

## Supporting Information

**Fig. S1.**  $^1\text{H}$  NMR(600 MHz) of **Probe NE** in DMSO  $-d_6$ .

**Fig. S2.**  $^{13}\text{C}$  NMR(150 MHz) **Probe NE** in DMSO  $-d_6$ .

**Fig. S3.** HR-MS spectrum of **Probe NE**.

**Fig. S4.** The fluorescence intensity of **Probe NE** toward  $\text{N}_2\text{H}_4$  under different pH conditions.

**Fig. S5.**  $^1\text{H}$  NMR titration of **Probe NE** towards  $\text{N}_2\text{H}_4$ .

**Fig. S6.** The HR-MS spectrum of [**Probe NE** +  $\text{N}_2\text{H}_4$ ].

**Fig. S7.** Cell viability values estimated by CCK-8 assay with HeLa cells.

**Fig. S8.** Viability of HeLa cells by treating with probe **Probe RE** and **Probe NE**.

### 1. Materials and instruments

The materials and reagents used are commercially available and have not been further purified. The solution of compounds was prepared of deionized water. UV-vis spectroscopy was performed using HITACHI U-3900 spectrophotometer, and fluorescence spectroscopy was performed using HITACHI F-7000 spectrophotometer. All related compounds were characterized by  $^1\text{H}$  NMR and  $^{13}\text{C}$  NMR using a Bruker AVANCE-600 MHz spectrometer, followed by mass spectrometry using an AB Triple TOF 5600plus System (AB SCIEX, Framingham, USA). The final bioimaging application were measured the Zeiss LSM880 Airyscan confocal laser scanning microscope.

### 2. Experimental Section

Precursors were synthesized using literature method. Synthesis of **Probe NE** (Scheme 1). In a 25.0 mL round bottom flask, 6-methoxyquinoline-2-carbaldehyde (0.245 g, 1.00 mmol) and 1,2,3,3-tetramethyl-3H-indol-1-ium (0.186 g, 1.00 mmol) were dissolved in ethanol (10.0 mL). Piperidine (0.150 mL) was added and the mixture was refluxed for 8 h. The reaction was cooled to room temperature, the crude product was purified by silica gel column chromatography using ethyl acetate and

petroleum ether (1:8) as eluent to afford pure compound Probe NE as a yellow solid (0.240 g, yield 58.5%). <sup>1</sup>H NMR (600 MHz, DMSO) δ 8.52, 8.48, 8.29, 8.09, 8.01, 7.93, 7.68, 7.54, 7.49, 4.22, 3.96, 1.84. (Fig. S1); <sup>13</sup>C NMR (151 MHz, DMSO) δ 163.51, 162.85, 156.13, 155.55, 149.80, 133.62, 132.01, 129.97, 128.93, 123.03, 120.84, 119.11, 118.59, 115.10, 61.96, 30.06, 20.27, 14.57, 14.21. (Fig. S2); HR-MS m/z: calcd for 343.18049; found: m/z: 343.18040(Fig. S3).

### 3. Spectral test preparation

**Probe NE** was dissolved in DMSO to make a 2.0 mM stock solution. Through testing, we finally chose DMSO / PBS (5/5, v/v, pH = 7.4) as the test system, the probe concentration was 10.0 μM. The concentration of interfering ions is 0.1 M.

