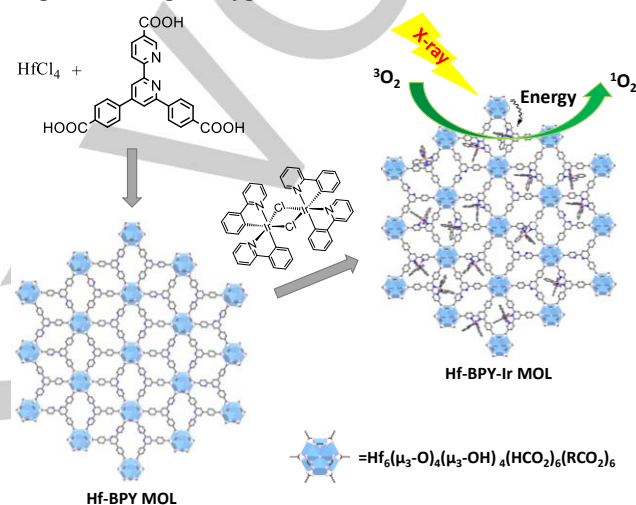
By combining three intrinsically nontoxic components--a photosensitizer (PS), light, and tissue oxygen--to generate cytotoxic reactive oxygen species (ROS), particularly singlet oxygen ($^1\text{O}_2$), photodynamic therapy (PDT) provides a highly effective phototherapy against cancer.^[1] Because ROS indiscriminately kill both diseased and normal cells, it is critical to selectively localize PSs in tumors in order to enhance PDT efficiency and minimize collateral damage to normal tissues.^[2-13] We and others reported a series of porphyrin- and chlorin-based nanoscale metal-organic frameworks (nMOFs) as PSs for effective PDT.^[14-19] Because the lifetime of ROS is short, it is not feasible for all the species generated to diffuse out of the 3-D structure of nMOFs to exert cytotoxicity on cellular organelles, thus limiting the overall efficacy of PDT *in vivo*.

We hypothesized that the *in vivo* PDT efficacy of nMOFs could be further improved by reducing the dimensionality to afford 2-D metal-organic layers (MOLs) and exciting the MOLs with more tissue-penetrating X-rays. We recently reported a new class of tunable and functionalizable MOLs that are composed of $[\text{Hf}_6\text{O}_4(\text{OH})_4(\text{HCO}_2)_6]$ secondary building units (SBUs) and benzene-1,3,5-tribenzoate (BTB) bridging ligands.^[20] The 2-D structure of MOLs allows ROS to diffuse freely, thus presenting an ideal platform for designing nanoscale PSs for efficient PDT.

$\text{Ir}[\text{bpy}(\text{ppy})_2]^+$ (bpy = 2,2'-bipyridine, ppy = 2-phenylpyridine) and $\text{Ru}(\text{bpy})_3^{2+}$ are two efficient PSs with the very high $^1\text{O}_2$ quantum yields (Φ_A) of 0.97 and 0.73, respectively.^[21-23] However, due to large Stokes shifts, they can only be excited with photons at short wavelengths, ~ 355 nm for $\text{Ir}[\text{bpy}(\text{ppy})_2]^+$ and ~ 450 nm for $\text{Ru}(\text{bpy})_3^{2+}$. Such UV-Vis photons cannot penetrate human tissue (penetration depth < 0.1 mm),^[24] which severely limits their application in PDT. Our previous work demonstrated that a Hf-based nMOF can absorb X-rays and transfer energy to coordinated anthracene-based ligands to luminesce in the visible spectrum.^[25] We believe that coordination between $\text{Ir}[\text{bpy}(\text{ppy})_2]^+$ or $\text{Ru}(\text{bpy})_3^{2+}$ -derived tricarboxylate ligands and heavy Hf-based

SBUs would enable direct excitation of the PSs by X-rays to achieve X-ray induced photodynamic therapy (X-PDT).^[26] Here we report the rational design of two MOLs, composed of $[\text{Hf}_6\text{O}_4(\text{OH})_4(\text{HCO}_2)_6]$ SBUs and $\text{Ir}[\text{bpy}(\text{ppy})_2]^+$ or $\text{Ru}(\text{bpy})_3^{2+}$ -derived tricarboxylate ligands, as potent PSs. The Hf-MOLs achieve greatly enhanced PDT efficacy both *in vitro* and *in vivo* upon X-ray irradiation.

