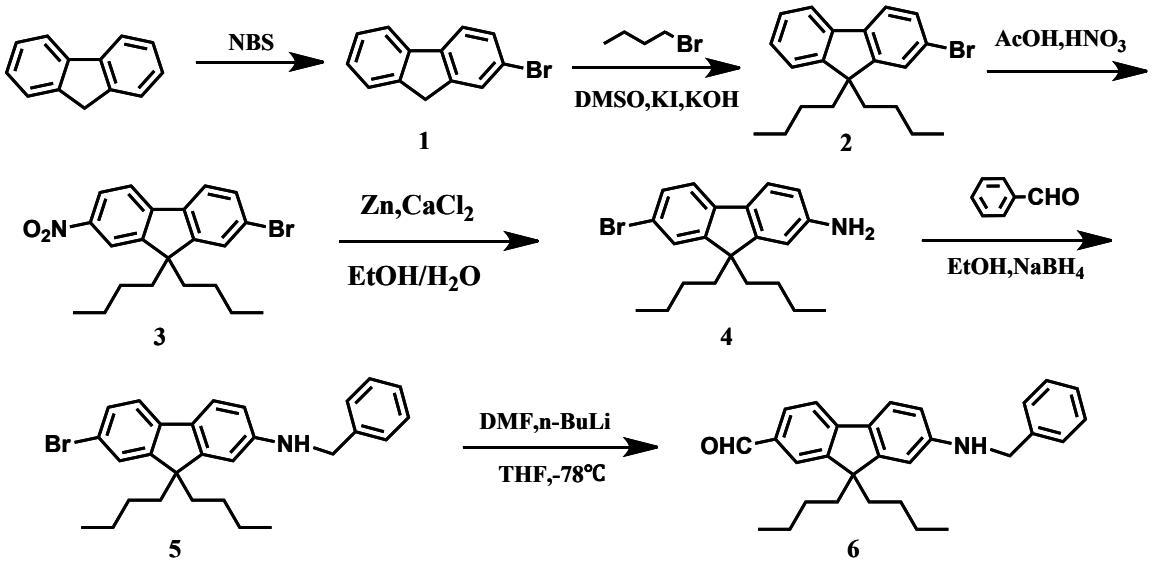
# Supplemental Material

## "Turn-on" fluorescent sensor for ultrasensitive detection of melamine based on a new fluorescence probe and AuNPs AuNPs

Qiujun Lu, Jiangna Zhao+, Shanyan Xue, Peng Yin, Youyu Zhang*, Shouzhuo Yao

*Key Laboratory of Chemical Biology and Traditional Chinese Medicine Research (Ministry of Education),College of Chemistry and Chemical Engineering, Hunan Normal University, Changsha 410081, PR China*

*Corresponding author. Tel: +86-731-8865515; fax: +86-731-8865515;
E-mail address: zhangyy@hunnu.edu.cn




Scheme S1 The synthetic route of BDFC.

*Synthesis of BDFC*

The synthetic steps of BFDC were shown in Scheme S1.

Synthesis of compound **1**.

N-bromosuccinimide (NBS, 5.34 g, 30 mmol) and fluorine (4.98 g, 30 mmol) was dissolved in propylene carbonate (40 mL), and then transferred to a water bath at 60 $^{o}$C for 2 min. The solid was dissolved and an orange solution was formed. The mixture was brought to room temperature and stirred for 30 min. The color of the solution turned to yellow, and a negative starch-iodide test was obtained. Then the mixture was poured into $H_2O$ (500 mL), the precipitate was filtered, and crystallization from ethanol. The compound **1** was obtained (6.7 g, yield: 91%). $^{1}$H NMR (500 MHz, $CDCl_3$) $\delta$(ppm): 7.76 (1H, d, $J$ = 7.5 Hz), 7.67 (1H, s), 7.64(1H, d, $J$ = 8.1 Hz), 7.54 (1H, d, $J$ = 7.3 Hz), 7.50 (1H, dd, $J$ = 1.9, 1.8 Hz), 7.38 (1H, t, $J$ = 7.0 Hz), 7.34 (1H, m), 3.88 (2H, s).

