

A Fast-Responsive Fluorescent Sensor for Hg²⁺ with High Selectivity and Sensitivity in Aqueous Media

Shanshan Zhang¹ · Qingfen Niu¹ · Xingxing Wu¹ ·
Tianduo Li¹ · Yuezhi Cui¹ · Xiaoyan Li²

Received: 21 July 2015 / Accepted: 14 September 2015 / Published online: 23 September 2015
© Springer Science+Business Media New York 2015

Abstract A fast-responsive fluorescent phenylamine-oligothiophene sensor 2TBDA was reported. This sensor exhibited highly selective and sensitive detection of Hg²⁺ ion in THF/H₂O (7/3, v/v) solution through fluorescence quenching. The detection was not affected by the coexistence of other competitive metal ions. In addition, the detection limit was found to be as low as 3.841×10^{-7} M.

Keywords Fluorescence sensor · Mercury ion · Oligothiophene · Quenching

Introduction

Among the metal cations, mercury has received considerable attention because of its high toxicity. Particularly, mercury is a bioaccumulative and extremely toxic heavy metal that could affect the immune and nervous systems, alter genetic expression, cause serious damage to human

health, and also lead to die even at low concentrations [1–5]. In order to protect public health, the US Environmental Protection Agency (EPA) has set the detection limit down to 10 nM Hg²⁺ in drinking water [6]. Therefore, the selective, sensitive and rapid detection of Hg²⁺ in the environment and food industry is in high demand.

The development of highly selective and sensitive fluorescent sensors for metal cations has received increasing attention due to their potential use in medicine, environment and biology in recent years [7, 8]. Importantly, such sensors for metal cation detection and measurement offer significant advantages such as high sensitivity, good selectivity, versatility, rapid response time, local observation and relatively simple handling [9, 10]. Therefore, fluorescent chemical sensors are elegant alternative to the traditional analytical instruments. Currently, a great number of efforts have been made to develop more fluorescent sensors for the detection of mercury and mercuric salts with sensitivity and selectivity [4, 11–18].

To date, many fluorescent chemosensors for detection of Hg²⁺ have been developed based on calixarenes [19, 20], quinolines [21–23], fluorescein [24–26], rhodamine [27–31], pyrene [32, 33], naphthalimide [34–36] and other structural moieties [11, 37–41], however, some of them still suffer from practical use. Thus, development of more practical, selective and sensitive fluorescent chemosensors is still a challenge.

In our previous studies, we reported the synthesis, photophysical and thin-film properties of a phenylamine-oligothiophene-based derivative 2TBDA [42] (Fig. 1). In order to expand our interest to the fluorescent chemosensor for metal-ion screening studies, we continue to explore its cation-sensing properties. However, the investigated results demonstrated that 2TBDA exhibited high selectivity, sensitivity and rapid fluorescence response to Hg²⁺ ion over other metal

Highlights

- A fast-responsive fluorescent sensor for Hg²⁺ in aqueous solution was reported.
- The sensor exhibited a high selectivity and sensitivity for Hg²⁺.
- This sensor could detect Hg²⁺ ion on-line and in real time.

✉ Qingfen Niu
qf_niu1216@qlu.edu.cn

¹ Shandong Provincial Key Laboratory of Fine Chemicals, Qilu University of Technology, Jinan 250353, People's Republic of China

² School of Chemistry and Chemical Engineering, Shandong University, Shanda Nanlu 27, Jinan 250100, People's Republic of China

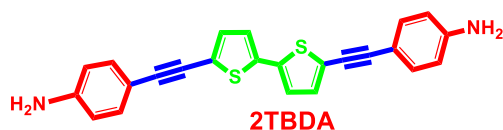


Fig. 1 The structure of sensor **2TBDA**

cations like Na^+ , K^+ , Ag^+ , Ca^{2+} , Al^{3+} , Co^{2+} , Cu^{2+} , Ni^{2+} , Zn^{2+} , Pb^{2+} , Cd^{2+} , Fe^{2+} and Cr^{3+} in aqueous media.

