ORGANIC LETTERS
2010
Vol. 12, No. 11
2566−2569

Hg^{2+}-Selective Ratiometric and "Off−On" Chemosensor Based on the Azadiene−Pyrene Derivative

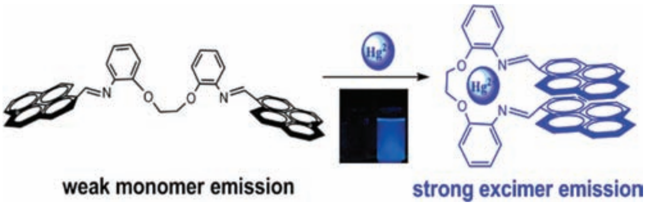
Yi Zhou,[†] Chao-Yun Zhu,[†] Xu-Sheng Gao,[†] Xue-Yan You,[†] and Cheng Yao*[,†,‡]

College of Science and State Key Laboratory of Materials-Oriented Chemical Engineering, Nanjing University of Technology, Nanjing 210009, P. R. China

yaochengnjut@126.com

Received April 4, 2010

ABSTRACT



A new pyrene-based derivative bearing an azadiene group was synthesized as a ratiometric chemosensor for Hg^{2+} in aqueous acetonitrile solution. The "off−on" type signaling behavior of the fluoroionophore is due to the metal ion induced conformational changes from the weak pyrene monomer emissions to strong pyrene excimer emission.

The development of artificial receptors for the sensing and recognition of environmentally and biologically important species has been actively investigated in recent years.[1] In this regard, chemosensors that can highly sensitively and selectively detect heavy and transition metal ions (HTM) such as Hg^{2+}, Pb^{2+}, Cd^{2+}, and Cu^{2+} are especially important.[2] Mercury is considered as a prevalent toxic metal in the environment because both elemental and ionic mercury can be converted by bacteria in the environment to methyl mercury, which subsequently bioaccumulates through the food chain.[3] When absorbed in the human body, mercury causes damage to the central nervous, DNA, mitosis, and endocrine system.[4] As a result, developing new and practical multisignaling chemosensors for Hg^{2+} is still a challenge.[5]

[†] College of Science.

[‡] State Key Laboratory of Materials-Oriented Chemical Engineering.

(1) (a) de Silva, A. P.; Gunaratne, H. Q. N.; Gunnlaugsson, T.; Huxley, A. J. M.; McCoy, C. P.; Rademacher, J. T.; Rice, T. E. *Chem. Rev* **1997**, *97*, 1515–1566. (b) Valeur, B.; Leray, I. *Coord. Chem. Rev.* **2000**, *205*, 3–40. (c) Amendola, V.; Fabbrizzi, L.; Forti, F.; Licchelli, M.; Mangano, C.; Pallavicini, P.; Poggi, A.; Sacchi, D.; Taglieti, A. *Coord. Chem. Rev.* **2006**, *250*, 273–299. (d) Ojida, A.; Nonaka, H.; Miyahara, Y.; Tamaur, S.; Sada, K.; Hamachi, I. *Angew. Chem., Int. Ed.* **2006**, *45*, 5518–5521.

(2) (a) Jiang, P. G.; Chen, L. Z.; Lin, J.; Liu, Q.; Ding, J.; Gao, X.; Guo, Z. J. *Chem. Commun.* **2002**, 1424–1425. (b) Zhang, H.; Han, L.-F.; Zachariasse, K. A.; Jiang, Y. B. *Org. Lett.* **2005**, *7*, 4217–4220. (c) He, Q.; Miller, E. W.; Wong, A. P.; Chang, C. J. *J. Am. Chem. Soc.* **2006**, *128*, 9316–9317. (d) Martinez, R.; Zapata, F.; Caballero, A.; Espinosa, A.; Tairraga, A.; Molina, P. *Org. Lett.* **2006**, *8*, 3235–3238. (e) Avirah, R. R.; Jyothish, K.; Ramaiah, D. *Org. Lett.* **2007**, *9*, 121–124. (f) Yoon, S.; Miller, E. W.; He, Q.; Do, P. H.; Chang, C. J. *Angew. Chem., Int. Ed.* **2007**, *46*, 6658–6661. (g) Shiraishi, Y.; Maehara, H.; Ishizumi, K.; Hirai, T. *Org. Lett.* **2007**, *9*, 3125–3128.