Scheme 1. Synthesis of Hf-based MOLs and MOL-enabled X-PDT to generate singlet oxygen.



Hf-BPY-Ir and Hf-BPY-Ru MOLs were synthesized by a postsynthetic metalation method. 4',6'-dibenzoato-[2,2'-bipyridine]-4-carboxylic acid (H_3BPY) was synthesized as shown in Figure S1 (SI) and treated with HfCl_4 in N,N -dimethylformamide (DMF), formic acid, and water to afford Hf-BPY-MOL as a white precipitate, which was then washed twice with DMF and once with ethanol. By optimizing the amounts of formic acid and H_2O , the size of Hf-BPY could be controlled to a diameter of ~ 500 nm, as verified by transmission electron microscopy (TEM) (Figure 1a). Hf-BPY was treated with $[\text{Ir}(\text{ppy})_2\text{Cl}]_2/\text{Ru}(\text{bpy})_2\text{Cl}_2$ to afford $\text{Hf-BPY-Ir}/\text{Hf-BPY-Ru}$ MOL as an orange/brown particulate. Due to the 2-D structure of Hf-BPY , the bpy coordination sites are highly accessible, resulting in efficient metalation. The Ir and Ru loadings were determined to be 67% and 59% for Hf-BPY-Ir and Hf-BPY-Ru , respectively, as determined by Inductively Coupled Plasma-Mass Spectrometry (ICP-MS).

In Hf-BPY , each Hf_6 cluster of 12-connectivity was capped by 6 formate groups (three at the top and three at the bottom), leaving the remaining six sites coordinated to 3-connected BPY ligands to form a 3,6-connected 2-D network of $[\text{Hf}_6(\mu_3\text{-O})_4(\mu_3\text{-OH})_4(\text{HCO}_2)_6(\text{BPY})_2]$ of kagome dual (kgd) topology (Scheme 1). High-resolution TEM (HRTEM) images of Hf-BPY , where Hf_6 clusters appear as dark spots, and fast Fourier transform (FFT) patterns (Figure 1b) of Hf-BPY were consistent with the kgd topology. The distance between two adjacent dark spots in the HRTEM was 2.0 nm, which matched the distance between two adjacent SBUs. The powder X-ray diffraction (PXRD) pattern of Hf-BPY was identical to that of the Hf-BTB-MOL (Figure 2a),^[20] which further confirmed the kgd structure of Hf-BPY . Atomic force microscopy (AFM) images (Figure 1e, 1f and S13) of Hf-BPY showed a 1.2 nm thickness, which is very close to the van der Waals size of the Hf_6 cluster capped by formate groups, indicating

[a] G. Lan, K. Ni, R. Xu, Dr. K. Lu, Z. Lin, C. Chan and Prof. W. Lin
Department of Chemistry, The University of Chicago
929 East 57th street, Chicago, Illinois 60637, United States
E-mail: wenbinlin@uchicago.edu

[‡] These authors contributed equally to this work.

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the monolayer structure of Hf-BPY. A nitrogen sorption study of Hf-BPY gave a BET surface area of 346 m²/g (Figure S15), indicating its porous structure. The ultrathin monolayer structure facilitates the diffusion of ¹O₂ as its diffusion length was estimated to be 20–220 nm in cells.^[27]

