

Supporting Information

A FRET-based fluorescent and colorimetric probe for the specific detection of picric acid

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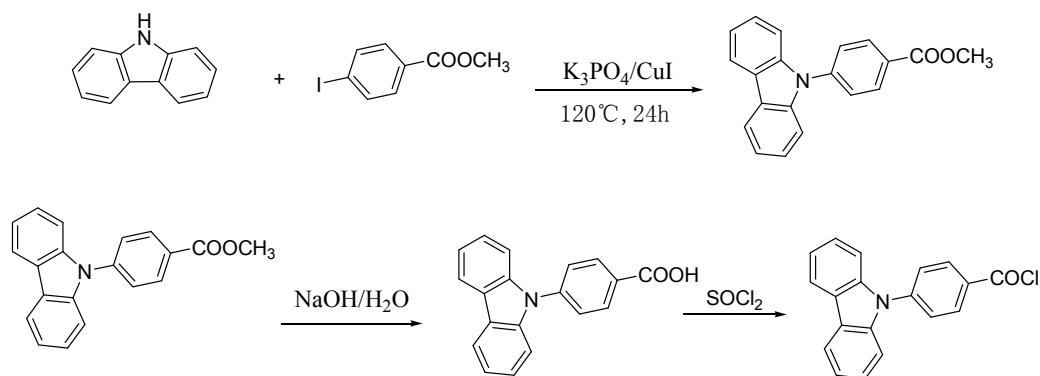
I- Synthesis and characterization of the intermediates

To a flask (25 ml) were added 9*H*-carbazole (184 mg, 1.1 mmol), methyl 4-iodobenzoate (262 mg, 1.0 mmol), K₃PO₄ (424 mg, 2 mmol), CuI (190 mg, 1 mmol) and DMF (5 ml). The mixture was stirred for 24 h at 120 °C until completion of the reaction (monitored by TLC). Upon cooling, the mixture was quenched with brine (40 ml) and the resulting solid was filtered and dried to furnish the desired product. The crude product was further purified by recrystallization from EtOAc-PE. Methyl 4-(9*H*-carbazol-9-yl)benzoate, white solid; ¹H NMR (400 MHz, DMSO-*d*₆) δ: ppm 8.27

(d, $J = 8.80$ Hz, 2H), 8.24 (d, $J = 8.80$ Hz, 2H), 7.83 (d, $J = 8.40$ Hz, 2H), 7.49 (t, $J = 6.00$ Hz, 2H), 7.45 (d, $J = 7.20$ Hz, 2H), 7.33 (t, $J = 6.80$ Hz, 2H), 3.93 (s, 3H); MS (EI): $m/z = 301[M]^+$, 270 $[M-OCH_3]^+$, 242 $[M-COOCH_3]^+$.

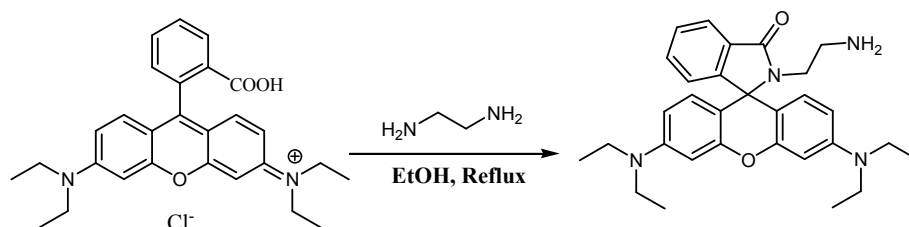
The purified methyl 4-(9*H*-carbazol-9-yl)benzoate was treated with concentrated sodium hydroxide at 80 °C for 12 h. After that the reaction mixture was neutralized with concentrated hydrochloric acid. The resulting solid was filtered and dried to furnish the desired product 4-(9*H*-carbazol-9-yl)benzoic acid, which was used without further purification.

To a flask (25 mL) were added 4-(9*H*-carbazol-9-yl)benzoic acid (247 mg, 1 mmol) and $SOCl_2$ (5 mL). The mixture was stirred at room temperature for 12 h, then concentrated under vacuum to furnish the crude product, which was used without further purification.



Scheme S1. The synthesis of 4-(9*H*-carbazol-9-yl)benzoyl chloride

N-(rhodamine-B)lactam-ethylenediamine was synthesized using Rhodamine-B and ethanediamine in ethanol according to the reported methods^[1].