(3) (a) Renzoni, A.; Zino, F.; Franchi, E. *Environ. Res.* **1998**, *77*, 68–72. (b) Harris, H. H.; Pickering, I. J.; George, G. N. *Science* **2003**, *301*, 1203–1203. (c) Boening, D. W. *Chemosphere* **2000**, *40*, 1335–1351. (d) Benoit, J. M.; Fitzgerald, W. F.; Damman, A. W. *Environ. Res.* **1998**, *78*, 118–133.

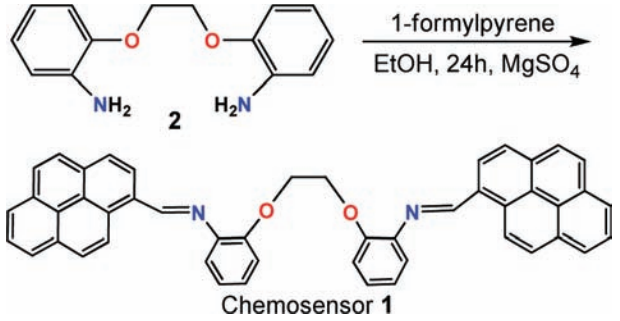
(4) (a) Clarkson, T. W.; Magos, L.; Myers, G. J. *N. Engl. J. Med.* **2003**, *349*, 1731–1737. (b) Von Burg, R. *J. Appl. Toxicol.* **1995**, *15*, 483–493. (c) Nolan, E. M.; Lippard, S. J. *Chem. Rev.* **2008**, *108*, 3443–3480.

(5) (a) Nolan, E. M.; Lippard, S. J. *J. Am. Chem. Soc.* **2003**, *125*, 14270–14271. (b) Zhang, G.; Zhang, D.; Yin, S.; Yang, X.; Shuai, Z.; Zhu, D. *Chem. Commun.* **2005**, 2161–2163. (c) Yoon, S.; Albers, A. E.; Wong, A. P.; Chang, C. J. *J. Am. Chem. Soc.* **2005**, *127*, 16030–16031. (d) Caballero, A.; Martinez, R.; Lloveras, V.; Ratera, I.; Vidal-Gancedo, J.; Wurst, K.; Tarraga, A.; Molina, P.; Veciana, J. *J. Am. Chem. Soc.* **2005**, *127*, 15666–15667. (e) Sasaki, D. Y.; Padilla, B. E. *Chem. Commun.* **2005**, 1581–1582. (f) Coskun, A.; Akkaya, E. U. *J. Am. Chem. Soc.* **2006**, *128*, 14474–14475. (g) Avirah, R. R.; Jyothish, K.; Ramaiah, D. *Org. Lett.* **2007**, *9*, 121–124. (h) Guliyev, R.; Coskun, A.; Akkaya, E. U. *J. Am. Chem. Soc.* **2009**, *131*, 9007–9013. (i) Coskun, A.; Yilmaz, D.; Akkaya, E. U. *Org. Lett.* **2007**, *9*, 607–609. (j) Zhu, M.; Yuan, M.; Liu, X.; Xu, J.; Lv, J.; Huang, C.; Liu, H.; Li, Y.; Wang, S.; Zhu, D. *Org. Lett.* **2008**, *10*, 1481–1484.