Experimental

Reagents

Unless otherwise stated, solvents and reagents were of analytical grade from commercial suppliers, and were used without

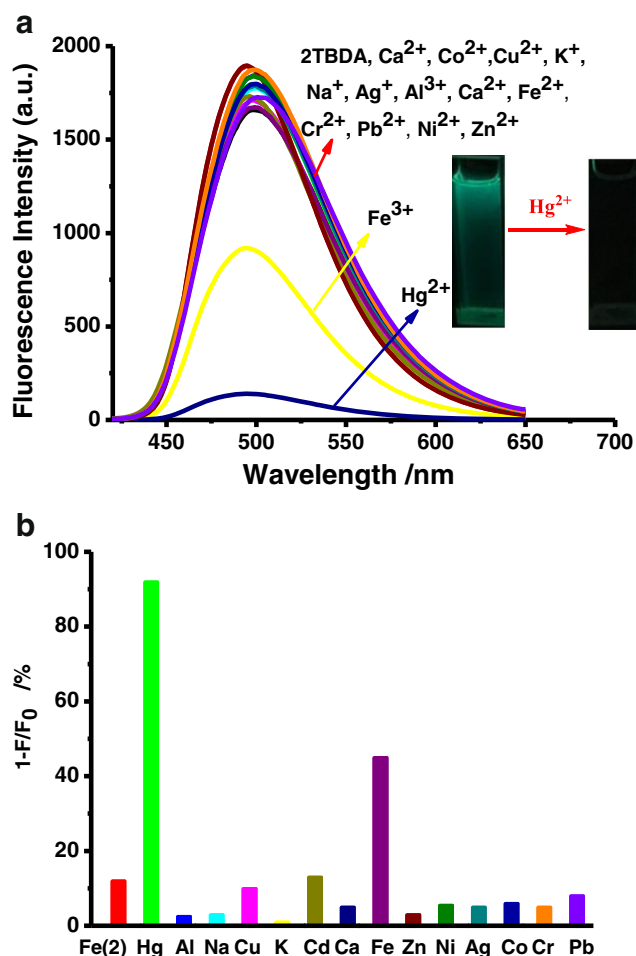


Fig. 2 **a** Fluorescence spectra of **2TBDA** (1.0×10^{-5} M) in aqueous solution (THF/ H_2O , 7/3, v/v) in the presence of various cations (2.0 equiv.). Inset: photos of **2TBDA** without and with addition of Hg^{2+} under the irradiation of UV light at 365 nm; **b** Fluorescence quenching degree of **2TBDA** (10 μM) in the presence of Hg^{2+} (20 μM) and other metal cations (each 20 μM). F_0 and F represented the fluorescence intensities of the **2TBDA**-based sensor system at 498 nm in the absence and presence of M^{n+} , respectively

further purification. Metal salts were purchased from Sigma–Aldrich and used as received. THF was spectrometric grade and purchased from Qingdao Yage Chemical Reagent Company. Water was deionized with a Milli-QSP reagent water system (Millipore) to a specific resistivity of 18.4 M Ωcm . THF and deionized water were used in all of the experiments. All other reagents are analytical grade and purchased from Beijing Chemical works. The salts used in stock solutions of metal cations were NaNO_3 , $\text{Ca}(\text{NO}_3)_2 \cdot 4\text{H}_2\text{O}$, $\text{Al}(\text{NO}_3)_3 \cdot 9\text{H}_2\text{O}$, $\text{Pb}(\text{NO}_3)_2$, $\text{Cu}(\text{NO}_3)_2 \cdot 3\text{H}_2\text{O}$, AgNO_3 , $\text{Zn}(\text{NO}_3)_2 \cdot 6\text{H}_2\text{O}$, $\text{Cd}(\text{NO}_3)_2 \cdot 4\text{H}_2\text{O}$, $\text{Fe}(\text{NO}_3)_3 \cdot 9\text{H}_2\text{O}$, $\text{Co}(\text{NO}_3)_2 \cdot 6\text{H}_2\text{O}$, $\text{Ni}(\text{NO}_3)_2 \cdot 6\text{H}_2\text{O}$, KNO_3 , $\text{Hg}(\text{NO}_3)_2 \cdot \text{H}_2\text{O}$, $\text{FeCl}_2 \cdot 4\text{H}_2\text{O}$ and $\text{CrCl}_3 \cdot 6\text{H}_2\text{O}$. **2TBDA** was prepared as reported previously [42].

Apparatus

All UV-vis absorption spectra were recorded with a Shimadzu UV-2600 spectrophotometer at room temperature. All fluorescence measurements were carried out on a Hitachi F-4600 fluorescence spectrophotometer with a scan rate at 1200 nm/min. The excitation wavelength was set at 320 nm. The slits for excitation and emission were set at 5 nm/5 nm, respectively.