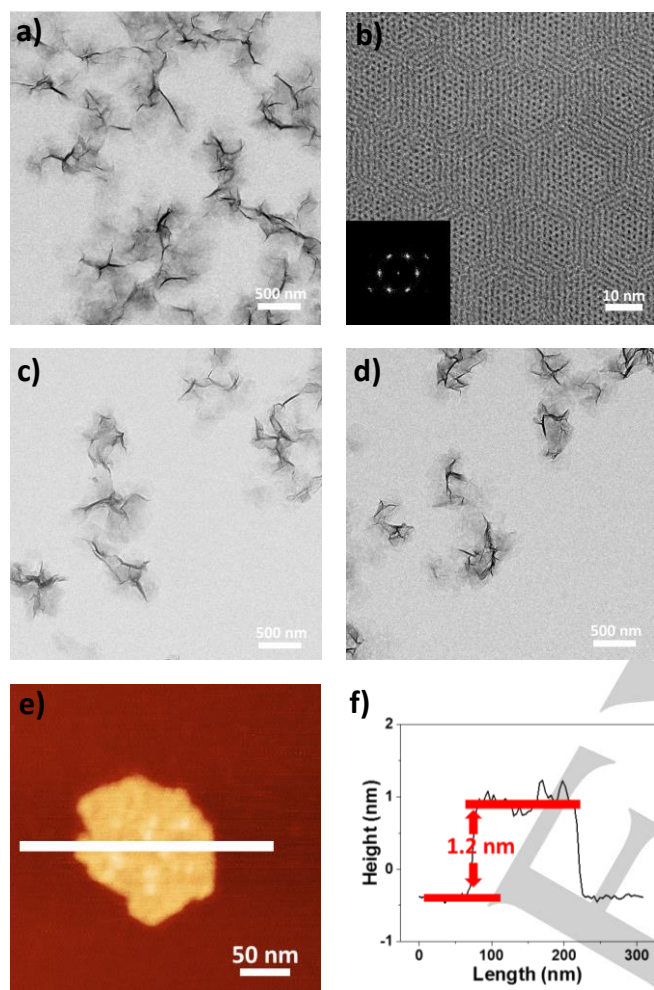


Figure 1. Morphology and structure of Hf-based MOLs. TEM image (a), HRTEM image, and FFT pattern (b) of Hf-BPY. TEM images of Hf-BPY-Ir (c) and Hf-BPY-Ru (d). Tapping-mode AFM topography (e) and the height profile along the white line (f) of Hf-BPY.

TEM imaging showed that Hf-BPY-Ir (Figure 1c) and Hf-BPY-Ru (Figure 1d) have similar morphologies and sizes as Hf-BPY. The retention of the MOL structure after metalation was supported by the similarity among the PXRD patterns of Hf-BPY-Ir, Hf-BPY-Ru, and Hf-BPY (Figure 2a). In addition, the HRTEM images and FFT patterns of Hf-BPY-Ir and Hf-BPY-Ru (Figure S10) were identical to those of Hf-BPY. TEM images and PXRD patterns of the MOLs remained unchanged after incubation in DMEM media for 12 h (Figures S16 and 17), suggesting that the MOLs are stable for biomedical applications.

To further confirm the metalation of Hf-BPY and to understand the coordination environments of Ir and Ru centers in Hf-MOLs, we synthesized [(H₃BPY)Ir(ppy)₂]Cl (H₃BPY-Ir) and [(H₃BPY)Ru(bpy)₂]Cl₂ (H₃BPY-Ru) as ligand controls (Figure S6–S9). The UV-visible absorption spectra of Hf-based MOLs exhibit similar MLCT bands as their corresponding ligands (Figure 2b). Importantly, X-ray absorption spectroscopy (XAS) indicated that Zr-BPY-Ir and Hf-BPY-Ru have the same Ir and Ru coordination environments as H₃BPY-Ir and H₃BPY-Ru, respectively (Figure

S20–S23). Due to similar energy between Ir L₃-edge (11215 eV) and Hf L₁-edge (11271 eV), XAS data was collected for Zr-BPY-Ir instead of Hf-BPY-Ir.