10.1021/ol1007636 © 2010 American Chemical Society
Published on Web 05/07/2010

The pyrene moiety is one of the most useful fluorophores for the construction of fluorogenic chemosensors for a variety of important chemical species.[6] Particularly, the introduction of two pyrene moieties can be situated closely enough to yield face-to-face orientation excimer emission. Upon coordination with a specific guest ion, the resulting compound could be fine-tuned to yield monomer and/or excimer emissions depending on the orientation of the two pyrene moieties.[7] Utilizing the pyrene moiety of monomer vs excimer emission, a variety of "on−off" type HTM ion recognition systems were successfully devised.[8] However, the pyrene excimer "off−on" type system with HTM ions has been rarely reported until now.[9] Furthermore, only a few ratiometric fluorescent probes for Hg^{2+} have been found in the literature, and most of them were working only in pure organic solvents.[10] Herein, we describe a new azadiene−pyrene derivative chemosensor **1** (Scheme 1), which shows a selective, sensitive, and reversible fluorescence enhancement response to the Hg^{2+} ion due to the two pyrene moieties' orientation excimer emission.

Scheme 1. Synthesis of Chemosensor **1**



2

Chemosensor 1

Chemosensor **1** was synthesized by the condensation of 2,2′-(ethylenedioxy)dianiline with 1-formylpyrene in 42% yield (Scheme 1). A 2,2′-(ethylenedioxy)-diimino-based molecular framework was designed as a platform for the construction of efficient ionophores via its ether-methylene O and imino N atoms.[11] This Shiff base was stable in neutral acetonitrile and water−acetonitrile solutions for at least 3 days. Thermogravimetric analyses showed that chemosensor **1** was stable up to 280 °C (Figure S1, see Supporting Information), at which point decomposition of the components occurred. The fluorescence emission at 406 nm was relatively unaffected by pH values between 4.13 and 9.14 (Figure S4, Supporting Information). These observations indicate that chemosensor **1**, which is thermally stable, pH-stable, and organic solvent-stable, may be useful as a potential chemosensor material.

The absorption spectrum of **1** shows the typical pyrene absorption bands at 238, 283, and 326 nm,[12] along with a slightly low-energy band centered at 395 nm attributed to the imino bridge. Upon addition of 1−1.2 equiv of Hg^{2+} ions to the solution of **1**, the most significant changes were broadening of the absorption bands around 350−520 nm, and a new red-shifted band was formed at 442 nm (Figure 1). The formation of the new low-energy band may be attributed to the interaction of Hg^{2+} ions with the imino nitrogens leading to the intramolecular charge transfer from the pyrene moieties to the imino groups. The new low-energy band (47 nm red shift) is responsible for the change of color, which is perceptible to the naked eye, from colorless to pale yellow.

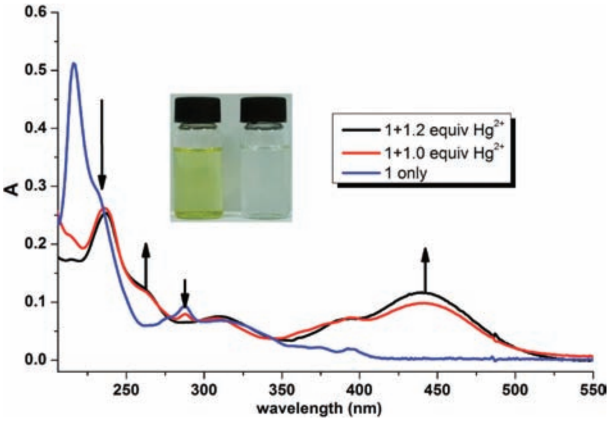


Figure 1. UV−vis spectra of chemosensor **1** (10 μM) in the presence of Hg^{2+} ions (1.0 equiv and 1.2 equiv) in CH_3CN. Inset shows the change in color of chemosensor **1** (10 μM) upon addition of Hg^{2+} ions (10 μM).