General Procedures for Spectral Determination

All tests described in this paper were carried out at room temperature. All the metal salts of Na^+ , K^+ , Ag^+ , Ca^{2+} , Fe^{3+} , Al^{3+} , Co^{2+} , Ni^{2+} , Cu^{2+} , Zn^{2+} , Pb^{2+} , Cd^{2+} , Hg^{2+} , Fe^{2+} and Cr^{3+} were dissolved in water to prepare the stock solution with the concentration of 1.0×10^{-3} M. **2TBDA** was dissolved in THF to give the stock solution (1.0×10^{-3} M) and diluted with a mixed solution of THF/ H_2O to prepare the analytical solution (1.0×10^{-5} M) (THF/ H_2O , 7/3, v/v). The stock solution of the

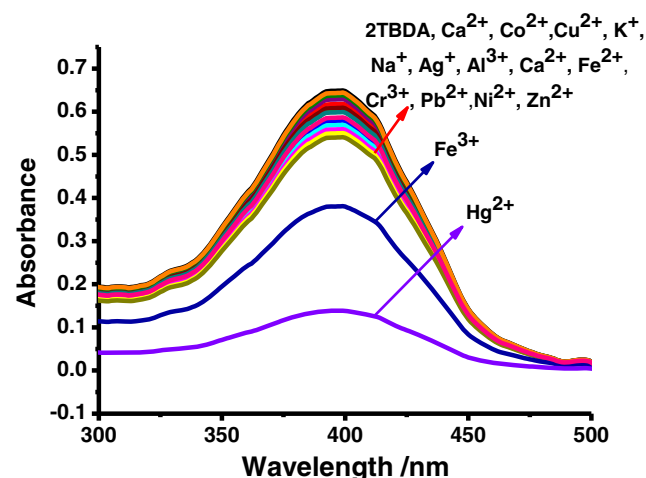


Fig. 3 UV-vis absorption spectra of **2TBDA** (10 μM) in the presence of 2.0 equiv. of different metal ions

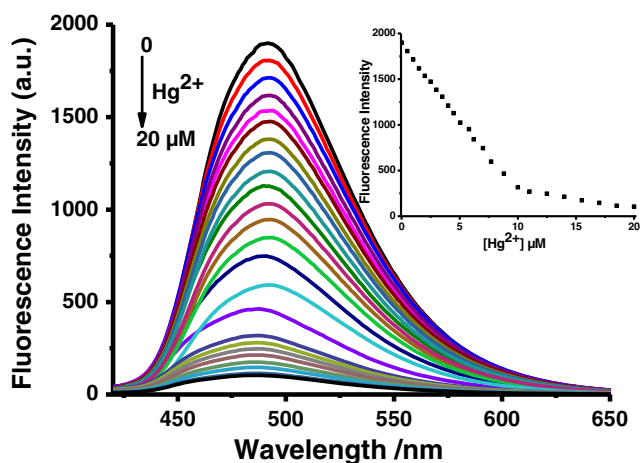


Fig. 4 Fluorescence emission spectra of 2TBDA (1.0×10^{-5} M) was titrated with Hg^{2+} (0–2.0 equiv.) in aqueous solution (THF/ H_2O , 7/3, v/v). Inset: plot of the fluorescence intensity at 498 nm as a function of Hg^{2+} concentration

metal cations and 2TBDA was used directly in the spectroscopic measurement. For the sensitivity measurement, different concentrations of Hg^{2+} ions were added to the assay solution, and the fluorescence spectra were recorded. The selectivity was checked by addition of Na^+ , K^+ , Ag^+ , Ca^{2+} , Fe^{3+} , Al^{3+} , Co^{2+} , Ni^{2+} , Cd^{2+} , Zn^{2+} , Pb^{2+} , Cu^{2+} , Fe^{2+} and Cr^{3+} in to the stock solution.

