Indium(III)-Induced Fluorescent Excimer Formation and Extinction in Calix[4]arene–Fluoroionophores

Sung Kuk Kim,[†] Su Ho Kim,[†] Hyun Jung Kim,[†] Seoung Ho Lee,[†] Soon W. Lee,[‡] Jaejung Ko,*,[§] Richard A. Bartsch,[||] and Jong Seung Kim*,[†]

Department of Chemistry, Dankook University, Seoul 140-714, Korea, Department of Chemistry, Sungkyunkwan University, Suwon 440-746, Korea, Department of Chemistry, Korea University, Chochiwon 339-700, Korea, and Department of Chemistry and Biochemistry, Texas Tech University, Lubbock, Texas 79409-1061

Received May 4, 2005

New fluorogenic or/and chromogenic calix[4]arenes **1**–**3** with two facing amide groups linked to fluorescent pyrene units are synthesized. Orientations of the pyrene units are remote from each other in **1** and face-to-face π-stacked in **2**, which produces different photophysical properties. In the excited state, the two pyrene units of **2** form a strong intramolecular excimer displaying an emission at 472 nm with a relatively weak monomer emission at 395 nm. In contrast, **1** exhibits only a monomer emission at 398 nm because intramolecular hydrogen bonding between the phenolic OH oxygens and the amide hydrogens prevents π-stacking of the two pyrene groups. Fluorescence changes upon addition of various metal ions show that **1** has a remarkably high selectivity for In^{3+} over the other metal ions tested. Compound **1** forms 2:1 (metal:ligand), as well as 1:1 complexes, with In^{3+}, with fluorescence varying uniquely with the complex stoichiometry. Compound **3**, which possess two pyrene units and two chromogenic azo groups, shows almost the same binding behavior toward metal ions as does **1**, together with additional bathochromic shifts of the absorption maximum. Compared with **1**, compound **3** emits a considerably weaker fluorescence, which is attributed to electron transfer from the pyrene units to the nitro groups of the phenylazo moieties.

Introduction

Design and synthesis of fluorescent sensors capable of selectively detecting chemical species including anions and cations[1–7] have attracted increasing attention due to several distinct advantages:[8,9] (1) high sensitivity and selectivity;[10–15] (2) "on–off" switchability, applicable for logic gates;[16–21] (3) convenient monitoring of molecular-level events through

* To whom correspondence should be addressed. Fax: +82-2-797-3277. E-mail: jongskim@dankook.ac.kr.
† Dankook University.
‡ Sungkyunkwan University.
§ Korea University.
|| Texas Tech University.

(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. (b) *Chemosensors of Ion and Molecule Recognition*; Desvergne, J.-P., Czarnik, A. W., Eds.; NATO ASI Series; Kluwer Academic: Dordrecht, The Netherlands, 1997; p 492. (c) Valeur, B.; Leray, I. *Coord. Chem. Rev.* **2000**, *205*, 3. (d) de Silva, A. P.; Fox, D. B.; Huxley, A. J. M.; Moody, T. S. *Coord. Chem. Rev.* **2000**, *205*, 41.
(2) (a) Zhang, X.-B.; Guo, C.-C.; Li, Z.-Z.; Shen, G.-L.; Yu, R.-Q. *Anal. Chem.* **2002**, *74*, 821. (b) Aragon, M. C.; et al. *Inorg. Chem.* **2002**, *41*, 6623. (c) Chen, C.-T.; Huang, W.-P. *J. Am. Chem. Soc.* **2002**, *124*, 6246. (d) Guo, X.; Qian, X.; Jia, L. *J. Am. Chem. Soc.* **2004**, *126*, 2272.
(3) (a) Hirano, T.; Kikuchi, K.; Urano, Y.; Higuchi, T.; Nagano, T. *Angew. Chem.*, *Int. Ed.* **2000**, *39*, 1052. (b) Burdette, S. C.; Walkup, G. K.; Spingler, B.; Tsien, R. Y.; Lippard, S. J. *J. Am. Chem. Soc.* **2001**, *123*, 7831.
(4) (a) Chen, C. T.; Huang, W. P. *J. Am. Chem. Soc.* **2002**, *123*, 6246. (b) Lee, S. H.; Kim, J. Y.; Kim, S. K.; Lee, J. H.; Kim, J. S. *Tetrahedron* **2004**, *60*, 5171.
(5) Fabbrizzi, L.; Poggi, A. *Chem. Soc. Rev.* **1995**, *24*, 197.
(6) Leray, I.; Lefevre, J.-P.; Delouis, J.-F.; Delaire, J.; Valeur, B. *Chem. Eur. J.* **2001**, *7*, 4590.
(7) Valeur, B.; Bardez, E. *Chem. Ber.* **1995**, *31*, 216.
(8) Bryan, A. J.; de Silva, A. P.; de Silva, S. A.; Rupasinghe, R. A. D. D.; Sandanayake, K. R. A. S. *Biosensors* **1989**, *4*, 169.
(9) Bissell, R. A.; de Silva, A. P.; Gunaratne, H. Q. N.; Lynch, P. L. M.; Maguire, G. E. M.; Sandanayake, K. R. A. S. *Chem. Soc. Rev.* **1992**, *21*, 187.
(10) Xie, X. S. *Acc. Chem. Res.* **1996**, *29*, 598.
(11) Goodwin, P. M.; Ambrose, W. P.; Keller, R. A. *Acc. Chem. Res.* **1996**, *29*, 607.
(12) Orrit, M.; Bernard, J. *Phys. Rev. Lett.* **1990**, *65*, 2716.
(13) Mets, U.; Rigler, R. *J. Fluoresc.* **1994**, *4*, 259.
(14) (a) Moerner, W. E.; Basche, T. *Angew. Chem.*, *Int. Ed. Engl.* **1993**, *32*, 457. (b) Moerner, W. E. *Acc. Chem. Res.* **1996**, *29*, 563.
(15) Yeung, E. S. *Acc. Chem. Res.* **1994**, *27*, 209.

light signals; and (4) sub-millisecond temporal resolution. Fluorescent chemosensors for cations typically consist of a cation recognition unit (ionophore) and a fluorogenic unit (fluorophore) linked by a spacer.[1c] The fluorophore (signaling moiety) makes human-molecule communication possible through a light signal resulting from its changes in photophysical characteristics, whereas the recognition unit (ionophore) linked to the fluorophore is responsible for the selectivity and cation binding efficiency of the entire chemosensor. Among fluorophores, pyrenes are one of the most useful fluorogenic units because they display not only a well-defined monomer emission at $370-430$ nm but also an efficient excimer emission at around 480 nm.[22,23] With an intensity ratio of excimer to monomer emission (I_E/I_M), which is sensitive to conformational changes of the pyrene-appended receptors, variation in I_E/I_M values upon metal-ion binding to the receptor can be an informative parameter in sensing systems.[1a,23,24]

Calixarenes functionalized with appropriate cation-ligating groups, such as carboxylic acid, amide, crown ether, and azacrown ether groups, are good candidates for cation recognition due to their high selectivity toward specific cations.[25-30] As one such ligand, we previously reported a Pb^{2+}-selective fluorescent calix[4]crown with two facing amide groups,[4b] in which the amide groups not only form a complex with the Pb^{2+} ion but also act as a PET (photoinduced electron-transfer) acceptor, causing the pyrene fluorescence to be quenched.[4b,31,32] For formation of a pyrene excimer, Reinhoudt et al.[33a] reported a Na^+-selective fluorescent calix[4]arene with four amide groups, where two pyrenes linked to amide groups form a strong intramolecular excimer through strong face-to-face π-stacking interaction. In this case, binding of Na^+ to the amide oxygen atoms of the ligand induces interdependent fluorescence changes of the excimer and monomer, which allows fluorescence ratiometric measurement. Jin et al.[33b] and Shinkai et al.[33c,d] independently reported fluorogenic calix[4]arene compounds with two or four ester groups in which the relative emission ratio of pyrene excimer to its monomer varies sensitively with the amount and the identity of added metal ions. Also, we reported a 1,3-alternate fluorescent calix[4]arene that complexes Pb^{2+} with two facing amide functions resulting in fluorescence quenching of both the pyrene excimer and its monomer by a combination of conformational change of the amide groups linking the pyrenes and reverse PET from the pyrenes to the metal-binding carbonyl groups.[34]

Indium, a heavy metal of group 3, is believed to interfere with iron metabolism at sites of absorption, transportation, utilization, and storage in cells.[35] According to several animal studies, acute intravenous administration of indium trichloride is extremely toxic to the liver and kidneys.[36] In^{3+} is more reactive toward biological membranes than Cd^{2+} and Hg^{2+},[37] but its functions in vivo remain unexplored. Development of an In^{3+}-selective fluorescent chemosensor would be beneficial in elucidating the cellular role of In^{3+} in living systems. Although a variety of effective fluorescent sensors for alkali metal cations, alkaline-earth metal cations,[1] Hg^{2+},[2] Zn^{2+},[3] and Pb^{2+} [4] have already been developed, none for In^{3+} has been reported.