The fluorescent properties of **1**−Hg^{2+} were surveyed in typical organic solvent systems including their aqueous

---

[6] (a) Winnick, F. M. *Chem. Rev.* **1993**, *93*, 587–614. (b) Nishizawa, S.; Kato, A.; Teramae, N. *J. Am. Chem. Soc.* **1999**, *121*, 9463–9464. (c) Sahoo, D.; Narayanaswami, V.; Kay, C. M.; Ryan, R. O. *Biochemistry* **2000**, *39*, 6595–6601. (d) Kim, S. K.; Kim, S. K.; Yoon, J. *J. Org. Chem.* **2003**, *68*, 597–600. (e) Yuasa, H.; Miyagawa, N.; Izumi, T.; Nakatani, M.; Izumi, M.; Hashimoto, H. *Org. Lett.* **2004**, *6*, 1489–1492.

[7] (a) Shiraishi, Y.; Tokitoh, Y.; Hirai, T. *Org. Lett.* **2006**, *8*, 3841–3844. (b) Kim, H.; Hong, J.; Hong, A.; Ham, S.; Lee, J.; Kim, J. S. *Org. Lett.* **2008**, *10*, 1963–1966. (c) Jung, H.; Park, M.; Han, D.; Kim, E.; Lee, C.; Ham, S.; Kim, J. S. *Org. Lett.* **2009**, *11*, 3378–3381.

[8] (a) Moon, S. Y.; Youn, N. J.; Park, S. M.; Chang, S. K. *J. Org. Chem.* **2005**, *70*, 2394–2397. (b) Kim, J. S.; Choi, M. G.; Song, K. C.; No, K. T.; Ahn, S.; Chang, S. K. *Org. Lett.* **2007**, *9*, 1129–1132. (c) Park, S. M.; Kim, M. H.; Choe, J. I.; No, K. T.; Chang, S. K. *J. Org. Chem.* **2007**, *72*, 3550–3553. (d) Xu, Z.; Yoon, J.; Spring, D. R *Chem. Commun.* **2010**, *46*, 2563–2565.

[9] (a) Xie, J.; Menand, M.; Maisonneuve, S.; Metivier, R. *J. Org. Chem.* **2007**, *72*, 5980–5985. (b) Zhang, H.; Han, L. F.; Zachariasse, K. A.; Jiang, Y. B. *Org. Lett.* **2005**, *7*, 4217–4220. (c) Yang, J. S.; Lin, C. S.; Hwang, C. Y. *Org. Lett.* **2001**, *3*, 889–892. (d) Hung, H. C.; Cheng, C. W.; Ho, I. T.; Chung, W. S. *Tetrahedron. Lett.* **2009**, *50*, 302–305. (e) Hung, H. C.; Cheng, C. W.; Wang, Y. Y.; Chen, Y. J.; Chung, W. S. *Eur. J. Org. Chem.* **2009**, 6360–6366.

[10] (a) Xiang, Y.; Tong, A.; Jin, P.; Ju, Y. *Org. Lett.* **2006**, *8*, 2863–2866. (b) Wu, J.; Hwang, I.; Kim, K.; Kim, J. *Org. Lett.* **2007**, *9*, 907–910. (c) Lee, M.; Wu, J.; Lee, J.; Jung, J.; Kim, J. *Org. Lett.* **2007**, *9*, 2501–2504. (d) Zhang, X.; Shiraishi, Y.; Hirai, T. *Tetrahedron Lett.* **2007**, *48*, 5455–5459. (e) Soh, J.; Swamy, K.; Kim, S.; Kim, S.; Lee, S.; Yoon, J. *Tetrahedron Lett.* **2007**, *48*, 5966–5969. (f) Zhang, X.; Shiraishi, Y.; Hirai, T. *Tetrahedron Lett.* **2008**, *10*, 4178–4181. (g) Lee, M. H.; Kim, H. J.; Yoon, S.; Park, N.; Kim, J. S. *Org. Lett.* **2008**, *10*, 213–216.