Scheme S2. The synthesis of N-(rhodamine-B)lactam-ethylenediamine

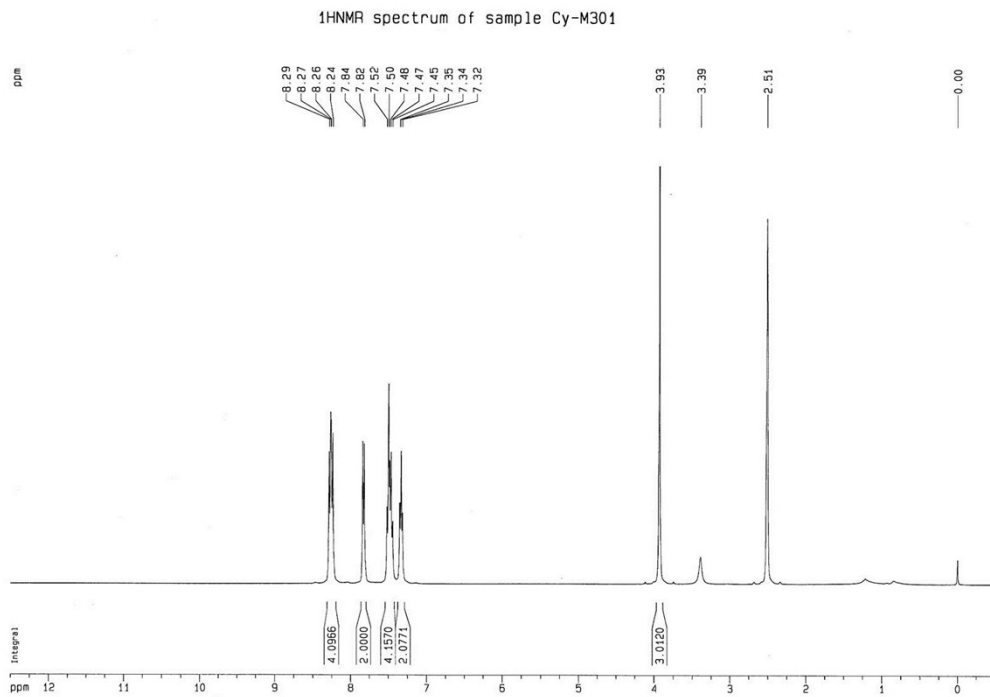


Fig. S1 ^1H NMR spectrum of methyl 4-(9*H*-carbazol-9-yl)benzoate in $\text{DMSO-}d_6$ (400 MHz)

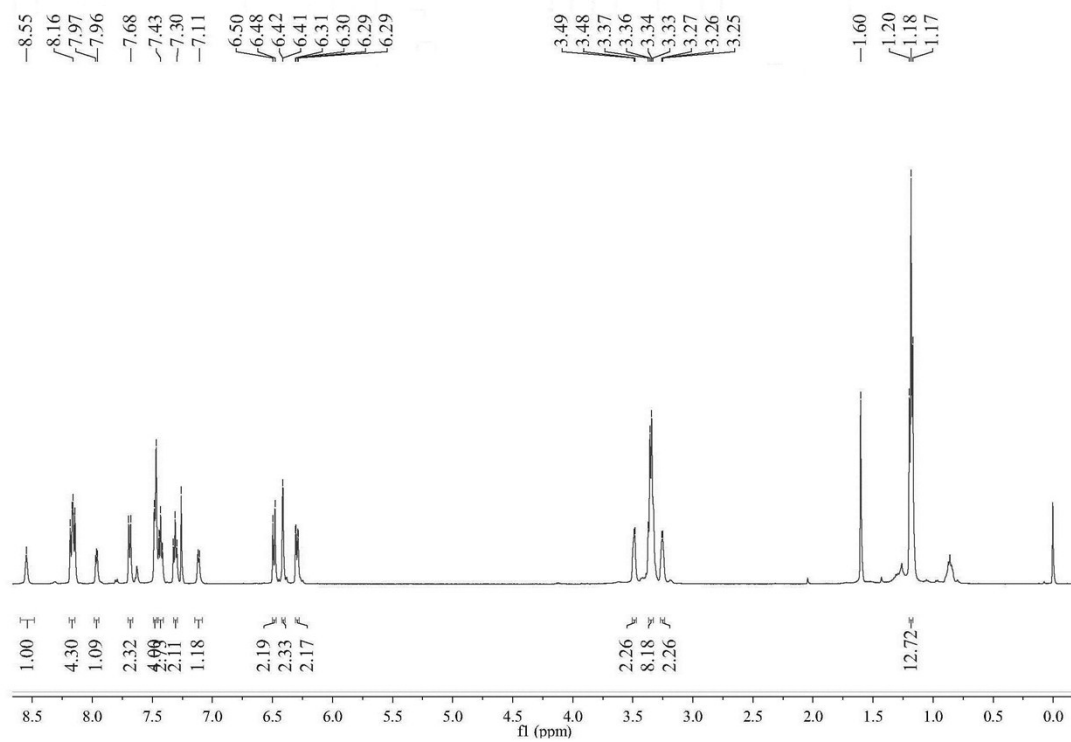


Fig. S2 ^1H NMR spectrum of probe (**L**) in CD_3Cl (500 MHz, CD_3Cl)

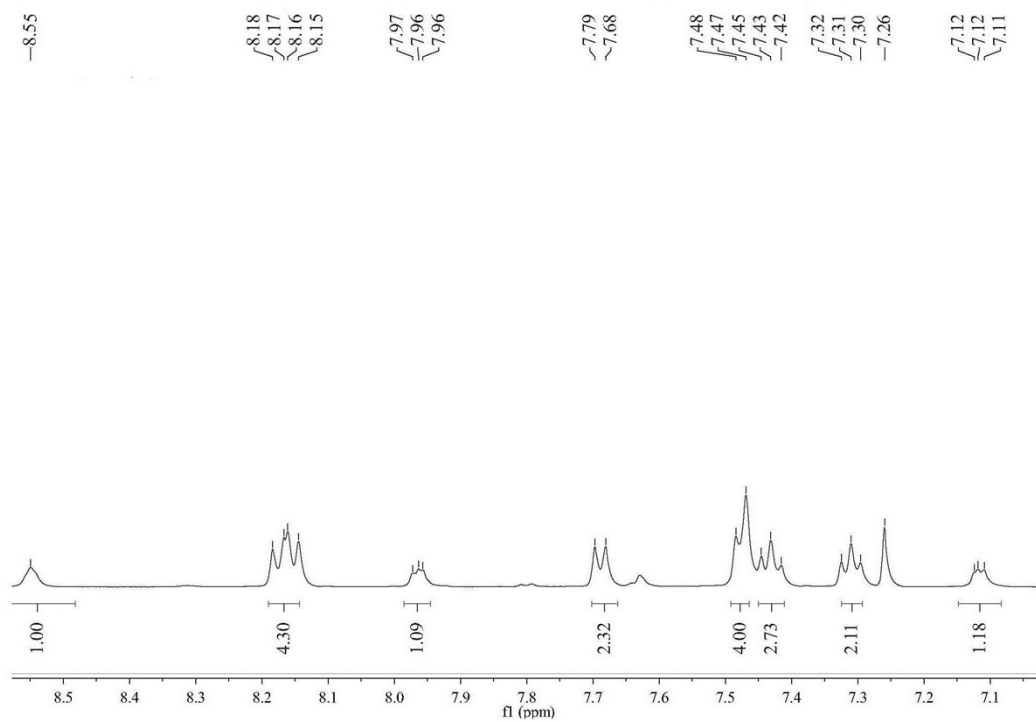


Fig. S3 ^1H NMR spectrum of probe (L) in CD_3Cl (500 MHz, CD_3Cl)

