

## Supporting Information

### **Simultaneous Two-Color Visualization of Lipid Droplets and Lysosomes for Cell Homeostasis Monitoring Using a Single Fluorescent Probe**

Mengxiao Liu<sup>1, a</sup>, Yingcui Bu<sup>1, a\*</sup>, Dongxiao Wang<sup>a</sup>, Lihua Tang<sup>a</sup>, Didi Hu<sup>a</sup>, Longchun Li<sup>a</sup>, Xiaoping Gan<sup>a, b\*</sup>

<sup>a</sup> School of Materials and Chemistry, Anhui Agricultural University, 230036 Hefei, P. R. China

<sup>b</sup> Key Laboratory of Agricultural Sensors, Ministry of Agriculture Rural Affairs, 230036 Hefei, P. R. China

\*Corresponding author Fax: +86-551-65786121

E-mail: gan-xp@ahau.edu.cn, byc@ahau.edu.cn.

<sup>1</sup>these authors contributed equally to this work and should be considered as co-first authors

## 1.1 Synthesis of the intermediate M1

0.82 g (2 mmol) 1-(4-bromophenyl)-1,2,2-triphenylethylene, 0.80 g (8 mmol) potassium acetate, 0.63 g (2.50 mmol) Bis(pinacolato) diboron and a catalytic amount of palladium triphenyl phosphine dichloride were dissolved in 20 mL of anhydrous 1,4-dioxane. The reaction was carried out at 80 °C under the protection of N<sub>2</sub> for 24 h. After the reaction was completed, 100 mL dichloromethane was added to the mixture to dilute it, and extracted with an equal volume of pure water for 3 times. After the organic phase was dried with anhydrous calcium chloride, the solvent was removed by vacuum distillation and white solid **M1** was obtained by column chromatography (0.77 g, 84 % yield). **M1**: <sup>1</sup>H NMR (600 MHz, CDCl<sub>3</sub>) δ 7.54-7.53 (d, *J*=6.0 Hz, 2H), 7.08 (t, *J*=6.0 Hz, 9H), 7.05-7.02 (d, *J*=12.0 Hz, 8H), 1.31 (s, 12H). <sup>13</sup>C NMR (151 MHz, CDCl<sub>3</sub>) δ 146.72, 143.68, 143.59, 143.51, 141.38, 140.88, 134.07, 131.31, 131.28, 131.27, 130.64, 127.69, 127.59, 126.48, 126.41, 126.39, 83.64, 24.87. The HRMS was seen from the previous literature <sup>[1]</sup>. Each of spectrum has impurity peak at 1.6 ppm, which might be due to the influence of the solvent itself.

## 1.2 Synthesis of the intermediate M2

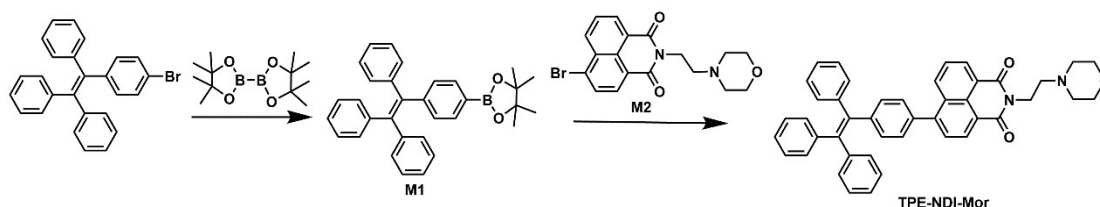
1 mmol (0.28 g) of 6-bromo-1H, 3H-benzo[d]isochromene-1, 3-dione was dissolved in 15 mL of ethanol, and 0.23 g of zinc acetate

