Dyes and Pigments 133 (2016) 248–254

Contents lists available at ScienceDirect

Dyes and Pigments

journal homepage: www.elsevier.com/locate/dyepig

A rapid and highly sensitive fluorescent imaging materials for thiophenols

Weijie Zhang [a, 1], Fangjun Huo [b, *], Tao Liu [a, 1], Yin Wen [a], Caixia Yin [a, **]

[a] Institute of Molecular Science, Key Laboratory of Materials for Energy Conversion and Storage of Shanxi Province, Shanxi University, Taiyuan, 030006, China

[b] Research Institute of Applied Chemistry, Shanxi University, Taiyuan, 030006, China

ARTICLE INFO

Article history:
Received 24 April 2016
Received in revised form
1 June 2016
Accepted 4 June 2016
Available online 6 June 2016

Keywords:
Thiophenols
Detection
Fluorescent probe
Bioimaging

ABSTRACT

Thiophenols, a class of toxic and polluting compounds, are widely used in industrial production. Meanwhile, some aliphatic thiols play important roles in living organisms. Therefore, the development of probes for specific thiophenol detection is of great importance. Herein, a novel highly sensitive and selective 'off-on' fluorescent probe for detecting thiophenols has been developed by an ICT mechanism through a rational design. The probe displays a large Stokes shift (140 nm) and 60-fold fluorescence intensity enhancement. More importantly, the probe features a rapid signal response time (within 100 s), a good linearity range and the detection limit is as low as 13 nM. In addition, the ability of the probe to detect thiophenols in living cells (HepG2 cells) via an enhancement of the fluorescence has also been demonstrated.

1. Introduction

Thiophenols, also named benzenethiols, are extensively used in organic synthesis for preparing various products in the agrochemical industry and pharmaceutical industry. However, thiophenols are poisonous to aquatic bio-organisms and animals, possessing a median lethal concentration (LC 50) of 0.01–0.4 mM in fish and a median lethal dose (LD 50) of 46.2 mg/kg in mouse. It was also reported that thiophenols can enter into the human body easily by inhalation and skin absorption to induce systemic injuries including shortness of breath, muscular weakness, nausea, vomiting, and even death [1–3]. Despite high toxicity, thiophenols are essential and widely used chemicals for the preparation of agrochemicals, pharmaceuticals, and various industrial products [4–6]. Considering their toxicity and the continuing environmental concerns, simple, rapid, sensitive, and selective detection of thiophenols is therefore of considerable interest in both environmental and biological science.

Fluorescence detection using fluorescent probes has been recognized as one of the most attractive methods to monitor and visualize molecules due to its simplicity, convenience, and great potential for use in a wide range of chemical, biological, and environmental applications. Since Wang et al. [7] reported the first reaction based fluorescent turn-on probe for selective detection of thiophenols, a number of fluorescent probes have been developed for these compounds [8–20]. Because of the similarity of chemical properties among aliphatic thiols and thiophenols, most of these probes are designed mainly for discrimination of aliphatic thiols such as cysteine and glutathione from other amino acids, and in general, they cannot clearly discriminate thiophenols over aliphatic thiols [21–24]. 2,4-Dinitrobenzenesulfonyl (DNBS) was first used by Maeda et al. as a sulfonate ester of a fluorophore for the detection of thiols [25]. The high level of electron deficiency enables the DNBS moiety to act as an electron sink and incur intramolecular charge transfer (ICT), resulting in the quenching of the fluorescence. The DNBS ester can easily undergo desulfonylation in the presence of thiophenols through a nucleophilic aromatic substitution (SNAr) mechanism, liberating SO_2 and the attached fluorophore, thus resulting in an increase in fluorescence.

Herein, we report a novel type of highly sensitive and selective fluorescent probe toward thiophenols which operates through an ICT pathway by selecting *N*-butyl-4-amino-1,8-naphthalimide as a fluorophore, the 2,4-dinitrobenzene- sulfonamide group as a recognition unit, and a piperazine moiety as a linker to extend the fluorophore and reactive sulfonamide bond. The masked

* Corresponding author.
** Corresponding author.
E-mail addresses: huofj@sxu.edu.cn (F. Huo), yincx@sxu.edu.cn (C. Yin).
[1] Weijie Zhang, Tao Liu contributed equally to this work.

http://dx.doi.org/10.1016/j.dyepig.2016.06.009
0143-7208/

sulfonamide moiety can be easily removed by a highly nucleophilic thiolate anion through the S_NAr process (see Scheme 1) [26]. More importantly, we found that this probe not only shows high selectivity and sensitivity (DL = 13 nM) but also offers a rapid fluorescence turn-on sensing process (100s) for detecting thiophenols. Furthermore, this probe was successfully applied in fluorescent imaging in living cells.