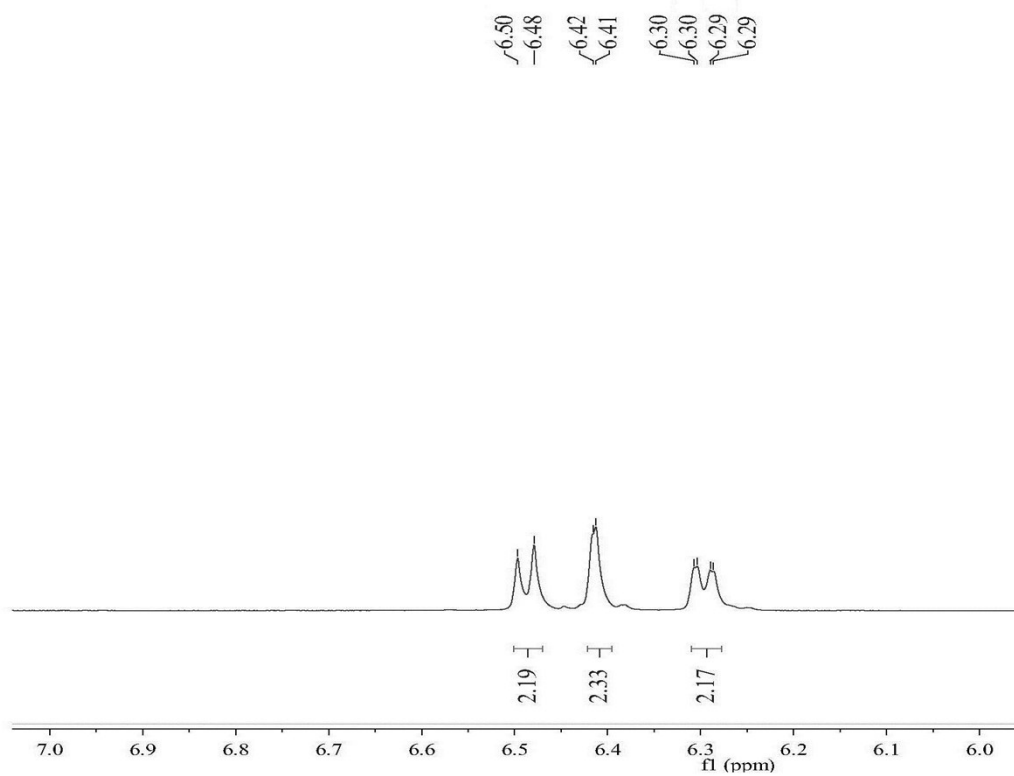


Fig. S4 ^1H NMR spectrum of probe (L) in CD_3Cl (500 MHz, CD_3Cl)

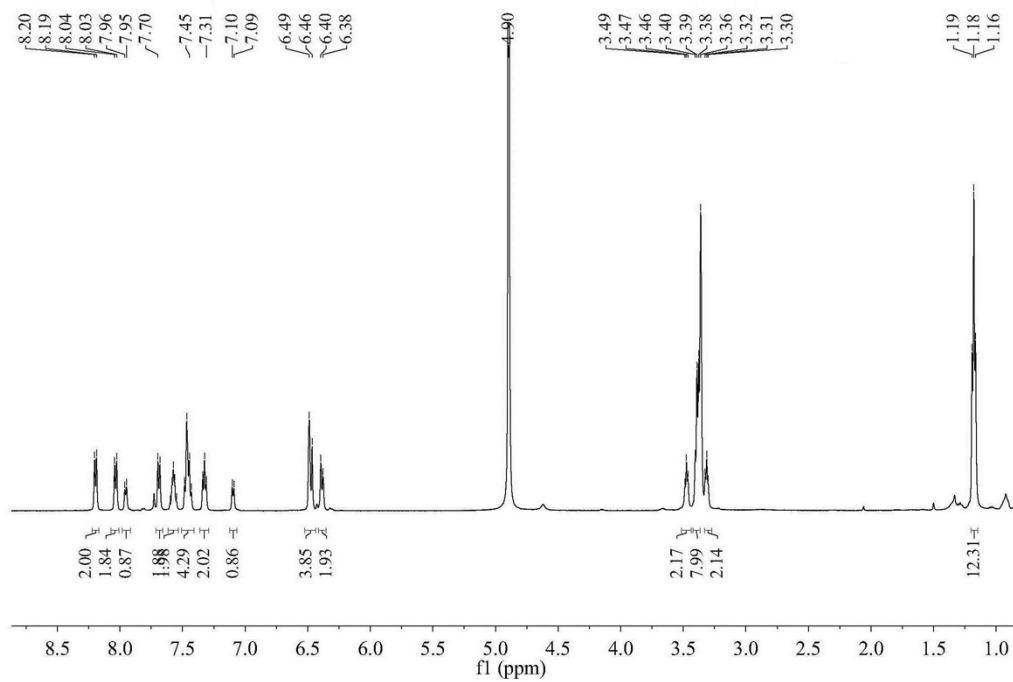


Fig. S5 ^1H NMR spectrum of probe (L) in CD_3OD (500 MHz, CD_3OD)

