Supporting Information

Cell-Penetrating Peptides Noncovalently Modified Red Phosphorescent Nanoparticles for High-Efficiency Imaging

Zihan Luo[a], Zhuofan Zhou[a], Yiwen Pan[a], Zece Zhu[b], Huanxiang Yuan[c], Yutao Li[d], Shumin Feng*[a], Yi Hong*[a], Li Xu*[a]

a. Department of Pharmacy, Hubei University of Chinese Medicine, Wuhan 430065, P. R. China.

b. School of Bioengineering and Health, State Key Laboratory of New Textile Materials and Advanced Processing Technologies, Wuhan Textile University, Wuhan 430200, P. R. China.

c. Department of Chemistry, College of Chemistry and Materials Engineering, Beijing Technology and Business University, Beijing 100048, P. R. China.

d. School of Laboratory Medicine, Hubei University of Chinese Medicine, Wuhan 430065, P. R. China.

*Corresponding author E-mail: 2997@hbtcm.edu.cn. (Li Xu)

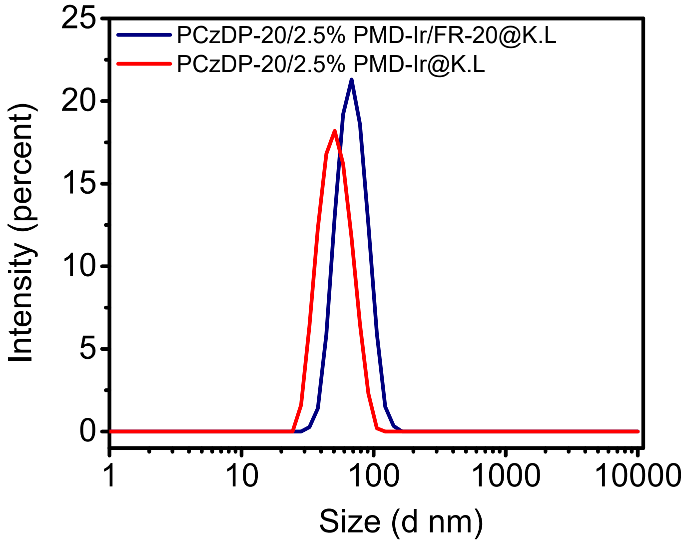


Figure S1. DLS histograms of PCzDP-20/2.5% PMD-Ir/FR-20@K.L and PCzDP-20/2.5% PMD-Ir@K.L nanoparticles. [PCzDP-20] = 200 μg/mL, [PMD-Ir] = 5 μg/mL, [FR-20] = 25 μg/mL.

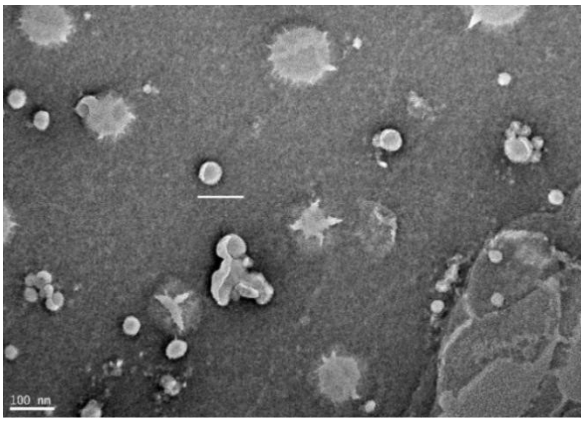
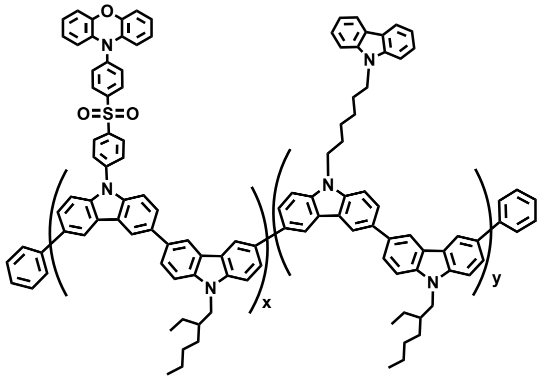


Figure S2. TEM images of PCzDP-20/2.5% PMD-Ir/FR-20@K.L nanoparticles. Scale bar = 100 nm for the images.