was added to the mixture, which was refluxed at 80 °C for 10 min until dissolved. Then 1 mmol (0.33 mL) of N-(2-amino-ethyl) morpholine was added to the mixture. The reaction was carried out at 80 °C for 24 h. After the reaction was completed, it was first cooled and crystallized, then filtered, and then a white solid was obtained. (0.32 g, yield: 83 %). **M2**: <sup>1</sup>H NMR (600 MHz, CDCl<sub>3</sub>) δ 8.65-8.64 (d, *J* = 6.0 Hz, 1H), 8.57-8.56 (d, *J* = 6.0 Hz, 1H), 8.41-8.39 (d, *J* = 12.0 Hz, 1H), 8.04-8.03 (d, *J* = 6.0 Hz, 1H), 7.84 (t, *J* = 12.0 Hz, 1H), 4.33 (s, 2H), 3.67 (s, 4H), 2.71 (s, 2H), 2.59 (s, 4H), 1.59 (s, 1H). <sup>13</sup>C NMR (151 MHz, CDCl<sub>3</sub>) δ 163.61, 133.28, 132.00, 131.19, 131.09, 130.69, 128.06, 122.23, 37.25. The HRMS was seen from the previous literature [2]. Each of spectrum has impurity peak at 1.6 ppm, which might be due to the influence of the solvent itself.

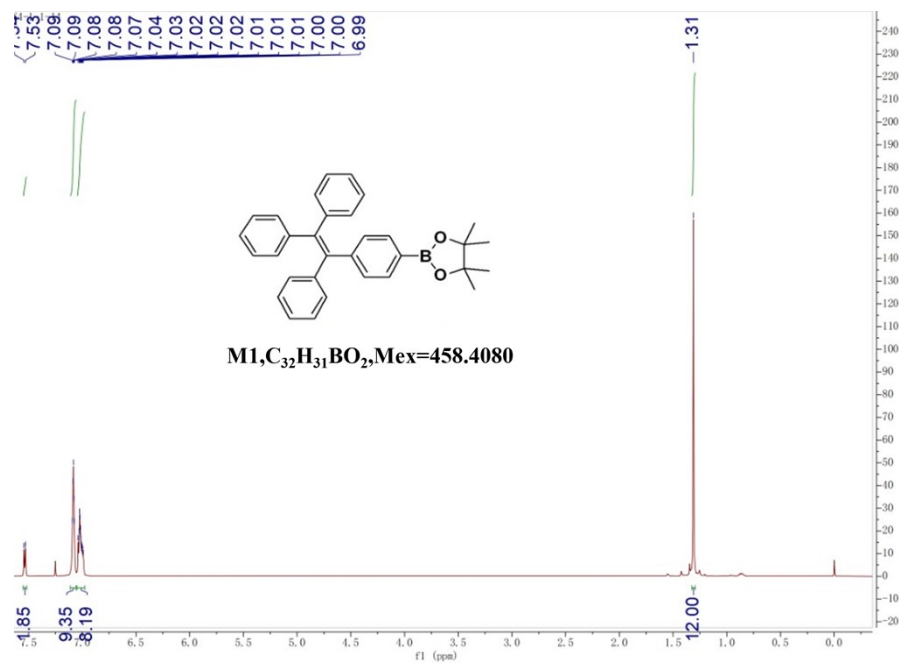
### 1.3 Synthesis of the compound TPE-NDI-Mor

0.46 g (1 mmol) **M1**, 0.39 g (1 mmol) **M2**, catalytic amount of palladium triphenyl phosphine dichloride, 0.69 g (5 mmol) potassium carbonate, 3 drops of trioctyl methyl ammonium chloride and 2.50 mL of pure water were dissolved in 20 mL of toluene. The reaction was carried out at 85 °C under the protection of N<sub>2</sub> for 24 h. After the reaction was complete, 100 mL dichloromethane was added into the mixture to dilute it, and extracted with an equal volume of pure water for 3 times. Then organic phase was dried with anhydrous calcium