We now describe the synthesis of fluorogenic calix[4]arene **1** (Chart 1), which exhibits a unique fluorescent response in the presence of In^{3+}. To probe the causative factors for this response, analogues **2**−**4** were also prepared and examined. A solid-state structure of metal-free **1** provides important insight.
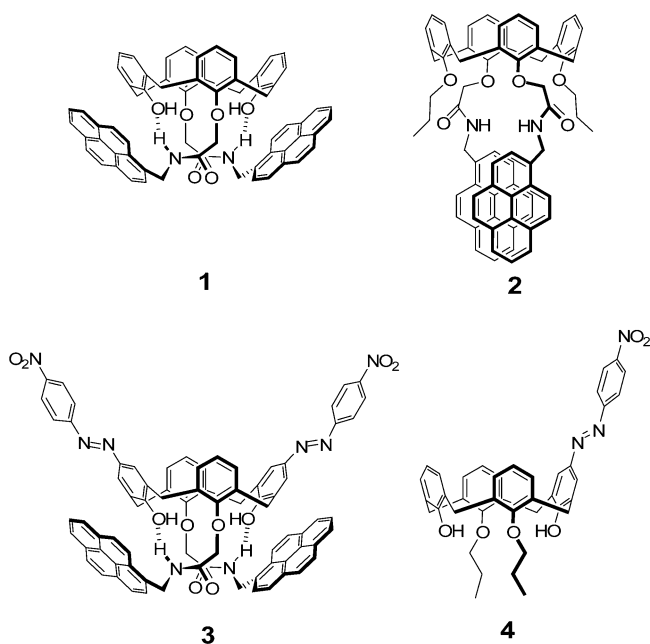
Experimental Section

Synthesis. Compounds **5**,[33a] **6**,[38] and **10**[39] were prepared following literature procedures.

25,27-Bis[(*N*-(1-pyrenylmethyl)aminocarbonyl)methoxy]calix[4]arene, Cone (1). To a mixture of 0.50 g (1.18 mmol) of calix[4]arene and 0.74 g (2.41 mmol) of *N*-(1-pyrenyl-

(16) Langford, S. J.; Yann, T. *J. Am. Chem. Soc.* **2003**, *125*, 11198.
(17) Ji, H.-F.; Dabestani, R.; Brown, G. M. *J. Am. Chem. Soc.* **2000**, *122*, 9306.
(18) de Silva, A. P.; Gunaratne, H. Q. N.; McCoy, C. P. *J. Am. Chem. Soc.* **1997**, *119*, 7891.
(19) Credi, A.; Balzani, V.; Langford, S. J.; Stoddart, J. F. *J. Am. Chem. Soc.* **1997**, *119*, 2679.
(20) (a) de Silva, A. P.; Dixon, I. M.; Gunaratne, H. Q. N.; Gunnlaugsson, T.; Maxwell, P. R. S.; Rice, T. E. *J. Am. Chem. Soc.* **1999**, *121*, 1393. (b) de Silva, A. P.; Gunaratne, H. Q. N.; McCoy, C. P. *Nature* **1993**, *364*, 42.
(21) Saghatelian, A.; Volcker, N. H.; Guckian, K. M.; Lin, V. S.-Y.; Ghadiri, M. R. *J. Am. Chem. Soc.* **2003**, *125*, 346.
(22) Birks, J. B. *Photophysics of Aromatic Molecules*; Wiley-Interscience: London, 1970.
(23) Winnik, F. M. *Chem. Rev.* **1993**, *93*, 587.
(24) (a) Lewis, F. D.; Zhang, Y.; Letsinger, R. L. *J. Am. Chem. Soc.* **1997**, *119*, 5451. (b) Lou, J.; Hatton, T. A.; Laibinis, P. E. *Anal. Chem.* **1997**, *69*, 1262. (c) Reis e Sousa, A. T.; Castanheira, E. M. S.; Fedorov, A.; Martinho, J. M. G. *J. Phys. Chem. A* **1998**, *102*, 6406. (d) Suzuki, Y.; Morozumi, T.; Nakamura, H.; Shimomura, M.; Hayashita, T.; Bartsch, R. A. *J. Phys. Chem. B* **1998**, *102*, 7910.
(25) Leray, I.; O'Reilly, F.; Habib Jiwan, J.-L.; Soumillion, J.-Ph.; Valeur, B. *Chem. Commun.* **1999**, 795.
(26) (a) Gutsche, C. D. *Calixarenes*; Royal Society of Chemistry: Cambridge, U.K., 1989. (b) Casnati, A.; Ungaro, R.; Asfari, Z.; Vicens, J. In *Calixarenes 2001*; Asfari, Z., Böhmer, V., Harrowfield, J., Vicens, J., Eds.; Kluwer Academic: Dordrecht, The Netherlands, 2001; pp 365−384.
(27) Andreetti, G. D.; Ugozzoli, F.; Ungaro, R.; Pochini, A. In *Inclusion Compounds*; Atwood, J. L., Davies, J. E. D., MacNicol, D. D., Eds.; Oxford University Press: New York, 1991; Vol. 4, pp 64−125.
(28) (a) Nishizawa, S.; Kaneda, H.; Uchida, T.; Teramae, N. *J. Chem. Soc., Perkin Trans. 2* **1998**, 2325. (b) Nishizawa, S.; Kato, Y.; Teramae, N. *J. Am. Chem. Soc.* **1999**, *121*, 9463.
(29) Hecht, S.; Vladimirov, N.; Fréchet, J. M. J. *J. Am. Chem. Soc.* **2001**, *123*, 18.
(30) (a) Kim, J. S.; Shon, O. J.; Rim, J. A.; Kim, S. K.; Yoon, J. *J. Org. Chem.* **2002**, *67*, 2348. (b) Kim, J. S.; Noh, K. H.; Lee, S. H.; Kim, S. K.; Kim, S. K.; Yoon, J. *J. Org. Chem.* **2003**, *68*, 597.

(31) (a) Ojida, A.; Mito-oka, Y.; Inoue, M.-A.; Hamachi, I. *J. Am. Chem. Soc.* **2002**, *124*, 6256. (b) de Silva, A. P.; Gunarante, H. Q. N.; Lynch, P. M. L. *J. Chem. Soc., Perkin Trans. 2* **1995**, 685. (c) Kubo, Y.; Ishihara, S.; Tsukahara, M.; Tokita, S. *J. Chem. Soc., Perkin Trans. 2* **2002**, 1455.
(32) (a) Chae, M.-Y.; Cherian, X. M.; Czarnik, A. W. *J. Org. Chem.* **1993**, *58*, 5797. (b) Bergonzi, R.; Fabbrizzi, L.; Licchelli, M.; Mangano, C. *Coord. Chem. Rev.* **2000**, *205*, 31.
(33) (a) van der Veen, N. J.; Flink, S.; Deij, M. A.; Egberink, R. J. M.; van Veggel, F. C. J. M.; Reinhoudt, D. N. *J. Am. Chem. Soc.* **2000**, *122*, 6112. (b) Jin, T.; Ichikawa, K.; Koyama, T. *J. Chem. Soc., Chem. Commun.* **1992**, 499. (c) Aoki, I.; Kawabata, H.; Nakashima, K.; Shinkai, S. *J. Chem. Soc., Chem. Commun.* **1991**, 1771. (d) Aoki, I.; Sakaki, T.; Tsutsui, S.; Shinkai, S. *Tetrahedron Lett.* **1992**, *33*, 89.
(34) Kim, S. K.; Lee, S. H.; Lee, J. Y.; Lee, J. Y.; Bartsch, R. A.; Kim, J. S. *J. Am. Chem. Soc.* **2004**, *126*, 16499.
(35) Moshtaghie, A. A.; Ghaffari, M. A. *Iran. Biomed. J.* **2003**, *7*, 73.
(36) Chapin, R. E.; Harris, M. W.; Sidney Hunter, H. E.; Davis, B. J.; Collins, B. J.; Lockhart, A. C. *Fundam. Appl. Toxicol.* **1995**, *27*, 140.

Chart 1

the solvent was evaporated in vacuo to give a white solid. Recrystallization from CH$_2$Cl$_2$–diethyl ether (1:9) gave 0.15 g (68%) of **5** as a white solid. Mp: 209–210 °C. IR (KBr pellet, cm^{-1}): 3316, 1743. ^{1}H NMR (400 MHz, CDCl$_3$): δ 8.14 (m, 18 H, Ar*H*, pyrene), 7.36 (br t, 2 H, CON*H*CH$_2$), 6.90–6.82 (m, 2 H, Ar*H*$_p$; 4 H, Ar*H*$_m$), 6.13 (d, 4 H, Ar*H*$_m$, *J* = 7.2 Hz), 5.96 (t, 2 H, Ar*H*$_p$, *J* = 6.8 Hz), 5.17 (d, 4 H, CONHC*H*$_2$, *J* = 6.0 Hz), 4.21 (s, 4 H, ArOC*H*$_2$CO), 3.82 (d, 4 H, ArC*H*$_2$Ar, *J* = 13.2 Hz), 3.54 (t, 4 H, ArOC*H*$_2$CH$_2$, *J* = 7.60 Hz), 3.18 (d, 4 H, ArC*H*$_2$Ar, *J* = 13.6 Hz), 1.77–1.73 (m, 4 H, CH$_2$C*H*$_2$CH$_3$), 0.91 (t, 6 H, CH$_2$CH$_2$C*H*$_3$, *J* = 7.4 and 7.6 Hz). ^{13}C NMR (50 MHz, CDCl$_3$): δ 166.4, 133.8, 130.3, 129.6, 129.2, 129.1, 128.3, 126.8, 126.2, 125.8, 125.4, 123.6, 120.4, 75.6, 32.9, 23.7, 10.7. FAB MS *m*/*z* (M$^+$). Calcd: 1051.2. Found: 1051.0. Anal. Calcd for C$_{72}$H$_{62}$N$_2$O$_6$: C, 82.26; H, 5.94. Found: C, 82.29; H, 5.99.