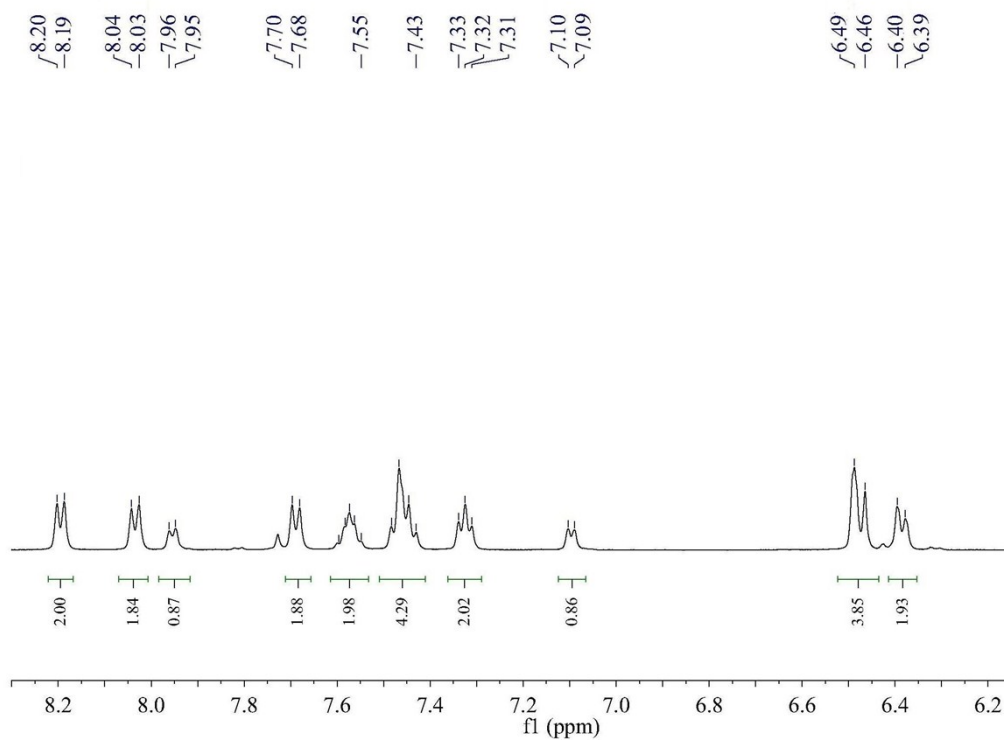


Fig. S6 ^1H NMR spectrum of probe (L) in CD_3OD (500 MHz, CD_3OD)

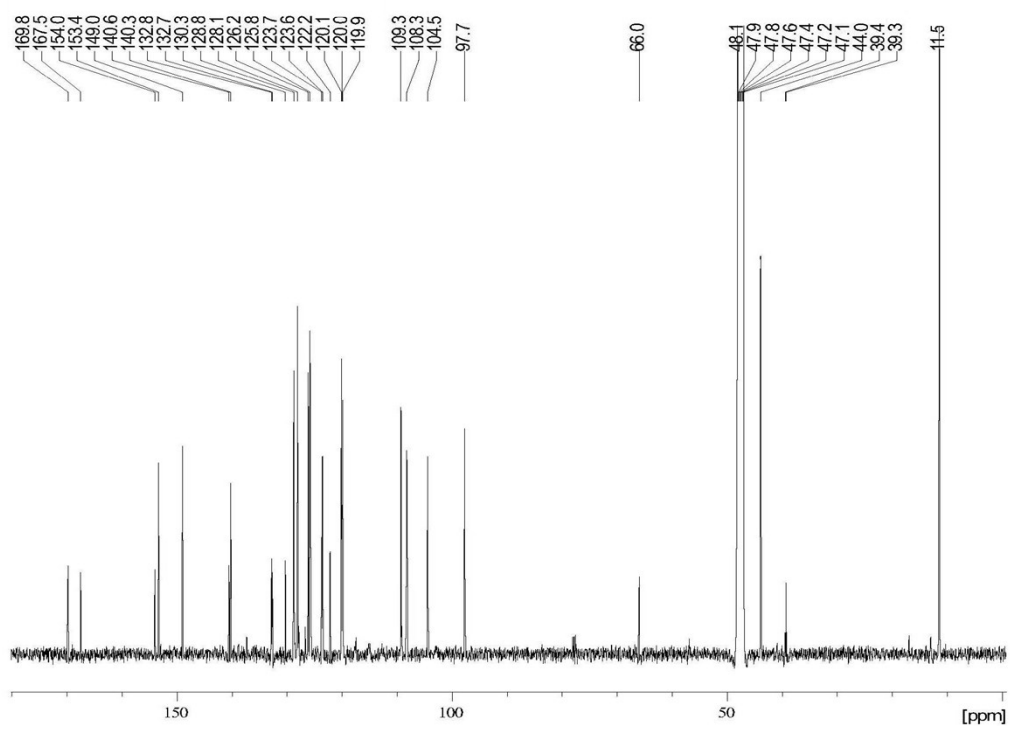


Fig. S7 ^{13}C NMR spectrum of probe (L) in CD_3OD (125 MHz, CD_3OD)