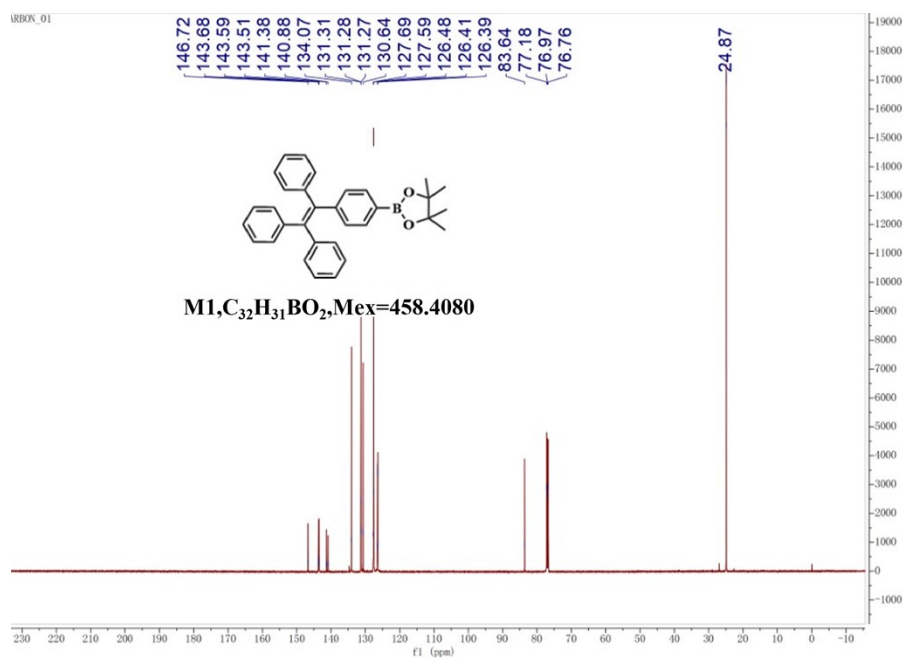
chloride, the solvent was removed by vacuum distillation and a yellowish-green solid was obtained by column chromatography (0.51 g, 80 % yield). **TPE-NDI-Mor**:  $^1\text{H}$  NMR (600 MHz,  $\text{CDCl}_3$ )  $\delta$  8.60 (t,  $J = 6.0$  Hz, 2H), 8.19-8.18 (d,  $J = 6.0$  Hz, 1H), 7.70-7.68 (d,  $J = 12.0$  Hz, 1H), 7.67-7.65 (d,  $J = 12.0$  Hz, 1H) 7.21-7.19 (d,  $J = 12.0$  Hz, 2H), 7.16 (s, 6H), 7.12 (s, 8H), 7.07 (s, 2H), 4.37 (s, 2H), 3.69 (s, 4H), 2.73 (s, 3H), 2.62 (s, 4H).  $^{13}\text{C}$  NMR (151 MHz,  $\text{CDCl}_3$ )  $\delta$  164.30, 164.10, 146.80, 144.19, 143.57, 143.36, 143.31, 141.99, 140.20, 136.64, 132.65, 131.53, 131.37, 131.31, 131.24, 131.10, 130.76, 130.03, 129.19, 128.75, 127.81, 127.73, 127.69, 126.69, 126.66, 126.63, 122.84, 121.55, 66.95, 56.14, 53.77, 37.16. MS (APCI):  $m/z$  641.2780  $[(\text{M} + \text{H})^+]$ , calcd 641.2799].



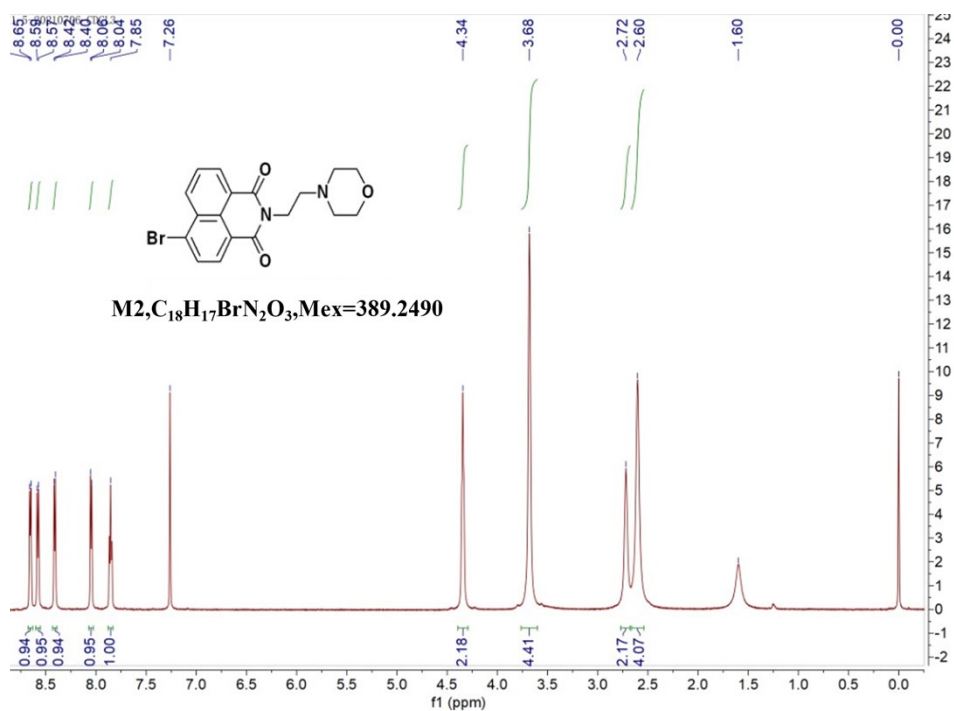
**Scheme S1** The synthetic routes of target compound **TPE-NDI-Mor**.