25,27-Bis(ethoxycarbonylmethoxy)-5,17-bis[(4-nitrophenylazo)phenyl]calix[4]arene, Cone (7). To a solution of **6** (1.00 g, 1.68 mmol) in tetrahydrofuran (THF; 300 mL), 4-nitrobenzenediazonium tetrafluoroborate (0.95 g, 4.02 mmol) was added. The reaction mixture was stirred for 30 min at 0 °C, and then pyridine (0.34 g, 3.68 mmol) was added dropwise. The reaction mixture was stirred for an additional 12 h at 0 °C, treated with 10% aqueous HCl solution (300 mL), and extracted with CH$_2$Cl$_2$ (300 mL). The organic layer was washed with 10% aqueous HCl solution (2 × 300 mL) and dried over MgSO$_4$. Removing the organic solvent in vacuo afforded a reddish solid. Column chromatography on silica gel with EtOAc–hexane (1:3) as eluent gave 0.89 g (42%) of **7**. Mp: 170–183 °C. IR (KBr pellet, cm^{-1}): 3259, 1669, 1521, 1343. ^{1}H NMR (200 MHz, CDCl$_3$): δ 8.67 (s, 2 H, ArO*H*), 8.36 (d, 4 H, NO$_2$Ar*H*$_o$, *J* = 8.60 Hz), 7.95 (d, 4 H, NO$_2$Ar*H*$_m$, *J* = 8.80 Hz), 7.80 (s, 4 H, Ar*H*$_m$), 7.04 (d, 4 H, Ar*H*$_m$, *J* = 7.60 Hz), 6.82 (t, 2 H, Ar*H*$_p$, *J* = 6.60 & 8.40 Hz), 4.77 (s, 4 H, ArOC*H*$_2$CO), 4.54 (d, 4 H, ArC*H*$_2$Ar, *J* = 13.2 Hz), 4.38 (q, 4 H, OC*H*$_2$CH$_3$), 3.58 (d, 4 H, ArC*H*$_2$Ar, *J* = 13.2 Hz), 1.40 (t, 6 H, OCH$_2$C*H*$_3$, *J* = 7.20 and 7.00 Hz). ^{13}C NMR (50 MHz, CDCl$_3$): δ 168.0, 157.3, 155.6, 151.5, 147.2, 145.0, 131.6, 128.9, 128.0, 124.0, 122.2, 71.7, 61.0, 30.3, 13.5. FAB MS *m*/*z* (M$^+$). Calcd: 894.29. Found: 894.0. Anal. Calcd for C$_{48}$H$_{42}$N$_6$O$_{12}$: C, 64.42; H, 4.73. Found: C, 64.43; H, 4.73.

25,27-Bis(hydroxycarbonylmethoxy)-5,17-bis[(4-nitrophenylazo)phenyl]calix[4]arene, Cone (8). A solution of **7** (0.50 g, 0.56 mmol) and NaOH (0.5 g, 12.5 mmol) in THF (5 mL), ethanol (10 mL), and water (5 mL) was refluxed for 12 h and evaporated in vacuo. The residue was dissolved in EtOAc, and the solution was washed twice with 20% HCl and then three times with water. The organic layer was dried over MgSO$_4$ and evaporated in vacuo to yield 0.40 g (85%) of **8** as a reddish solid. Mp: 218–225 °C. IR (KBr, cm^{-1}): 3304, 1693, 1521, 1343. ^{1}H NMR (200 MHz, CDCl$_3$): δ 8.35 (d, 4 H, NO$_2$Ar*H*$_o$, *J* = 8.60 Hz), 7.95 (d, 4 H, NO$_2$-Ar*H*$_m$, *J* = 9.20 Hz), 7.81 (s, 4 H, Ar*H*$_m$), 7.10 (d, 4 H, Ar*H*$_m$, *J* = 7.00 Hz), 6.91 (t, 2 H, Ar*H*$_p$, *J* = 6.20 and 7.20 Hz), 4.79 (s, 4 H, ArOC*H*$_2$CO), 4.30 (d, 4 H, ArC*H*$_2$Ar, *J* = 13.6 Hz), 3.61 (d, 4 H, ArC*H*$_2$Ar, *J* = 13.2 Hz). ^{13}C NMR (50 MHz, CDCl$_3$): δ 168.7, 158.1, 156.3, 152.2, 147.9,

methyl)chloroacetamide (**5**) in 50 mL of dry MeCN, anhydrous K$_2$CO$_3$ (0.16 g, 1.18 mmol) and a catalytic amount of NaI were added. The reaction mixture was refluxed for 2 days. After removal of the solvent in vacuo, the resulting solid was dissolved in CH$_2$Cl$_2$ (100 mL) and the organic layer was washed three times with water. The organic layer was dried over MgSO$_4$ and evaporated in vacuo to give the crude product, which was purified by recrystallization from diethyl ether to give 0.82 g (73%) of **1** as a white solid. Mp: 234–240 °C. IR (KBr pellet, cm^{-1}): 3322, 1780. ^{1}H NMR (200 MHz, CDCl$_3$): δ 8.61 (br s, 2 H, CON*H*CH$_2$), 8.36–7.62 (m, 18 H, Ar*H*, pyrene), 6.40–6.32 (m, 4 H, Ar*H*$_m$; 2 H, Ar*H*$_p$), 6.05 (d, 4 H, Ar*H*$_m$, *J* = 7.60 Hz), 5.84 (t, 2 H, Ar*H*$_p$, *J* = 7.40 and 7.20 Hz), 5.63 (s, 2 H, ArO*H*), 5.44 (br d, 4 H, CONHC*H*$_2$), 4.13 (s, 4 H, ArOC*H*$_2$CO), 2.65 (broad d, 4 H, ArC*H*$_2$Ar), 2.46 (d, 4 H, ArC*H*$_2$Ar, *J* = 13.60 Hz). ^{13}C NMR (50 MHz, CDCl$_3$): δ 169.1, 151.4, 132.7, 132.5, 131.8, 130.7, 129.9, 129.4, 129.1, 128.9, 127.9, 126.8, 126.6, 126.5, 126.1, 125.6, 125.2, 123.0, 120.1, 75.5, 31.4. FAB MS *m*/*z* (M$^+$). Calcd: 967.1. Found: 967.0. Anal. Calcd for C$_{66}$H$_{50}$N$_2$O$_6$: C, 81.97; H, 5.21. Found: C, 81.84; H, 5.24.

25,27-Bis[(*N*-(1-pyrenylmethyl)aminocarbonyl)methoxy]-26,28-dipropyloxycalix[4]arene, Cone (2). Under nitrogen, a mixture of **1** (0.20 g, 0.21 mmol), 1-iodopropane (0.080 g, 0.21 mmol), and K$_2$CO$_3$ (0.09 g, 0.65 mmol) in MeCN (100 mL) was refluxed for 24 h, and the solvent was removed in vacuo. To the resulting white solid, 5% aqueous HCl solution (100 mL) and CH$_2$Cl$_2$ (50 mL) were added, and the organic layer was separated and washed with water (3 × 50 mL). The organic layer was dried over MgSO$_4$, and

(37) Suzuki, Y.; Matsushita, H. *Ind. Health* **1969**, *7*, 143.
(38) Cillins, E. M.; McKervey, M. A.; Madigan, E.; Moran, M. B.; Owens, M.; Ferguson, G.; Harris, S. J. *J. Chem. Soc., Perkin Trans. 1* **1991**, 3137.
(39) (a) Kim, J. S.; Lee, W. K.; No, K. H.; Asfari, Z.; Vicens, J. *Tetrahedron Lett.* **2000**, *41*, 3345. (b) Koh, K. N.; Araki, K.; Shinkai, S.; Asfari, Z.; Vicens, J. *Tetrahedron Lett.* **1995**, *36*, 6095.

145.8, 132.2, 129.5, 128.6, 124.7, 122.9, 72.4, 31.4 ppm. FAB MS m/z (M⁺). Calcd: 838.77. Found: 838.0. Anal. Calcd for $C_{44}H_{34}N_6O_{12}$: C, 63.01; H, 4.09. Found: C, 63.04; H, 4.11.

25,27-Bis(chlorocarbonylmethoxy)-5,17-bis[(4-nitrophenylazo)phenyl]calix[4]arene, Cone (9). A solution of **8** (0.30 g, 0.36 mmol) and $SOCl_2$ (3 mL) in dry toluene (5 mL) was refluxed with stirring for 4 h. The solvent and excess $SOCl_2$ were removed by simple distillation. The residue was dissolved in dry THF (5 mL), and the solution was evaporated in vacuo to remove the residual $SOCl_2$. The resulting white solid was used directly for the next reaction.

