



Naphthalimide-based fluorescent Zn²⁺ chemosensors showing PET effect according to their linker length in water

Soon Young Kim, Jong-In Hong*

Department of Chemistry, College of Natural Sciences, Seoul National University, Seoul 151-747, Republic of Korea

ARTICLE INFO

Article history:

Received 13 February 2009

Revised 24 March 2009

Accepted 26 March 2009

Available online 28 March 2009

ABSTRACT

We have developed naphthalimide-based fluorescent chemosensors that exhibit fluorescence enhancement upon binding Zn²⁺ ion in 10 mM HEPES buffer (pH 7.4) at 25 °C. The fluorescence enhancement was induced by a PET inhibition process in which electron transfer from the nitrogen lone pair electrons of the Dpa unit to naphthalimide was blocked upon the binding of the sensor to Zn²⁺. The longer the linker length ($n = 1-3$) of the sensor, the less the PET efficiency becomes. Among the sensors (**1**, **2**, and **3**) examined, **1** shows the highest selectivity and sensitivity for Zn²⁺ over other transition metal ions and alkali metal ions in water.

© 2009 Elsevier Ltd. All rights reserved.

1,8-Naphthalimide has high photostability, a large Stokes shift, and strong fluorescence, and therefore has a range of applications in the fields of polymers,¹ optical storage,² photophysical dyads,³ nucleic acids intercalators,⁴ and DNA photocleavage.⁵ Due to its favorable characteristics and numerous applications, naphthalimide-based fluorescent chemosensors have been developed by several research groups.⁶⁻⁹ Most of these chemosensors, which were developed for the detection of transition metal ions, were constructed by introducing the binding site at 4-^{6a,c,d} or 4,5-positions^{6b} of naphthalimide. In addition, most of them operate in an organic^{6e,8} or organic/water environment.^{6a,b,7,9}

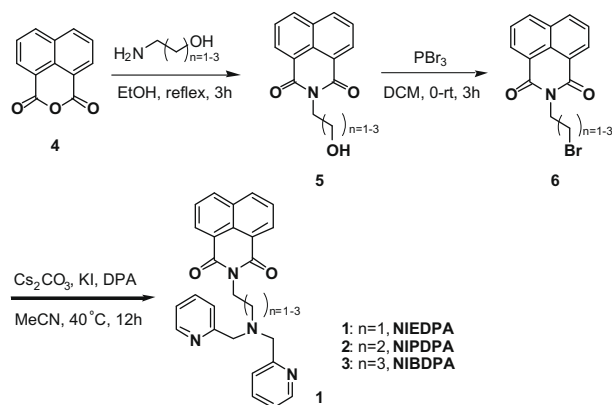
A few fluorescent chemosensors based on naphthalimide have been reported concerning pH sensing⁷ or the trend of PET efficiency according to the linker length between the fluorophore and the metal ion binding moiety.⁸ However, there has been no report on naphthalimide-based fluorescent Zn²⁺ selective sensors that are able to control the PET effect in 100% aqueous solution according to the distance between the donor and acceptor.⁹

Zn²⁺ plays diverse roles in medicinal, chemical, and biological events. In the human body, protein scaffolds contain Zn²⁺ ions (e.g., carbonic anhydrase, zinc finger protein),¹⁰ which play crucial roles in neurotransmission,¹¹ metalloenzymes, and gene transcription.^{10b} Also, Zn²⁺ induces apoptosis,¹² and the formation of β -amyloid¹³ which is important in the development of Alzheimer's disease. Due to the importance of Zn²⁺ ion in numerous biological systems, there is a great emphasis placed on the development of fluorogenic chemosensors for Zn²⁺ in water.¹⁴

Herein, we report fluorescent chemosensors (**1-3**) for Zn²⁺ which show different PET effects according to the length of the linker existing between the imide N and dipicolylamine (Dpa). The

synthesis of **1-3** is described in Scheme 1. Compounds **5** and **6** ($n = 1-3$) were synthesized according to the reported methods.¹⁵ Finally, **1**, **2**, and **3** were prepared by the S_N2 reaction of **6** with dipicolylamine in the presence of Cs₂CO₃ and KI in MeCN at 40 °C.¹⁶

