Electronic Supplementary Information:

Oxygen self-supplying porphyrinic MOFs to alleviate tumor hypoxia for starvation-amplified photodynamic therapy

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1. Experimental Section

1.1 Materials

All the chemical reagents and solvents in our research were of analytical grade and used directly without further purification. Zirconyl chloride octahydrate ($ZrOCl_2 \cdot 8H_2O$, 98%), Fe(III)-5,10,15,20-Tetrakis(4-carboxyphenyl)porphyrin (Fe-TCPP), N,Ndimethylformamide (DMF), Pentadecafluorooctanoic acid (PFOA) were purchased from Aladdin Chemical Reagent Co. Ltd. China. Methylthiazolyldiphenyl-tetrazolium bromide (MTT), Glucose oxidase (GOx) was purchased from BIOFROX. Beyotime sold Calcein acetoxymethyl ester (calcein AM), propidium iodide (PI). FEIYUBIO was the supplier of singlet oxygen sensor green (SOSG). Dulbecco's modified Eagle's medium (DMEM), TRYPSIN 0.25 % (1X) Solution were purchased from HyClone. Fetal bovine serum (FBS) was purchased from EVERY GREEN.

1.2 Characterization

Scanning electron microscopy (SEM) images were acquired on a ZEISS Gemini SEM 300. Transmission electron microscopy (TEM) observations were conducted on a FEI Talox F200X electron microscope at an acceleration voltage of 300 kV. The UV-Vis-NIR spectroscopy was recorded by using a Shimadzu UV3600 spectrophotometer. XRD was carried out by means of a Rigaku D/max-2550pc instrument with monochromatized CuKa radiation and a scanning step of 0.028. The zeta potential and dynamic light scattering (DLS) analysis were performed through a Zetasizer Nano-ZS LA-960 instrument (HORIBA, Japan). Confocal laser scanning microscope (CLSM) images were acquired by Leica TCS SP8 confocal laser scanning microscope.

1.3 Synthesis of PCN(Fe), PCN(Fe/F), and PCN(Fe/F)@GOx

Synthesis of PCN(Fe): The synthesis of PCN(Fe) was mainly referred to the previous work.[1] 18.6 μmol of $ZrOCl_2 \cdot 8H_2O$ and 2.5 μmol Fe-TCPP were ultrasonically dissolved in 2 mL of DMF solution and transferred to the round-bottom flask. After sonication for 30 min, the mixture was heated to 90 °C for 5 hours. Finally, the products were collected by centrifugation and washed with DMF and ethanol. The samples of PCN(Fe) are dried overnight in an oven at 70 °C. The synthesis process of PCN was similar to that of PCN(Fe), except the ligand of Fe-TCPP was replaced by TCPP.

Synthesis of PCN(Fe/F): The perfluoroalkane-functionalized PCN(Fe) was synthesized according to the literature method.[2] 20 mg PCN(Fe) was dispersed in 30 mL DMF and sonication for 30 min. Then, 4 mL of PFOA was added to the above solution and magnetic stirring for 24 h at 60 ºC. The supernatant of the reaction mixture was decanted and the sample was soaked into fresh hot DMF which was then filtered, washed sequentially with DMF, acetone and diethyl ether, and finally dried at 80 ºC under vacuum for 12 h.

Synthesis of PCN(Fe/F)@GOx: Briefly, 10 mg PCN(Fe/F) and 5 mg GOx were dissolved in 10 mL deionized water. Then the mixture was stirred at room temperature for 12 h. After that, the product was collected by centrifugation, washed with water for several times and dried for used.

1.4 Extracellular real-time O_2 concentration measurement

PCN (Fe/F) was added to the aqueous solution and oxygen was passed through for half an hour, and then the solid was collected by centrifugation. Then, 100 μg/mL of PCN, or PCN(Fe), or PCN(Fe/F), or PCN(Fe/F)@GOx was added into 400 μM H_2O_2 solution or glucose solution, and the oxygen concentrations of the solution can be detected by the portable dissolved oxygen instrument.

