Hg^{2+}-Selective Fluorescent Chemosensor Based on Cation-π Interaction

YANG Huixiao[1,2], HU Qinzhi[1,2], MA Guochun[1,2], CHEN Guofeng[1,2],
TAO Minli[1,2]* and ZHANG Wenqin[1,2]

*1. Department of Chemistry, School of Science, 2. Collaborative Innovation Center of
Chemical Science and Engineering(Tianjin), Tianjin University, Tianjin 300072, P. R. China*

Abstract   Two sulphur-containing 4-aminonaphthalimide derivatives were investigated as Hg^{2+} fluorescent chemosensors. In CH$_3$CN, both sensors present a remarkable fluorescence enhancement to Cu^{2+} and Fe^{3+}, but a selective fluorescence quenching to Hg^{2+} among the other metal ions. A cation-π interaction between Hg^{2+} and the naphthalimide moiety was proposed and confirmed by the density functional theory(DFT).

Keywords   4-Aminonaphthalimide; Fluorescent chemosensor; Cation-π interaction; Hg^{2+}; Density functional theory (DFT)

1   Introduction

The development of selective chemosensors for the determination of heavy-metal ions, which are very toxic in the living systems and result in serious environmental problems, is one of the most crucial research topics in the field of supramolecular chemistry[1]. Among heavy-metal ions, Hg^{2+} is considered to be highly dangerous because both elemental and ionic mercury can be converted into methyl mercury by certain bacteria, which subsequently bioaccumulates through the food chain[2—5]. Therefore, there is an urgent demand for the detection of Hg^{2+} both in the environmental analysis and in the industrial waste treatment. Recently, for Hg^{2+}-selective detection, many fluorescent chemosensors[6—12] using the principles of photoinduced electron transfer(PET)[13], internal charge transfer (ICT)[14] and the formation of new chemical bonds[15, 16] have been reported.

Cation-π interactions between metal ions and arenes as one of the fundamental bonding motives of key importance in molecular recognition have been paid much attention recently[17—21]. Most of these cation-π systems were based on the interactions between alkali[22,23], alkali earth[24] or transition metal ions[25] and π systems, but only a few involved in the heavy metal ions[26,27]. Fages *et al.*[28] and Ishikawa *et al.*[29] reported sensors for the recognition of silver ions with cation-π interaction design principle, respectively. Yi *et al.*[30] predicted theoretically that an Hg^{2+}-Bz complex favors a $\pi_C(\eta^1)$ type of cation-π interaction. However, direct experimental evidence of the application of the Hg^{2+}-selective chemosensor based on the cation-π interaction mechanism is rather rare. 1,8-Naphthalimide is a frequently used fluorophore, due to its high photostability, large Stokes shift and high quantum yield[31—33]. Herein

two sulfur-containing 4-aminonaphthalimide derivatives as fluorescent chemosensors were investigated for Hg^{2+} detection based on cation-π interaction.

2   Experimental

2.1   Materials and Measurements

All the chemicals and solvents used in the synthesis process were of analytical grade and used without further purification. The solvents for the spectrum analysis were further purified by standard methods. Melting points were measured on a Yanagimoto MP-500 apparatus and uncorrected. FTIR spectra were taken on a BIO-RAD FTS 3000 infrared spectrometer. UV spectra were recorded on an HP 8453 UV-visible spectrophotometer and fluorescent spectra were obtained on a Varian Cary Eclipse spectrometer. ^{1}H and ^{13}C NMR spectra were recorded on a Varian Oxford 500(500 MHz) instrument and the elemental analyses were carried out with a Perkin- Elmer 240C elemental analyzer.

2.2   Preparation of *N*-Butyl-4-bis(2-chloroethyl) Amino-1,8-naphthalimide(5)

As the method of the literature[34], 1.6 g(4.5 mmol) of *N*-butyl-4-bis(2-hydroxyethyl) amino-1,8-naphthalimide(4) and 4 g(26 mmol) of POCl$_3$ were refluxed in benzene for 3 h. After the removal of the solvent, the mixture was dissolved in CH$_2$Cl$_2$ and washed with NH$_3$ solution. After the removal of CH$_2$Cl$_2$, compound **5** was obtained as a light-green crystal by column chromatography(CH$_2$Cl$_2$, R_f=0.5). Yield 73.5%, m. p. 99—100 °C. IR(KBr), $\tilde{v}$/cm^{-1}: 2960, 1693, 1655, 1592, 1366; ^{1}H NMR(CDCl$_3$), δ: 0.98(t, J=7.5 Hz, 3H), 1.45(m, 2H),