Results and discussion

To explore the sensing ability of 2TBDA, 15 metal cations including Na^+ , K^+ , Ca^{2+} , Co^{2+} , Ni^{2+} , Cu^{2+} , Zn^{2+} , Fe^{3+} , Pb^{2+} , Ag^+ , Cd^{2+} , Hg^{2+} , Al^{3+} , Cr^{3+} and Fe^{2+} was investigated in THF/ H_2O (7/3, v/v) by fluorescence and UV-vis measurements. The results indicated that the fluorescence intensity of 2TBDA strongly quenched in the presence of the Hg^{2+}

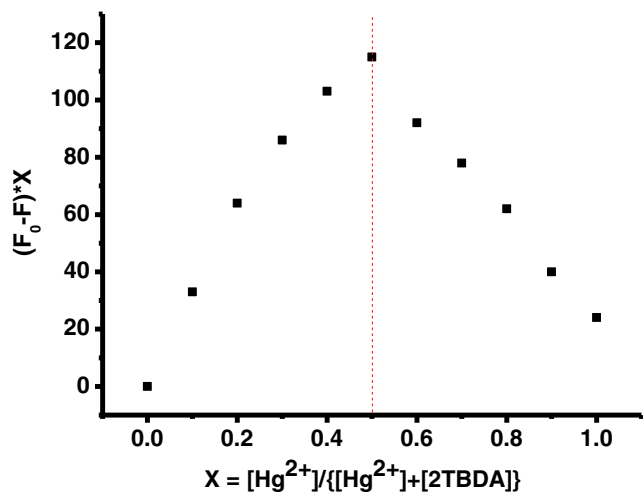


Fig. 5 Job's plot for determining the stoichiometry for 2TBDA and Hg^{2+} in THF/ H_2O (7/3, v/v) solution. The total concentration was 10 μ M

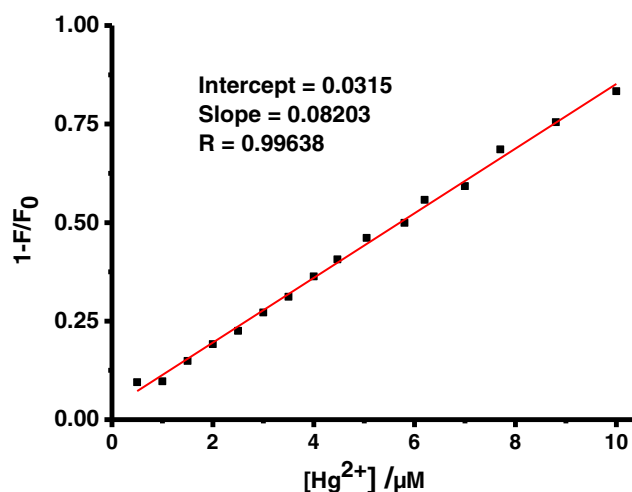


Fig. 6 Calibration curve of 2TBDA– Hg^{2+} in THF/ H_2O (7/3, v/v) solution

ion (2.0 equiv.) in THF/ H_2O (7:3, v/v) (Fig. 2a), accompanied by a green-yellow color fluorescence turn-off response (Fig. 2a, inset), while Fe^{3+} induced a relative higher fluorescence quenching. Other metal cations did not affect the fluorescence intensity of 2TBDA significantly. The observed quenching efficiency ($(1-F/F_0) \times 100\%$) at 498 nm was nearly 92 % by Hg^{2+} , where the other metal cations caused small quenching in fluorescence (except Fe^{3+} : 46 %) (Fig. 2b). As shown in Fig. 3, the free 2TBDA has strong absorption in the range 350–450 nm, and the absorption maxima located at 398 nm in aqueous solution (THF/ H_2O , 7/3, v/v). The strong absorption intensity of peak at 398 nm highly quenched upon the addition of Hg^{2+} ion (2.0 equiv.) to the solution, whereas other metal ions (2.0 equiv.) make insignificant spectral change in this region (except Fe^{3+}). These obvious