Synthesis of compound **2**.

A mixture of compound **1** (686 mg, 2.8 mmol), 1-bromobutane (805 mg, 5.8 mmol), KI (50 mg, 0.3 mmol), and DMSO (6.5 mL) was stirred at room temperature, then the powdered KOH (665 mg, 12 mmol) was slowly added under $N_2$. The color of the mixture changed from bright yellow to dark green immediately after KOH

addition. The mixture was stirred at room temperature for 1 h. The mixture was poured into water and extracted with hexanes. The organic extract was washed with water, dried with $NaSO_4$. Purification was accomplished by column chromatography using silica gel (hexanes), providing 700 mg of yellow solid (yield: 70%). $^1$H NMR (500 MHz, $CDCl_3$) $\delta$(ppm): 7.73 (1H, d, $J$ = 1.1 Hz), 7.48 (1H, m), 7.37(5H, m), 2.00 (4H, m), 1.13 (4H, m), 0.71 (6H, t, $J$ = 7.3 Hz), 0.61 (4H, m).

Synthesis of compound **3**.

Compound **2**（357 mg，1 mmol）was dissolved in 8 mL AcOH and heated to 60 $^o$C. And then 1.2 mL of the concentrated $HNO_3/H_2SO_4$ mixed acid (v/v = 5:1) was slowly added into the mixture. After being stirred at 80 $^o$C for 2 h, the reaction was stopped. The solution was extracted with $CH_2Cl_2$. The organic layer was dried. Using Hex and EA as the eluent and silica gel as the adsorbent, the residue was purified by column chromatography (Hex/EA=25:1). The Compound **3** was obtained (280 mg, yield: 70%). $^1$H NMR (500 MHz, $CDCl_3$) $\delta$(ppm): 8.27 (1H, dd, $J$ = 2.2, 2.0 Hz), 8.19 (1H, s), 7.64 (1H, d, $J$ = 2.1 Hz), 7.78 (1H, d, $J$ =8.4 Hz), 7.74 (2H, m), 7.53 (1H,d, $J$ = 8.4 Hz), 2.01 (4H, m), 1.11 (4H, m), 0.68 (6H, t, $J$ = 7.3 Hz), 0.58 (4H, m).

Synthesis of compound **4**.

Compound **3** was dissolved in 78% AcOH. After being stirred into dilute paste, then 0.1 g of calcium chloride powder (dissolved in 1.5 mL of water) and 3g of Zinc powder were added in the dilute paste. After being stirred for 3 h under backflow condition, the reaction was stopped. The reaction mixture was extracted with $CH_2Cl_2$. The organic extract was washed with water, and dried with anhydrous $NaSO_4$. Purification was accomplished by column chromatography using silica gel (Hex:EA=20:1), providing 330 mg of compound **4** (yield: 65%). $^1$H NMR (500 MHz, $CDCl_3$) $\delta$(ppm): 7.59 (2H, m), 7.45 (1H, d, $J$ = 7.9 Hz), 7.37 (1H, s), 7.30 (1H, d, $J$ = 6.3 Hz), 6.66 (2H, m), 3.78 (2H, s), 1.89 (4H, m), 1.11 (4H, m), 0.71 (6H, t, $J$ = 7.3 Hz), 0.64 (4H, m).