---

1.73(m, 2H), 3.59(t, J=6.5 Hz, 4H), 3.80(t, J=6.5 Hz, 4H), 4.18(t, J=7.5 Hz, 2H), 7.46(d, J=8.0 Hz, 1H), 7.75(t, J=7.5 Hz, 1H), 8.55(t, J=8.5 Hz, 2H), 8.61(d, J=7.5 Hz, 1H); ^{13}C NMR(CDCl$_3$), δ: 14.08, 20.61, 30.47, 40.39, 41.55, 55.99, 118.79, 119.92, 123.60, 126.65, 128.36, 130.27, 130.44, 131.74, 131.81, 164.00, 164.50. Elemental anal.(%). calcd. for C$_{20}$H$_{22}$Cl$_2$N$_2$O$_2$: C 61.08, H 5.64, Cl 18.03, N 7.12; found: C 61.22, H 5.79, Cl 18.11, N 6.90.

## 2.3   Preparation of *N*-Butyl-4-bis(2-ethylsulfanyl-ethyl) Amino-1,8-naphthalimide(1)

As the method of the literature[35], 0.37 g(6 mmol) of ethanethiol and 0.24 g(6 mmol) of NaOH were stirred in 30 mL of ethanol at room temperature under the protection of nitrogen atmosphere for 1 h and a clear solution was obtained. Then 30 mL of tetrahydrofuran(THF) solution containing 1.0 g(2.54 mmol) of compound **5** was slowly added to the above mixture under stirring. After another 4 h of stirring and 1 h of refluxing, the solvent was removed and the residue was dissolved in CH$_2$Cl$_2$ and washed with water. After the removal of CH$_2$Cl$_2$, product **1** was obtained as a yellow oil by column chromatography(cyclohexane/EtOAc, gradient elution). Yield: 79.7%. IR(KBr), $\tilde{v}$/cm^{-1}: 2962, 1687, 1646, 1584, 1351; ^{1}H NMR (DMSO-d$_6$), δ: 0.88(t, J=7.5 Hz, 3H), 1.06(t, J=7.5 Hz, 6H), 1.30(m, 2H), 1.55(m, 2H), 2.42(q, J=7.5 Hz, 4H), 2.69(t, J=7.5 Hz, 4H), 3.56(t, J=7.5 Hz, 4H), 3.97(t, J=7.5 Hz, 2H), 7.41(d, J=8.0 Hz, 1H), 7.75(dd, J=8.5, 7.0 Hz, 1H), 8.33(d, J=8.0 Hz, 1H), 8.41(dd, J=7.5, 1.0 Hz, 1H), 8.46(dd, J=8.5, 1.0 Hz, 1H); ^{13}C NMR(DMSO-d$_6$), δ: 14.39, 15.40, 20.48, 25.74, 29.03, 30.39, 53.63, 116.13, 118.27, 123.22, 126.42, 127.11, 130.08, 131.34, 131.37, 132.26, 154.26, 163.61, 164.16. Elemental anal.(%) calcd. for C$_{24}$H$_{32}$N$_2$O$_2$S$_2$: C 64.83, H 7.25, N 6.30, S 14.42; found: C 64.95, H 7.11, N 6.51, S 14.33.