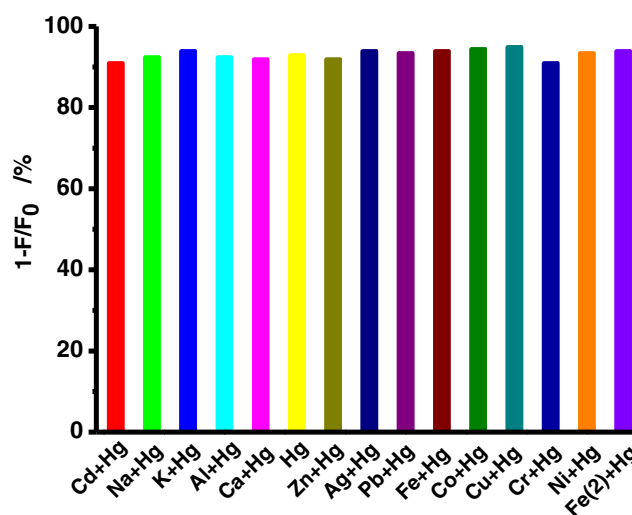


Fig. 7 Metal specificity: the concentration of 2TBDA was 10 μ M, the concentration of Hg^{2+} was 20 μ M and the other metal cations were used at 20 μ M. Mix: the mixture of Ni^{2+} , Na^+ , Ca^{2+} , Fe^{3+} , K^+ , Zn^{2+} , Cd^{2+} , Pb^{2+} , Ag^+ , Al^{3+} , Co^{2+} , Cu^{2+} , Fe^{2+} and Cl^{3+}

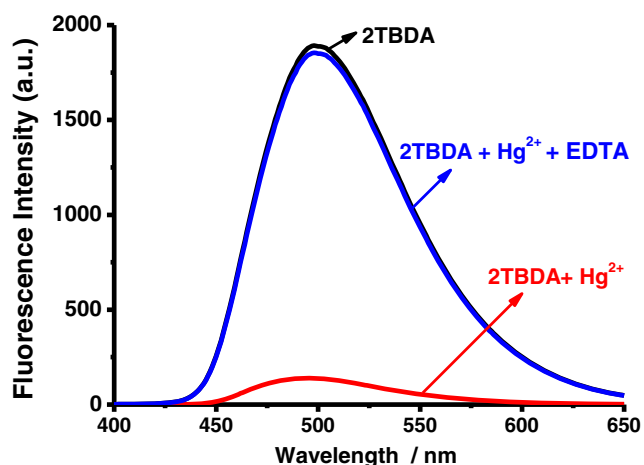


Fig. 8 Fluorescence spectra of 2TBDA in the absence and presence of Hg^{2+} and EDTA. $[\text{Hg}^{2+}] = 20 \mu\text{M}$, $[\text{2TBDA}] = 10 \mu\text{M}$, $[\text{EDTA}] = 100 \mu\text{M}$

observations indicate that 2TBDA displays clear ‘turn-off’ fluorescence response and considerably high selectivity towards Hg^{2+} ion in aqueous solution.

The fluorescence titrations of 2TBDA ($10.0 \mu\text{M}$) was performed in the presence of various concentrations of Hg^{2+} ions in THF/ H_2O (7:3, v/v) in Fig. 4. Upon addition of Hg^{2+} leads to a continuous fluorescence quenching around 498 nm, and the fluorescence intensity remains constant until upon addition of Hg^{2+} (1.0 equiv.). A good linear relationship between the fluorescence intensity at 498 nm and the Hg^{2+} concentration was found in the range of 0–10 μM (Fig. 4, inset). These observations above indicate that 2TBDA and Hg^{2+} have a 1:1 binding ratio. Furthermore, a 1:1 stoichiometry between 2TBDA and Hg^{2+} was obtained from Job’s plot (Fig. 5). The binding model was in good agreement with that of 3TDDA and Hg^{2+} [43]. In addition, the fluorometric titration data was also used to obtain the detection limit (DL) of 2TBDA for Hg^{2+} shown in Fig. 6. The detection limit was thus found to be $3.841 \times 10^{-7} \text{ M}$ for Hg^{2+} ion. The DL was

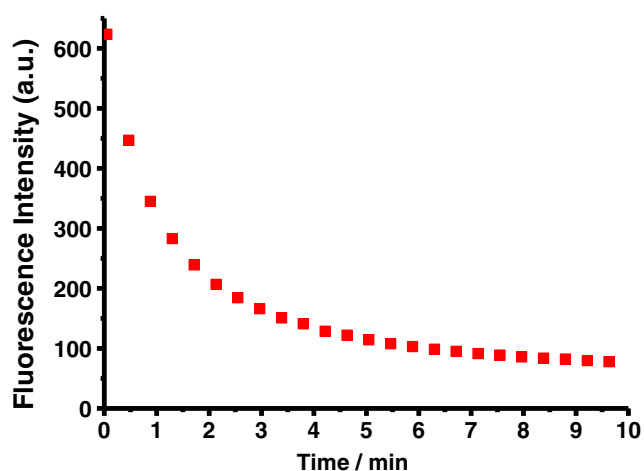


Fig. 9 Fluorescence quenching profile of addition Hg^{2+} ($20 \mu\text{M}$) to 2TBDA ($10 \mu\text{M}$) in THF/ H_2O (7/3, v/v) solution from 1 to 10 min

sufficiently low to detect submillimolar concentration of the Hg^{2+} ion, which belongs the range found in many chemical and biological systems.