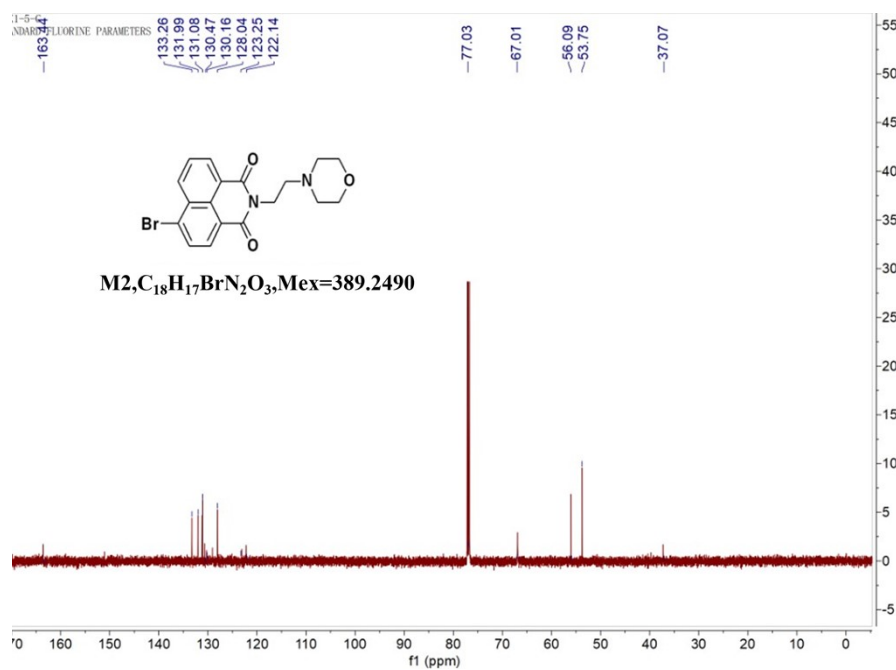
**Fig. S1** The <sup>1</sup>H NMR (600 MHz) spectrum of intermediate **M1** in CDCl<sub>3</sub>