[11] (a) Niasari, M. S. *J. Coord. Chem.* **2009**, *62*, 980–995. (b) Shanker, K.; Rohini, R.; Ravinder, V.; Reddy, P. M.; Ho, Y. P. *Spectrochim. Acta, Part A* **2009**, *73*, 205–211.

[12] Forster, T.; Kasper, K. Z. *Elektrochemistry* **1955**, *59*, 976–980.

solutions. **1**−Hg^{2+} exhibited a weak monomer emission around 395−410 nm with characteristic, but a very intense, excimer emission of the pyrene centered at 462 nm (Figure S3, Supporting Information). The fluorescence characteristics of **1**−Hg^{2+} were found to be strongly dependent on the nature of the employed medium. **1**−Hg^{2+} in aqueous acetonitrile (HEPES−CH_3CN, 10:90, v/v) displayed a relatively weak excimer emission at 462 nm. The pyrene excimer region increased significantly as the water content increased, particularly in the vicinity of 80% water composition, and then the changes were not so significant up to 90% aqueous solution (Figure S2, Supporting Information). This observation implies that complexation of **1** with Hg^{2+} ions presumably changes from its chair conformation to stacked or folded conformation resulting in the switch of the pyrene monomer emissions to an excimer emission.

The fluorescence spectrum of chemosensor **1** with several metal cations (Na^+, K^+, Mg^{2+}, Ca^{2+}, Cd^{2+}, Mn^{2+}, Fe^{3+}, Ni^{2+}, Ag^+, Pb^{2+}, Co^{2+}, Zn^{2+}, Cr^{3+}, Cu^{2+}, and Hg^{2+}) using their perchlorate salts in CH_3CN/HEPES (pH 7.2, 80:20, v/v) is investigated by titration studies (Figure 2). A 15.2-fold enhancement of fluorescence and a continuous red shift of the emission peak from 406 to 462 nm were observed upon the addition of 10 equiv of Hg^{2+} compared to that of only **1** in CH_3CN/HEPES mixture solutions. Upon interaction with varying metal ions of alkali (Na^+, K^+), alkaline earth (Mg^{2+}, Ca^{2+}), and transition-metal ions (Cd^{2+}, Mn^{2+}, Fe^{3+}, Ni^{2+}, Ag^+, Pb^{2+}, Co^{2+}, Zn^{2+}, and Cr^{3+}), a much weaker response is given compared to Hg^{2+} at the same concentration (10 μM), and the fluorescence signal of **1**−Hg^{2+} in the presence or absence of these contrast ions also exhibited only a mild difference. With Cu^{2+} ions, however, a slightly response of the pyrene excimer emission was observed, consistent with the earlier reported alkylamine−pyrene derivative compounds,[13] and the fluorescence intensity increased 3.2-fold upon added 10 μM Cu^{2+} ions (8.3-fold with 100 equiv of Cu^{2+} ions). The discrimination between two oppositely responding Hg^{2+} and Cu^{2+} ions could be realized ratiometrically by using the large changes in Hg^{2+} to trigger the monomer−excimer switching of **1**. To test whether the proposed complex could be reversed, we added Na_2S (0.1 M) to the solutions of **1**−Hg^{2+} species. The addition of 0.55 mL of Na_2S could restore the initial value of free probes due to the K_d value of 10^{-50} M^2 for Hg^{2+} at a standard condition in the form of $[HgS_2]^{2-}$.[14] Thus, the probes could be revived on addition of Hg^{2+} (Figure S5, Supporting Information).