To further explore the possibility of using 2TBDA as a practical ion-selective fluorescent chemosensor for Hg^{2+} , competition experiments were carried out. As can be seen from Fig. 7, 2TBDA exhibited a significant selectivity towards Hg^{2+} ion. The deviation of the fluorescence quenching in the presence of other metal cations was less than 6 %. This indicated that Hg^{2+} ion detection by 2TBDA was unaffected by the presence of other competitive metal cations. According to the results, it could be assumed that 2TBDA had potential prospects as a selective detector of Hg^{2+} ion in aqueous environment. Moreover, the EDTA experiments were conducted to examine the reversibility of 2TBDA towards Hg^{2+} ion in aqueous solution (THF/ H_2O , 7/3, v/v) as shown in Fig. 8, the solution changed from colorless to green-yellow when EDTA (5.0 equiv.) was added to the solution of 2TBDA/ Hg^{2+} , and the fluorescence was turned on in several seconds. These results indicate that the coordination process is reversible and thus 2TBDA is a reversible chemosensor for Hg^{2+} in aqueous solutions.

Reaction time is an important factor for sensors, thus the effect of the reaction time on the binding process of Hg^{2+} ion to 2TBDA was investigated (Fig. 9). Following the addition of Hg^{2+} ion ($20 \mu\text{M}$) to 2TBDA ($10 \mu\text{M}$), the fluorescence intensity of 2TBDA was quenched rapidly, reaching a stable value within 3 min and then remaining constant from 3 to 10 min. The rapid, stable complexation of Hg^{2+} ion by 2TBDA and the resulting quick response profile are important features for robust, real time detection of Hg^{2+} ion by portable device in field. In contrast, many previously reported fluorescent sensors showed responses to Hg^{2+} ion in the time range of tens of minutes, generally attributed to slower sensor reaction processes [44, 45].

Conclusions

In summary, a phenylamine-oligothiophene-based fluorescent sensor 2TBDA was reported and its sensing ability for a wide range of metal cations was investigated. 2TBDA displayed highly selective, sensitive and rapid response to Hg^{2+} ion through fluorescence quenching in aqueous solution (THF/ H_2O , 7/3, v/v). The detection limit was found to be as low as $3.841 \times 10^{-7} \text{ M}$. The coordination process of sensor 3TDDA and Hg^{2+} was chemically reversible with EDTA. In addition, this sensor could detect Hg^{2+} ion on-line and in real time, permitting its incorporation into a portable mercury detection kit in aqueous environment.

Acknowledgments We gratefully acknowledge the support by NSF China No. 21176147/21276149 and Program for Scientific Research Innovation Team in Colleges and Universities of Shandong Province.