We next examined singlet oxygen generation efficiencies of MOLs using 4-nitroso-N,N-dimethylaniline (RNO) assay. We also synthesized Zr-MOLs (Zr-BPY-Ir and Zr-BPY-Ru) using similar processes (Figure S11 and S12) and used them for comparison. Upon irradiation with a Xe lamp using a 400 nm long-pass filter or X-rays (225 Kvp, 13 mA), the ¹O₂ generated by MOLs reacted with RNO in the presence of histidine, leading to a decrease of absorbance at 440 nm in the UV-visible spectra (Figure S24 and S25). By linearly fitting difference in RNO peak absorbance [$\Delta(\text{OD})$] against irradiation doses (which scale linearly with exposure times upon visible light or X-ray dose, $Y = Ax + B$), the RNO assay provides a quantitative measure of ¹O₂ generation efficiencies, with a more positive slope indicating more efficient ¹O₂ generation. Upon visible light irradiation, the linear fitting results showed that Ir-based Zr- and Hf-MOLs generated ¹O₂ more efficiently than Ru-based Zr- and Hf-MOLs (Figures 2c, Table S3), consistent with the difference in ¹O₂ generation quantum yields between [Ir(bpy)(ppy)₂]⁺ ($\Phi_A = 0.97$) and [Ru(bpy)₃]²⁺ ($\Phi_A = 0.73$). Furthermore, only very slight differences were observed between two Ir-based MOLs ($A = 1.09 \times 10^{-2}$ for Hf-BPY-Ir and $A = 0.88 \times 10^{-2}$ for Zr-BPY-Ir) or two Ru-based MOLs ($A = 4.1 \times 10^{-3}$ for Hf-BPY-Ru and $A = 2.4 \times 10^{-3}$ for Zr-BPY-Ru), suggesting minor effects of the SBUs in the ¹O₂ generation efficiency through spin-orbit coupling.^[14, 28] The efficient singlet oxygen generation by Hf-BPY-Ir upon light irradiation was also demonstrated by Singlet Oxygen Sensor Green (SOSG) assay (Figure S26). Comparisons with DBP-Hf, a porphyrin-based nMOF reported by us previously, indicated that Hf-BPY-Ir is more effective in generating ¹O₂ than DBP-Hf upon light irradiation (Figure S27), suggesting the facile diffusion of ROS through the MOL monolayer.

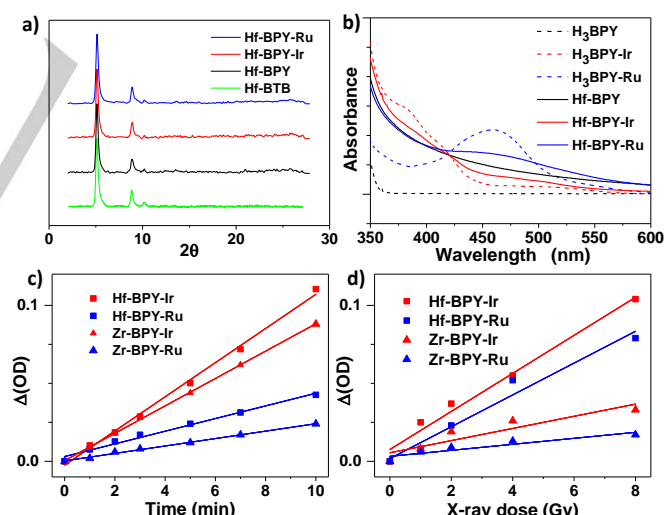


Figure 2. (a) PXRD patterns of new Hf-MOLs and the Hf-BTB MOL. (b) UV-visible absorption spectra of Hf-MOLs and the bridging ligands. Singlet oxygen generation of Hf- and Zr-MOLs upon visible light irradiation (c) or X-ray irradiation (d), as determined by RNO. Linear fit of $\Delta(\text{OD})$ at 440 nm against irradiation dose.

However, upon X-ray irradiation, there was a drastic difference in ¹O₂ generation efficiencies in Zr- and Hf-MOLs (Figure 2d, Table S4). Both Hf-MOLs ($A = 1.22 \times 10^{-2}$ for Hf-BPY-Ir and $A = 1.0 \times 10^{-2}$ for Hf-BPY-Ru) possessed much higher ¹O₂ generation efficiency than their corresponding Zr-MOLs ($A = 0.39 \times 10^{-2}$ for Hf-BPY-Ir and $A = 0.19 \times 10^{-2}$ for Zr-BPY-Ir), supporting our hypothesis that the X-ray energy was first absorbed by SBUs and

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then transferred to the PSs in the bridging ligands to lead to the X-PDT effect. Because the heavier Hf atoms absorb X-rays more efficiently than the Zr atoms, the Hf-MOLs are expected to be more effective at X-PDT. Additionally, Ir-based MOLs showed only slightly better X-PDT efficiency than Ru-based MOLs, suggesting different energy transfer processes involved in X-PDT and PDT.