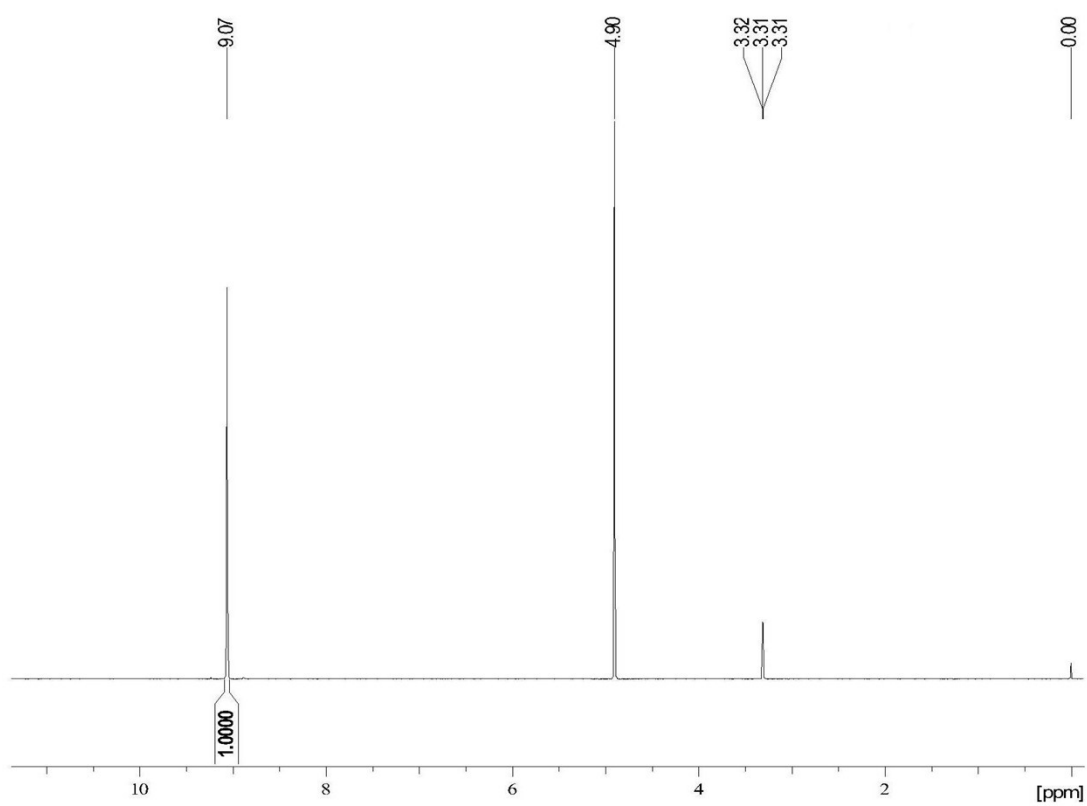


Fig. S8 ^1H NMR spectrum of PA in CD_3OD (500 MHz, CD_3OD)

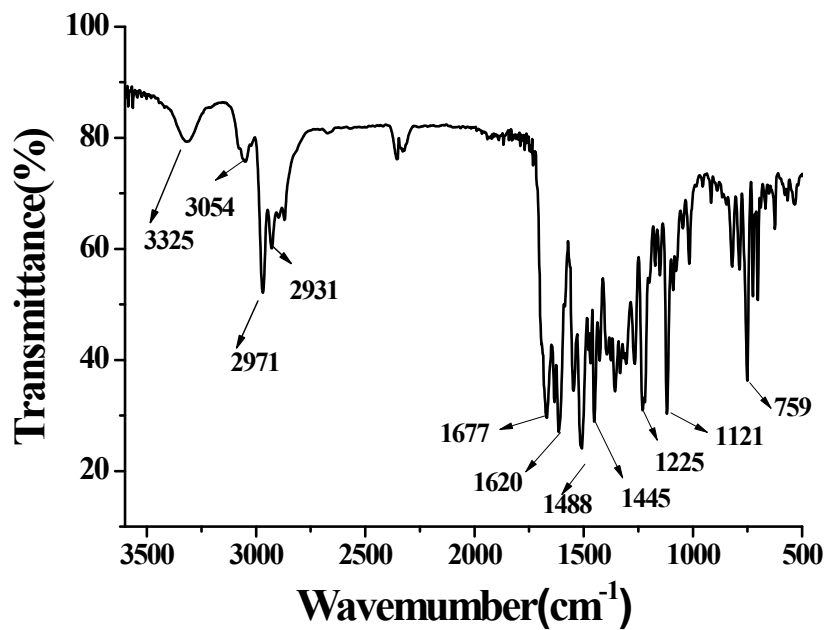


Fig. S9 FT-IR spectrum of probe (L)

II- Response time study of fluorescence and colorimetric titrations.

Probe solution (20 μM) was treated with PA (200 μM) and the spectra were recorded every 5 minutes.

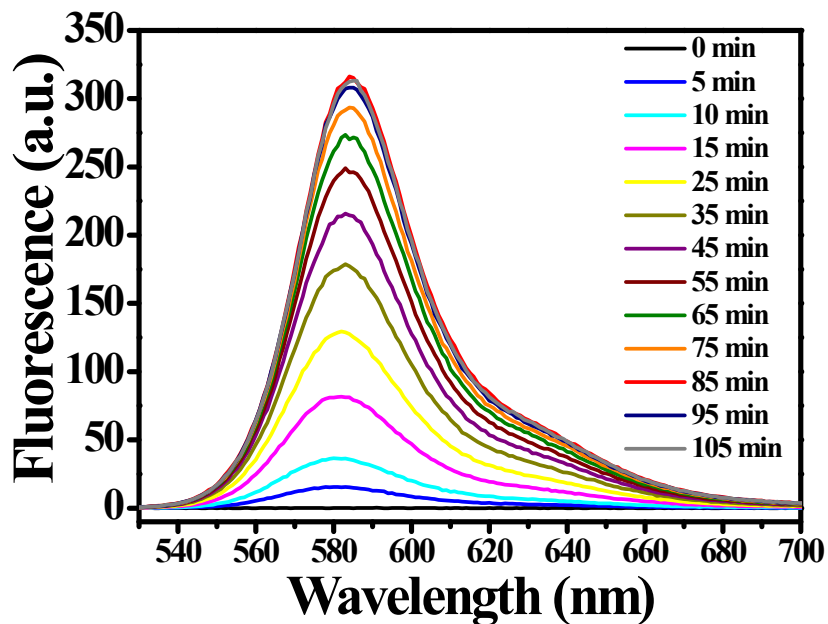


Fig. S10 Fluorescence spectra of probe (20 μM) with PA (200 μM) recorded every 5 min after mixed.