References

- Zahir F, Rizwi SJ, Haq SK, Khan RH (2005) Low dose mercury toxicity and human health. *Environ Toxicol Pharmacol* 20:351–360
- Magos L, Clarkson TW (2006) Overview of the clinical toxicity of mercury. *Ann Clin Biochem* 43:257–268
- Basu N, Kwan M, Chan HM (2006) Mercury but not organochlorines inhibits muscarinic cholinergic receptor binding in the cerebrum of ringed seals (*Phocahispida*). *J Toxicol Environ Health* 69:1133–1143
- Nolan EM, Lippard SJ (2008) Tools and tactics for the optical detection of mercuric ion. *Chem Rev* 108:3443–3480
- Dorea JG, Donangelo CM (2006) Early (in utero and infant) exposure to mercury and lead. *Clin Nutr* 25:369–376
- (2001) Office of Water, Mercury update: impact on fish advisories, EPA Fact Sheet EPA-823-F-01-011, U.S. EPA, Washington, DC
- de Silva AP, Gunaratne HQN, Gunnlaugsson T, Huxley AJM, McCoy CP, Rademacher JT, Rice TE (1997) Signaling recognition events with fluorescent sensors and switches. *Chem Rev* 97:1515–1566
- Valeur B, Leray I (2000) Design principles of fluorescent molecular sensors for cation recognition. *Coord Chem Rev* 205:3–40
- Zhang J, Campbell RE, Ting AY, Tisen RY (2002) Creating new fluorescent probes for cell biology. *Nat Rev Mol Cell Biol* 3:906–918
- de Silva AP, Eilers J, Zlokarnik G (1999) Emerging fluorescence sensing technologies: from photophysical principles to cellular applications. *Proc Natl Acad Sci U S A* 96:8336–8337
- Udhayakumari D, Velmathi S (2013) Colorimetric and fluorescent sensor for selective sensing of Hg^{2+} ions in semi aqueous medium. *J Lumin* 136:117–121
- Childress ES, Roberts CA, Sherwood DY, LeGuyader CLM, Harbron EJ (2012) Ratiometric fluorescence detection of mercury ions in water by conjugated polymer nanoparticles. *Anal Chem* 84:1235–1239
- Kwon NY, Kim D, Son JH, Jang GS, Lee JH, Lee TS (2011) Simultaneous detection and removal of mercury ions in aqueous solution with fluorescent conjugated polymer-based sensor ensemble. *Macromol Rapid Commun* 32:1061–1065
- Guo LQ, Yin N, Nie DD, Gan JR, Li MJ, Fu FF, Chen GN (2011) Label-free fluorescent sensor for mercury(II) ion by using carbon nanotubes to reduce back ground signal. *Analyst* 136:1632–1636
- Zhang LB, Tao L, Li BL, Jing L, Wang EK (2010) Carbon nanotube-DNA hybrid fluorescent sensor for sensitive and selective detection of mercury(II) ion. *Chem Commun* 46:1476–1478
- Yang Y-K, Ko S-K, Shin I, Tae J (2009) Fluorescent detection of methylmercury by desulfurization reaction of rhodamine hydrazide derivatives. *Org Biomol Chem* 7:4590–4593
- Liu W, Xu L, Zhang H, You J, Zhang X, Sheng R, Li H, Wu S, Wang P (2009) Dithiolane linked thiorhodamine dimer for Hg^{2+} recognition in living cells. *Org Biomol Chem* 7:660–664
- Ko S-K, Yang Y-K, Tae J, Shin I (2006) In vivo monitoring of mercury ions using a rhodamine-based molecular probe. *J Am Chem Soc* 128:14150–14155
- Othman AB, Lee JW, Wu JS, Kim JS, Abidi R, Thuéry P, Strub JM, Dorselaer AV, Vicens J (2007) Calix[4]arene-based, Hg^{2+} -induced intramolecular fluorescence resonance energy transfer chemosensor. *J Org Chem* 72:7634–7640
- Dhir A, Bhalla V, Kumar M (2008) Ratiometric sensing of Hg^{2+} based on the Calix[4]arene of partial cone conformation possessing a dansyl moiety. *Org Lett* 10:4891–4894
- Zhang H, Han LF, Zachariasse KA, Jiang YB (2005) 8-Hydroxyquinoline benzoates as highly sensitive fluorescent chemosensors for transition metal ions. *Org Lett* 7:4217–4220
- Song KC, Kim JS, Park SM, Chung K, Ahn S, Chang S (2006) Fluorogenic Hg^{2+} -selective chemodosimeter derived from 8-hydroxyquinoline. *Org Lett* 8:3413–3416
- Avirah RR, Jyothish K, Ramaiah D (2007) Dual-mode semisquaraine-based sensor for selective detection of Hg^{2+} in a micellar medium. *Org Lett* 9:121–124
- Yoon S, Miller EW, He Q, Do PH, Chang CJ (2007) A bright and specific fluorescent sensor for mercury in water, cells, and tissue. *Angew Chem Int Ed* 46:6658–6661