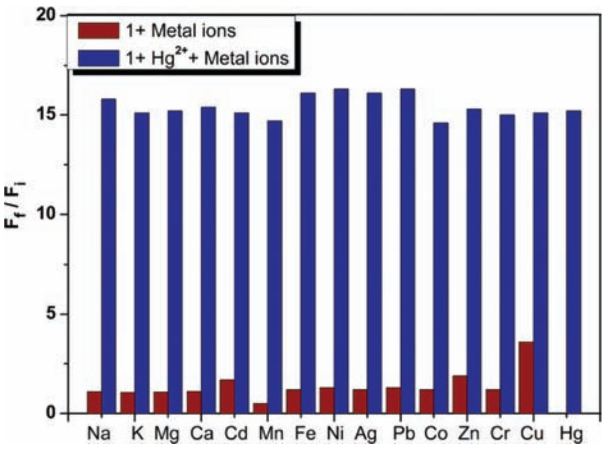


Figure 2. Fluorescence responses of 1 μM **1** to various 10 μM metal ions. Bars represent the final (F_f) over the initial (F_i) integrated emission. Spectra were acquired in (HEPES−CH_3CN, 80:20, v/v) pH 7.2. The red bars represent the addition of the competing metal ion to a 1 μM solution of **1**. The blue bars represent the change of the emission that occurs upon the subsequent addition of 10 μM Hg^{2+} to the above solution. Excitation was provided at 365 nm, with emission integrated over 380−600 nm.

The titration reaction curve showed a steady and smooth increase until a plateau was reached (15.0 μM Hg^{2+}) with an 18-fold increase at the plateau (Figure 3), suggesting the completion of the monomer−excimer switch of **1**. The chemosensor exhibited very efficient fluorescence responding, and over 65% of the total fluorescence intensity increase was observed with **1** equiv of Hg^{2+} ions. The association constant (K_a) of **1** with Hg^{2+} is 4.32 × 10^5 M^{-1} (error <10%), obtained by a nonlinear curve fitting of the fluorescence titration results.[15] The inset in Figure 3 exhibits the dependence of the intensity ratios of emission at 462 nm (F_f/F_i) on Hg^{2+}. This curve can be served as the calibration curve for the detection of Hg^{2+}.

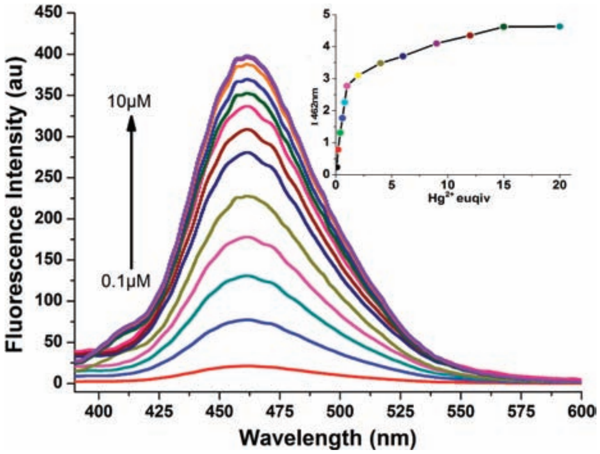


Figure 3. Fluorescence titration of **1** (1 μM) in HEPES buffer (50 mM, pH = 7.2) aqueous buffer solutions in the presence of different amounts of Hg^{2+}. Excitation was performed at 365 nm. Inset: fluorescence intensity at 462 nm as a function of Hg^{2+} concentration.

---

(13) Bhalla, V.; Tejpal, R.; Kumar, M.; Sethi, A. *Inorg. Chem.* **2009**, *48*, 11677–11684.

(14) (a) Findlay, D. M.; McLean, R. A. N. *Environ. Sci. Technol.* **1981**, *15*, 1388–1390. (b) Armstrong, R. D.; Porter, D. F.; Thirsk, H. R. *J. Phys. Chem.* **1968**, *72*, 2300–2306. (c) Huang, W.; Song, C.; He, C.; Lv, G. J.; Hu, X.; Zhu, X.; Duan, C. *Inorg. Chem.* **2009**, *48*, 5061–5072.