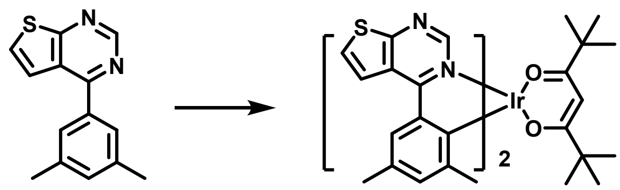
PCzDP-20 Structure

PCzDP-20 was synthesized by Yang group, and reported in literature[1]. The structure is as follows:



PCzDP-20 x:y=2:3

PMD-Ir Synthesis



PMD-L PMD-Ir

PMD-Ir was synthesized according to the previous work[2]. PMD-L (2.5 mmol), $IrCl_3 \cdot 3H_2O$ (1 mmol), 2-ethoxyethanol (15 mL) and H_2O (5 mL) was refluxed at 120 °C for 24 h under an argon atmosphere. The resulting mixture was cooled, filtered, and washed by water, ethanol, and diethyl ether to obtain the chloride-bridged dimer. Without further purification, a mixture of the dimer (0.4 mmol), Na_2CO_3 (2 mmol) and 2,2,6,6-tetrametylheptane-3,5-dionate (2 mmol) was added into 2-ethoxyethanol (20 mL). The reaction was heated at 90 °C for 12 h under an argon atmosphere. After cooling to room temperature, the reaction was quenched with water and extracted with

CH_2Cl_2. The product was purified by column chromatography on silica gel using CH_2Cl_2 as the eluent and then was recrystallized from CH_2Cl_2 and hexane. The desired compound as a red solid (Yield 39%): ^1H NMR (400 MHz, $CDCl_3$, δ): 8.67 (s, 2H), 8.20 (d, J = 6.3 Hz, 2H), 7.96 (s, 2H), 7.67 (d, J = 6.2 Hz, 2H), 6.63 (s, 2H), 5.39 (s, 1H), 2.33 (s, 6H), 1.46 (s, 6H), 0.71 (s, 18H); MS m/z: $[M+Na]^+$ calcd for $C_{39}H_{41}IrN_4NaO_2S_2$, 877.2198; found, 877.2196.

Calculation of transfer efficiency

In the case of receptor doped (F_{DA}) and undoped (F_D), the relative fluorescence intensity of the donor is usually used to calculate the transfer efficiency (η_{EnT}), and the formula is as follows[3]:

$$\eta_{EnT} = 1 - \frac{F_{DA}}{F_D}$$

Determination of quantum yield

The fluorescence quantum yields were calculated by comparison of the integrated area of the emission spectrum with a standard of $Ru(bpy)_3Cl_2$ in H_2O (0.02 mM, Φ = 2.8%)[4, 5]. The quantum yields were calculated with the following expression:

$$\Phi_x = \Phi_{st} \cdot \frac{I_x}{I_{st}} \cdot \frac{A_{st}}{A_x}$$

Φ_{st} is the reported standard quantum yield, I is the area under the emission spectrum, and A is the absorbance at 355 nm of the excitation wavelength. Fluorescence spectra were measured by Hitachi F-4600 spectrophotometer at the same room temperature.

Table S1. Photophysical properties of PMD-Ir, PCzDP-20, and their nanoparticles

	medium	λ_{abs} [nm]	$\lambda_{em, max}$ [nm]	Φ_{em}
PMD-Ir	Mill H_2O		622	1.9 %
	CH_2Cl_2		611	15.1 %
PCzDP-20	Mill H_2O		509	23.3 %
	CH_2Cl_2		525	29.1 %
PMD-Ir/FR-20@K.L	Mill H_2O	< 300	598	3.7 %
PCzDP-20/FR-20@K.L	Mill H_2O		514	43.6 %
PCzDP-20/2.5% PMD-Ir/FR-20@K.L	Mill H_2O	< 300	609	30.8 %

$Ru(bpy)_3Cl_2$ as reference

References

1. Xie, G.; Luo, J.; Huang, M.; Chen, T.; Wu, K.; Gong, S.; Yang, C., Inheriting the Characteristics of TADF Small Molecule by Side-Chain Engineering Strategy to Enable Bluish-Green Polymers with High PLQYs up to 74% and External Quantum Efficiency over 16% in Light-Emitting Diodes. *Adv. Mater.* **2017,** *29* (11), 1604223.
2. Jiang, B.; Zhao, C.; Ning, X.; Zhong, C.; Ma, D.; Yang, C., Using Simple Fused-Ring Thieno[2,3-d]pyrimidine to Construct Orange/Red Ir(III) Complexes: High-Performance Red Organic Light-Emitting Diodes with EQEs up to Nearly 28%. *Adv. Opt. Mater.* **2018,** *6* (14), 1800108.
3. J. R. Lakowicz, *Principles of Fluorescence Spectroscopy*, Springer, New York, **2006,** 873-953
4. Zheng, X.; Tang, H.; Xie, C.; Zhang, J.; Wu, W.; Jiang, X., Tracking Cancer Metastasis In Vivo by Using an Iridium-Based Hypoxia-Activated Optical Oxygen Nanosensor. *Angew Chem Int Ed Engl* **2015,** *54* (28), 8094-8099.
5. Trofymchuk, K.; Reisch, A.; Didier, P.; Fras, F.; Gilliot, P.; Mely, Y.; Klymchenko, A. S., Giant light-harvesting nanoantenna for single-molecule detection in ambient light. *Nat Photonics* **2017,** *11* (10), 657-663.