2.4   Preparation of *N*-Butyl-4-bis(2-diethylthio-carbamoylsulfanylethyl) Amino-1,8-naphthalimide (2)

Compound **5** of 1.0 g(2.54 mmol) and 1.71 g(7.6 mmol) of sodium diethyldithiocarbamate were stirred in 60 mL of aceto-nitrile at room temperature under the protection of nitrogen atmosphere for 24 h. After the removal of the solvent, the residue was dissolved in CH$_2$Cl$_2$ and washed with water. Product **2**(light-yellow oil) was obtained by column chromatography (cyclohexane/CH$_2$Cl$_2$, gradient elution). Yield: 82.7%. IR(KBr), $\tilde{v}$/cm^{-1}: 2958, 2933, 1695, 1649, 1355; ^{1}H NMR (DMSO-d$_6$), δ: 0.90(t, J=7.5 Hz, 3H), 1.05—1.11(m, 12H), 1.32(m, 2H), 1.57(m, 2H), 3.47(t, J=7.0 Hz, 4H), 3.62(q, J=7.0 Hz, 4H), 3.71(t, J=7.0 Hz, 4H), 3.86(q, J=7.0 Hz, 4H), 4.00(t, J=7.5 Hz, 2H), 7.56(d, J=8.0 Hz, 1H), 7.73(dd, J=8.5, 7.5 Hz, 1H), 8.34(d, J=8.0 Hz, 1H), 8.42(dd, J=7.5, 1.0 Hz, 1H), 8.51(dd, J=8.5, 1.0 Hz, 1H); ^{13}C NMR(DMSO-d$_6$), δ: 11.93, 12.93, 14.41, 20.49, 27.03, 30.42, 34.28, 47.14, 49.75, 52.33, 116.34, 118.83, 123.12, 126.47, 127.21, 130.04, 131.34, 131.55, 132.19, 154.26, 163.67, 164.23, 193.99. Elemental anal.(%) calcd. for C$_{30}$H$_{42}$N$_4$O$_2$S$_4$: C 58.22, H 6.84, N 9.05, S 20.72; found: C 58.00, H 6.66, N 9.41, S 20.63.

3   Results and Discussion

The syntheses of the two sulfur-containing 4-aminonaph-thalimide sensors **1** and **2** were shown in Scheme 1.
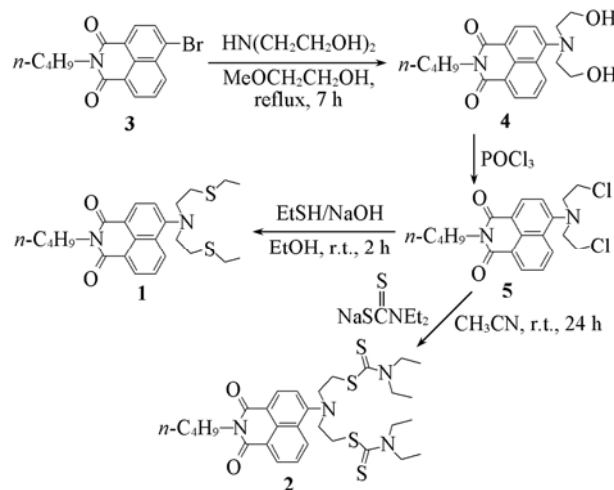
Scheme 1   Syntheses of sensors 1 and 2

In both sensors, 4-aminonaphthalimide was chosen as a fluorophore. A pair of sulfur-containing groups, *i.e.*, 2-ethylsulfanyl for compound **1** and dithiocarbamate(DTC) for compound **2**, respectively, were introduced to 4-aminona-phthalimide fluorophore *via* the —CH$_2$CH$_2$— linker and used as metal ion chelating groups. The fluorescence intensity of sensor **1** was reported to be enhanced by Hg^{2+} in the ethanol/water system[36]. In our case of CH$_3$CN system, fluorescence quenching(FQ) was observed during the sensing of Hg^{2+}, but fluorescence enhancement(FE) for the sensing of Cu^{2+}. For sensor **2**, similar results were observed for Fe^{3+} and Hg^{2+}. This implied that the recognition of transition and heavy metal ions may undergo different mechanisms.

Ions of Na$^+$, K$^+$, Mg^{2+}, Ca^{2+}, Zn^{2+}, Cu^{2+}, Ni^{2+}, Fe^{3+}, Co^{2+}, Mn^{3+}, Ag$^+$, Hg^{2+} and Pb^{2+} were used to evaluate the binding properties between metal ion and sensor **1** in acetonitrile. As shown in Fig.1, the fluorescence spectrum of sensor **1** shows a strong emission attributed to the characteristic emission of the 4-aminonaphthalimide moiety. Pronounced FE and blue-shift were observed upon the addition of Cu^{2+}. This can be explained
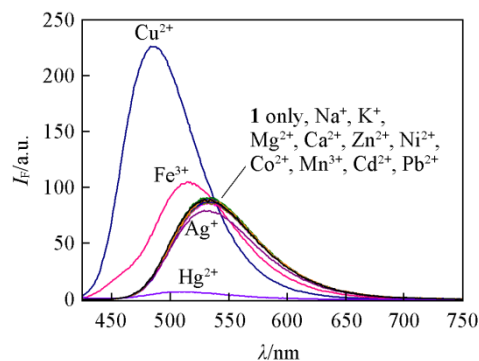