1.5 Catalytic activity of PCN(Fe/F)@GOx

100 mg PCN(Fe/F)@GOx was incubated with 1mg/mL glucose solution. The pH value of the mixture was recorded with a real-time pH meter. Moreover, the concentration of glucose in different samples was measured by a glucometer.

1.6 Extracellular $^1O_2$ generation measurement

A DPBF probe was used to measure the generation of extracellular singlet oxygen. Briefly, 150 μL of ethanol solution containing DPBF (100 μM) was added to 2 mL of PCN(Fe), or PCN(Fe/F), or PCN(Fe/F)@GOx (200 μL, 2 mg/mL), and then the mixture was kept in the dark under stirring and irradiated by the 660 nm laser (50 mW cm^{-2}) at various time periods. The UV−vis absorption spectra of DPBF (λ = 410 nm) were monitored.

1.7 Intracellular $^1O_2$ measurement

HeLa cells were seeded in a 6-well cell culture plate at a density of 1×10^5 cells per

well and incubated for 24 h. PCN, PCN(Fe/F), and PCN(Fe/F)@GOx were incubated with HeLa cells for 6 hours, followed by adding 10 μL of SOSG working solution and incubating for 30 min. Then, the cells were irradiated for 10 min (660 nm, 50 mW cm^{-2}). The incubation medium was removed. The images were obtained using a fluorescent microscope.

1.8 In Vitro Cytotoxicity

The hypoxic environment was built by incubation adherent cells in hypoxia chamber with 1 % O_2, 5 % CO_2, and 94 % N_2 gas for 4 h. First, HeLa cells were seeded in culture dishes (1×10^4 cells/mL) and then divided into two groups with different treatments. (1) Normoxia group. The cells were incubated in normoxia. (2) Hypoxia group. The adherent cells were incubated in hypoxia chamber for 4 h.

In brief, HeLa cells were seeded into 96-well plates (1×10^4 cells per well) for 24 h, and then incubated with different concentrations of PCN, PCN(Fe), PCN(Fe/F), and PCN(Fe/F)@GOx ranging from 10 to 160 μg/mL for another 48 h. Thereafter, the culture medium was replaced with DMEM (1 mL) and MTT solution (10 μL, 5 mg/mL). After 4 h incubation, the medium was replaced with DMSO (150 μL) and shaken for 10 min to dissolve blue formazan. The absorbance at 450 nm of each well was measured using microplate reader.

2. Figures

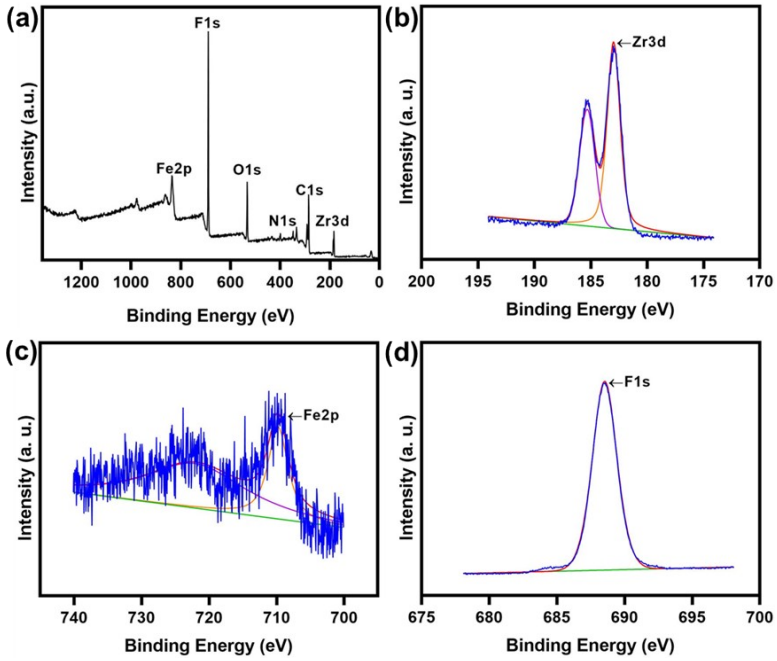

Fig. S1 (a) XPS spectra of PCN(Fe/F) nanoshuttles. (b) Zr 3d characteristic peaks, (c) Fe 2p characteristic peaks, and (d) F 1s characteristic peaks in the XPS high-resolution spectra of PCN(Fe/F).