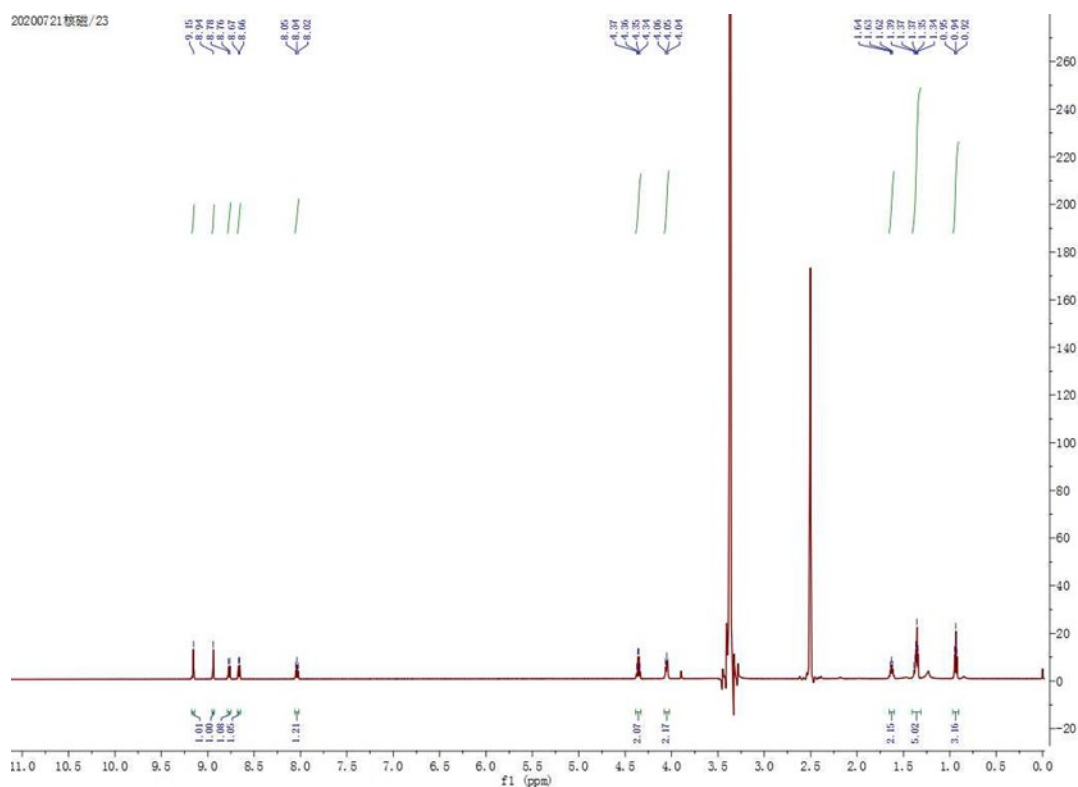
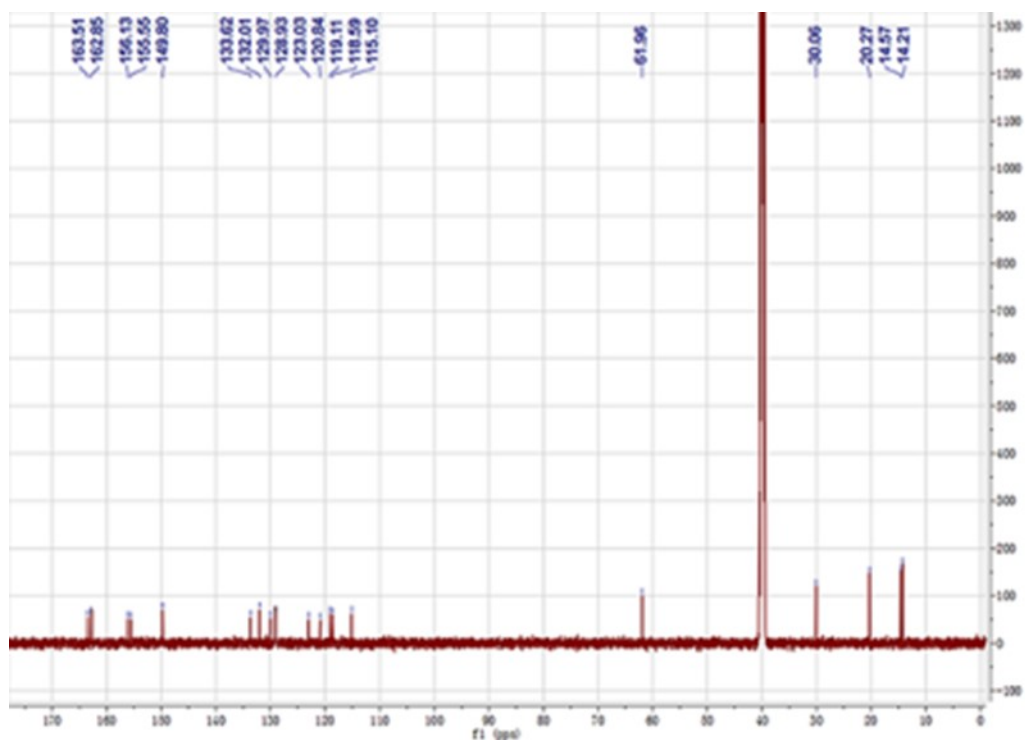