5,17-Bis[(4-nitrophenylazo)phenyl]-25,27-bis[*N*-(1-pyrenylmethyl)aminocarbonylmethoxy]calix[4]arene, Cone (3). A solution of **9** (0.31 g, 0.36 mmol), 1-pyrenemethylamine hydrochloride (0.91 g, 0.72 mmol), and Et_3N (1.0 mL, 1.38 mmol) in 20 mL of dry THF was refluxed with stirring for 24 h and then evaporated in vacuo. The resulting solid was dissolved in CH_2Cl_2 (100 mL), and the organic layer was washed three times with water, dried over $MgSO_4$, and evaporated in vacuo. The crude product was chromatographed on silica gel with EtOAc−hexane (2:1) as eluent to give 0.070 g (16%) of **3** as a reddish solid. Mp: 228−236 °C. IR (KBr, cm⁻¹): 3304, 1650, 1521, 1343. ¹H NMR (200 MHz, CDCl₃): δ 8.45−7.68 (m, 18 H, ArH, pyrene; 8 H, NO₂ArH; 4 H, ArH_m), 6.68 (br s, 2 H, CONHCH₂), 6.37−6.27 (m, 4 H, ArH_m; 2 H, ArH_p), 5.97 (s, 2 H, ArOH), 5.45 (s, 4 H, CONHCH_2), 4.22 (s, 4 H, ArOCH_2CO), 2.67 (br d, 4 H, ArCH_2Ar), 2.45 (br d, 4 H, ArCH_2Ar). ¹³C NMR (50 MHz, CDCl₃): δ 167.7, 156.1, 154.7, 150.2, 148.9, 148.0, 144.8, 131.5, 130.7, 130.1, 129.5, 129.1, 128.6, 128.0, 126.8, 125.8, 125.4, 124.8, 124.5, 123.9, 123.3, 122.7, 121.8, 74.7, 44.3, 30.5. FAB MS m/z (M⁺). Calcd: 1265.3. Found: 1265.0. Anal. Calcd for $C_{78}H_{56}N_8O_{10}$: C, 74.04; H, 4.46. Found: C, 74.02; H, 4.45.

5-[(4-Nitrophenylazo)phenyl]-25,27-bis(1-propyloxy)-calix[4]arene, Cone (4). The same procedure as that for **7** was performed with **5** (2.00 g, 3.93 mmol), 4-nitrobenzenediazonium tetrafluoroborate (1.12 g, 4.73 mmol), pyridine (0.34 g, 4.33 mmol), and THF (300 mL). Column chromatography on silica gel with CHCl₃−hexane (1:4) as eluent gave 0.54 g (21%) of **3**. Mp: 267−268 °C. IR (KBr pellet, cm⁻¹): 3405, 1521, 1343. ¹H NMR (400 MHz; CDCl₃): δ 9.26 (s, 1 H, ArOH), 8.36 (d, 2 H, NO₂ArH_o, J = 8.20 Hz), 8.29 (s, 1 H, ArOH), 7.96 (d, 2 H, NO₂ArH_m, J = 8.12 Hz), 7.80 (s, 2 H, ArH_m), 7.07 (d, 2 H, ArH_m, J = 7.24 Hz), 7.00−6.91 (m, 4 H, ArH_m), 6.77 (t, 2 H, ArH_p, J = 6.76 Hz), 6.66 (t, 1 H, ArH_p, J = 7.08 Hz), 4.37−4.30 (dd, 4 H, ArCH_2Ar), 4.01 (t, 4 H, ArOCH_2CH₂, J = 5.36 Hz), 3.53 (d, 2 H, ArCH_2Ar, J = 13.00 Hz), 3.41 (d, 2 H, ArCH_2Ar, J = 12.72 Hz), 2.12−2.07 (m, 4 H, CH₂CH_2CH₃), 1.34 (t, 6 H, CH₂CH₂CH_3, J = 7.04 Hz). ¹³C NMR (CDCl₃): δ 159.3, 157.1, 154.0, 152.5, 148.6, 146.3, 134.2, 133.1, 130.2, 129.3, 128.8, 128.6, 125.4, 123.6, 119.8, 119.6, 79.1, 32.2, 24.3, 11.7. FAB MS m/z (M⁺). Calcd. 657.8. Found: 657.0. Anal. Calcd for $C_{40}H_{39}N_3O_6$: C, 73.04; H, 5.98. Found: C, 71.48; H, 5.76.

General Procedure for Fluorescence Studies. Fluorescence spectra were recorded with a RF-5301PC spectrofluorophotometer. Stock solutions (1.00 mM) of metal perchlorate salts were prepared in MeCN. Stock solutions of free **1**−**3** (0.060 mM) were prepared in MeCN. Excitations were carried out at 343 nm (for **1** and **2**) and 346 nm (for **3**) with all excitation and emission slit widths at 3 nm. Titration experiments were performed with 6.0 μM solutions of **1**−**3** in MeCN and various concentrations of metal perchlorates in MeCN. From the calculated concentrations of the free ligands and complexed forms of **1**−**3** in the fluorescence titration experiments, association constants were obtained with the computer program ENZFITTER.[40]

Job Plot Experiment. Compound **3** (40 μM) in MeCN and $In(ClO_4)_3$ (40 μM) in MeCN were prepared as stock solutions. The concentrations of each MeCN solution were varied, but the total volume was fixed at 4.0 mL. After the mixture was shaken for 2 h, the UV/vis absorbance at 375 nm was recorded.
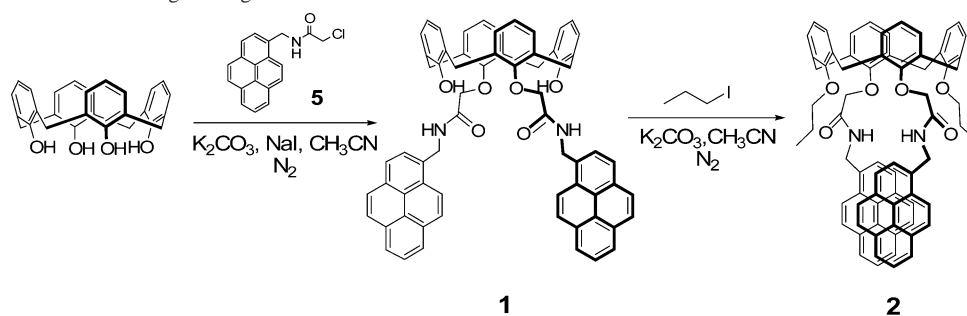
Solid-State Structure Determination for 1. All X-ray data were collected with a Siemens P4 diffractometer equipped with Mo Kα radiation (λ = 0.071 033 Å). The orientation matrix and unit-cell parameters were determined by least-squares analyses of the setting angles for 50 reflections in the range $10.0° < 2\theta < 25.0°$. Absorption corrections were not made. A brown, block-shaped crystal of **1**·CH₂Cl₂, of approximate dimensions 0.50 × 0.40 × 0.28 mm was used for crystal- and intensity-data collection. The unit-cell parameters and systematic absences unambiguously identified *Pbca* as the space group. The structure was solved by direct methods. Because of the poor crystal quality of **1**·CH₂Cl₂ and the consequence of the limited number of reflections, only oxygen, nitrogen, and chlorine atoms were refined anisotropically, and carbon atoms were refined isotropically. All hydrogen atoms were generated in ideal positions and refined in a riding mode.
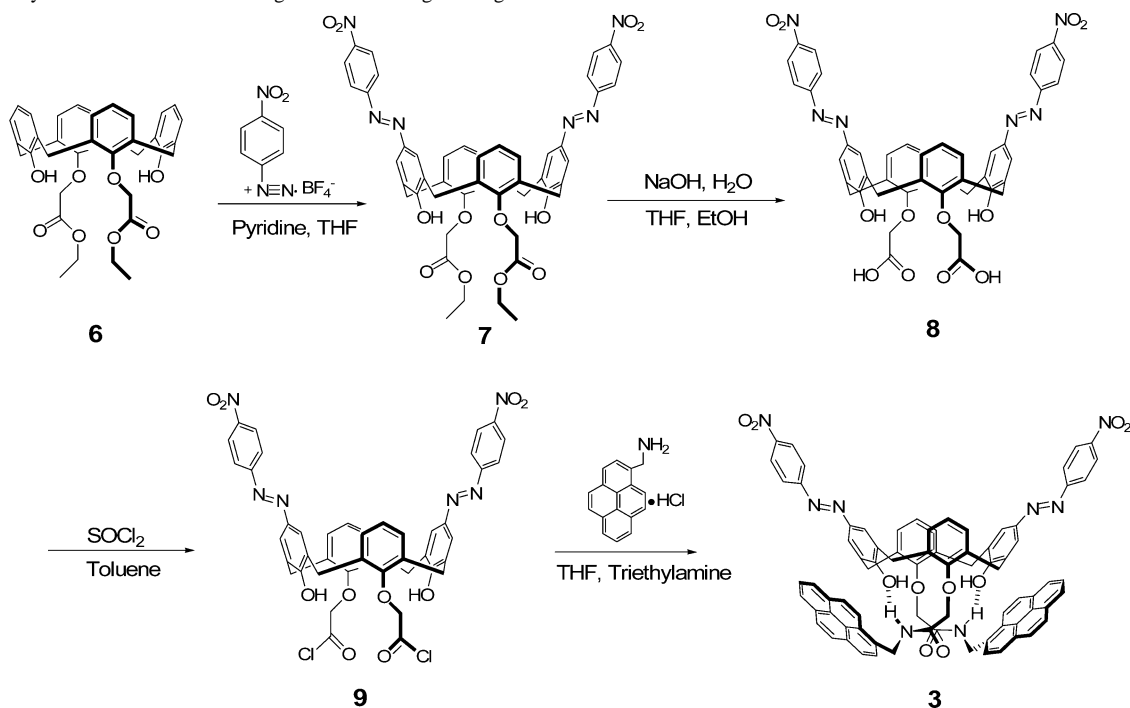
Results and Discussion

