## **Electronic Supplementary Information** (ESI)

## A novel pH probe based on rhodamine-rhodanine platform

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## 1. Part of optical spectroscopic data.



Fig. S1 UV-vis absorbance of **RER** (10  $\mu$ M) to pH variation in aqueous solvents containing ethanol (B-R buffer-EtOH, v/v = 7:3)



Fig. S2 The time courses of fluorescence intensity of **RER** in aqueous solvents containing ethanol (B-R buffer-EtOH, v/v = 8:2) at various pH values (4.1, 5.3 and 7.0, respectively). ( $\lambda_{ex} = 561 \text{ nm}, \lambda_{em} = 583.5 \text{ nm}$ )



Fig. S3 Cytotoxicity of **RE** ( $\blacksquare$ ) and **RER** ( $\blacksquare$ ) to HeLa cells. HeLa cells were incubated with 1, 5 and 10  $\mu$ M **RE** or **RER** respectively for 24 h. The cell viability was determined by SRB assays.



Fig. S4 Photostability of **RER** (5  $\mu$ M). (a) Fluorescence images (0-300 s) were achieved by means of time-sequential scanning of the HeLa cells. (b) Normalized fluorescence intensity of per cell of (a) from 0 to 300 s.



2. Characterization data (IR, <sup>1</sup>H & <sup>13</sup>C NMR and HRMS) of RER.

Fig. S5 IR spectrum of **RER** 





Fig. S7 <sup>13</sup>C NMR spectrum of **RER** in DMSO



Fig. S8 HRMS spectrum of RER