First, the fluorescence emission changes ($\lambda_{\text{ex}} = 335$ nm, $\lambda_{\text{em}} = 394$ nm) of NIEDPA (**1**, 5 μM) according to the Zn²⁺ (perchlorate salt) concentration were measured in 10 mM HEPES buffer (pH 7.4) at 25 °C (Fig. 1). Upon the addition of Zn²⁺ ions, the fluorescence emission intensity of **1** gradually increased. When 1 equiv of Zn²⁺ ions was added to **1**, the emission intensity ratio (I/I_0) and FE value¹⁷ of **1** showed 5.4- and 5.0-fold enhancements, respectively (SI). The association constant (K_a) between **1** and Zn²⁺ was estimated to be $1.22 \times 10^6 \text{ M}^{-1}$ by fluorescence titration curve fitting (SigmaPlot Program 2002 by Windows Version 8.0).¹⁸ The job



Scheme 1. Synthesis of chemosensors **1-3**.

* Corresponding author. Tel.: +82 28806682; fax: +82 28891568.
E-mail address: jihong@snu.ac.kr (J.-I. Hong).

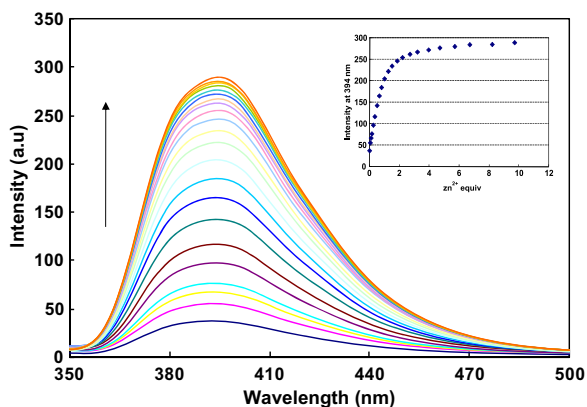


Figure 1. The fluorescence emission changes of **1** (5 μM) upon the addition of Zn^{2+} (perchlorate salt) in 10 mM HEPES buffer (pH 7.4) at 25 $^{\circ}\text{C}$: $[\text{Zn}^{2+}] = 0, 0.25, 0.50, 0.74, 1.22, 1.69, 2.38, 3.05, 3.70, 4.55, 5.56, 6.52, 7.81, 9.01, 10.6, 12.1, 14.2, 16.0, 18.2, 20.1, 22.5, 24.6 \mu\text{M}$. (Inset: Changes in the fluorescence intensity of **1** at 394 nm with the addition of Zn^{2+}).

plot showed 1:1 stoichiometry between **1** and Zn^{2+} in 10 mM HEPES buffer at 25 $^{\circ}\text{C}$ (Fig. S3).

However, upon the addition of 1 equiv of Cd^{2+} into **1**, the fluorescence emission ($\lambda_{\text{em}} = 394 \text{ nm}$) intensity ratio (I/I_0) and FE value of **1** showed 2.7- and 2.6-fold increases, respectively (Tables S1 and S2). The association constant (K_a) of **1** for Cd^{2+} was measured to be $7.21 \times 10^4 \text{ M}^{-1}$.¹⁷ Other transition metal ions such as Fe^{2+} , Ag^+ , Mn^{2+} , Hg^{2+} , Cu^{2+} , Co^{2+} , and Ni^{2+} , barely showed any emission increase after the addition of 1 equiv of each metal ion (Fig. 2).