As shown in Scheme 1, the reaction of calix[4]arene with 2.1 equiv of *N*-(1-pyrenylmethyl)chloroacetamide (**5**)[33a] in the presence of a catalytic amount of NaI and 1.0 equivalent of K_2CO_3 in MeCN afforded 25,27-bis[(*N*-(1-pyrenylmethyl)-aminocarbonyl)methoxy]calix[4]arene (**1**) in quantitative yield. Subsequently, fluorogenic calix[4]arene **1** was treated with 1-iodopropane and K_2CO_3 in MeCN to produce **2** in 68% yield. Scheme 2 shows the synthetic route to calix[4]-arene derivative **3** having both chromogenic azo groups and fluorogenic pyrene groups. Calix[4]arene diethyl ester **6** in the cone conformation was synthesized by reaction of calix-[4]arene with 4.0 equiv of ethyl bromoacetate and 1.0 equiv of K_2CO_3 in MeCN.[38] Subsequently, the diazo-coupling reaction of **6** with *p*-nitrobenzenediazonium tetraborate in the presence of pyridine in THF gave chromogenic diester **7** in moderate yield. Hydrolysis of **7** with NaOH THF/H₂O/

(40) (a) Association constants were obtained with the computer program: *ENZFITTER*; Elsevier-BIOSOFT: Cambridge, U.K. (b) Connors, K. A. *Binding Constants*; Wiley: New York, 1987.
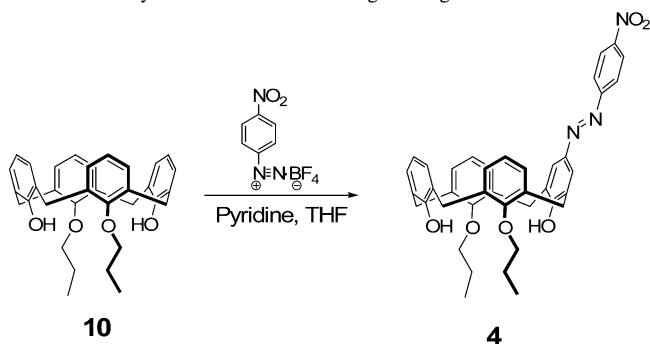
Scheme 1. Synthetic Route to Fluorogenic Ligands **1** and **2**

5

K$_2$CO$_3$, NaI, CH$_3$CN
N$_2$

K$_2$CO$_3$, CH$_3$CN
N$_2$

1　　　　**2**

Scheme 2. Synthetic Scheme to Chromogenic and Fluorogenic Ligand **3**

NO$_2$

+ N≡N· BF$_4^-$
Pyridine, THF

NaOH, H$_2$O
THF, EtOH

6　　　**7**　　　**8**

SOCl$_2$
Toluene

NH$_2$
·HCl

THF, Triethylamine

9　　　**3**

Scheme 3. Synthetic Route to Chromogenic Ligand **4**

NO$_2$

N≡N·BF$_4^-$
Pyridine, THF

NO$_2$

10　　　**4**

EtOH solution afforded the corresponding calix[4]arene dicarboxylic acid **8** in high yield. Reaction of **8** with SOCl$_2$ gave the corresponding acid chloride **9**, which underwent amination with 1-pyrenemethylamine hydrochloride to give the colorimetric and fluorogenic chemosensor **3** in 16% yield. As another model compound, chromogenic calix[4]arene **4** with two propyl groups in place of the amide groups was prepared by the reaction of **10** [39] with *p*-nitrobenzenediazonium tetrafluoroborate in THF (Scheme 3). The calix[4]arene frameworks in **1**−**4** are in the cone conformation, as confirmed by their NMR spectra: (a) two doublets with an AB pattern for methylenic protons of ArC*H*$_2$Ar in the ^{1}H NMR spectra and (b) singlets at about 32 ppm for the Ar*C*H$_2$-Ar bridge carbons in the ^{13}C NMR spectra. Also, the solid-state structure of **1** (Figure 1) provides convincing evidence for its cone conformation.[41]

Excitation of **2** at 343 nm gave both a strong monomer emission (λ_{em} = 396 nm) and a strong excimer emission (λ_{em} = 472 nm) (Figure 2), suggesting that the two pyrene units are face-to-face π-stacked and that formation of the excimer
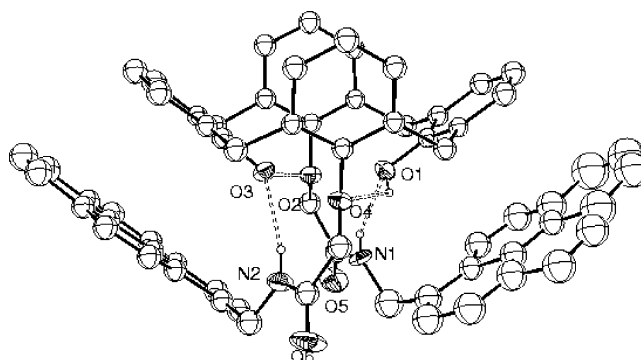
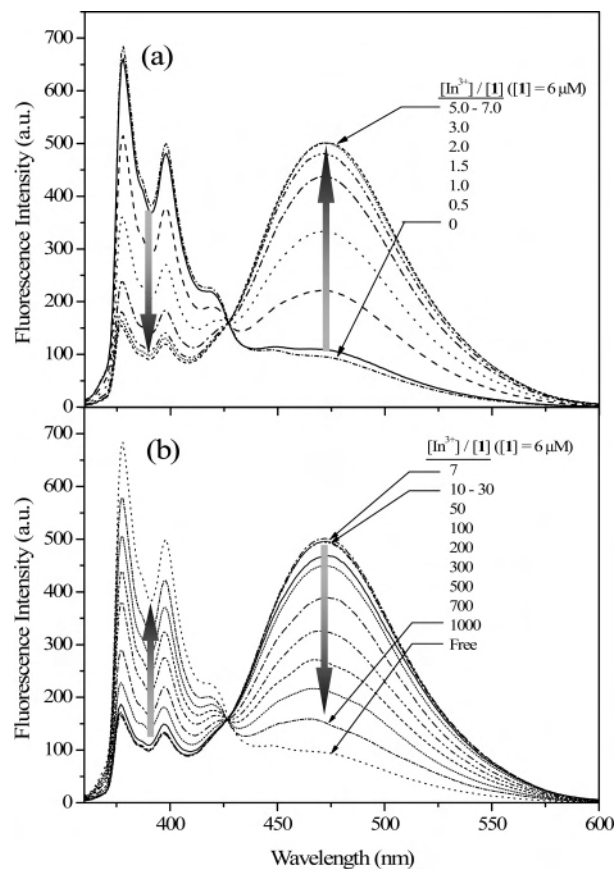**Figure 1.** X-ray crystal structure of **1** with hydrogen bonds represented by dotted bonds.