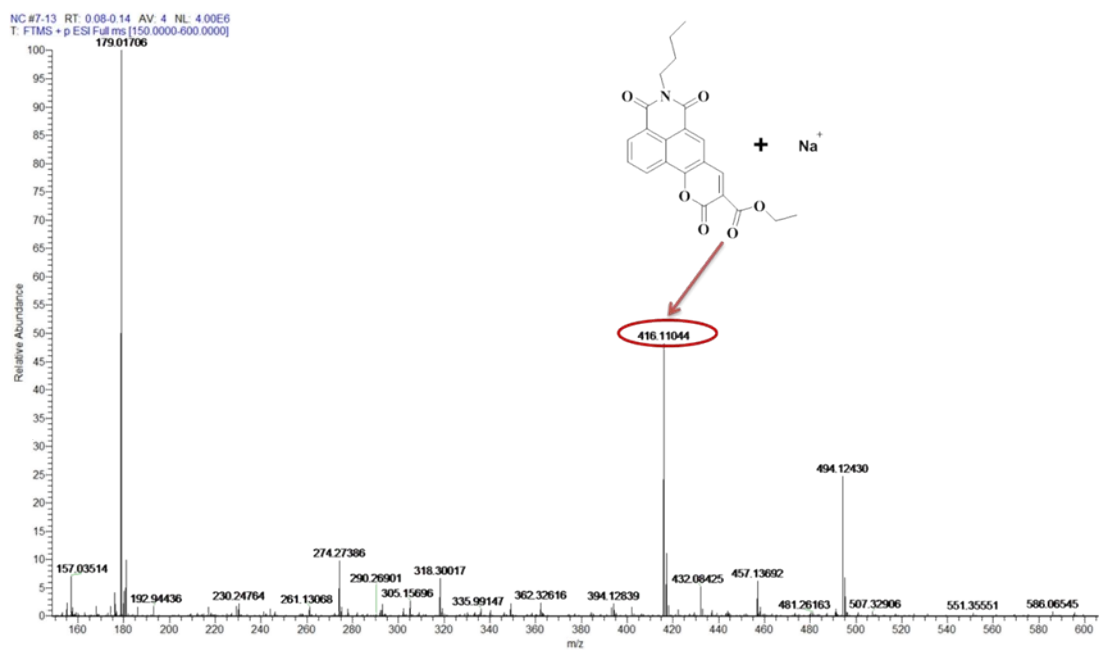