In the clinic, PDT is typically applied to superficial malignant tumors such as skin lesions and esophageal cancer due to the limited penetration of light (~3 mm at 800 nm). We sought to examine the potential of MOL-mediated X-PDT in the treatment of deeply seated tumors. Two types of murine colon adenocarcinoma cells, CT26 and MC38, were used for *in vitro* and *in vivo* studies. The cellular uptake was evaluated on CT26 cells incubated with Hf-BPY-Ir, Hf-BPY-Ru, or Hf-BPY at a Hf concentration of 50 μM for 1, 4, 8, and 24 h. At each time point, cells were digested and the Hf contents were determined by ICP-MS. Hf-BPY-Ru showed higher uptake ($6580 \pm 1770 \text{ ng}/10^5 \text{ cells}$) than Hf-BPY-Ir ($3317 \pm 665 \text{ ng}/10^5 \text{ cells}$) and Hf-BPY ($1930 \pm 716 \text{ ng}/10^5 \text{ cells}$), presumably because of the higher positive charge of Hf-BPY-Ru, which favors interacting with the negatively charged cell membrane to facilitate endocytosis (Figure S28).

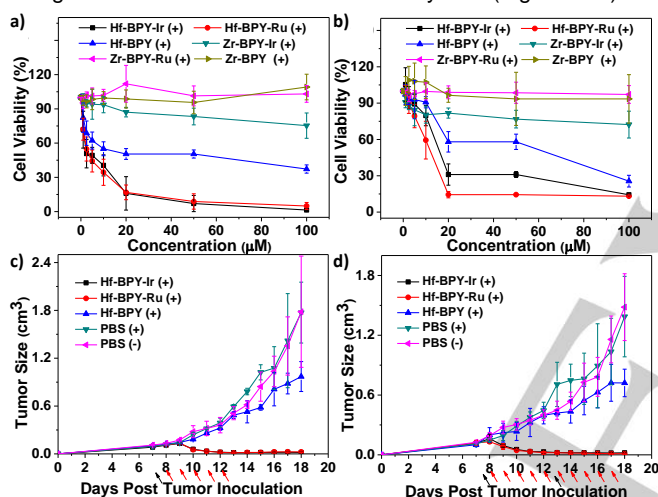


Figure 3. *In vitro* and *in vivo* anticancer efficacy of Hf-MOLs. Cytotoxicity of Hf- and Zr-MOLs in CT26 cells (a) and MC38 cells (b). Tumor growth inhibition curves after X-PDT treatment in the CT26 (c) and MC38 (d) models. Black arrows refer to injection of MOLs and red arrows refer to X-ray irradiation.

We next investigated the *in vitro* anticancer efficacy of three different Hf-based MOLs against CT26 (Figure 3a) and MC38 (Figure 3b) cells. To elucidate the key role of Hf in efficient absorption of X-rays, three corresponding Zr-MOLs were used as controls. MOLs were incubated with cells at various concentrations for 8 h, followed by irradiation with an X-ray irradiator at a dose of 2 Gy. Hf-BPY-Ir and Hf-BPY-Ru outperformed Hf-BPY and three Zr-MOLs. The IC_{50} values for Hf-BPY-Ir, Hf-BPY-Ru, and Hf-BPY against CT26 cells were calculated to be 3.82 ± 1.80 , 3.63 ± 2.75 , and $24.90 \pm 7.87 \mu\text{M}$, respectively. Against MC38 cells, the IC_{50} values were 11.66 ± 1.84 , 10.72 ± 2.92 , and $37.80 \pm 6.57 \mu\text{M}$, respectively. IC_{50} values exceeded 100 μM for Zr-BPY-Ir, Zr-BPY-Ru, and Zr-BPY in both CT26 and MC38 cell lines. No cytotoxicity was observed in dark control groups (Figure S29). We also tested cell viability with fixed Hf-MOL concentrations based on Ir, Ru, or BPY of 20 μM , respectively, and various X-ray doses (Figure S30). All of the results showed greatly enhanced X-PDT potency of $[\text{Ir}(\text{bpy}(\text{ppy})_2)]^+$ and $[\text{Ru}(\text{bpy})_3]^{2+}$ in Hf-MOLs. Importantly, X-ray induced cytotoxicity of Hf-BPY-Ir and Hf-BPY-Ru remained essentially unchanged when the cells were covered with a beef block of 1 cm