Fig.1   Fluorescence spectra of free sensor 1 and sen-sor 1 in the presence of various metal ions with concentration of 2×10^{-4} mol/L in acetonitrile
[**1**]=1×10^{-5} mol/L; λ_{ex}=415 nm. Bandwidth: E_x=2.5 nm, E_m=5 nm.

by the co-effect of PET and ICT processes origined from the coordination of Cu^{2+} with one N and two S atoms in the receptor cavity. The coordination of Cu^{2+} with N atom precludes the PET process which increases the fluorescence intensity of sensor **1**, and inhibits the ICT process, which contributes to the blue-shift of fluorescence of sensor **1**. In the presence of water as reported in the literature[36], Cu^{2+} will coordinate with H_2O and shows no influence on sensor **1**. It is of special interest that sensor **1** exhibits distinctive fluorescence quenching upon the addition of Hg^{2+}(92%, quenching rate) in CH_3CN, which suggests that the sensing of Hg^{2+} may undergo a different process from that of Cu^{2+}.

This FQ effect of Hg^{2+} may origin from the cation-π interactions between Hg^{2+} and the naphthalene moiety. Compared to the coordination of Cu^{2+} with the receptor, Hg^{2+} adopts a different complexation manner. Due to the softness and larger volume, Hg^{2+} is unable to be embedded in the receptor cavity but suspends over the naphthalene moiety with two S atoms coordinated(Fig.2). The eight-membered ring formed by Hg^{2+} and the receptor leans to one side of the fluorophore to form a chair-like conformation, which fixes Hg^{2+} above the naphthalene ring. The softness and location of Hg^{2+} facilitate the formation of the cation-π interaction, which quenches the fluorescence of sensor **1**. While in the presence of water, this kind of cation-π interaction may be destroyed due to the coordination of metal ions with water, which results in a reasonable fluorescence enhancement[36].
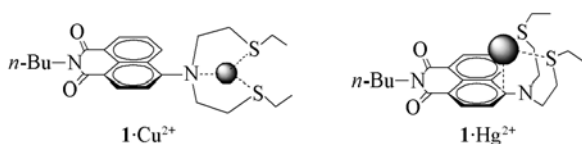

Fig.2 Sketch of the interaction of metal ions with sensor 1 in acetonitrile

In order to enhance the FQ selectivity for Hg^{2+}, sensor **2** with a pair of DTC as receptor was designed. The introduction of DTC enlarges the cryptand of receptors. The fluorescence titration of sensor **2** demonstrated our hypothesis. Fig.3 shows that sensor **2** recognitize Fe^{3+} with a pronounced FE effect which can be easily attributed to the suppression of the PET process in which the N atom in the receptor cavity participates the coordination. Besides Fe^{3+}, some other metal ions, such as
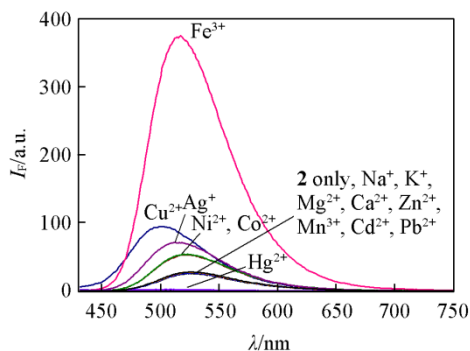

Fig.3 Fluorescence spectra of free sensor 2 and sensor 2 in the presence of various metal ions with concentration of 2×10^{-4} mol/L in acetonitrile

[**2**]=1×10^{-5} mol/L; λ_{ex}=420 nm. Bandwidth E_x=2.5 nm, E_m=5 nm.

Cu^{2+}, Ag^+, Ni^{2+} and Co^{2+} also show observable FE due to the same coordination manner. A remarkable FQ effect of Hg^{2+}(93%, quenching rate) is also observed which further demonstrates the existence of the cation-π interaction.