2. Materials and methods

2.1. Materials

4-(2-Hydroxyethyl)-1-piperazineethanesulfonic acid (HEPES) was purchased from Sigma-Aldrich (St. Louis, MO). Sodium hydroxide solution (0.1 mol/L) was added to aqueous HEPES (10 mmol/L) to adjust the pH to 7.4. Amino acids were purchased from Shanghai Experiment Reagent Co., Ltd (Shanghai, China). All other chemicals used were of analytical grade.

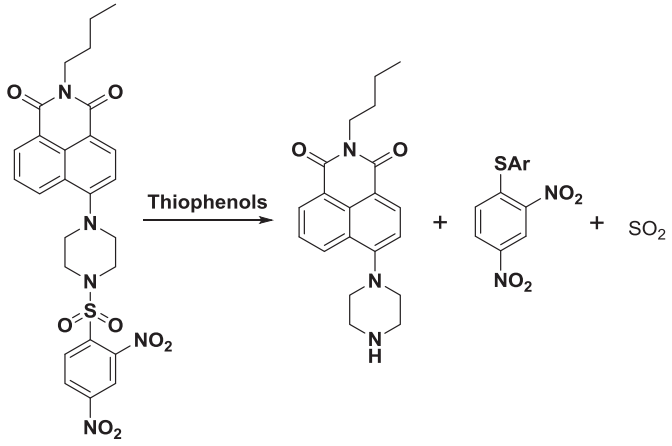
2.2. Instruments

A pH meter (Mettler Toledo, Switzerland) was used to determine the pH. Ultraviolet–visible (UV–vis) spectra were recorded on an Agilent 8453 UV–Visible spectrophotometer. Fluorescence spectra were measured on F-7000 FL Spectrophotometer. A PO-120 quartz cuvette (10 mm) was purchased from Shanghai Huamei Experiment Instrument Plants, China. ^{1}H NMR, ^{13}C NMR spectra were recorded on a Bruker AVANCE-600 MHz and 150 MHz NMR spectrometer, respectively (Bruker, Billerica, MA). ESI determinations were carried out on AB Triple TOF 5600plus System (AB SCIEX, Framingham, USA). The ability of the probe to react with thiophenols in living cells was also evaluated by laser confocal fluorescence imaging using an Olympus FV1000 laser scanning microscope.

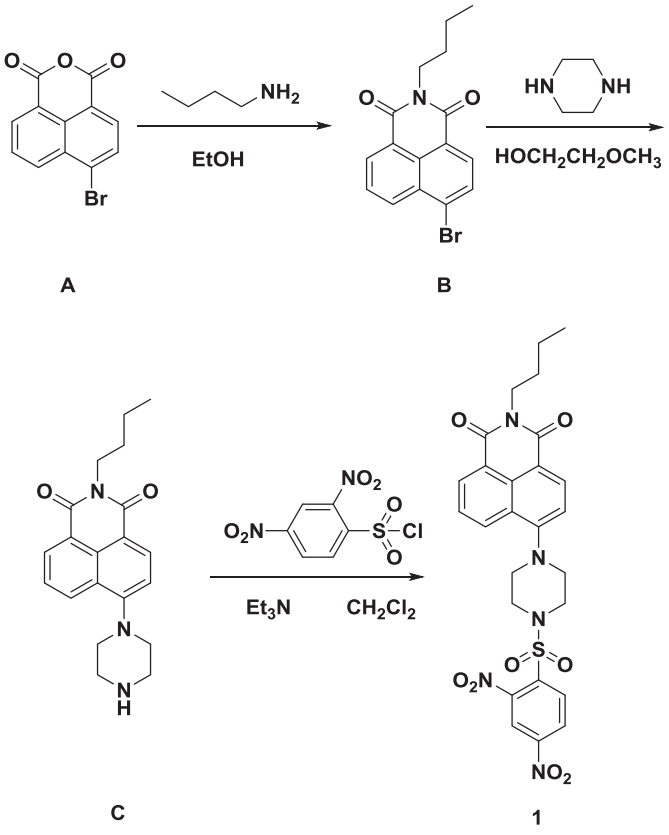
2.3. Preparation and characterization of the probe