in thickness during X-ray irradiation (Figure S41). In contrast, light induced cytotoxicity of Hf-BPY-Ir and Hf-BPY-Ru was completely lost when the cells were covered with the same beef block during light irradiation (Figure S40). These results support our hypothesis that MOL-mediated X-PDT can be used to treat deep-seated tumors. Interestingly, control experiments with Hf-QPDC-Ir nMOF, a UiO nMOF built from $\text{Hf}_6(\mu_3\text{-O})_4(\mu_3\text{-OH})_4$ SBUs and QPDC-Ir bridging ligands, further support the enhanced X-PDT efficacy of the MOLs due to facile ROS diffusion; upon X-ray irradiation, Hf-QPDC-Ir nMOF exhibited much higher IC_{50} values of $32.85 \pm 3.02 \mu\text{M}$ for CT26 cells and $26.08 \pm 2.38 \mu\text{M}$ for MC38 cells, respectively (Figure S39).

We then explored the mechanism of X-ray induced cytotoxicity on CT26 cells. $^1\text{O}_2$ generation in live cells was examined by SOSG and detected by confocal laser scanning microscopy (CLSM) (Figure S31). After preloading cells with SOSG and incubating them with PBS, Hf-MOLs, or H_2BPY ligand for 8 h at a concentration of 20 μM based on Ir, Ru, or BPY, respectively, they were irradiated with X-rays at a dose of 2 Gy, immediately followed by CLSM imaging. Both Hf-BPY-Ir- and Hf-BPY-Ru-treated cells showed strong green SOSG fluorescence, indicating the efficient generation of $^1\text{O}_2$ in the MOLs upon X-ray irradiation. In contrast, PBS, Hf-BPY and H_2BPY ligand-treated groups showed no SOSG signal after X-ray induced $^1\text{O}_2$ generation, which supported our proposed X-PDT process using Hf-BPY-Ir and Hf-BPY-Ru MOLs. We also performed $\gamma\text{-H}_2\text{AFX}$ assay (Life technology, USA) on CT26 cells to determine DNA double-strand breaks (DSBs) caused by MOLs upon X-ray irradiation. As shown in Figure S37, CT26 cells treated with three Hf-based MOLs showed significant red fluorescence, indicating DSBs induced by hydroxyl radical from X-ray irradiation. This result suggests that Hf_6 SBUs are capable of radiosensitization to further enhance cytotoxicity of MOL-mediated X-PDT.

Encouraged by *in vitro* results, we carried out *in vivo* anticancer efficacy experiments on subcutaneous flank tumor-bearing mouse models of CT26 and MC38. When tumors reached 100–150 mm^3 in volume, Hf-BPY-Ir, Hf-BPY-Ru, or Hf-BPY with amount of 0.5 nmol based on Ir, Ru or BPY, respectively, or PBS was intratumorally injected followed by daily X-ray irradiation at a dose of 1 Gy/fraction (120 kVp, 20 mA, 2 mm-Cu filter) for a total of 5 fractions on the CT26 model (Figure 3c) or 10 fractions on the MC38 model (Figure 3d) on consecutive days. Tumor sizes and body weights were measured every day. All mice were sacrificed 18 days after tumor inoculation, and the excised tumors were photographed and weighed (Figure S33–S34). To rule out any radiotherapy effects from the low dose X-ray, we used PBS-treated mice without X-ray irradiation as a dark control. The PBS groups with or without irradiation did not show any difference in tumor growth curves, indicating that low dose X-rays alone had no radiotherapeutic effects. The Hf-BPY groups appeared to show slight inhibition of tumor growth ($P=0.047$ or 0.048 for CT26 or MC38, respectively), consistent with the radiosensitization effects of the Hf_6 SBUs. In stark contrast, Hf-BPY-Ir and Hf-BPY-Ru treatments led to effective tumor regression in CT26 with 5 fractions of X-ray irradiation (5 Gy total; total volume reduction of 83.6% or 77.3%, respectively) and in MC38 with 10 fractions of X-ray irradiation (10 Gy; total total volume reduction of 82.3% or 90.1%, respectively). The weights and sizes of tumors treated with Hf-BPY-Ir and Hf-BPY-Ru at the end point were significantly smaller than the other groups (Table S5 and S6). Histology of frozen tumor slices confirmed MOL-assisted X-PDT caused apoptosis/necrosis in tumors (Figure S35). No abnormalities were observed on histological images of frozen organ slices, which indicated that X-PDT with intratumoral injection of MOLs was not systemically toxic (Figure S36). The lack of systemic toxicity was further supported by steady body weights and similar weight gain patterns in all groups (Figure S32).

