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A New Fluorescent Probe of Hydrazine: 2-(4-(Imidazo[1,2-a]pyridin-3-ylethynyl)benzylidene)malononitrile

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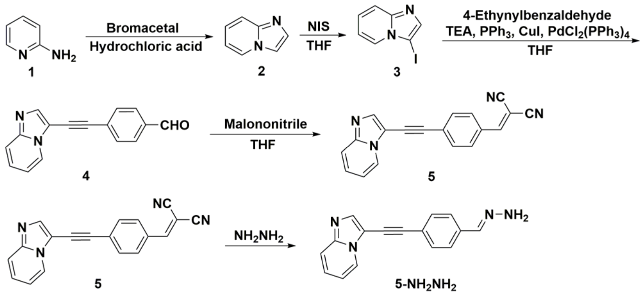
Hydrazine is widely used in plently of fields as a raw material and closely related to our healthy. A new fluorescent probe of hydrazine was designed and synthesized, choosing imidazo[1,2-a]pyridine as the electron acceptor, based on the intrarmolecular charge transfer (ICT) effect. The probe, 2-(4-(imidazo[1,2-a]pyridin-3-ylethynyl)benzylidene)malononitrile (**5**), responds rapidly toward hydrazine and exhibits obvious color changes from yellow to colorless, indicating its use as a color indicator for hydrazine. Sensing mechanism of probe **5** toward hydrazine was deduced through HOMO-LUMO energy levels and the interfacial plots of the molecular orbitals. And its physical and chemical properties were demonstrated by UV, fluorescence and single-crystal X-ray diffraction via computation and experiment. The calculated data are consistent with experimental results.

Hydrazine, a colorless and transparent oily liquid with strong alkaline and hygroscopicity, can corrode glass, rubber, leather, cork and so on. It is widely used in the industry of aerospace, agriculture, pharmaceutical, chemical, photography as missile systems, weapons for mass destruction, pesticides, plant growth regulators, antimicrobial drugs, fuel cells, catalysts, metal corrosion inhibitor, emulsifiers, dyes, photography chemicals and so on.[1-5] Nevertheless, since it's toxic effects, hydrazine will potentially leads to serious environmental pollution and health hazards during its manufacture, use, transport and disposal procedures.[6] The research indicates that long-term exposure to the atmosphere of hydrazine can induce steatosis,[1] damage of the liver and central nervous system,[7] mutation and cancer. Thus, the concentration of hydrazine in the environment needs to be strictly monitored and sophisticated methods are always required.[8] The generally described efforts devoted to determine hydrazine include chemiluminescence, electrochemistry, flow detectionand spectrofluorimetry technique. [9-14] In recent years, the fluorescent has been widely used as a simple but reliable detection method for the rapid and sensitive detection of hydrazine both qualitatively and quantitatively.[15] So we tried to design a new fluorescent probe for detecting trace amounts of hydrazine.

In our work, the photophysical properties, synthesis, sensitivity, crystal structure and calculation of **5** are reported (Scheme 1). Compound **5** responded rapidly toward hydrazine and exhibited distinct color changes from bright yellow to colorless, indicating a good sensitivity as a color indicator for hydrazine. Its molecular structure was determined by ^{1}H NMR, ^{13}C NMR, MS and IR. We also determined its crystal structure by single-crystal X-ray diffraction analysis and calculated its geometric data using Gaussian 09 program package[16] based on density functional theory (DFT) calculations. The crystal structure was extremely similar to calculated geometries.



Scheme 1. Synthesis and proposed sensing mechanism of probe **5**.

The synthesis of imidazo[1,2-a]pyridine (**2**): The solution of bromacetal (50 g, 0.254 mol) in 1 N hydrochloric acid (200 mL) was refluxed for 30 minutes at 80 ^{o}C. The pH of the solution was adjusted to 7 by sodium bicarbonate and to the mixture was added 2-aminopyridine(15.90 g, 0.169 mol) followed by stirring for 2 h at room temperature. Adjust the pH to 10 with sodium hydroxide and the product was extracted by ethyl acetate. The organic phase was collected and the solvent was evaporated to get the brown oil (12.5 g, 55.9%).