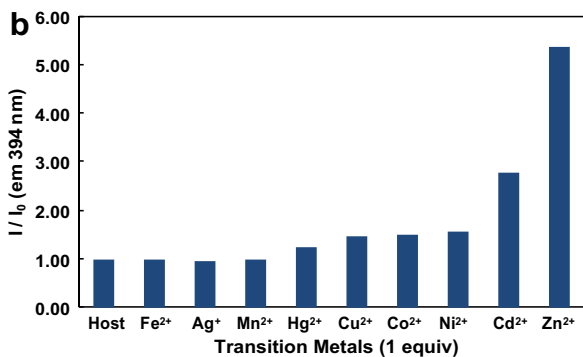
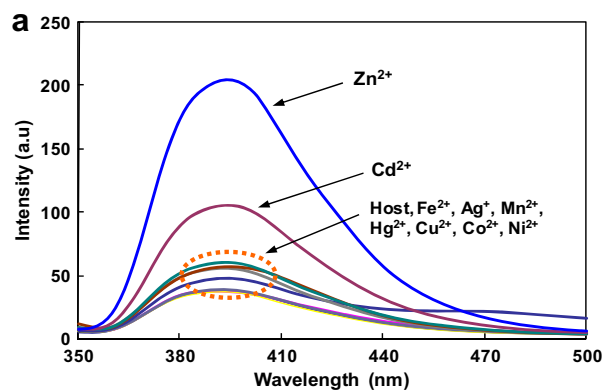


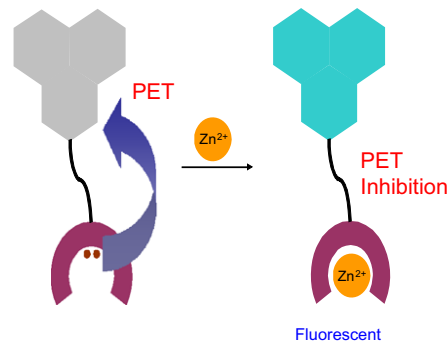
Figure 2. (a) The fluorescence emission spectra of **1** (5 μM) after the addition of 1 equiv of each transition metal ion (perchlorate salt) in 10 mM HEPES buffer (pH 7.4) at 25 $^{\circ}\text{C}$. (b) The comparison of the fluorescence emission intensity (I) at 394 nm of **1** after the addition of 1 equiv of each transition metal ion with the emission intensity (I_0) at 394 nm of **1** (5 μM) before the addition of each metal ion.

Upon the addition of 1 equiv of each transition metal ion, the fluorescence emission changes of **NIPDPA** (**2**, 5 μM) were examined in 10 mM HEPES buffer solution (Fig. S1). After the addition of Zn^{2+} and Cd^{2+} , the fluorescence emission intensity of **2** increased less compared to that of **1**. When 1 equiv of Zn^{2+} ions was added to the solution of **2**, the fluorescence emission intensity ratio (I/I_0) and FE value of **2** showed 3.9- and 3.8-fold increases, respectively (Tables S1 and S2). Upon the addition of 1 equiv of Cd^{2+} , the fluorescence emission intensity ratio (I/I_0) and FE value of **2** exhibited 2.5- and 2.6-fold enhancements, respectively (Tables S1 and S2). As expected, the addition of 1 equiv of other transition metal ions rendered the fluorescence emission changes of **2** barely observable (Fig. S1).

A sensing mechanism for the fluorescence OFF–ON of chemosensors is shown in Scheme 2. When Zn^{2+} ions were absent in the host solution, the fluorescence emission of chemosensors was quenched by a PET (Photo-induced Electron Transfer) process, which takes place through electron transfer from the nitrogen lone pair (donor) of the Dpa moiety to the naphthalimide (acceptor).

By the strong coordination of Zn^{2+} to the Dpa unit,^{6c,9,14,19} the PET effect is blocked, and as a result, fluorescence revives (fluorescence–ON). The critical factor of the PET efficiency is the distance between the donor and acceptor. Therefore, the longer the linker length ($n = 1–3$), the less effective the PET becomes. As a result, the fluorescence emission efficiency due to PET is largest in **1** among the chemosensors **1–3** (Fig. 3).