- Nolan EM, Lippard SJ (2007) Turn-on and ratiometric mercury sensing in water with a red-emitting probe. *J Am Chem Soc* 129:5910–5918
- Yoon S, Albers AE, Wong AP, Chang CJ (2005) Screening mercury levels in fish with a selective fluorescent chemosensor. *J Am Chem Soc* 127:16030–16031
- Zheng H, Qian ZH, Xu L, Yuan FF, Lan LD, Xu JG (2006) Switching the recognition preference of Rhodamine B spirolactam by replacing one atom: design of Rhodamine B thiohydrazide for recognition of $\text{Hg}(\text{II})$ in aqueous solution. *Org Lett* 8:859–861
- Kim HN, Lee MH, Kim HJ, Kim JS, Yoon J (2008) A new trend in rhodamine-based chemosensors: application of spirolactam ring-opening to sensing ions. *Chem Soc Rev* 37:1465–1472
- Wu JS, Hwang IC, Kim KS, Kim JS (2007) Rhodamine-based Hg^{2+} -selective chemodosimeter in aqueous solution: fluorescent off-on. *Org Lett* 9:907–910
- Yang YK, Yook KJ, Tae JS (2005) A rhodamine-based fluorescent and colorimetric chemodosimeter for the rapid detection of Hg^{2+} ions in aqueous media. *J Am Chem Soc* 127:16760–16761
- Yang H, Zhou ZG, Huang KW, Yu MX, Li FY, Yi T, Huang CH (2007) Multisignaling optical-electrochemical sensor for Hg^{2+} based on a Rhodamine derivative with a ferrocene unit. *Org Lett* 9:4729–4732
- Kim JS, Choi MG, Song KC, No KT, Ahn S, Chang S (2007) Ratiometric determination of Hg^{2+} ions based on simple molecular motifs of pyrene and doxaocanediamide. *Org Lett* 9:1129–1132
- Martinez R, Espinosa A, Tàrraga A, Molina P (2005) New Hg^{2+} and Cu^{2+} selective chromo- and fluorionophore based on a bichromophoric azine. *Org Lett* 7:5869–5872
- Wang JB, Qian XH (2006) Two regioisomeric and exclusively selective $\text{Hg}(\text{II})$ sensor molecules composed of a naphthalimide fluorophore and an *o*-phenylenediamine derived triamide receptor. *Chem Commun* 109–111
- Liu B, Tian H (2005) A selective fluorescent ratiometric chemodosimeter for mercury ion. *Chem Commun* 3156–3158
- Wang JB, Qian XH (2006) A series of polyamide receptor based PET fluorescent sensor molecules: positively cooperative Hg^{2+} ion binding with high sensitivity. *Org Lett* 8:3721–3724
- Zhu M, Yuan M, Liu X, Xu J, Lv J, Huang C, Liu H, Li Y, Wang S, Zhu D (2008) Visible near-infrared chemosensor for mercury ion. *Org Lett* 10:1481–1484
- Ha-Thi M, Penhoat M, Michelet V, Leray I (2007) Highly selective and sensitive phosphane sulfide derivative for the detection of Hg^{2+} in an organoaqueous medium. *Org Lett* 9:1133–1136
- Yuan MJ, Li YL, Li JB, Li CH, Liu XF, Lv J, Xu JL, Liu HB, Wang S, Zhu DB (2007) A colorimetric and fluorometric dual-modal assay for mercury ion by a molecule. *Org Lett* 9:2313–2316
- Shiraishi Y, Maehara H, Ishizumi K, Hirai T (2007) $\text{Hg}(\text{II})$ -selective excimer emission of a bisnaphthyl azadiene derivative. *Org Lett* 9:3125–3128
- Chen QY, Chen C-F (2005) A new Hg^{2+} -selective fluorescent sensor based on a dansyl amide-armed calix[4]-aza-crown. *Tetrahedron Lett* 46:165–168

42. Niu QF, Sun HJ, Li XY (2014) Synthesis, photophysical and thin-film self-assembly properties of novel fluorescent molecules with carbon–carbon triple bonds. *Spectrochim Acta A* 133:229–240
43. Niu QF, Wu XX, Zhang SS, Li TD, Cui YZ, Li XY (2016) A highly selective and sensitive fluorescent sensor for the rapid detection of Hg^{2+} based on phenylamine-oligothiophene derivative. *Spectrochim Acta A* 153:143–146
44. Ando S, Koide K (2011) Development and applications of fluorogenic probes for mercury(II) based on vinyl ether oxymercuration. *J Am Chem Soc* 133:2556–2566
45. Miyake Y, Togashi H, Tashiro M, Yamaguchi H, Oda S, Kudo M, Tanaka Y, Kondo Y, Sawa R, Fujimoto T, Machinami T, Ono A (2006) Mercury(II)-mediated formation of thymine-Hg(II)-thymine base pairs in DNA duplexes. *J Am Chem Soc* 128:2172–2173