2.3.1. Synthesis of N-butyl-4-Br-1,8-naphthalimide

The route employed to synthesise of the probe is summarized in Scheme 2. 4-Bromo-1,8-naphthalic anhydride (2.76 g, 10 mmol) and *N*-butylamine (0.8 g, 11 mmol) was taken in EtOH (20 mL) and stirred at 55 °C monitoring with TLC. After the reaction was complete, the reaction mixture was evaporated in vacuum and the residue was washed with water and dried to give a solid product. The solid obtained was purified by column chromatography (silica gel dichloromethane) to give a white product in 88% yield. mp, 104°C–106 °C. IR (KBr): 3389.40, 3340.32, 3308.66, 3069.14, 2954 45, 2930.41, 2871.51, 1700.09, 1658.69, 1616.26, 1590.03, 1569.12, 1502.94, 1460.23, 1435.22, 1402.59, 1380.89, 1359.43, 1342.57, 1259.15, 1229.36, 1191.63, 1151.33, 1116.74, 1098.79, 1075.34, 1042.95, 996.06, 940.45, 899.29, 873.13, 858.94, 848.36, 780.97, 748.66, 730.67, 711.70, 659.48, 567.65, 424.73. ^{1}H NMR (DMSO-d_6, 600 MHz): δ (ppm): 8.54 (d, J = 7.2 Hz, 1H), 8.51 (d, J = 8.7 Hz, 1H), 8.30 (d, J = 7.6 Hz, 1H), 8.19 (d, J = 7.7 Hz, 1H), 7.98 (t, J = 7.8 Hz, 1H), 4.02 (t, J = 7.3 Hz, 1H), 1.61 (m, 1H), 1.35 (m, 1H), 0.93 (t, J = 7.4 Hz, 2H); ^{13}C NMR (DMSO-d_6, 150 MHz): δ 163.22, 163.17, 132.95, 131.94, 131.74, 131.32, 130.14, 129.51, 129.17, 128.61, 123.10, 122.32, 30.02, 20.27, 14.17. ESI–MS *m/z*: $[B + H]^+$ Calcd. for $C_{16}H_{15}BrNO_2$ 332.0286, Found 332.0286 (Fig. S1).



Scheme 1. Proposed detection mechanism of the probe to thiophenol.



Scheme 2. The synthesis of the probe 1.

2.3.2. Synthesis of N-butyl-4-(piperazin-1-yl)-1,8-naphthalimide

N-butyl-4-Br-1,8-naphthalimide (1.65 g, 5 mmol) and Piperazine(0.52 g, 6 mmol) was taken in 20 mL of 2-methoxyethanol. The mixture was stirred at 130 °C with monitoring by TLC. After the reaction was complete, the clear solution obtained was concentrated and left to cool. The brown solid was collected by filtration, washed with water and dried to give a solid product. After crystallization from MeCN, a solid was obtained to give a brown product in 82% yield. mp, 102°C–104 °C. IR (KBr): 3374.44, 3324.79, 3291.76, 3066.96, 2956.74, 2927.13, 2862.04, 2837.13, 2748.11, 1691.30, 1652.99, 1613.22, 1588.15, 1512.96, 1456.36, 1427.33, 1384.64, 1355.30, 1287.47, 1243.90, 1181.73, 1164.87, 1137.39, 1118.77, 1089.48, 1070.96, 1024.50, 1001.53, 949.56, 929.37, 893.57, 873.89, 848.70, 835.18, 783.44, 758.58, 746.45, 671.98, 656.48, 412. 20. ^{1}H NMR (600 MHz, DMSO) δ 8.41 (dd, J = 11.9, 8.0 Hz, 2H), 8.35 (d, J = 7.9 Hz, 1H), 7.76 (t, J = 7.6 Hz, 1H), 7.27 (d, J = 7.9 Hz, 1H), 4.00 (t, J = 6.7 Hz, 2H), 3.13 (s, 4H), 3.00 (s,