Fig. S1. <sup>1</sup>H NMR(600 MHz) of **Probe NE** in DMSO -*d*<sub>6</sub>.



**Fig. S2.**  $^{13}\text{C}$  NMR (150 MHz) of Probe NE in  $\text{DMSO-}d_6$



**Fig. S3.** HR-MS spectrum of Probe NE.

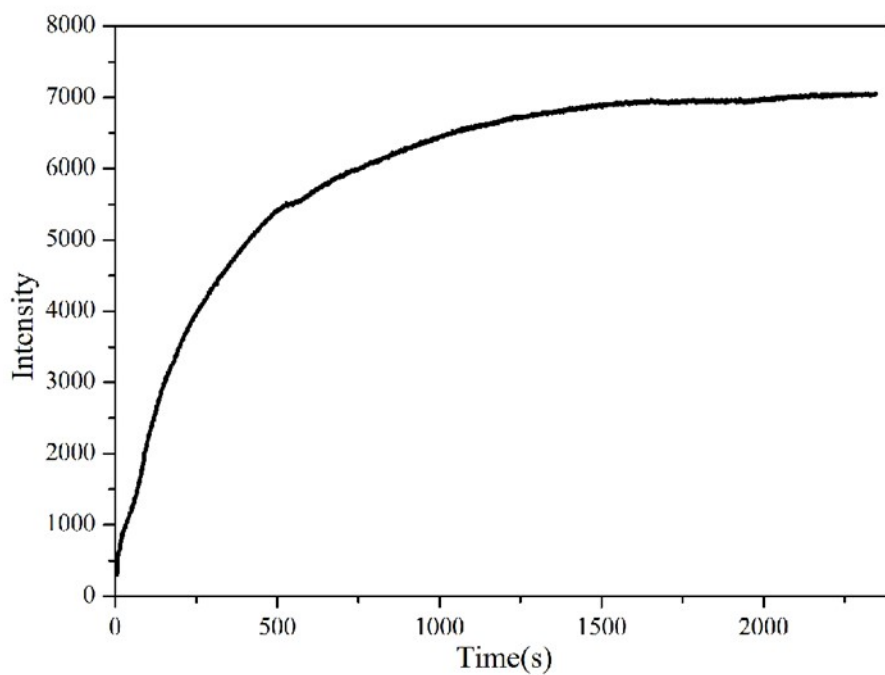


Fig. S4. Kinetics study of **Probe NE** towards  $N_2H_4$ .