To further validate the existence of the cation-π interaction, the molecular geometries of a model of compound **6** and a complex of compound **6** with Hg^{2+} were optimized by the restricted Becke3LYP(B3LYP) functional[37,38]. The 6-311+G* basis set was employed for C, H, N, O and S atoms, and the effective core potentials(ECPs) of Hay and Wadt with a double-ξ basis set(LanL2DZ)[39—41] were used to describe the Hg atom. All the calculations were performed with the Gaussian 03 software package[42].

The optimized structures of compound **6** and complex **6**·Hg^{2+} are shown in Fig.4. Complex **6**·Hg^{2+} favors an asymmetric $\pi_C(\eta^2)$ or $\pi_C(\eta^1)$ type of cation-π interaction, which is similar as that reported by Yi et al.[30]. In complex **6**·Hg^{2+}, the two shortest bond distances of Hg—C1 and Hg—C2 are 0.2614 and 0.2986 nm, while bond distances of Hg—S1 and Hg—S1 are 0.2675 and 0.2643 nm, respectively. The natural bond orbital (NBD) charge in compound **6** and complex **6**·Hg^{2+} were also calculated and the net charge of the p-formylphenyl moiety in compound **6** is +0.08, while in complex **6**·Hg^{2+}, it increases to +0.17, which also suggests that the electron flows from the benzene ring to Hg^{2+} in complex **6**·Hg^{2+}.
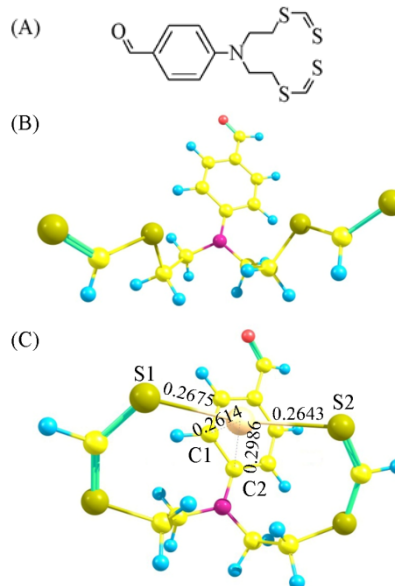

Fig.4 Structure of model compound 6(A) and the optimized structures of compound 6(B) and complex 6·Hg^{2+}(C)

The selective and sensitive signal responses to both emission and absorption of sensors **1** and **2** toward Hg^{2+} were investigated. With the gradual addition of mercury ion, the emission intensities of both sensor **1** and sensor **2** obviously decrease. For example, in Fig.5, the characteristic emission band of sensor **2** at 525 nm gradually decreases upon the addition of Hg^{2+}. Its fluorescence intensity decreases linearly upon the addition of Hg^{2+} with the concentration increasing from 0 to 1.0×10^{-5} moL/L(linearly dependent coefficient: R^2=0.9973) until the molar ratio of sensor **2** to Hg^{2+} is 1:1, then the fluorescence intensity keeps constant. The associated constant(K_a) estimated

by the nonlinear curve fitting[43] was found to be 5.2×10^5 L/mol. The complexation of sensor **2** with Hg^{2+} results in a remarkable absorption change of sensor **2** upon the addition of Hg^{2+} with increasing amounts(Fig.6). Notably, upon the addition of Hg^{2+}, the characteristic absorption band at 415 nm decreases, and a new band centered at 350 nm appears, corresponding to a blue-shift of $\lambda_{max(abs)}$ of 65 nm and an isobestic point at 393 nm, which indicates the formation of a complex $\mathbf{2}\cdot Hg^{2+}$ with 1:1 binding stoichiometry. At the same time, an apparent color fading from yellow-green to colorless can be observed by 'naked eye'(Fig.7).
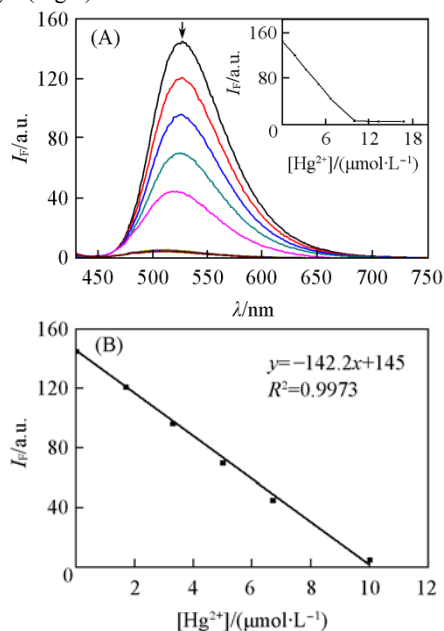