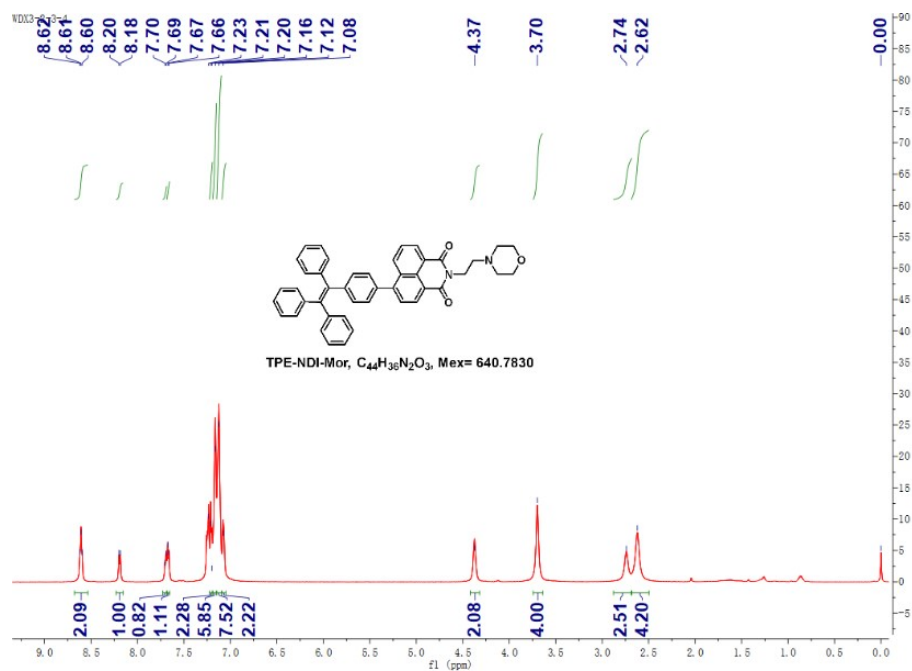
**Fig. S2** The <sup>13</sup>C NMR (151 MHz) spectrum of intermediate **M1** in CDCl<sub>3</sub>



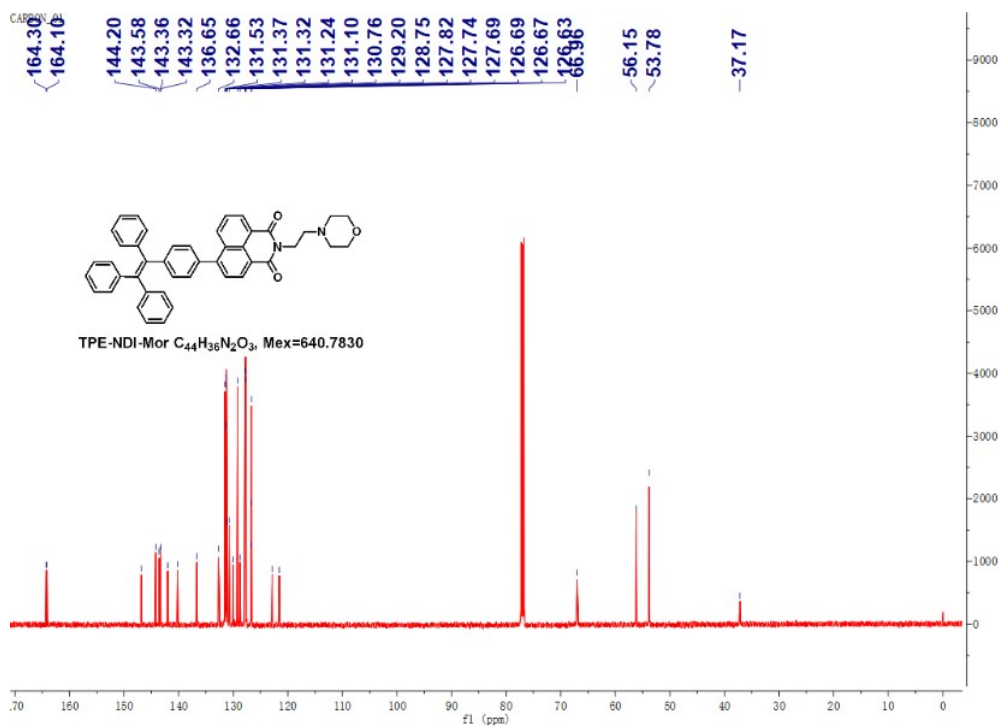
**Fig. S3** The <sup>1</sup>H NMR (600 MHz) spectrum of intermediate **M2** in CDCl<sub>3</sub>