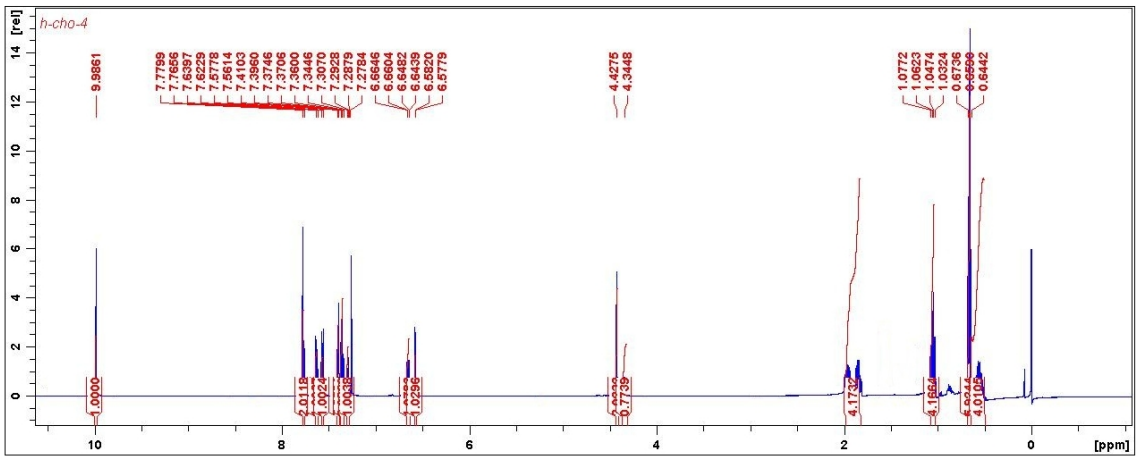
Synthesis of compound **5**.

Compound **4** (200 mg，0.5 mmol) was dissolved in 10 mL of AcOH. Benzaldehyde (0.1ml, 1mmol) was added into this solution. After being backflow at

75 °C for 5 h, $NaBH_4$ (74mg, 2 mmol) was added into this mixture. After being backflow at room temperature for 5 h, the reaction was stopped. The end product was dried by rotary evaporation instrument. Purification was accomplished by column chromatography using silica gel (hexanes), providing 120 mg of compound **5** (yield: 50%). $^1$H NMR (500 MHz, $CDCl_3$) $\delta$(ppm): 7.44 (1H, d, $J$ = 8.1 Hz), 7.39 (1H, d, $J$ = 1.4 Hz), 7.37 (1H, s), 7.35 (5H, m), 7.27 (1H, m), 4.37 (2H, s), 4.15(1H, s), 1.84 (4H, m), 1.09 (4H, m), 0.67 (6H, t, $J$ = 7.3 Hz), 0.61 (4H, m).

Synthesis of compound **6**.

Compound **5** was dissolved in THF (10 mL) under a $N_2$ atmosphere. 2 mL of n-BuLi (2.4 M in hex) was slowly added into this solution at -78 °C. After being reacted for 10 min at -78 °C, excess of DMF was slowly added into the mixture at -78 °C. After being reacted for 2 h at -78 °C, the mixture was brought to room temperature and stirred overnight, then adding water to quench the reaction. Purification was accomplished by column chromatography using silica gel (hex：EA=100:1), providing compound 6 (18%). $^1$H NMR (500 MHz, $CDCl_3$) $\delta$(ppm): 9.98 (1H, s), 7.78 (1H, s), 7.76 (1H, s), 7.63 (1H, d, J = 8.4 Hz), 7.57 (2H, d, J = 8.2 Hz), 7.41 (2H, d, J = 7.1 Hz), 7.36 (2H, t, J = 7.3 Hz). 7.29 (1H, t, J = 8.5 Hz), 6.66 (2H, dd, J = 2.1, 2.2 Hz), 6.58 (1H, d, J =2.1 Hz), 4.42 (2H, s), 4.34 (1H, s), 1.99 (4H, m), 1.07 (4H, m), 0.66 (6H, t, J = 7.3 Hz), 0.59 (4H, m). $^{13}$C NMR (126 MHz, $CDCl_3$). $\delta$(ppm): 192.16, 154.14, 150.44, 149.71, 148.69, 138.59, 133.37, 131.09, 128.53, 128.01, 126.86, 126.50, 122.44, 121.89, 117.88, 111.39, 106.58, 54.84, 54.70, 51.62, 40.14, 29.53, 25.82, 22.92, 20.34, 13.94, 13.73.