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In summary, we rationally designed and synthesized two Hf-MOLs as powerful PSs for effective X-PDT of colon cancer models. Upon X-ray irradiation, Hf atoms in the SBUs absorb X-rays and transfer energy to Ir[bpy(ppy)₂]⁺ or [Ru(bpy)₃]²⁺ in the ligands to generate ¹O₂, demonstrated by both RNO assay and *in vitro* ¹O₂ detection as well as cytotoxicity studies. As a result of deep tissue penetration of X-rays, high ¹O₂ quantum yields of Ir[bpy(ppy)₂]⁺ or [Ru(bpy)₃]²⁺, and efficient ROS diffusion through ultrathin MOLs, X-PDT treatment led to an impressive 90% reduction in tumor volumes. MOLs thus represent a novel class of 2-D materials with great potential for cancer treatment and other biomedical applications.

Acknowledgements

We thank Dr. Chunbai He and Wenbo Han for experimental help. We acknowledge the National Cancer Institute (U01-CA198989), the University of Chicago Materials Research Science and Engineering Center (DMR-1420709), the CBI Training Grant (NIH 5T32GM008720-15), and the Ludwig Institute for Metastasis Research for funding support. XAFS data were collected at the APS at ANL on Beamline 10BM-A, B, supported by the Materials Research Collaborative Access Team (MRCAT). MRCAT operations are supported by the Department of Energy and the MRCAT member institutions. This research used resources of the Advanced Photon Source, a U.S. Department of Energy (DOE) Office of Science User Facility operated for the DOE Office of Science by Argonne National Laboratory under Contract No. DE-AC02-06CH11357.

Keywords: Nanoscale Metal-Organic Layer • Photodynamic Therapy • X-ray • Cancer Treatment•