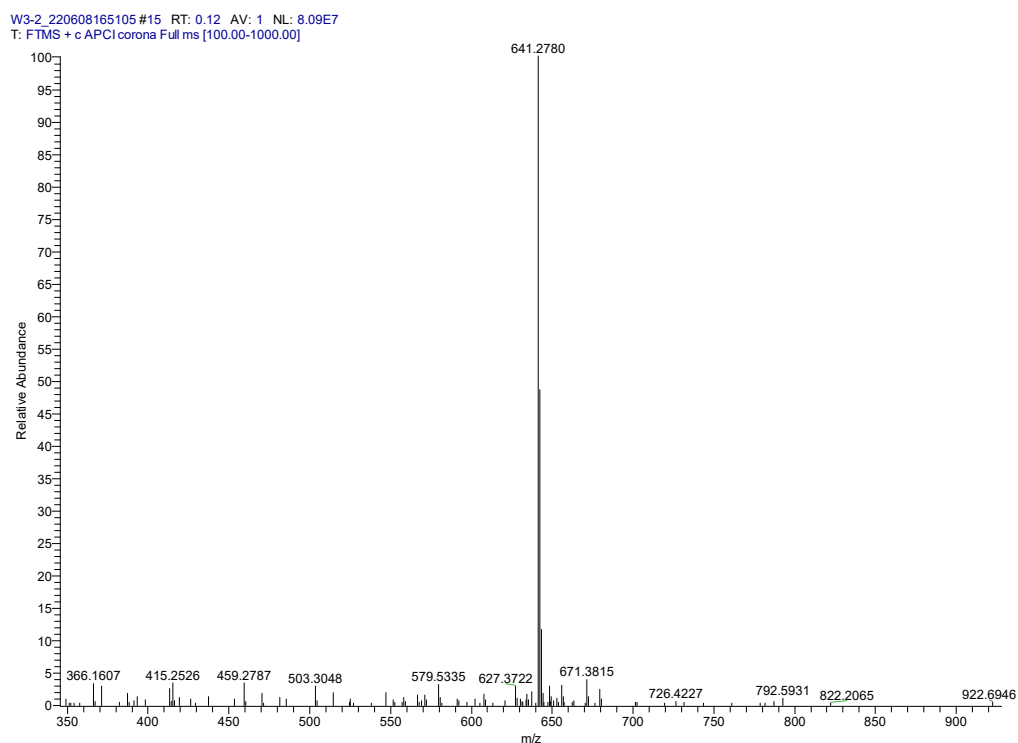
**Fig. S4** The <sup>13</sup>C NMR (151 MHz) spectrum of intermediate **M2** in CDCl<sub>3</sub>



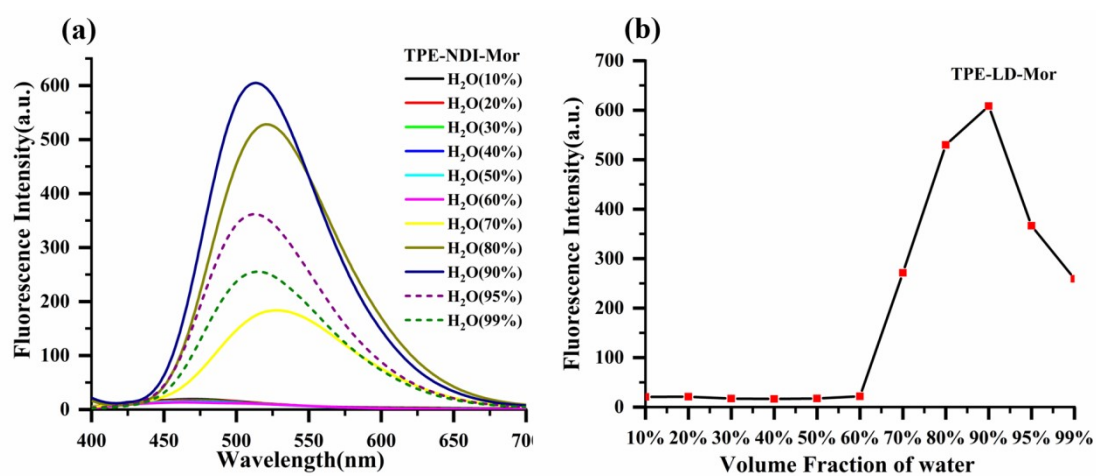
**Fig. S5** The <sup>1</sup>H NMR (600 MHz) spectrum of compound **TPE-NDI-Mor** in CDCl<sub>3</sub>