(15) (a) Conners, K. A. *Binding Constants*; Wiley: New York, 1987. (b) Valeur, B. *Molecular Fluorescence. Principles and Applications*; Wiley-VCH: Weinheim, 2002.

The detection limit[16] was also estimated from the titration results and was 2×10^{-7} M.

As shown in the ^{1}H NMR spectra of Figure 4a and 4b, treatment of 1 equiv of Hg^{2+} resulted in a considerable downfield shift of imino protons by δ 1.22 ppm. There was also a slight upfield shift of ether−methylene protons by δ 0.12 ppm. The corroborative evidence for the **1**−Hg^{2+} complex was observed in their solid state reaction, which showed that after one minute grinding in the mortar the light yellow solid **1** turned to an obvious reddish-brown color solid in the presence of 1 equiv of Hg^{2+}, and other ions of our interest showed little interference (Figure S6, Supporting Information). These ^{1}H NMR and grind reaction results suggest that Hg^{2+} coordinated with the imino of the Schiff base forms Hg−N bonding and that O atoms of ether-methylene forms Hg−O bonding.

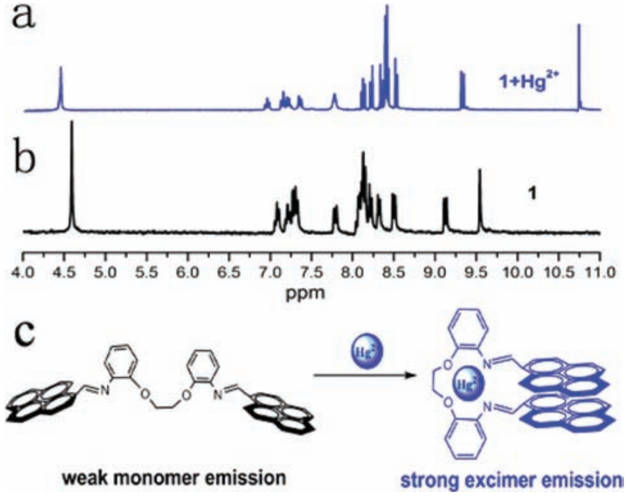


Figure 4. ^{1}H NMR spectra of **1** (a) and in the presence of Hg^{2+} (b) in DMSO-d_6. (c) Proposed mechanism of binding mode of **1** with Hg^{2+}.

On the basis of the ^{1}H NMR spectroscopy, the plausible binding mechanism of **1** in the present system is schematically depicted in Figure 4c. The transformation into folded conformation might be partly attributed to the participation of the imino of the Schiff base and O atoms of ether-methylene in the complex formation with the guest metal ions, which forces the two pyrene moieties to become relatively folded with face to face orientations. The conformational changes result in the weak pyrene monomer emission to strong pyrene excimer emission switch.

In summary, we have developed a new fluorescent sensor for Hg^{2+} on the basis of the pyrene monomer vs excimer emission with high sensitivity and selectivity. Moreover, this simple molecule makes it possible to detect the Hg^{2+} ratiometrically. This chemosensor was easily prepared and found to be stable in both alkaline and acidic solutions. The spectral response toward Hg^{2+} was established to be reversible. The design strategy and remarkable photophysical properties of the two pyrene moieties would help to extend the development of fluorescent sensors for HTM ions.

Acknowledgment. This work is supported by the Open Fund of the State Key Laboratory of Materials-Oriented Chemical Engineering (KL09-9) and the National Natural Science Foundation of China (Grants 20971017). We also thank Dr. Jie Song (China Pharmaceutical University) for his kind help in NMR characterization.

Supporting Information Available: Synthesis, experimental details, and additional spectroscopic data. This material is available free of charge via the Internet at http://pubs.acs.org.

OL1007636

---

[16] Shortreed, M.; Kopelman, R.; Kuhn, M.; Hoyland, B. *Anal. Chem.* **1996**, *68*, 1414–1418.