Table 1. Fluorescence Changes ($I - I_0$) for **1** and **2** upon Addition of Various Metal Ion Species[a]

ligand	λ_{em} (nm)	Fluorescence changes ($I - I_0$)									
		Li⁺	Na⁺	K⁺	Cs⁺	Ag⁺	Mg²⁺	Ca²⁺	Pb²⁺	Zn²⁺	In³⁺
1	398	29.2	33.2	27.1	29.1	26.4	39.5	54.4	−147.5	16.6	−243.9
	470	−10.4	−7.0	−10.4	−9.3	−5.0	23.6	13.8	−52.5	3.8	304.7
2	396	−64.6	−29.8	11.5	−3.6	−37.2	−37.7	−15.1	−103.9	−9.4	−63.1
	472	11.0	57.8	13.9	18.1	42.5	71.3	135.7	−361.0	30.2	−258.6

[a] Conditions: **1** and **2**, 6.0 μM in MeCN, excitation at 343 nm; metal ions, 500 equiv in MeCN. I_0, fluorescence emission intensity of free **1** and **2**; I, fluorescence emission intensity of metal-ion-complexed **1** or **2**.
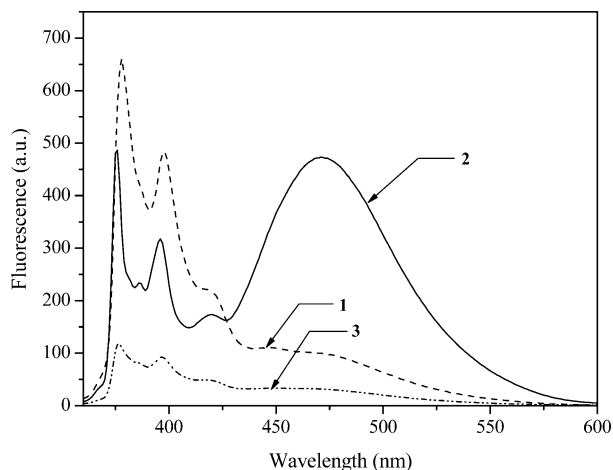
Figure 2. Fluorescence emission spectra of free ligands **1−3** (6.0 μM) in MeCN. The excitation wavelengths were 343 nm for **1** and **2** and 346 nm for **3**.
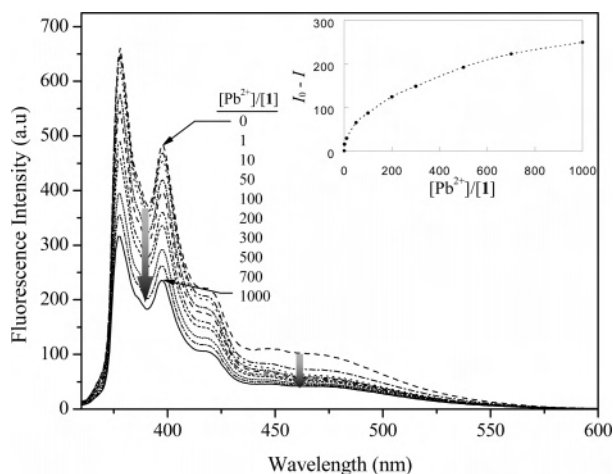
Figure 3. Fluorescence emission spectra of **1** (6.0 μM) for Pb²⁺ ion titration in MeCN. The excitation wavelength was 343 nm.


Figure 4. Fluorescence emission spectra of **1** (6.0 μM) upon the addition of various amounts of In³⁺ in MeCN. The excitation wavelength was 343 nm.

is dynamic. The intensity ratio ($I_{excimer}/I_{monomer}$) for **2** with concentration variation ($10^{-7}-10^{-4}$ M) remained unchanged, indicating that the excimer formation results from intramolecular interactions, not from intermolecular interactions. In contrast, unexpectedly, free **1** exhibited only a monomer emission at around 398 nm with small excimer emission (Figure 2). This phenomenon should be distinguished from that found by Broan,[42] who reported a fluorogenic calix[4]-arene chemosensor with two facing ester groups and two pendant pyrene groups linked to the ester groups, which forms a strong intramolecular excimer. Compared with those of Broan's compound and **2**, this unique fluorescence behavior of **1** can be rationalized by its solid-state structure.

As shown in the X-ray crystal structure (Figure 1), intramolecular hydrogen bonds are formed between the phenolic OH groups and the amide hydrogen atoms, which is believed to preclude face-to-face π-stacking of the pyrene units and

(41) Crystal data for **1**·CH₂Cl₂: C₆₇H₅₂Cl₂N₂O₆, M_r = 1052.01, brown block 0.50 × 0.40 × 0.28 mm, orthorhombic, space group *Pbca*, a = 23.788(4) Å, b = 17.453(3) Å, c = 25.592(5) Å, V = 10625(3) Å³, ρ_{calcd} = 1.315 g cm⁻³, μ = 0.180, $F(000)$ = 4400, Z = 8, Mo Kα radiation, (λ = 0.710 73 Å, graphite monochromator), T = 293 K, ω scan range 2.07 < θ < 21.50°, 5928 independent eflections, 2242 observed reflections [$I > 2\sigma(I)$], 362 refined parameters, R = 0.1413, wR_2 = 0.3444, largest difference peak and hole = 0.837 and −0.358 e·Å⁻³. The structure was solved by direct methods. Because of the poor crystal quality of **1**·CH₂Cl₂ and consequently the limited number of reflections, only oxygen, nitrogen, and chlorine atoms were refined anisotropically, and carbon atoms were refined isotropically. All hydrogen atoms were generated in ideal positions and refined in a riding mode. CCDC-255617 contains the supplementary crystallographic data for this paper. These data can be obtained free of charge via www.ccdc.cam.ac.uk/conts/retrieving.html (or from the Cambridge Crystallographic Data Center, Cambridge, U.K.; deposit@ccdc.cam.ac.kr).
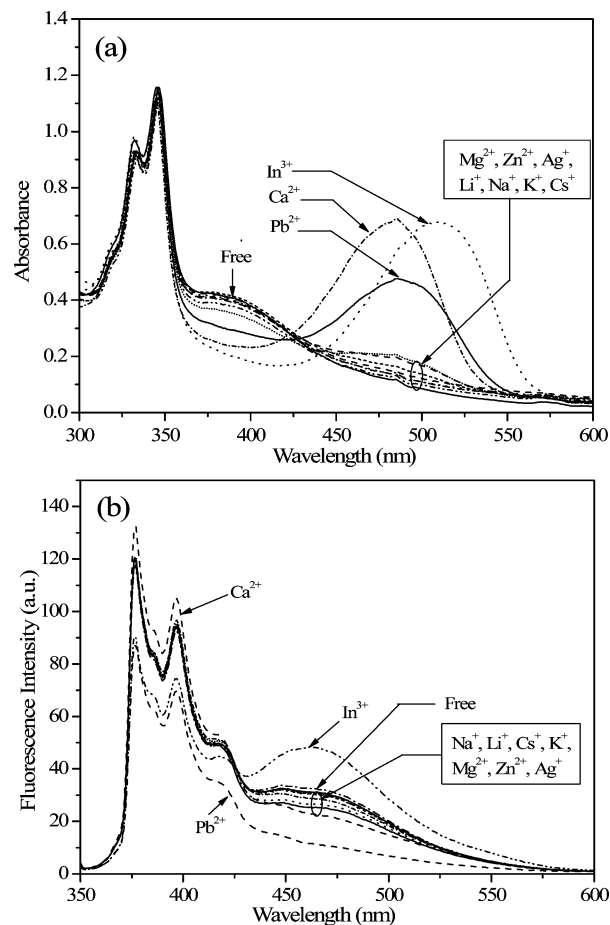(42) Broan, C. J. *Chem. Commun.* **1996**, 699.
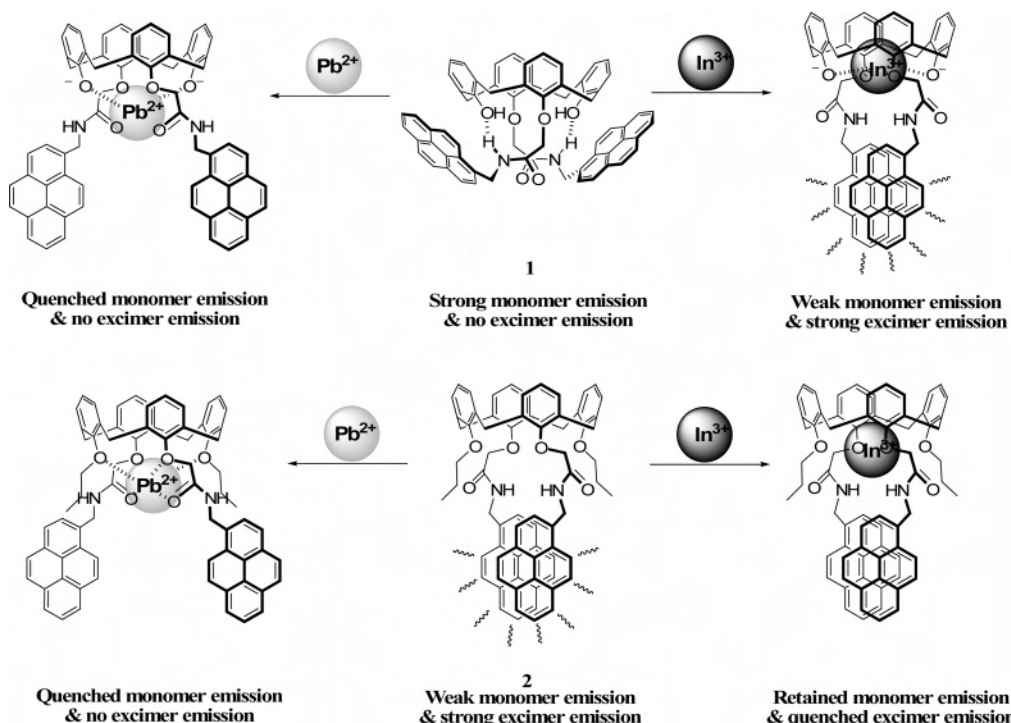
Figure 5. Proposed conformational changes of **1** and **2** upon complex formation with Pb^{2+} and with In^{3+}.

consequently to a strongly quenched excimer emission from **1**. In the solid-state structure of **1**, the N1···O1 and N2···O3 distances are 3.316 and 3.024 Å, respectively. The N1−H···O1 and N2−H···O3 angles are 162.89 and 167.11°, respectively. These parameters indicate moderate hydrogen bonding. The fluorescence spectrum of **3** suggests that its conformation is the same as that of **1** (Figure 2). This is supported by similar ^{1}H NMR spectra for **1** and **3** in which all methylenic protons of the ArCH_2Ar fragments unusually appear as two broad doublets at around 2.5 ppm in the AB pattern. Despite the structural similarity between **1** and **3**, compound **3** emits a considerably weaker fluorescence with an intensity of only 18% that of **1** (Figure 2), which is attributed to quenching by electron transfer from the pyrene units to the nitro groups of the phenylazo moieties.[43] It is also believed that FRET (fluorescence resonance energy transfer) from the pyrene units to the azo groups is partially involved in weakening the emission from **3**. In general, FRET occurs by an interaction between the excited states of two fluorophores or chromophores when the emission spectrum of a fluorophore (a FRET donor) is overlapped with the absorption spectrum of another fluorophore or chromophore (a FRET acceptor), by which excitation energy of the donor is transferred to the acceptor without emission of photon.[44] In **3**, the pyrene units and azo groups can act as FRET donors and acceptors, respectively, and FRET between them probably quenches the emission of the pyrene donors.