The selectivity for Zn^{2+} versus alkali metal ions was also investigated. After the addition of 0–40 equiv of each Na^+ , K^+ , Mg^{2+} , and Ca^{2+} (perchlorate salts), the fluorescence emission intensity of **1**



Scheme 2. Sensing mechanism.

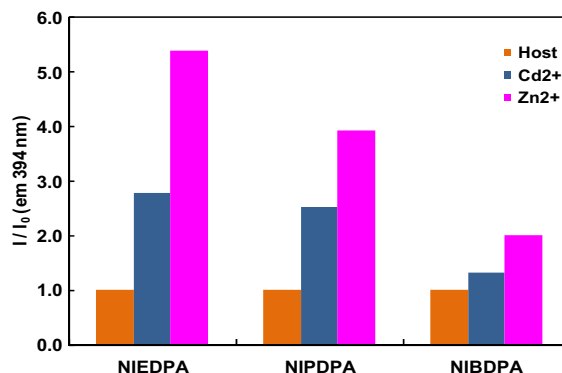


Figure 3. The comparison of the emission intensity (I) at 394 nm of **1–3** (5 μM) after the addition of 1 equiv of each Zn^{2+} and Cd^{2+} ion with the emission intensity (I_0) at 394 nm of **1–3** (5 μM) before the addition of each metal ion in 10 mM HEPES buffer (pH 7.4) at 25 $^{\circ}\text{C}$.

did not change. However, the fluorescence emission intensities of **1** displayed about twofold increase after the addition of 2 equiv of Zn^{2+} (perchlorate salt) and 40 equiv of each cation (Fig. S4). Therefore, **1** can selectively detect Zn^{2+} ion in the presence of excess alkali metal ions under physiological conditions. The selectivity for Zn^{2+} is due to the high affinity of Zn^{2+} for three nitrogen atoms of the Dpa unit.

In conclusion, we have developed naphthalimide-based fluorescent chemosensors **1–3** which exhibit fluorescence enhancement upon binding with Zn^{2+} ions in 10 mM HEPES buffer (pH 7.4) at 25 °C. Chemosensors **1–3** exhibited a different fluorescence emission response according to the length of the spacer between the donor (nitrogen lone pair electrons of the Dpa moiety) and acceptor (naphthalimide); the longer the linker length of chemosensors is, the less efficient the PET process becomes. As a result, **1** ($n = 1$) shows the highest PET efficiency, high selectivity, and sensitivity for Zn^{2+} over other transition metal ions and alkali metal ions in water.

Acknowledgments

This work was supported by the KRF (2008-312-C00206) and Seoul R&BD. S.Y.K. is grateful to the Ministry of Education for the award of the BK 21 fellowship.

Supplementary data

Supplementary data (synthesis, 1H NMR, ^{13}C NMR and HRMS data for chemosensors **1–3**, fluorescence emission spectra of **2**, **3** for various transition metal ions, I/I_0 ratios and FE values of **1–3**, competition experiments for excess alkali metal ions, Job's plot) associated with this article can be found, in the online version, at doi:10.1016/j.tetlet.2009.03.179.