$^{1}$H NMR (500 MHz, $CDCl_3$) $\delta$(ppm): 9.98 (1H, s), 7.78 (1H, s), 7.76 (1H, s), 7.63 (1H, d, J = 8.4 Hz), 7.57 (2H, d, J = 8.2 Hz), 7.41 (2H, d, J = 7.1 Hz), 7.36 (2H, t, J = 7.3 Hz). 7.29 (1H, t, J = 8.5 Hz), 6.66 (2H, dd, J = 2.1, 2.2 Hz), 6.58 (1H, d, J =2.1 Hz), 4.42 (2H, s), 4.34 (1H, s), 1.99 (4H, m), 1.07 (4H, m), 0.66 (6H, t, J = 7.3 Hz), 0.59 (4H, m).

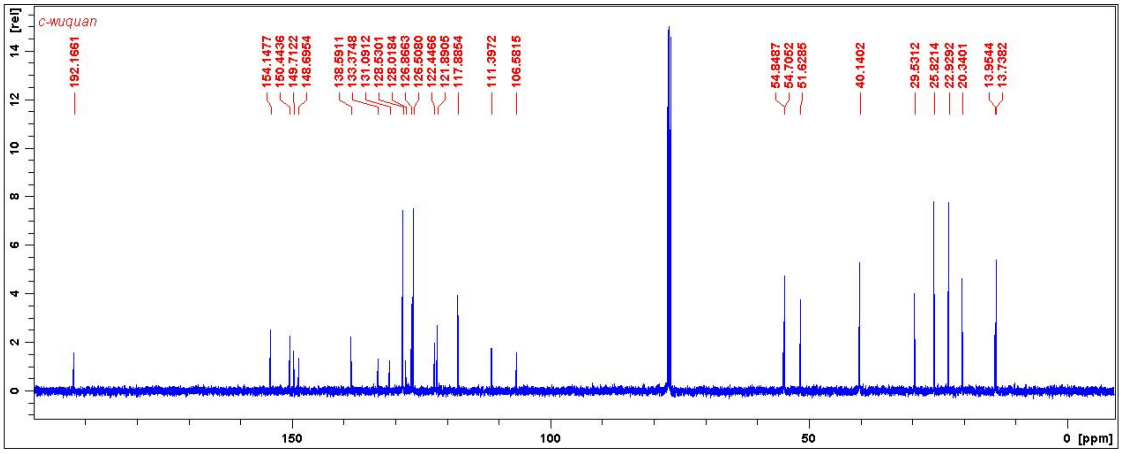



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**Fig.S1:** 1HNMR spectra and $^{13}$C NMR spectra of probe.