Fig.5 **Fluorescence spectral changes of senor 2 (1.0×10⁻⁵ mol/L) upon the addition of Hg²⁺ in acetonitrile(A) and fluorescence intensity at $\lambda_{max(em)}$ as a function of Hg²⁺ concentration(B)**

Inset of (A): fluorescence intensity at $\lambda_{max(em)}$ as a function of Hg^{2+} concentration. $\lambda_{ex}=420$ nm; bandwidth: $E_x=5$ nm, $E_m=5$ nm.
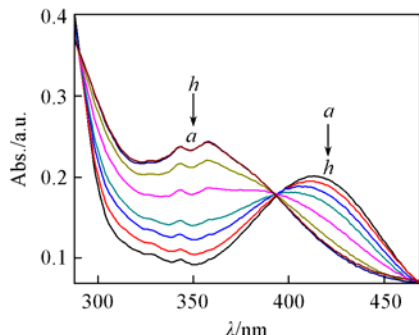

Fig.6 **Absorption spectral changes of sensor 2 (1.0×10⁻⁵ mol/L) upon the addition of Hg²⁺ in acetonitrile**

$[Hg^{2+}]/(\mu mol\cdot L^{-1})$ from *a* to *h*: 0, 1, 1.7, 2.5, 5.0, 6.7, 10.0, 13.3.

To further explore the utility of sensor **2** as an ion-selective fluorescence chemosensor for Hg^{2+}, the competition experiments were conducted in the presence of Hg^{2+} with a concentration of 1.0×10^{-5} mol/L mixed with Na^+, K^+, Mg^{2+},

Ca^{2+}, Zn^{2+}, Cu^{2+}, Ni^{2+}, Fe^{3+}, Co^{2+}, Mn^{3+}, Ag^+ and Pb^{2+} with a concentration of 3.0×10^{-5} mol/L, as well as the mixture of the metal ions, respectively. The results are depicted in Fig.8. No significant variation in the quenching of fluorescence intensity can be observed by comparison with that of Hg^{2+}. Moreover, no evident interference can be observed in the fluorescence titrations with Hg^{2+} in different mixtures of metal ions(5.0×10^{-5} mol/L). The above results suggest that the selectivity of sensor **2** for Hg^{2+} is remarkable.
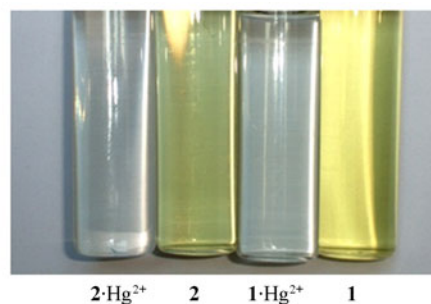

Fig.7 **Color response of senor 1 and sensor 2(1.0×10⁻⁵ mol/L) to Hg²⁺(1.0×10⁻⁵ mol/L) in acetonitrile**

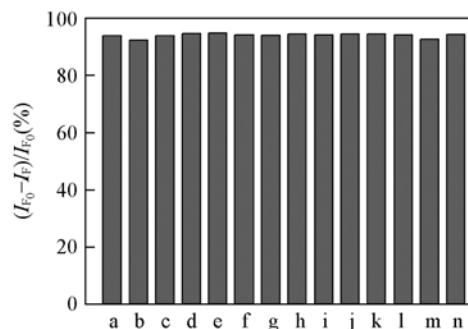**Fig.8** **Integrated fluorescence response of the solution containing sensor 2(1×10⁻⁵ mol/L) and Hg²⁺(1×10⁻⁵ mol/L) to various metal ions (3× 10⁻⁵ mol/L)**

a. Free of metal ion; b. Na^+; c. K^+; d. Mg^{2+}; e. Ca^{2+}; f. Zn^{2+}; g. Cu^{2+}; h. Ni^{2+}; i. Fe^{3+}; j. Co^{2+}; k. Mn^{2+}; l. Cd^{2+}; m. Ag^+; n. Pb^{2+}. I_F represents integrated fluorescence intensity of the solution upon the addition of the other metal ions, I_{F_0} represents integrated fluorescence intensity of the solution containing sensor **2** and Hg^{2+}.