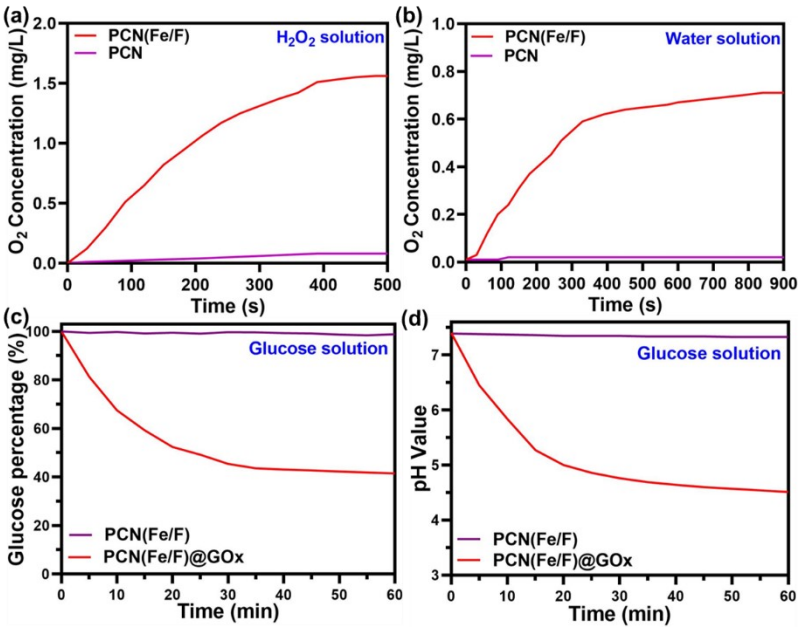

Fig. S2 Oxygen concentration changes: (a) in H_2O_2 solution (400 μM) after treating with PCN and PCN(Fe/F), (b) in water solution after treating with PCN and PCN(Fe/F). (c) The glucose concentration and (d) pH changes in PCN(Fe/F)@GOx solution with the addition of glucose (1.0 mg mL^{-1}).

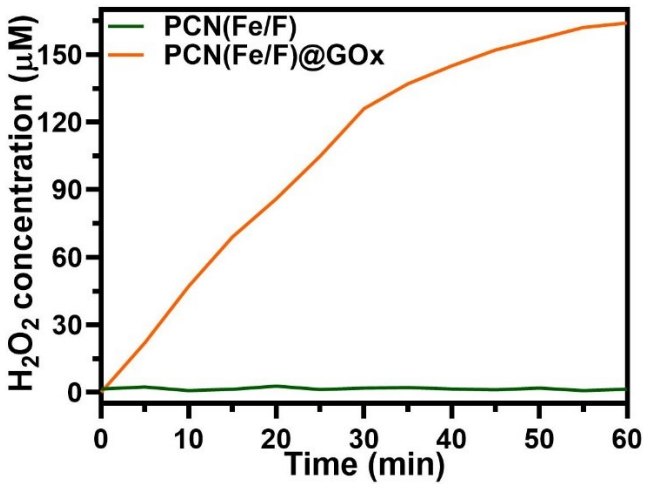

Fig. S3 The change of H_2O_2 concentrations in the glucose solution (1.0 mg/mL) with the addition of PCN(Fe/F)@GOx (100 μg/mL) and PCN(Fe/F).