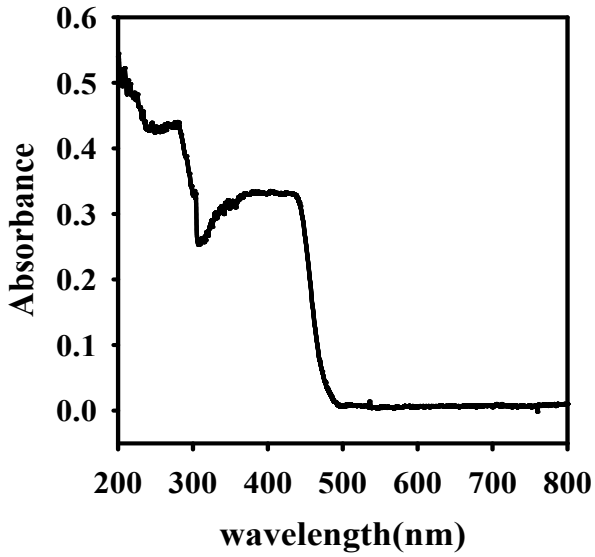


**Fig. S2:** UV-vis spectra of BDFC

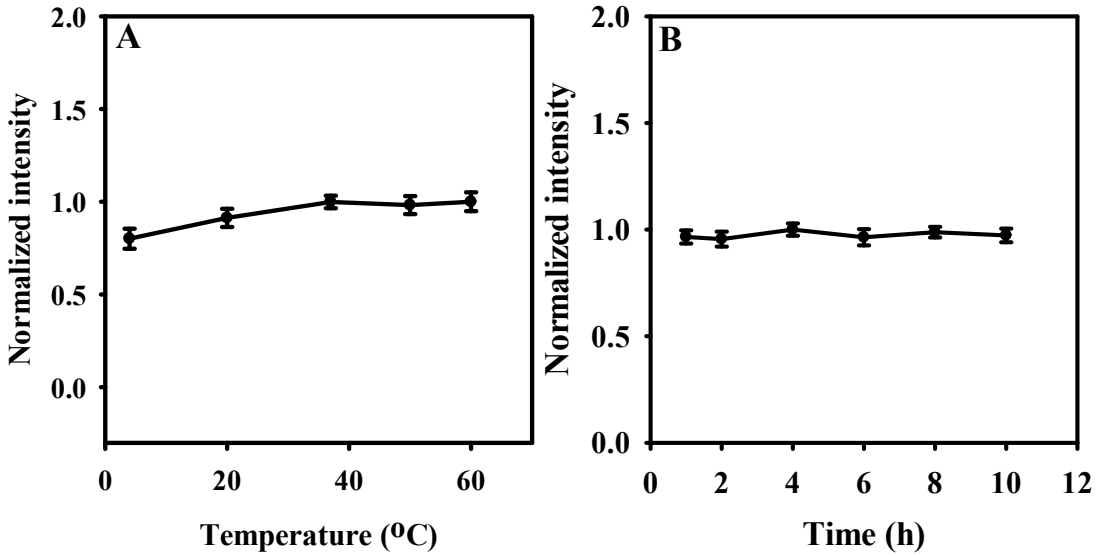


**Fig. S3:** Effects of temperature (A) and time (B) on the normalized FL intensity of the BDFC solution.

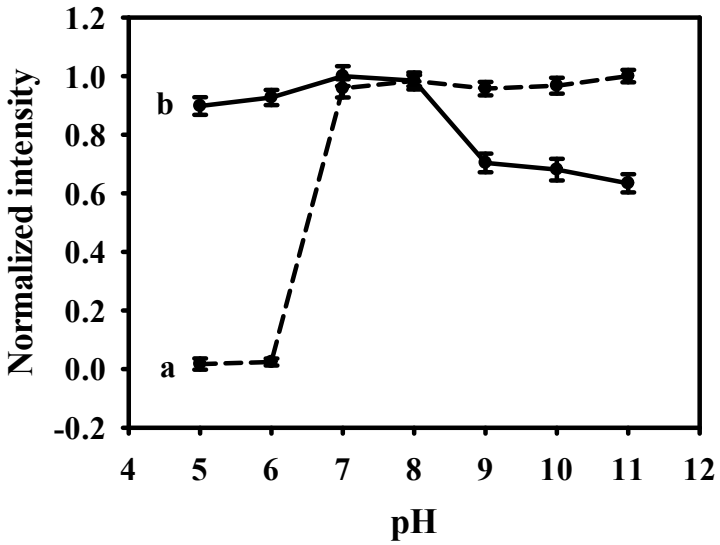


**Fig. S4** Effects of pH on the normalized FL intensity of the fluorescein (a) and BDFC (b).

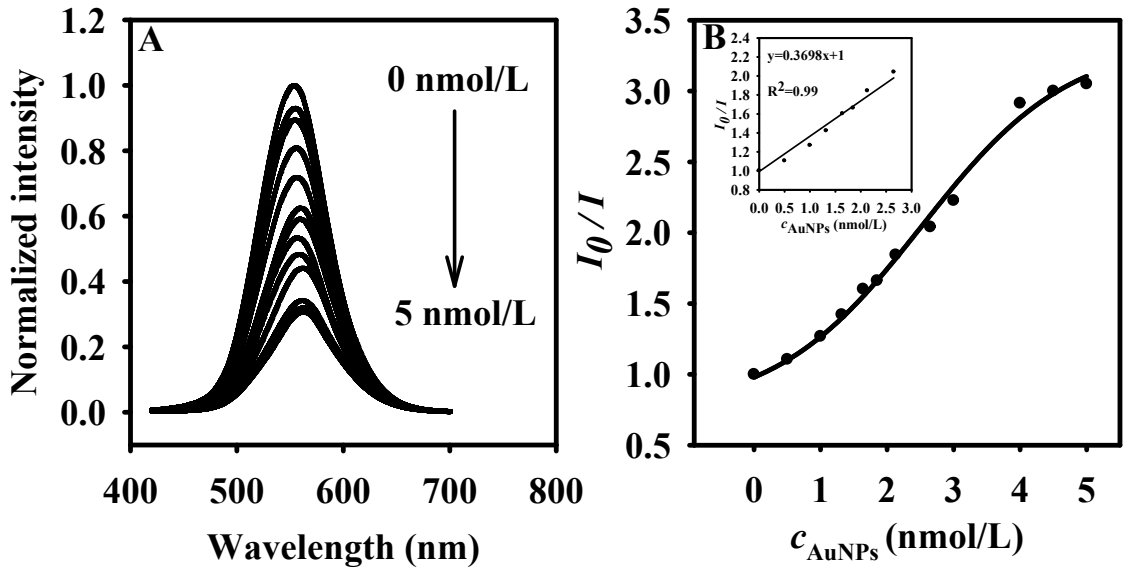


**Fig. S5 :**(A)Fluorescence emission spectra of BFDC as a function of AuNPs concentration. The concentration of probe is $7\times10^{-7}$ mol/L. (B) Plot of the normalized value of fluorescence at 550 nm versus the concentration of Au NPs. The concentration of probe is $7\times10^{-7}$ mol/L. Where $I_0$ and I are the fluorescent intensity of BDFC in the absence and presence of AuNPs, respectively.

TableS1.Comparisons of analytical performances of various typical techniques for melamine analysis.

| Method | Linear range (μM) | Detection limit (μM) | Reference |
|---|---|---|---|
| reversed phase high-performance liquid chromatography | 7.930−634.000 | 0.793 | [1] |
| mass spectrometry | | 1.35 | [2] |
| ELISA | 0.060−5.800 | 0.021 | [3] |
| gas chromatography−mass spectrometric (GC-MS) method | 0.396−7.936 | 0.040 | [4] |