- [21] S.-y. Takizawa, R. Aboshi, S. Murata, *Photochem. Photobiol. Sci.* **2011**, *10*, 895.
- [22] P. I. Djurovich, D. Murphy, M. E. Thompson, B. Hernandez, R. Gao, P. L. Hunt, M. Selke, *Dalton Trans.* **2007**, *34*, 3763.
- [23] D. Garcia - Fresnadillo, Y. Georgiadou, G. Orellana, A. M. Braun, E. Oliveros, *Helv. Chim. Acta* **1996**, *79*, 1222.
- [24] A. M. Smith, M. C. Mancini, S. Nie, *Nat. Nanotechnol.* **2009**, *4*, 710.
- [25] C. Wang, O. Volotskova, K. Lu, M. Ahmad, C. Sun, L. Xing, W. Lin, *J. Am. Chem. Soc.* **2014**, *136*, 6171.
- [26] H. Chen, G. D. Wang, Y.-J. Chuang, Z. Zhen, X. Chen, P. Biddinger, Z. Hao, F. Liu, B. Shen, Z. Pan, J. Xie, *Nano Lett.* **2015**, *15*, 2249.
- [27] J. Moan, K. BERG, *Photochem. photobiol.* **1991**, *53*, 549.
- [28] F. Scandola, C. Chiorboli, A. Prodi, E. Iengo, E. Alessio, *Coord. Chem. Rev.* **2006**, *250*, 1471.
- [1] J. P. Celli, B. Q. Spring, I. Rizvi, C. L. Evans, K. S. Samkoe, S. Verma, B. W. Pogue, T. Hasan, *Chem. Rev.* **2010**, *110*, 2795.
- [2] D. Bechet, P. Couleaud, C. Frochot, M.-L. Viriot, F. Guillemin, M. Barberi-Heyob, *Trends biotechnol.* **2008**, *26*, 612.
- [3] K. K. Ng, G. Zheng, *Chem. Rev.* **2015**, *115*, 11012.
- [4] E. Huynh, B. Y. Leung, B. L. Helfield, M. Shakiba, J.-A. Gandier, C. S. Jin, E. R. Master, B. C. Wilson, D. E. Goertz, G. Zheng, *Nat. Nanotechnol.* **2015**, *10*, 325.
- [5] K. A. Carter, S. Shao, M. I. Hoopes, D. Luo, B. Ahsan, V. M. Grigoryants, W. Song, H. Huang, G. Zhang, R. K. Pandey, *Nat. Comm.* **2014**, *5*.
- [6] J. F. Lovell, C. S. Jin, E. Huynh, H. Jin, C. Kim, J. L. Rubinstein, W. C. Chan, W. Cao, L. V. Wang, G. Zheng, *Nat. Mater.* **2011**, *10*, 324.
- [7] N. M. Idris, M. K. Gnanasammandhan, J. Zhang, P. C. Ho, R. Mahendran, Y. Zhang, *Nat. Med.* **2012**, *18*, 1580.
- [8] C. He, D. Liu, W. Lin, *ACS Nano* **2015**, *9*, 991.
- [9] C. He, X. Duan, N. Guo, C. Chan, C. Poon, R. R. Weichselbaum, W. Lin, *Nat. Comm.* **2016**, *7*.
- [10] Y. Cheng, C. Samia A, J. D. Meyers, I. Panagopoulos, B. Fei, C. Burda, *J. Am. Chem. Soc.* **2008**, *130*, 10643.
- [11] D. K. Chatterjee, L. S. Fong, Y. Zhang, *Adv. Drug Deliv. Rev.* **2008**, *60*, 1627.
- [12] I. Roy, T. Y. Ohulchanskyy, H. E. Pudavar, E. J. Bergey, A. R. Oseroff, J. Morgan, T. J. Dougherty, P. N. Prasad, *J. Am. Chem. Soc.* **2003**, *125*, 7860.
- [13] X. Zheng, L. Wang, Q. Pei, S. He, S. Liu, Z. Xie, *Chem. Mater.* **2017**, *29*, 2374.
- [14] K. Lu, C. He, W. Lin, *J. Am. Chem. Soc.* **2014**, *136*, 16712.
- [15] K. Lu, C. He, W. Lin, *J. Am. Chem. Soc.* **2015**, *137*, 7600.
- [16] K. Lu, C. He, N. Guo, C. Chan, K. Ni, R. R. Weichselbaum, W. Lin, *J. Am. Chem. Soc.* **2016**, *138*, 12502.
- [17] J. Park, Q. Jiang, D. Feng, L. Mao, H. Zhou, *J. Am. Chem. Soc.* **2016**, *138*, 3518.
- [18] J. Liu, Y. Yang, W. Zhu, X. Yi, Z. Dong, X. Xu, M. Chen, K. Yang, G. Lu, L. Jiang, Z. Liu, *Biomaterials* **2016**, *97*, 1.
- [19] M. Lismont, L. Dreesen, S. Wuttke, *Adv. Funct. Mater.* **2017**, *27*, 1606314.
- [20] L. Cao, Z. Lin, F. Peng, W. Wang, R. Huang, C. Wang, J. Yan, J. Liang, Z. Zhang, T. Zhang, L. Long, J. Sun, W. Lin, *Angew. Chem. Int. Ed.* **2016**, *55*, 4962.

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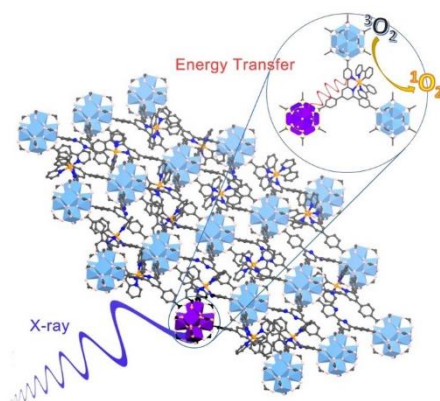
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COMMUNICATION

X-ray induced PDT

Heavy Hf atoms in the SBUs efficiently absorb X-rays and transfer energy to Ir[bpy(ppy)₂]⁺ or Ru(bpy)₃²⁺ moieties in ultrathin metal-organic layers to induce PDT by generating reactive oxygen species (ROS). Deep tissue penetration of X-rays, high ¹O₂ quantum yields, and efficient ROS diffusion allow for regression of colorectal tumors via MOL-mediated, X-ray induced PDT treatments.



Guangxu Lan, Kaiyuan Ni, Ruoyu Xu,
Kuangda Lu, Zekai Lin, Christina Chan,
and Wenbin Lin*

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**Nanoscale Metal-Organic Layers for
Deeply Penetrating X-ray Induced
Photodynamic Therapy**