**Fig. S6** The <sup>13</sup>C NMR (151 MHz) spectrum of compound **TPE-NDI-Mor** in CDCl<sub>3</sub>

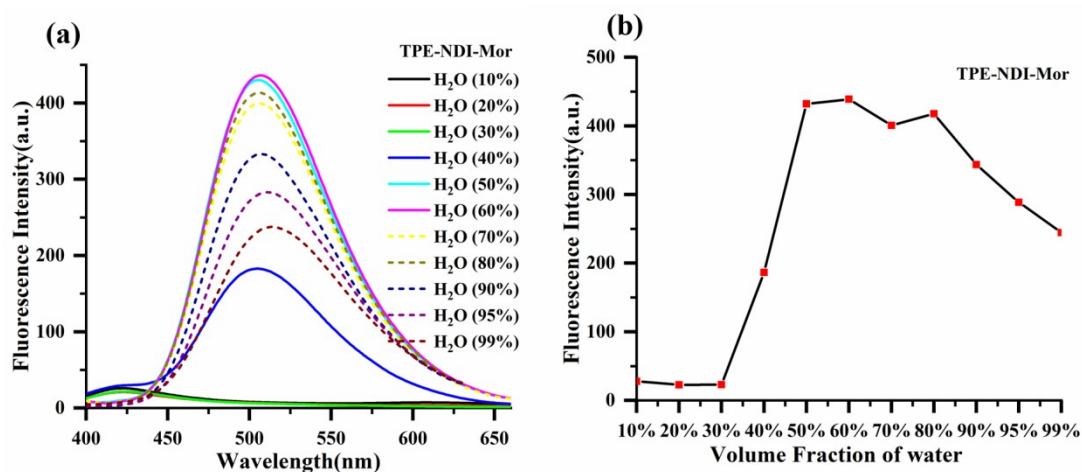


**Fig. S7** HRMS of compound **TPE-NDI-Mor**.

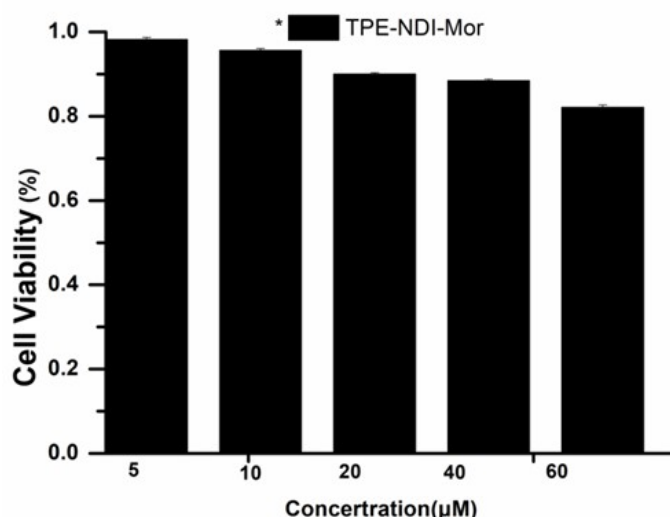


**Fig. S8** a) Fluorescence spectra of **TPE-NDI-Mor** in ACN/H<sub>2</sub>O mixtures with different water fractions. b) Fitted curve of maximum fluorescence intensity versus water fractions for the probe **TPE-NDI-Mor**.