4 Conclusions

In summary, selective chromogenic chemosensors for Hg^{2+} in CH_3CN were developed based on the principle of cation-π interactions. The probes with S ligands appended to the 4-aminonaphthalimide subunits exhibited a prominent Hg^{2+}-selective FQ effect over other metal ions and its selectivity for Hg^{2+} was not affected under the competition condition with other metal ions. Such a novel design strategy would be of great interest in the development of other chemosensors for the detection of hazardous heavy metal ions.

References

[1] De Silva A. P., Gunaratne. H. N., Gunnlaugsson T., Huxley A. J., McCoy C. P., Rademacher J. T., Rice T. E., *Chem. Rev.*, **1997**, *97*(5), 1515

[2] Boening D. W., *Chemosphere*, **2000**, *40*(12), 1335

[3] Benoit J. M., Fitzgerald W. F., Damman A. W. H., *Environ. Res.*, **1998**, *78*(2), 118

[4] He S., Liu Q., Li Y., Wei F., Cai S., Lu Y., Zeng X., *Chem. Res. Chinese Universities*, **2014**, *30*(1)*,* 32

[5] Su W. Q., Yang B. Q., *Chem. Res. Chinese Universities*, **2013**, *29*(4)，657

[6] Caballero A., Martínez R., Lloveras V., Ratera I., Vidal-Gancedo J., Wurst K., Tárraga A., Molina P., Veciana J., *J. Am. Chem. Soc.*, **2005**, *127*(45), 15666

[7] Coskun A., Akkaya E. U., *J. Am. Chem. Soc.*, **2006**, *128*(45), 14474

[8] Avirah R. R., Jyothish K., Ramaiah D., *Org. Lett.*, **2007**, *9*(1), 121

[9] Kim S. H., Kim J. S., Park S. M., Chang S. K., *Org. Lett.*, **2006**, *8*(3), 371

[10] Choi M. J., Kim M. Y., Chang S. K., *Chem. Commun.*, **2001**, *17*, 1664

[11] Youn N. J., Chang S. K., *Tetrahedron Lett.*, **2005**, *46*(1), 125

[12] Moon S. Y., Youn N. J., Park S. M., Chang S. K., *J. Org. Chem.*, **2005**, *70*(6), 2394

[13] Cheng Y. F., Zhao D. T., Zhang M., Liu Z. Q., Zhou Y. F., Shu T. M., Li F. Y., Yi T., Huang C. H., *Tetrahedron Lett.*, **2006**, *47*(36), 6413

[14] Park S. M., Kim M. H., Choe J. I., No K. T., Chang S. K., *J. Org. Chem.*, **2007**, *72*(9), 3550

[15] Lee M. H., Wu J. S., Lee J. W., Jung J. H., Kim J. S., *Org. Lett.,* **2007**, *9*(13)*,* 2501

[16] Wu J. S., Hwang I. C., Kim K. S., Kim J. S., *Org. Lett.*, **2007**, *9*(5), 907

[17] Suelter C., *Science,* **1970**, *168*(3933)*,* 789

[18] McRee D. E., *Nat. Struct. Biol.*, **1998**, *5*, 8

[19] Agranoff D. D., Krishna S., *Mol. Microbiol.*, **1998**, *28*(3), 403

[20] Pyle A., *J. Biol. Inorg. Chem.,* **2002**, *7*(7/8), 679

[21] Masson E., Schlosser M., *Org. Lett.*, **2005**, *7*(10), 1923

[22] Murayama K., Aoki K., *Inorg. Chim. Acta*, **1998**, *281*(1), 36

[23] De Wall S. L., Meadows E. S., Barbour L. J., Gokel G. W., *J. Am. Chem. Soc.*, **1999**, *121*(23)*,* 5613

[24] Reddy A. S., Zipse H., Sastry G. N., *J. Phys. Chem. B*, **2007**, *111*(39), 11546