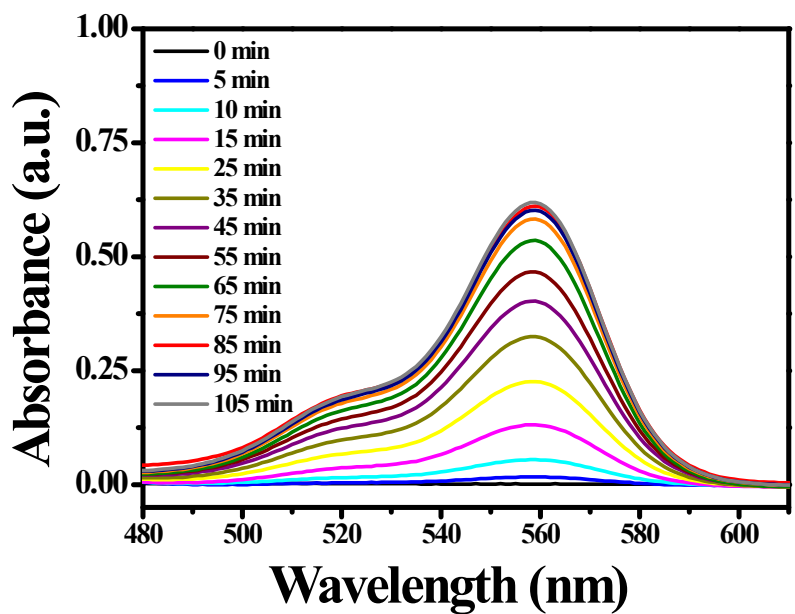


Fig. S11 Absorbance spectra of probe (20 μM) with PA (200 μM) recorded every 5 min after mixed.

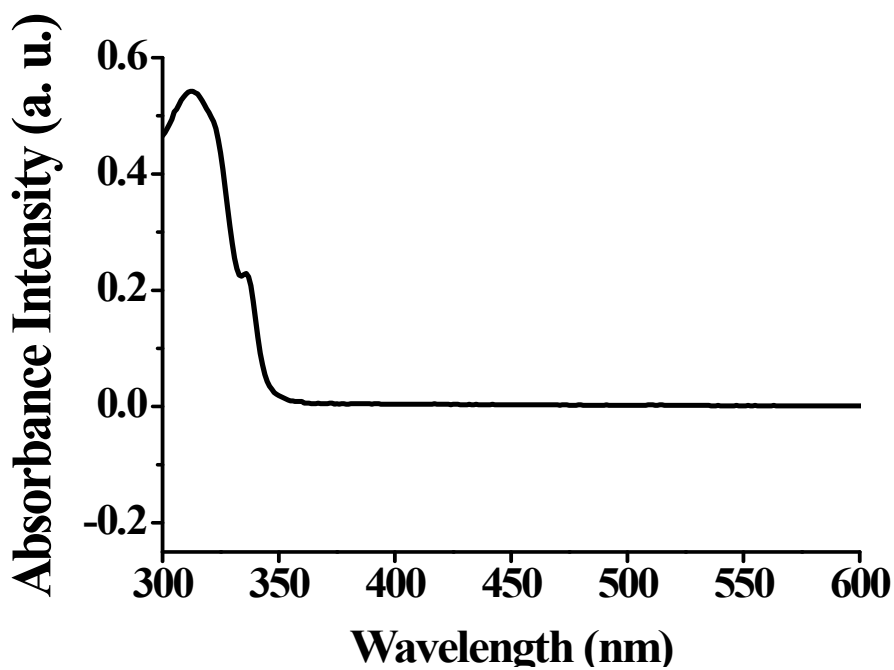


Fig. S12 Absorbance spectra of probe (L, 20 μM).

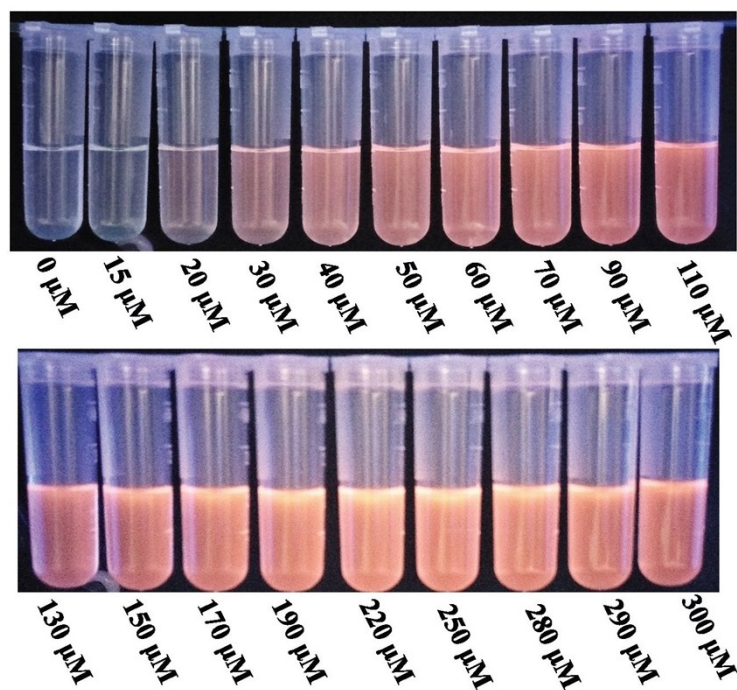


Fig. S13 Photograph of probe solutions (L) in the presence of different concentration of PA under 365 nm UV light.