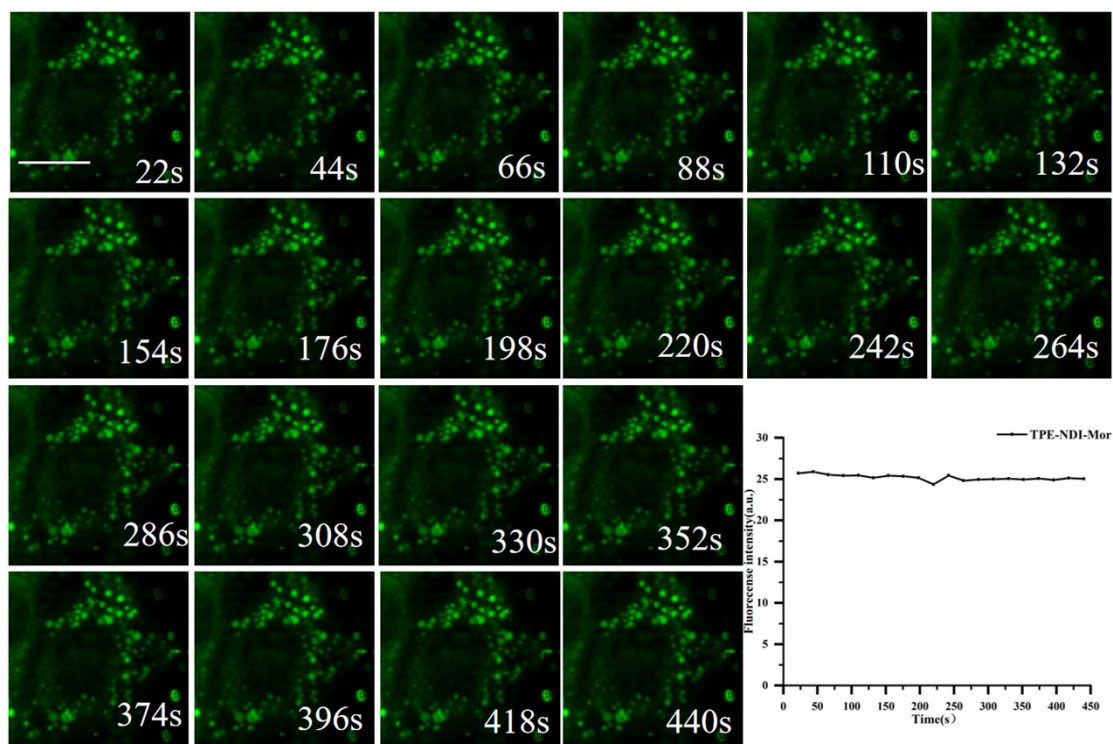




**Fig. S9** Fluorescence spectrum of **TPE-NDI-Mor** in DMSO/H<sub>2</sub>O mixtures with different water fractions (a). Polygraph of **TPE-NDI-Mor** in DMSO/H<sub>2</sub>O mixture with different water fractions (b).



**Fig. S10** MTT assay of HepG2 cells treated with **TPE-NDI-Mor** at different concentrations for 24 h.



**Fig. S11** Confocal fluorescence images of HepG2 cells treated with the **TPE-NDI-Mor** ( $\lambda_{\text{ex}} = 405 \text{ nm}$ ) under confocal laser scanning microscope with different irradiation time. Scale bar: 20  $\mu\text{m}$ .

**Table S1:** Photophysical properties of **TPE-NDI-Mor** in different solvents.

		Toluene	Dioxane	EtOAc	$\text{CHCl}_3$	DMSO	ACN
<b>TPE-NDI-Mor</b>	$\lambda_{\text{abs}} \text{ (nm)}$ <sup>[a]</sup>	327, 371	241, 369	251, 362	242, 382	260, 372	239, 361
	$\lambda_{\text{em}} \text{ (nm)}$ <sup>[b]</sup>	498	509	538	555	614	608
	$E_{\text{T}}(30)$ <sup>[c]</sup>	33.9	36	38.1	39.1	45	46
	Stokes shift (nm)	127	140	176	173	242	247
	$\Phi_{\text{F}}$ <sup>[c]</sup>	4.7 %	7.2 %	7.9 %	30.6 %	2.3 %	2.3 %

[a] represented the wavelength of UV-vis absorption; [b] represented the wavelength of Fluorescence emission [c] represented the

Empirical polarity parameter <sup>[e]</sup> represented the Quantum yield of fluorescent light.

### **Reference**

- S1. Y. Lin, C.M. Deng, L.Tang, A.J. Qin, R.R. Hu, J.Z. Sun, B.Z. Tang. *J. Am. Chem. Soc.*, 2011, **113**, 660-663.
- S2. J. Yin, M. Peng, W. Y. Lin. *Chem. Commun.*, 2019, **55**, 11063-11066.