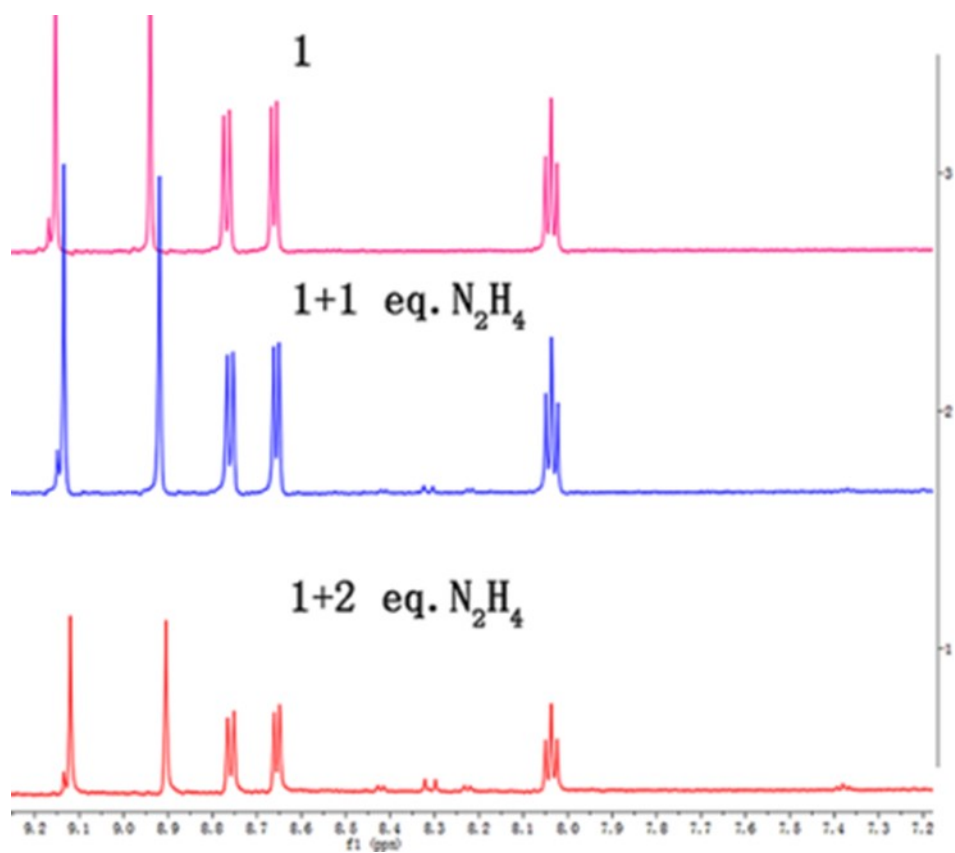


Fig. S5.  $^1H$  NMR titration of **Probe NE** towards  $N_2H_4$ .

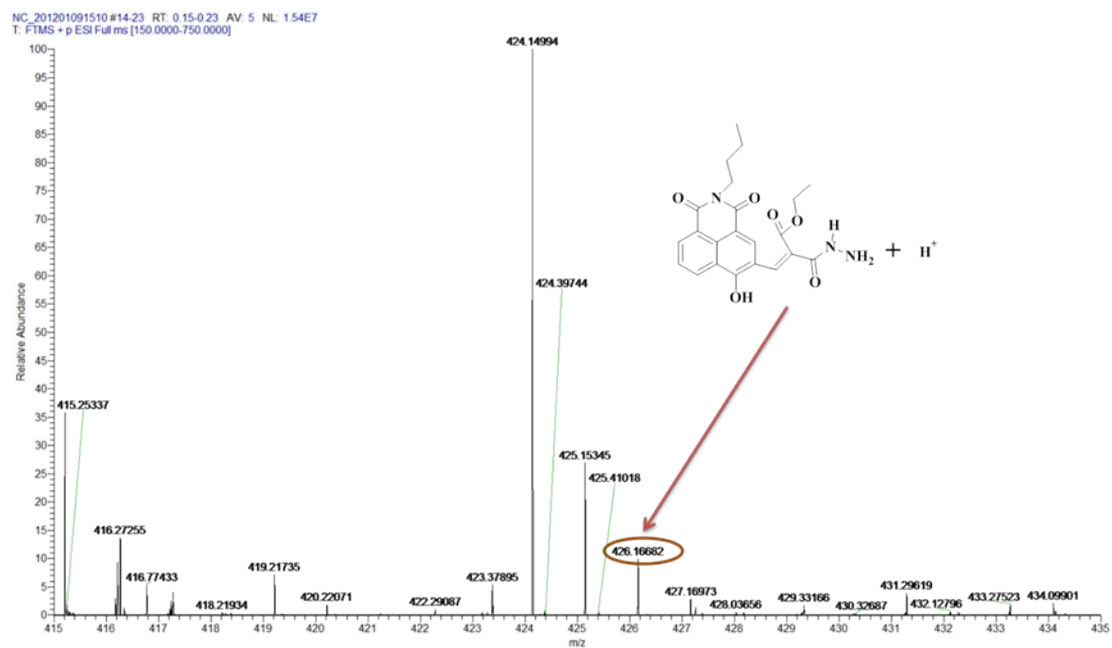


Fig. S6. The HR-MS spectrum of [Probe NE + N<sub>2</sub>H<sub>4</sub>].