The synthesis of 3-iodoimidazo[1,2-a]pyridine (**3**): To the solution of imidazo[1,2-a] pyridine (6.50 g, 0.055 mol) in 80 mL THF was added 1-iodopyrrolidine-2,5-dione (14.85 g, 0.066 mol) slowly. The reaction mixture was stirred for 1 h at room temperature and extracted by ethyl acetate. The organic phase was collected and the solvent was evaporated to afford the yellow solid (6.10 g, 45.5%).

The synthesis of 4-(imidazo[1,2-a]pyridin-3-ylethynyl) benzaldehyde (**4**): To a 100 mL round bottom flask, 3-iodoimidazo[1,2-a]pyridine (6.10 g, 24.4 mmol) was dissolved in 100 mL THF followed by adding 4-ethynylbenzaldehyde (3.28 g, 0.244 mmol), triethylamine (3.81 g, 36.6 mmol), triphenylphosphane (0.066 g, 0.244 mmol), copper iodide (0.047 g, 0.244 mmol) and bis(triphenylphosphine) palladium(II) chloride (0.175 g, 0.244 mmol) in order under nitrogen atmosphere. The reaction was heated to 65 °C to stir for 1 h and the mixture was poured into water after the completion of the reaction that no starting material could be detected via TLC. THF was removed by a rotary evaporator and solid precipitation was filtered, washed with dichloromethane. The aqueous phase was extracted with dichloromethane and the organic phase was collected and evaporated to afford the yellow solid (3.05 g, 49.6%). ^{1}H NMR (400 MHz, $CDCl_3$) δ 10.04 (s, 1H), 8.39 (d, J = 6.8 Hz, 1H), 7.99 (s, 1H), 7.91 (d, J = 8.4 Hz, 2H), 7.77 – 7.68 (m, 3H), 7.39 – 7.31 (m, 1H), 7.03 (t, J = 6.4 Hz, 1H).

The synthesis of 2-(4-(imidazo[1,2-a]pyridin-3-ylethynyl)benzylidene)malononitrile (**5**): To the solution of compound **4** (3.00 g, 11.44 mmol) in 50 mL THF was added malononitrile (0.79 g, 17.16 mmol) and five drops of tetrahydropyrrole. The mixture was heated to 65 °C and stirred for 1 h. The yellow solid (2.1 g, 62.4%) was got by filtration after the reaction was cooled to room temperature. m.p.: 204.6 - 205.8 °C. IR (KBr pellets): ν 3452.3, 3037.1, 2226.7, 2196.4, 1582.8, 1256.8, 1154.2, 835.5, 756.1 cm^{-1}. ^{1}H NMR (400 MHz, DMSO-d_6) δ 8.74 (s, 1H), 8.54 (s, 1H), 8.33 – 7.69 (m, 6H), 7.49 (s, 1H), 7.19 (s, 1H). ^{13}C NMR (100 MHz, DMSO-d_6) δ 160.04, 145.73, 139.45, 139.16, 131.36, 130.87, 130.75, 129.67, 127.62, 127.16, 126.14, 117.52, 114.24, 114.12, 113.25, 107.14, 99.11, 81.80, 81.62. MS (ESI+): m/z = 295.0 [M + H]$^+$.

The crystal of probe **5** was grown by slow evaporation of chloroform under ambient conditions, and suitable crystal was obtained for crystallographic analysis. The measured values reveals that **5** possesses triclinic crystal system having P-1 space group [unit cell: a = 4.9408(16) Å, b = 12.394(4) Å, c = 15.052(5) Å]. ORTEP diagrams of the molecular structure and the atomic numbering schemes of **5** is shown in Figure 1. The hydrogen atoms were omitted for clarity. The crystallographic and refinement data are shown in Supplementary Material Table S1. The crystal structure of **5** was compared with the DFT-optimized structure. Some selected experimental and calculated geometry parameters for **5** are listed in Table S2. As expected, most of the calculated geometry parameters for the probe are close to the X-ray data.

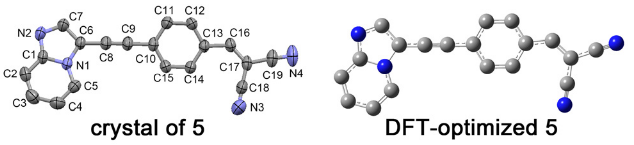


Figure 1. DFT-optimized and crystal structure of probe **5**.