References and notes

- Grabchev, I.; Petkov, C.; Bojinov, V. *Macromol. Mater. Eng.* **2002**, *287*, 904–908.
- Jiang, G.; Wang, S.; Yuan, W.; Jiang, L.; Song, Y.; Tian, H.; Zhu, D. *Chem. Mater.* **2006**, *18*, 235–237.
- Tasior, M.; Gryko, D. T.; Cembor, M.; Jaworski, J. S.; Ventura, B.; Flamigni, L. *New J. Chem.* **2007**, *31*, 247–259.
- Wamberg, M. C.; Warczak, K.; Andersen, L.; Hassan, A. A.; Pedersen, E. B. *Helv. Chim. Acta* **2006**, *89*, 1826–1839.
- Ryan, G. J.; Quinn, S.; Gunnlaugsson, T. *Inorg. Chem.* **2008**, *47*, 401–403.
- (a) Guo, X.; Qian, X.; Jia, L. *J. Am. Chem. Soc.* **2004**, *126*, 2272–2273; (b) Xu, Z.; Qian, X.; Cui, J. *Org. Lett.* **2005**, *7*, 3029–3032; (c) Fan, J.; Wu, Y.; Peng, X. *Chem. Lett.* **2004**, *33*, 1392–1393; (d) Wang, J.; Qian, X. *Chem. Commun.* **2006**, 109–111; (e) Chovelon, J.-M.; Grabchev, I. *Spectrochim. Acta Part A* **2007**, *67*, 87–91.
- (a) de Silva, A. P.; Nimal Gunaratne, H. Q.; Habib-Jiwan, J.-L.; McCoy, C. P.; Rice, T. E.; Soumillion, J.-P. *Angew. Chem., Int. Ed.* **1995**, *34*, 1728–1731; (b) Cui, D.; Qian, X.; Liu, F.; Zhang, R. *Org. Lett.* **2004**, *6*, 2757–2760; (c) de Silva, A. P.; Rice, T. E. *Chem. Commun.* **1999**, 163–164.
- (a) Ramachandram, B.; Saroja, G.; Sankaran, N.; Samanta, A. *J. Phys. Chem. B* **2000**, *104*, 11824–11832; (b) Wen, G.-T.; Zhu, M.-Z.; Wang, Z.; Meng, X.-M.; Hu, H.-Y.; Guo, Q.-X. *Chin. J. Chem.* **2006**, *24*, 1230–1237. In the above papers, the highest PET efficiency was observed for systems where the fluorophore and the metal ion binding moiety are separated by two methylene groups.
- Wang, J.; Xiao, Y.; Zhang, Z.; Qian, X.; Yang, Y.; Xu, Q. *J. Mater. Chem.* **2005**, *15*, 2836–2839.
- (a) Vallee, B. L.; Falchuk, K. H. *Physiol. Rev.* **1993**, *73*, 79–118; (b) Berg, J. M.; Shi, Y. *Science* **1996**, *271*, 1081–1085.
- Fredrickson, C. J. *Int. Rev. Neurobiol.* **1989**, *31*, 145–238.
- Truong-Tran, A. Q.; Ho, L. H.; Chai, F.; Zalewski, P. D. *J. Nutr.* **2000**, *130*, 1459S–1466S.
- Bush, A. I.; Pettingell, W. H.; Multhaup, G.; Paradis, M. D.; Vonsattel, J. P.; Gusella, J. F.; Beyreuther, K.; Masters, C. L.; Tanzi, R. E. *Science* **1994**, *265*, 1464–1467.
- (a) Maruyama, S.; Kikuchi, K.; Hirano, T.; Urano, Y.; Nagano, T. *J. Am. Chem. Soc.* **2002**, *124*, 10650–10651; (b) Kiyose, K.; Kojima, H.; Urano, Y.; Nagano, T. *J. Am. Chem. Soc.* **2006**, *128*, 6548–6549; (c) Woodroffe, C. C.; Lippard, S. J. *J. Am. Chem. Soc.* **2003**, *125*, 11458–11459; (d) Nolan, E. M.; Ryu, J. W.; Jaworski, J.; Feazell, R. P.; Sheng, M.; Lippard, S. J. *J. Am. Chem. Soc.* **2006**, *128*, 15517–15528.
- Hossain, S. U.; Sengupta, S.; Bhattacharya, S. *Bioorg. Med. Chem.* **2005**, *13*, 5750–5758.