To obtain insight into the metal ion binding properties of ligands **1** and **2**, we investigated fluorescence changes upon the addition of various metal perchlorates to MeCN solutions

(43) Aoki, I.; Sakaki, T.; Shinkai, S. *J. Chem. Soc.*, *Chem. Commun.* **1992**, 730.
(44) Lakowicz, J. R. *Principles of Fluorescence Spectroscopy*, 2nd ed.; Plenum: New York, 1999.

Figure 6. UV/vis absorption spectra (a) and fluorescence emission spectra (b) of **3** (20 μM for a and 6 μM for b) upon the addition of various metal ions (500 equiv) in MeCN. The excitation wavelength for b was 346 nm.
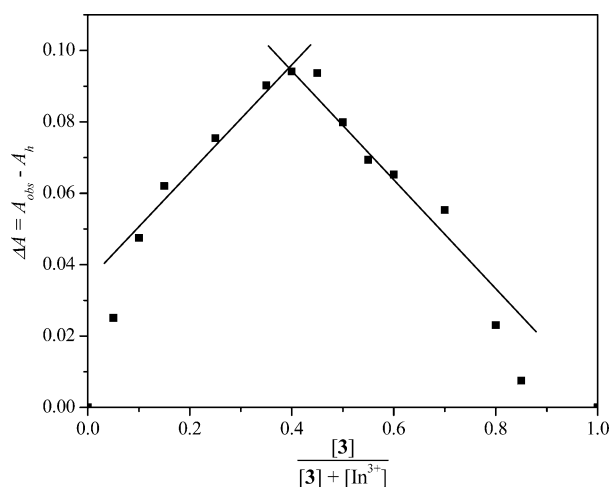
of **1** and **2**. The results are presented in Table 1. Compound **1** shows pronounced selectivity and sensitivity for Pb^{2+} and
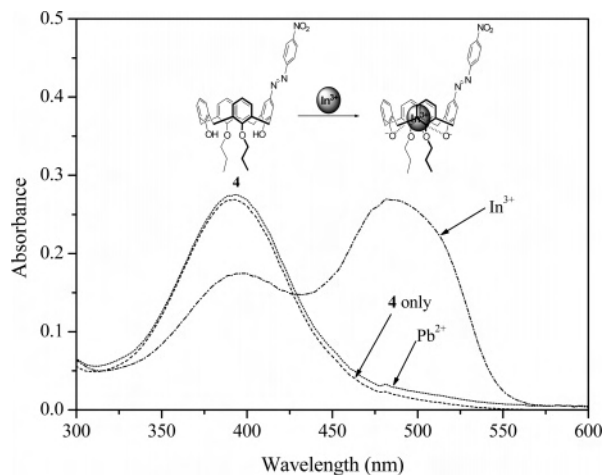
Figure 7. UV/vis absorption spectra of **4** (10 μM) upon addition of Pb^{2+} (10 equiv) and In^{3+} (10 equiv) in MeCN.

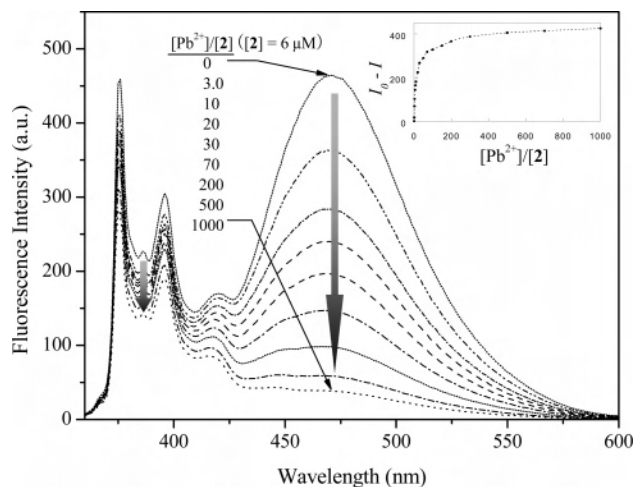**Figure 9.** Fluorescence spectra of **2** (6.0 μM) upon addition of Pb^{2+} in MeCN. The excitation wavelength was 343 nm.

Figure 8. Job plot of **3** with In^{3+} in MeCN. $\Delta A = A_{obs} - A_h$, where A_{obs} and A_h denote the absorbance at 375 nm upon the In^{3+} ion binding and that of free ligand **3**, respectively.
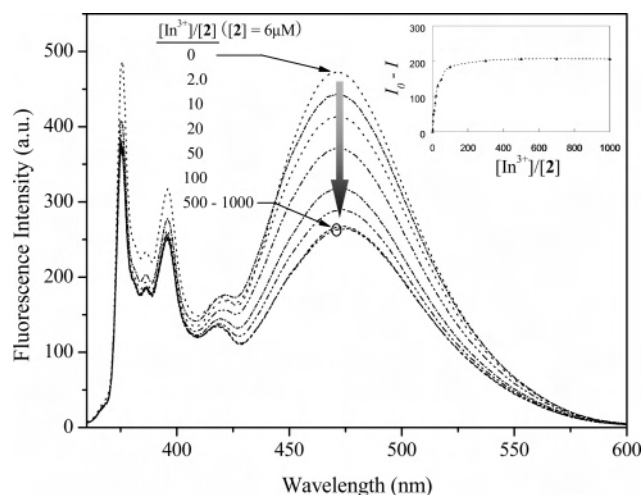

Figure 10. Fluorescence changes of **2** (6.0 μM) with In^{3+} in MeCN. The excitation wavelength was 343 nm.

even greater response to In^{3+}, whereas compound **2** responds to most metal ions tested, in particular, to Ca^{2+}, Pb^{2+}, and In^{3+}. This implies that hydrogen bonding between the phenolic OH groups and the amide hydrogen atoms plays a major role in the high selectivity of **1** for In^{3+}. For other metal ions, we speculate that binding by **1** is not sufficiently strong to break the hydrogen bonding.

From the fluorescence changes, we found that **1** binds metal ions in different modes. When **1** is complexed by Pb^{2+}, the fluorescence intensity is strongly quenched, which is due not only to reverse PET[4b,31] from the pyrene groups to the electron-deficient carbonyl oxygen atoms, but also to a heavy-metal ion effect.[4b,32] Figure 3 shows the fluorescence changes of **1** as a function of [Pb^{2+}] from which an association constant (K_a) with **1** is calculated to be 540 M^{-1}.[40]

Interestingly, unusual fluorescence changes were observed upon the addition of In^{3+} to a solution of **1**. The titration of In^{3+} with **1** displays a ratiometry with an isoemissive point at 428 nm dependent on the In^{3+} concentration (Figure 4). Upon the addition of up to 7 equiv of In^{3+} to **1** the excimer emission gradually increased and the monomer emission decreased (Figure 4a). This behavior can be ascribed to In^{3+}-

promoted deprotonation of the phenolic OH groups, which disrupts the hydrogen bonding (Figure 5). The oxygen atoms involved in hydrogen bonding with the amide hydrogen atoms form a complex with In^{3+}, in which the hydrogen bonding is broken and the OH groups are deprotonated. As a result, the two pyrene pendants overlap with face-to-face π-stacking and therefore display a strong excimer emission with a relatively quenched monomer emission. This is opposite for complex formation of Pb^{2+} with **1** through amide oxygen atoms, which causes the excimer emission to be strongly quenched. Thus, the binding mode of **1** with Pb^{2+} is quite different from that with In^{3+}.