[25] Zarić S. D., *Chem. Phys. Lett.*, **1999**, *311*(1), 77

[26] Lau W., Huffman J. C., Kochi J. K., *J. Am. Chem. Soc.*, **1982**, *104*(20), 5515

[27] Lau W., Kochi J., *J. Org. Chem.*, **1986**, *51*(10), 1801

[28] Fages F., Desvergne J. P., Bouas-Laurent H., Marsau P., Lehn J. M., Kotzyba-Hibert F., Albrecht-Gary A. M., Al-Joubbeh M., *J. Am. Chem. Soc.*, **1989**, *111*(23), 8672

[29] Ishikawa J., Sakamoto H., Nakao S., Wada H., *J. Org. Chem.*, **1999**, *64*(6), 1913

[30] Yi H. B., Lee H. M., Kim K. S., *J. Chem. Theory Comput.*, **2009**, *5*(6), 1709

[31] Lee S., Lee J. H., Pradhan T., Lim C. S., Cho B. R., Bhuniya S., Kim S., Kim J. S., *Sens. Actuator B*, **2011**, *160*(1), 1489

[32] Xu S., Li W., Chen K. C., *Chin. J. Chem.*, **2007**, *25*(6), 778

[33] Guo X. F., Zhu B. C., Liu Y. Y., Zhang Y., Jia L. H., Qian X. H., *Chin. J. Org. Chem.*, **2006**, *26*(4), 504

[34] Springer C. J., Dowell R., Burke P. J., Hadley E., Davies D. H., Blakey D. C., Melton R. G., Niculescu-Duvaz I., *J. Med. Chem.*, **1995**, *38*(26), 5051

[35] Tanaka M., Nakamura M., Ikeda T., Ikeda K., Ando H., Shibutani Y., Yajima S., Kimura K., *J. Org. Chem.*, **2001**, *66*(21), 7008

[36] Zhang Z. Y., Chen Y. H., Xu D. M., Yang L., Liu A. F., *Spectroc. Acta Pt. A*, *Molec. Biomolec. Spectr.*, **2013**, *105*, 8

[37] Miehlich B., Savin A., Stoll H., Preuss H., *Chem. Phys. Lett.*, **1989**, *157*(3), 200

[38] Lee C., Yang W., Parr R. G., *Phys. Rev. B*, **1988**, *37*(2), 785

[39] Hay P. J., Wadt W. R., *J. Chem. Phys.*, **1985**, *82*(1), 270

[40] Hay P. J., Wadt W. R., *J. Chem. Phys.*, **1985**, *82*(1), 299

[41] Hay P. J., Wadt W. R., *J. Chem. Phys.*, **1985**, *82*(1), 284

[42] Frisch M. J., Trucks G. W., Schlegel H. B., Scuseria G. E., Robb M. A., Cheeseman J. R., Montgomery J. A. Jr., Vreven T., Kudin K. N. B. J. C., Millam J. M., Iyengar S. S., Tomasi J., Barone V. M. B., Cossi M., Scalmani G., Rega N., Petersson G. A. N. H., Hada M., Ehara M., Toyota K., Fukuda R., Hasegawa J. I. M., Nakajima T., Honda Y., Kitao O., Nakai H., Klene M., Li X., Knox J. E., Hratchian H. P., Cross J. B., Bakken V., Adamo C. J. J., Gomperts R., Stratmann R. E., Yazyev O., Austin A. J. C. R., Pomelli C., Ochterski J. W., Ayala P. Y., Morokuma K. V. G. A., Salvador P., Dannenberg J. J., Zakrzewski V. G., Dapprich S., Daniels A. D., Strain M. C., Farkas O., Malick D. K., Rabuck A. D. R. K., Foresman J. B.,Ortiz J. V., Cui Q., Baboul A. G. C. S., Cioslowski J., Stefanov B. B., Liu G., Liashenko A., Piskorz P., Komaromi I., Martin R. L., Fox D. J., Keith T., Al-Laham M. A. P. C. Y., Nanayakkara A., Challacombe M., Gill P. M. W., Johnson B., Chen W., Wong M. W., Gonzalez C., Pople J. A., *Gaussian 03*, Walligford, CT, **2004**

[43] Kuzmič P., *Anal. Biochem.*, **1996**, *237*(2), 260