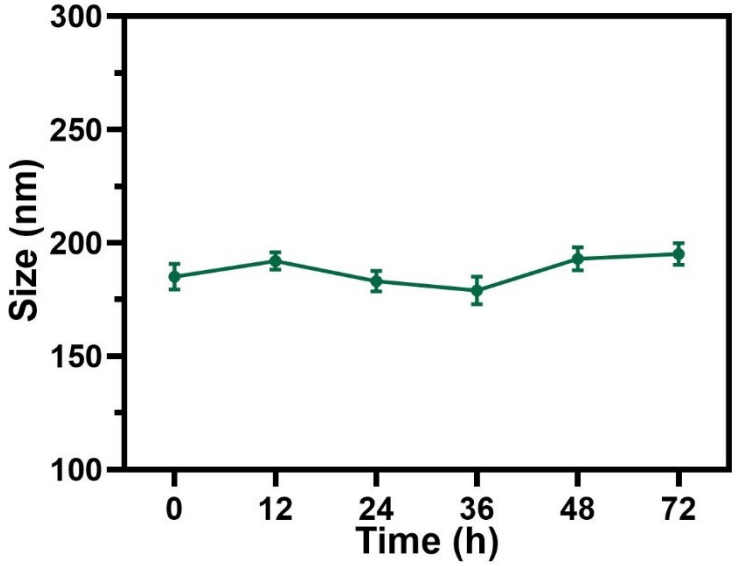

Fig. S4 The mean particle size changes of PCN(Fe/F)@GOx within 3 days in PBS solution (pH=7.4).

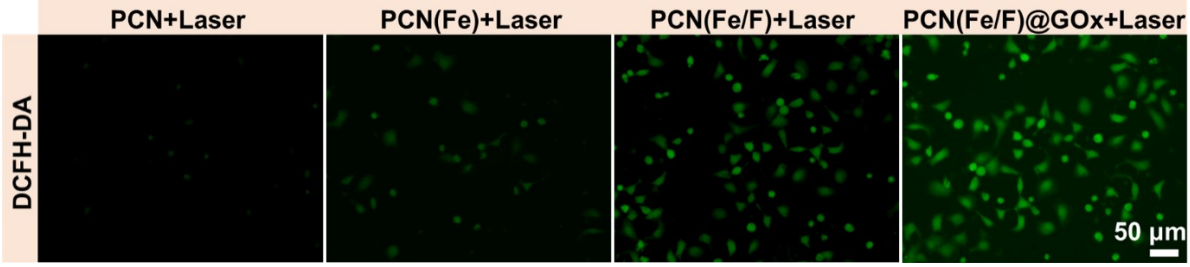

Fig. S5 Fluorescent images of intracellular ROS generation (DCFH-DA) stained HeLa cells treated with PCN, PCN(Fe), PCN(Fe/F), and PCN(Fe/F)@GOx under 660 nm

laser irradiation for 5 min (50 mW cm^{-2}).

References.

1. Zhou, Z.; Li, M.; Zhao, J.; Di, Z.; Di, C.; Liu, B.; Zhang, C.; Yan, C.-H.; Li, L., Trace water mediated growth of oriented single-crystalline mesoporous metal–organic frameworks on gold nanorods. *Chem. Commun.* 2018, **54**, 8182-8185.

2. (a) Huang, Y.-B.; Shen, M.; Wang, X.; Huang, P.; Chen, R.; Lin, Z.-J.; Cao, R., Water-medium C–H activation over a hydrophobic perfluoroalkane-decorated metal-organic framework platform. *J. Catal.* 2016, **333**, 1-7. (b) Deria, P.; Mondloch, J. E.; Tylianakis, E.; Ghosh, P.; Bury, W.; Snurr, R. Q.; Hupp, J. T.; Farha, O. K., Perfluoroalkane Functionalization of NU-1000 via Solvent-Assisted Ligand Incorporation: Synthesis and CO_2 Adsorption Studies. *J. Am. Chem. Soc.* 2013, **135**, 16801-16804.