To elucidate the role of the phenolic OH groups in metal-ion binding, we synthesized analogue **3** with added 4-nitrophenylazo groups. Free **3** shows absorption peaks at 356 nm (pyrene) and 380 nm (azo). Upon addition of metal ions, the spectral changes in UV/vis absorption and fluorescence for **3** show that it responds to metal ions in essentially the same manner as does **1** (Figure 6). Consistent with our expectation, the addition of In^{3+} to **3** gave rise to a marked red-shift from about 380 to 507 nm (Figure 6a), which supports In^{3+} binding to the phenoxide oxygens formed by
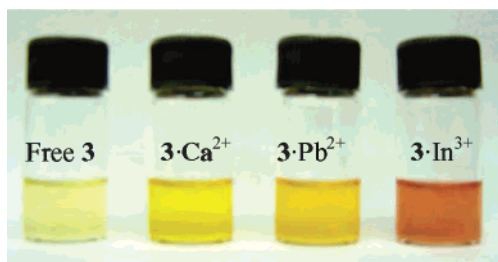
Figure 11. Color changes of **3** (40 μM) in MeCN upon addition of 100 equiv of Ca^{2+}, Pb^{2+}, and In^{3+}.

deprotonation of the phenolic OH groups to facilitate the quinine−hydrazone tautomerization. Similar bathochromic shifts were observed upon the addition of Ca^{2+} and Pb^{2+} to **3,** suggesting that these metal ions also bind to the deprotonated phenoxy groups, but more weakly than does In^{3+} (Figure 6a). Unfortunately, we cannot clearly explain the different fluorescence responses of **1** to In^{3+} and Pb^{2+} only by the metal-induced hydrogen-bonding disappearance observed for **3**.

To probe the effect of the amide groups on the metal-ion binding of **1**, which exhibits unique fluorescence behaviors toward In^{3+} and Pb^{2+}, we prepared a monoazo-coupled calix-[4]arene **4** with two propyl groups in place of the two amide groups. Upon addition of In^{3+} to solutions of **4**, a marked bathochromic shift from λ_{max} = 393 to 498 nm ($\Delta\lambda_{max}$ = 105 nm) was observed in the UV/vis spectra (Figure 7). This indicates that the hydroxyl groups of the calix[4]arene predominate the In^{3+} binding over amide oxygen atoms. On the other hand, addition of Pb^{2+} to **3** gave no change in the UV/vis spectrum (Figure 7), which supports the contention that in Pb^{2+} binding by **1** and **2** the two facing amide groups play a more important role than do the phenoxide oxygen atoms (Figure 5). Consequently, we believe that In^{3+} binds to the phenoxy oxygen atoms (not to the amide groups) after deprotonation of the phenolic OH groups, by which the hydrogen bonding between the amides and the phenoxy groups is no longer retained and the two pyrene units are allowed to overlap. In contrast, Pb^{2+} binding to the facing amide groups of **1** is well-known to prevent pyrene excimer formation and monomer emission via reverse PET and a heavy-metal ion effect.[34]

Surprisingly, the addition of more than 7 equiv of In^{3+} to **1** gave an excimer extinction with monomer revival (Figure 4b). These unique fluorescence changes are believed to occur because **1** can form a complex with In^{3+} in a mole ratio of 1:2 (ligand:metal). In this case, binding of the first In^{3+} ion to **1** results in the deprotonation of the phenolic OH to destroy the hydrogen bonding, leading to excimer formation by the two pyrene units. However, the second In^{3+} ion binding reduces the excimer emission with a concurrently enhanced monomer emission. It is worth noting that the incoming In^{3+} ions in the solution of **1** are entrapped first by the two phenolic oxygens and the second In^{3+} subsequently binds to the amide oxygen atoms in a 1:2 mole ratio. A Job plot experiment with **3** supports 1:2 complex formation of **1** with In^{3+} (Figure 8). The maximum point at the mole fraction of 0.4, which is a little higher than that at the typical ligand
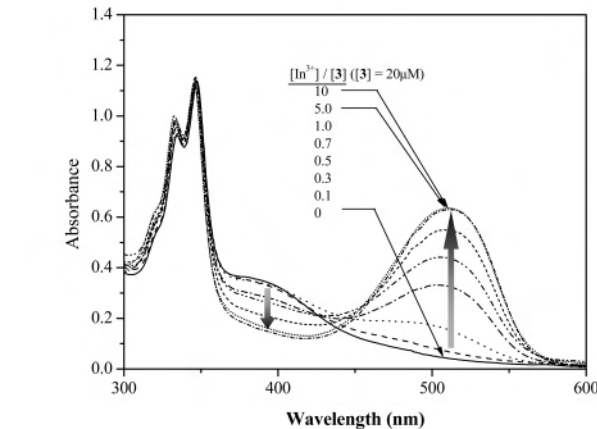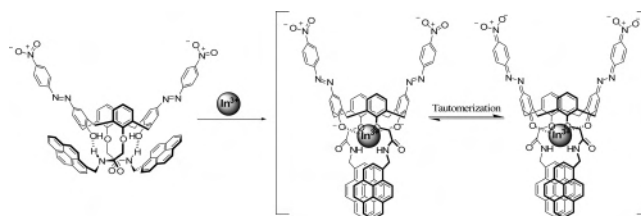
Figure 12. UV/vis absorption spectra of **3** (20 μM) upon the addition of various amounts of In^{3+} in MeCN.

mole fraction (0.33) for 1:2 stoichiometric complexes. This is probably because the binding constants of **3** with the first In^{3+} and the second In^{3+} are different as shown in Figure 4, with the first In^{3+} changing the fluorescence rapidly and the second In^{3+} rather slowly.

Figure 9 shows a variation in the fluorescence changes for **2** with the concentrations of Pb^{2+}. The excimer emission gradually weakened with increasing Pb^{2+} concentration and was completely quenched by addition of 1000 equiv of Pb^{2+}. Extinction of the excimer emission from **1** is attributed to a conformational change of **1** on metal-ion binding, by which two outward-facing amide carbonyl groups turn inward to bind Pb^{2+}.[34] In contrast, upon addition of In^{3+} to **2**, the excimer emission was only partially quenched (Figure 10), indicating that the excimer between the two pyrene units is retained even upon addition of 1000 equiv of In^{3+} (see Figure 5). This means that In^{3+} binds to the phenolic oxygen atoms, not to the amide groups, where the metal-ion binding would induce a conformational change of the ligand. Accordingly, a decrease in the excimer emission induced by In^{3+} is ascribed to a heavy-metal effect[4b,32] and reverse PET[4b,31,34] from the pyrene units to In^{3+}, rather than a conformational change for **2**.[33b−d] From these fluorescence titration experiments (Figures 9 and 10), association constants for **2** with Pb^{2+} and In^{3+} in MeCN were calculated to be 1.2 × 10^4 and 7.4 × 10^3 M^{-1}, respectively.[40] A relatively low K_a value of **1** for Pb^{2+}, compared with that of **2**, can also be ascribed to the intramolecular hydrogen bonding between the phenolic OH's and the amide groups.

For **3**, as mentioned above, the fluorescence responses to metal ions are similar to those of **1** with additional absorption band shifts. In^{3+} and Pb^{2+} complexation induced red shifts for **3** to 507 and 485 nm, respectively (Figure 6a), by which

different colors are displayed by the metal complexes of **3** (Figure 11).

Figure 12 shows the UV/vis spectra for titration of **3** with In³⁺, which was similar to that observed for **1** (Figure S1 of the Supporting Information). Figures 2b and S3b (Supporting Information) show fluorescence spectra in the presence of varying amounts of Pb²⁺ and Ca²⁺, respectively. The sensitive optical response of **3** to In³⁺ over other metal ions suggests that **3** can serve effectively as a fluorogenic and colorimetric chemosensor for In³⁺.

Conclusions

New fluorogenic calix[4]arenes **1** and **2** with two facing amide groups linked to pyrene units were synthesized. Relative orientations of these pyrene groups in the two compounds are quite different. They are divergent in **1**, but show face-to-face π-stacking in **2**. Unlike the strong excimer-emitting **2**, **1** exhibits only monomer emission due to the intramolecular hydrogen bonding between the phenolic OH groups and the amide hydrogen atoms, which prevents the two pyrene units from overlapping. The fluorescence changes upon addition of various metal ions show that **1** is highly selective for In³⁺ over the other metal ion species tested. Addition of In³⁺ to the solution of **1** enhances the excimer emission with quenching of the monomer emission, which can be explained by In³⁺-induced deprotonation of the phenolic OH groups, leading to the destruction of hydrogen bonding between the phenolic OH groups and the amide groups. Interestingly, addition of excess In³⁺ to **1** produced

exciter extinction with monomer revival. These unique fluorescence changes are believed to occur because **1** forms a 1:2 (ligand:metal) complex with In³⁺. However, when Pb²⁺ was added to **1**, the fluorescence emission was strongly quenched because of the reverse PET from the pyrene units to the amide groups, as well as a heavy-metal ion effect. For **2**, addition of Pb²⁺ or In³⁺ quenches the excimer emission due to a conformational change caused by two outward-facing amide carbonyl groups turning inward to bind to the metal ion. In the resulting conformation, face-to-face π-stacking of the two pyrene units is no longer retained. Compound **3** showed almost the same binding properties for the metal ions tested as does **1**, which provides crucial evidence for the mechanism of binding of **1** to In³⁺ and Pb²⁺. Our results suggest that **1** can be used as a powerful sensor for In³⁺ in vivo and in vitro systems.

Supporting Information Available: Listings of preparative procedures and analytical data for compounds **1**−**4**, additional fluorescence spectra, and additional tables (Table S1−S6) of crystal data for **1**. This material is available free of charge via the Internet at http://pubs.acs.org.

IC050702V