| Electrochemical | 5.000−200.000 | 0.800 | [5] |
| Colorimetric assay | 79.365-634.920 | 3.175 | [6] |
| Fluorescence assay | 0.0075−0.3500 | 0.00089 | [7] |
| Fluorescence assay | 1.984-60.079 | 1.587 | [8] |
| Fluorescence assay | 0.100-4.000 | 0.001 | [9] |
| **Fluorescence assay** | **0.01-4** | **0.003** | **This work** |

## References


1. G. Venkatasami and J. R. Sowa, *Anal. Chim. Acta*, 2010, **665**, 227.
2. L. Vaclavik, J. Rosmus, B. Popping and J. Hajslova, *Journal of Chromatography A*, 2010, **1217**, 4204.
3. H. Lei, Y. Shen, L. Song, J. Yang, O. P. Chevallier, S. A. Haughey, H. Wang, Y. Sun and C. T. Elliott, *Anal. Chim. Acta*, 2010, **665**, 84.
4. X. Zhu, S. Wang, Q. Liu, Q. Xu, S. Xu and H. Chen, *J. Agric. Food Chem.*, 2009, **57**, 11075.
5. C.-W. Liao, Y.-R. Chen, J.-L. Chang and J.-M. Zen, *J. Agric. Food Chem.*, 2011, **59**, 9782.
6. L. Li, B. Li, D. Cheng and L. Mao, *Food Chem.*, 2010, **122**, 895.
7. F. Gao, Q. Ye, P. Cui and L. Zhang, *J. Agric. Food Chem.*, 2012, **60**, 4550.
8. Y.-Y. Zhou, J. Yang, M. Liu, S.-F. Wang and Q. Lu, *Journal of Luminescence*, 2010, **130**, 817.
9. L. Guo, J. Zhong, J. Wu, F. Fu, G. Chen, Y. Chen, X. Zheng and S. Lin, *Analyst*, 2011, **136**, 1659.