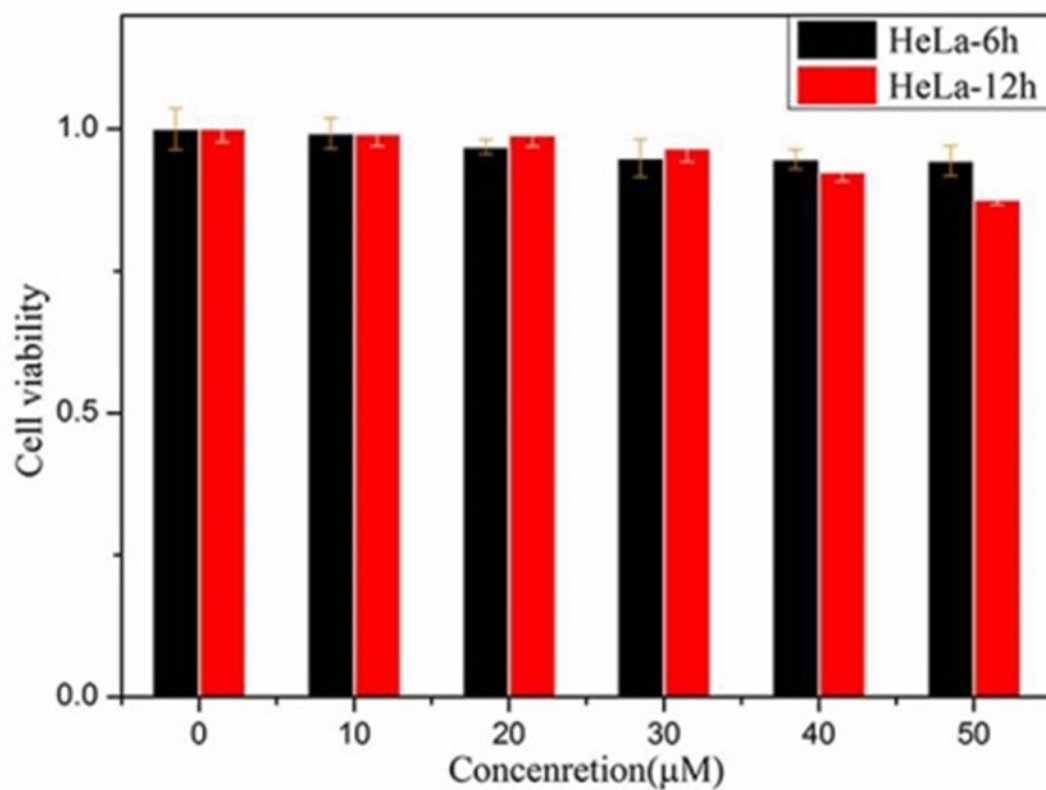


Fig. S7. Cell viability values estimated by CCK-8 assay with HeLa cells.

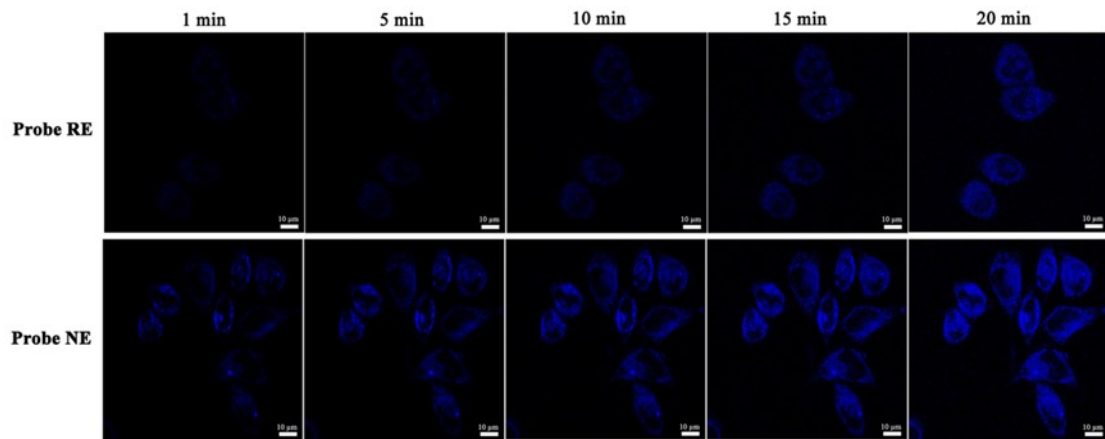


Fig. S8. Viability of HeLa cells by treating with probe **Probe RE** and **Probe NE**.