III— FRET effect study

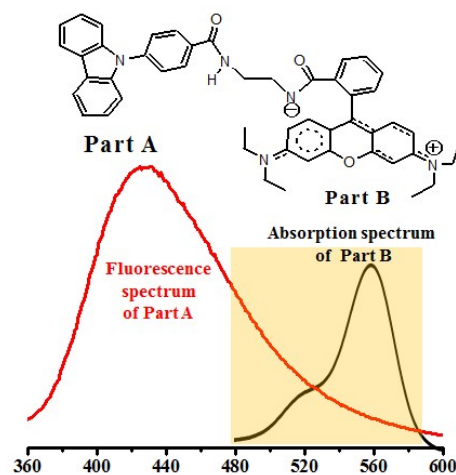


Fig. S14 The fluorescence spectrum of Part A (the energy donor unit) and the absorption spectrum of Part B (the acceptor unit) in probe L

Fluorescence titration of N-(rhodamine-B)lactam-ethylenediamine (L_1) and PA

In order to reveal the intramolecular FRET effect between 4-(9*H*-carbazol-9-yl)benzamide (Donor) and the spiro-lactam ring-opening product rhodamine fluorophore (Acceptor), fluorescence titration control experiments were performed using N-(rhodamine-B)lactam-ethylenediamine (L_1) and PA in ethanol. The fluorescence spectra of L_1 solution (20 μ M) were recorded at 85 min after treated with

different concentration of PA (40 μM , 60 μM and 100 μM). As depicted in **Fig. 13S**, the relative fluorescence intensity of L_1 solution (20 μM) with 40 μM PA is at about 5, which reveals a low efficiency using the 314 nm excitation wavelength. However, the relative fluorescence intensity of probe (L , 20 μM) solution in ethanol with 40 μM PA is at about 250 ($\lambda_{\text{ex}} = 314 \text{ nm}$, $\lambda_{\text{em}} = 586 \text{ nm}$; slits 2.5 nm/5.0 nm). These results reveal that the 4-(9*H*-carbazol-9-yl)benzamide subunit might serve as a donor for the excitation of delocalized xanthane fluorophore in the spiro-lactam ring opening products **Fig. S14**

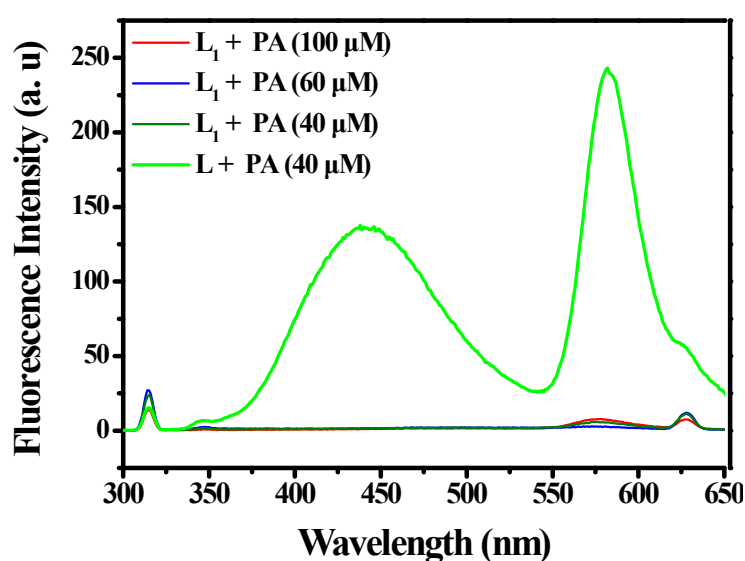


Fig. S15 Fluorescence titration of N-(rhodamine-B)lactam-ethylenediamine (L_1) and PA ($\lambda_{\text{ex}} = 314 \text{ nm}$, $\lambda_{\text{em}} = 586 \text{ nm}$; slits 2.5 nm/5.0 nm).

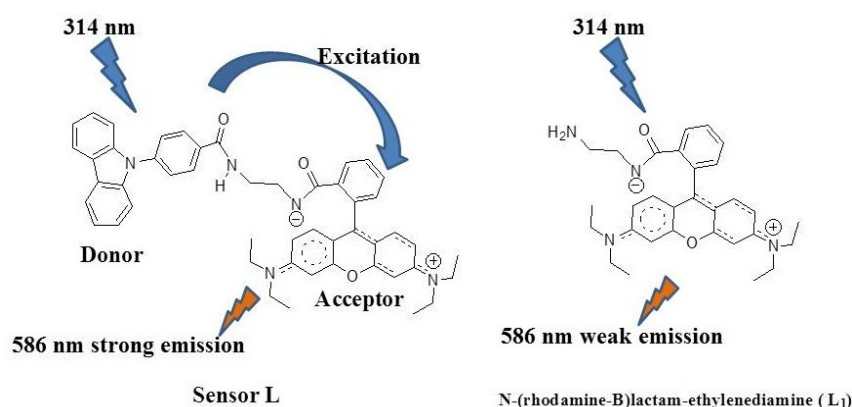


Fig. S16 Intramolecular FRET effect of the probe L .

As shown in Fig S17, the luminescent lifetime of the Part A (energy donor unit) reduced from 5.31 ns to 3.52 ns in the presence of part B (the energy acceptor unit,

which was formed after the addition of PA). The decrease of fluorescence lifetime for energy donor unit (Part A) revealing that a FRET process occurred between part A (energy donor unit) and part B (energy acceptor unit) in the probe molecular.

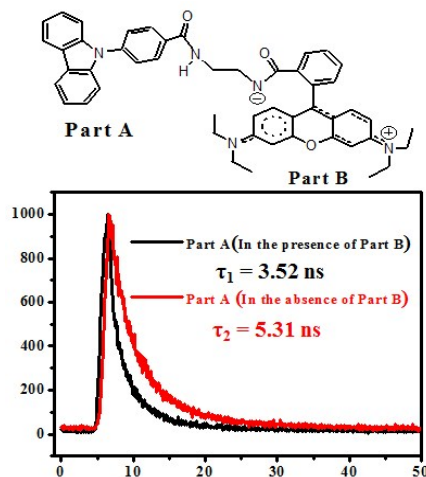


Fig. S17 The fluorescence decays assay of part A in the presence and absence of part B

IV- Benesi-Hildebrand equation used for the binding mode investigation

According to the Job's plot of L + PA complex in ethanol (Fig. 8a), a 1:1 stoichiometry for PA and probe (L) was observed. With the initial experiment results in hand, the Benesi-Hildebrand polts was performed according to the colorimetric titration experiments. The following Benesi-Hildebrand equation^[2-3] was used.

$$\frac{1}{(A-A_0)} = \frac{1}{K_a(A_{max}-A_0)(C_{PA})} + \frac{1}{(A_{max}-A_0)}$$
 . In this equation, A_0 is the initial absorbance intensity of probe solution (centered at 560 nm) in the absence of PA and A is the absorbance intensity of probe solution with different concentration of PA. A_{max} is the absorbance intensity of probe solution with excess PA, C_{PA} represents the concentration of PA. K_a is the association constant between probe and PA.