- NIEDPA (1)** To a solution of compound **6** (300 mg, 0.99 mmol) in MeCN were added successively Cs_2CO_3 (355 mg, 1.09 mmol), KI (181 mg, 1.09 mmol), and di-2-picolyamine (217 mg, 1.09 mmol). The reaction mixture was stirred at 40 °C overnight, and then all the volatile components were evaporated. The residue was partitioned between CH_2Cl_2 and brine ($\times 2$). The combined organic phase was washed with water, and then dried in anhydrous Na_2SO_4 . Flash chromatographic purification (CH_2Cl_2 to $CH_2Cl_2/MeOH = 20:1$) yielded **1** (220 mg, 53% yield). 1H NMR (300 MHz, acetone- d_6): δ 2.89 (t, $J = 6$ Hz, 2H), 3.84 (s, 4H), 4.37 (t, $J = 6$ Hz, 2H), 7.02 (t, $J = 5.4$ Hz, 2H), 7.27 (t, $J = 7.3$ Hz, 2H), 7.35 (d, $J = 7.7$ Hz, 2H), 7.87 (t, $J = 7.7$ Hz, 2H), 8.33 (d, $J = 4.3$ Hz, 2H), 8.42–8.49 (m, 4H). ^{13}C NMR (75 MHz, acetone- d_6): δ 37.48, 51.53, 60.07, 121.61, 122.51, 122.90, 127.04, 128.01, 130.66, 131.84, 133.95, 135.70, 148.61, 159.86, 163.54. HRMS (FAB): m/e calcd for $C_{26}H_{22}N_4O_2$ [M+H] $^+$: 423.1821, found: 423.1821. **NIPDPA (2)** **2** was similarly prepared using the same procedure as used in the synthesis of **1** (56% yield). 1H NMR (300 MHz, acetone- d_6): δ 1.98 (t, $J = 7.0$ Hz, 2H), 2.67 (t, $J = 6.9$ Hz, 2H), 3.83 (s, 4H), 4.18 (t, $J = 7.6$ Hz, 2H), 7.13 (t, $J = 11.7$ Hz, 2H), 7.62–7.70 (m, 4H), 7.80 (t, $J = 7.8$ Hz, 2H), 8.34 (d, $J = 8.2$ Hz, 2H), 8.41 (d, $J = 4.7$ Hz, 2H), 8.46 (d, $J = 7.3$ Hz, 2H). ^{13}C NMR (75 MHz, acetone- d_6): δ 25.50, 38.32, 51.46, 59.87, 121.76, 122.72, 122.84, 126.95, 127.87, 130.56, 131.73, 133.89, 136.07, 148.89, 159.97, 163.51. HRMS (FAB): m/e calcd for $C_{26}H_{22}N_4O_2$ [M+H] $^+$: 423.1821, found: 423.1821. **NIBDPA (3)** **3** was similarly prepared using the same procedure as used in the synthesis of **1** (53% yield). 1H NMR (300 MHz, acetone- d_6): δ 1.67–1.79 (m, 4H), 2.06 (t, $J = 4.3$ Hz, 2H), 3.83 (s, 4H), 4.12 (t, $J = 7.1$ Hz, 2H), 7.17 (t, $J = 5.7$ Hz, 2H), 7.62–7.73 (m, 4H), 7.86 (t, $J = 7.8$ Hz, 2H), 8.39–8.45 (m, 4H), 8.53 (d, $J = 7.2$ Hz, 2H). ^{13}C NMR (75 MHz, acetone- d_6): δ 24.76, 25.71, 39.77, 53.92, 60.24, 121.79, 122.63, 122.69, 126.93, 127.79, 130.56, 131.65, 133.87, 136.19, 148.70, 160.11, 163.48. HRMS (FAB): m/e calcd for $C_{27}H_{24}N_4O_2$ [M+H] $^+$: 437.1978, found: 437.1972.
- Quantitatively, FE value is the factor of the final integrated fluorescence emission intensity over the initial integrated fluorescence emission intensity upon the addition of 1 equiv of transition metal ions.
- Binding Constants, The Measurement of Molecular Complex Stability*; Connors, K. A., Ed.; John Wiley and Sons: New York, 1987; pp 175–183.
- (a) Lee, D. H.; Im, J. H.; Son, S. U.; Chung, Y. K.; Hong, J.-I. *J. Am. Chem. Soc.* **2003**, *125*, 7752–7753; (b) Ojida, A.; Miko-oka, Y.; Sada, K.; Hamachi, I. *J. Am. Chem. Soc.* **2004**, *126*, 2454–2463.