We have investigated the changes of fluorescence spectra and the UV-vis spectra for the probe **5** in the absence and presence of hydrazine. The color change following by the addition of hydrazine are detectable easily by the naked eye of sensor. As shown in Figure 2(**a**), the green line showed a weak absorption at 419 nm, which was attributed to the intramolecular charge transfer (ICT) transition in the molecule. Upon the addition of hydrazine, the absorption intensity at 419 nm moved left immediately and became stronger. And as we can see, Figure 2(**b**) indicated that the absorption peak moved to 416 nm from 596 nm when adding hydrazine in the fluorescence spectra. The calculated UV absorption spectra and fluorescence titration spectra of probe **5** and its reacted form with hydrazine **5**-N_2H_4 are consistent with experimental value (Figure 3).

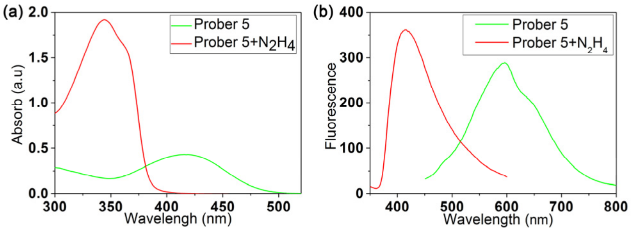


Figure 2. Experimental UV absorption spectra (**a**) and fluorescence titration spectra (**b**) of probe **5** and its reacted form with hydrazine **5**-N_2H_4.

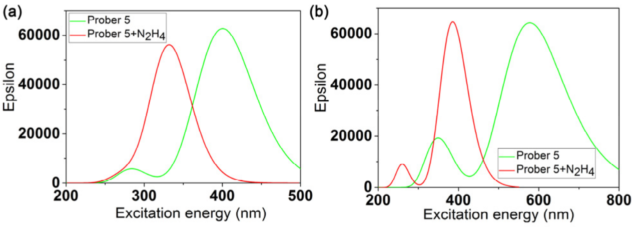


Figure 3. Calculated UV absorption spectra (**a**) and fluorescence titration spectra (**b**) of probe **5** and its reacted form with hydrazine **5**-N_2H_4.

We have performed the computations on the probe with and without hydrazine based on density functional theory (DFT) and had gotten insight into the sensing mechanism of compound **5** toward hydrazine. As shown in Figure 4, comparing the level changes of the HOMO and LUMO in compound **5** and compound **5**-N_2H_4 , the ICT effect in **5**-N_2H_4 is stronger than in compound **5**. Thus we can see an obvious blue-shift in the absorption spectra and an intense fluorescence enhancement with the adding of N_2H_4. In these process, the imidazo[1,2-a]pyridine group plays a role of donor and its electron transfers to the malononitrile accepter group after excitation. And after reacted with hydrazine, the malononitrile group is substituted by electron-abundant hydrazine to form **5**-N_2H_4, which shows a strong ICT effect.

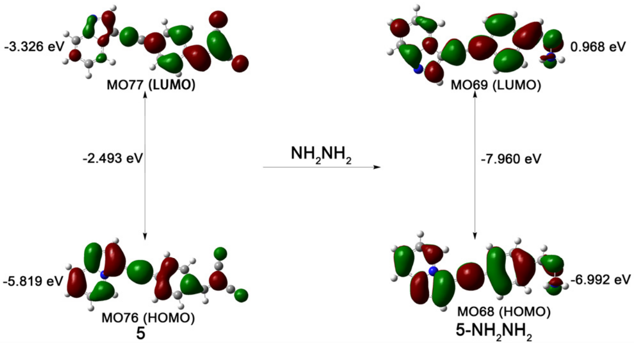


Figure 4. HOMO-LUMO energy levels and the interfacial plots of the molecular orbitals for probe **5** and its reacted form with hydrazine **5**-N_2H_4.

In summary, we have designed and synthesized a fluorescence probe of hydrazine. The probe showed a high sensitivity character during the experimental verification. The crystal of probe **5** was grown and its crystal structure was determined by single-crystal X-ray diffraction analysis, which showed a extremely similar data to calculated geometries using Gaussian 09 program package based on DFT calculations. We have also studied its character via UV and fluorescence. Comparing the experimental UV absorption spectra and fluorescence titration spectra of probe **5** and its reacted form with hydrazine with calculated value, the results are in agreement.

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Supporting Information is available electronically on J-STAGE.

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