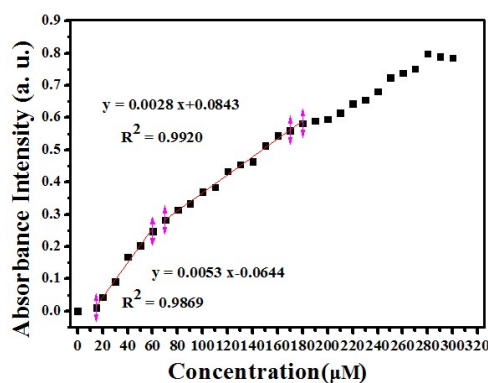


Fig. S18 Absorbance of probe solution with different concentration of PA

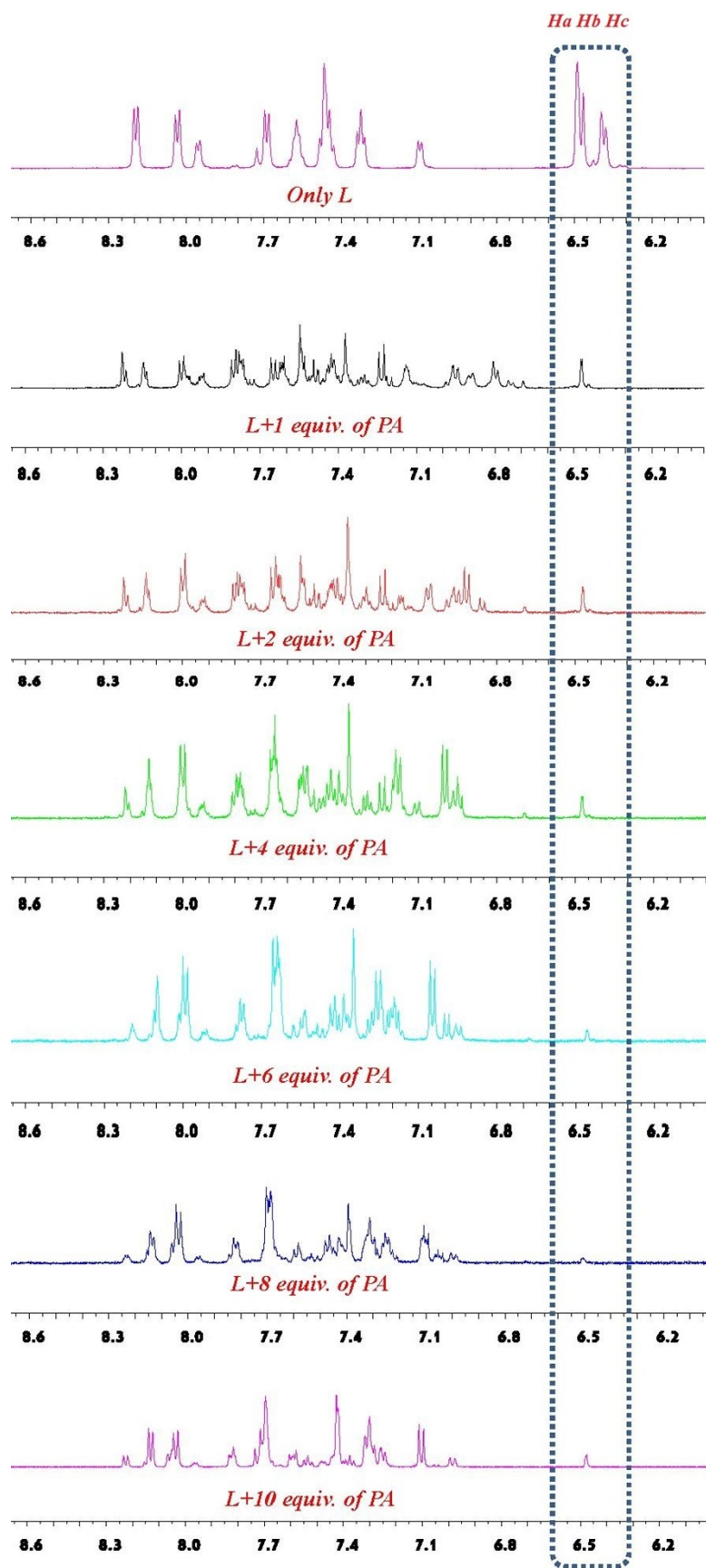


Fig. S19 ^1H NMR titrations of probe with different equiv. of PA

V-References

- [1] Li, Z.; Xue, Z.; Wu, Z.; Han, J.; Han, S. Chromo-fluorogenic detection of aldehydes with a rhodamine based sensor featuring an intramolecular deoxylactam. *Org. Biomol. Chem.*, **2011**, *9*(22), 7652-7654 (and references there).
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- [3] Chowdhury, B.; Khata, S.; Dutta, R.; Chakraborty, S.; Ghosh, P. Bis-heteroleptic ruthenium (II) complex of a triazole ligand as a selective probe for phosphates. *Inorg. Chem.*, **2014**, *53*(15), 8061-8070 (and references there).