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An Effective Fluorescent Chemosensor for Mercury Ions

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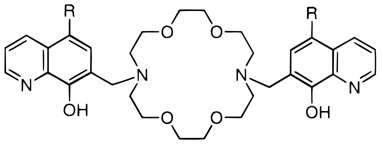
Recognition of the detrimental effects of certain transition and post transition metal ions on humans and higher animals[1] has, in part, inspired work to develop compounds that selectively respond to specific metal ions for use as ion sensors.[2] Ion-selective sensors could find use in monitoring metal ion concentrations in solutions including waste effluent streams and drinking water. Emphasis has been placed on development of compounds that selectively respond to the presence of specific metal ions through a change in one or more properties of the system, such as redox potentials,[3] absorption[4] or fluorescence spectra.[2]

We have prepared and studied several series of macrocyclic ligands with appended chromophores and fluorophores for use as selective metal ion chemosensors.[5] We recently reported ligand **1** as an ion-selective chemosensor for Mg^{2+}.[6] We now report that replacement of the chlorine atoms on the 8-hydroxyquinoline moieties in **1** with nitro groups yields a compound (**2**)[5a] that is an effective fluorescent chemosensor for Hg^{2+}, even in the presence of other metal cations, including Mg^{2+}, that might interfere with its detection.



1 R = -Cl
2 R = -NO$_2$

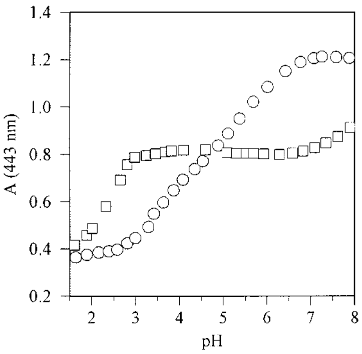


Figure 1. Absorbance of **2** (2.5 × 10^{-5} M) in methanol−water (1:1 v/v) at various pH values: ○, **2** alone; □, **2** with Hg^{2+} (2.5 × 10^{-5} M).

Few potential chemosensors for Hg^{2+} have been developed.[2] Notable exceptions include the work of Czarnik and co-workers,[7] who developed an anthracene-based chemosensor that selectively responded to Hg^{2+} and Cu^{2+} in aqueous solution. Association with these ions caused fluorescence quenching of the anthracene. In contrast, chemosensor **2** responds to Hg^{2+} via a fluorescent enhancement, a feature that would reduce the likelihood of false positive signals. Recently, Rurack et al.[8] reported a fluoroionophore for Hg^{2+}, Ag^{+}, and Cu^{2+} that gave significant fluorescence enhancements upon complexation with these ions in acetonitrile. However, in water/acetonitrile mixtures, fluorescence of the metal ion complexes was significantly attenuated. In aqueous media, a medium in which ion-sensing applications would likely be carried out, **2** displays a strong fluorescent response to Hg^{2+}.

Neutral 8-hydroxyquinolines are typically nonfluorescent in protic solvents; intra- and intermolecular excited-state proton transfers are efficient mechanisms for a radiationless deactivation to the ground state.[9] In some cases, complexation of 8-hydroxyquinoline derivatives with metal cations causes a decrease in the apparent pK_a of the phenolic group, and deprotonation concomitant with ion complexation yields fluorescent complexes in protic solvents.[6]

Because the photophysical properties of the 8-hydroxyquinoline derivatives are influenced by their protonation states, it is possible to determine pK_a values of the quinoline nitrogen and the phenolic group by observing changes in UV absorption at various pHs. To determine the effects of metal ion complexation on the pK_a values of **2**, we measured the UV absorbance as a function of pH with **2** alone and in the presence of various metal cations. Our experiments showed that the pK_a values for the quinoline nitrogens and the phenolic groups are approximately 3.7 and 5.4, respectively (Figure 1[10]).

As reported for other lariat ethers containing hydroxyquinoline derivatives, **2** was able to complex a large variety of metal ions. In particular, in methanol−water (1:1 v/v)[11] solutions, it formed stable 1:1 complexes with Hg^{2+}, Cu^{2+}, Cd^{2+}, Zn^{2+}, Ni^{2+}, and Mg^{2+}. Much lower association constants in this solvent (log K_a < 2.5) were observed with the other alkaline earth ions,

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(10) pK_a values were assigned as the maxima of the first derivative of the data shown in Figure 1. Plots of the derivatives of the titrations in Figure 1 are included in the Supporting Information. These data are in fair agreement with the data reported for 5-nitro-8-hydroxyquinoline in water (pK_a values of 2.5 and 6.4 for the nitrogen and the phenolic group, respectively (Klofutar, C.; Paljk, S.; Krasovec, F.; Horvat, I. *Mikrochim. Acta* **1973**, 559)), particularly if the different solvent and potential hydrogen bonding interactions with the macrocycle are considered.

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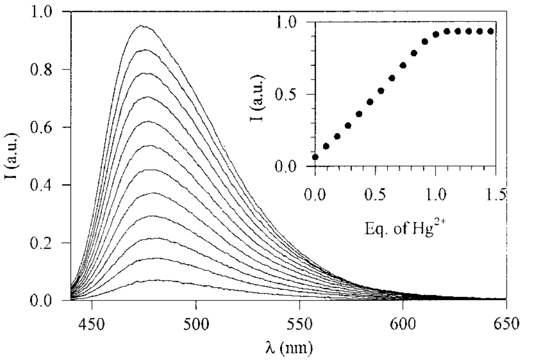

Figure 2. Fluorescence spectra ($\lambda_{exc} = 423$ nm) of **2** (2.5×10^{-5} M) in methanol−water (1:1 v/v, pH 7.0) with increasing amounts of Hg^{2+} ions. Inset: Fluorescence intensity values ($\lambda_{exc} = 423$ nm, $\lambda_{em} = 476$ nm) vs equivalents of added Hg^{2+}.

whereas no complexation was detected (log $K_a < 1.5$) for alkali metal ions.

Analogous to previous observations,[6] the complexation process caused strong changes in the absorption spectrum of **2** upon addition of increasing amounts of specific metal ions.[12] More importantly, changes were also observed in the fluorescence spectrum of **2** upon metal ion complexation. As already observed for the other 8-hydroxyquinoline derivatives, uncomplexed **2** gave only a very weak fluorescence band ($\lambda_{max} = 540$ nm, $\Phi = 5 \times 10^{-5}$, $\tau < 0.2$ ns). Complexes with Cu^{2+} and Ni^{2+} were not luminescent as expected because these metal ions provide pathways for energy- and electron-transfer processes which cause fluorescence quenching. A small increase of the luminescence intensity was detected upon addition of Cd^{2+} (with an enhancement factor (EF) = 3.0; $\lambda_{max} = 466$ nm), Zn^{2+} (EF = 3.4; $\lambda_{max} = 467$ nm) or Mg^{2+} (EF = 1.7; $\lambda_{max} = 470$ nm). A much higher luminescence increase (EF = 12; $\lambda_{max} = 476$ nm, $\Phi = 1 \times 10^{-3}$, $\tau < 0.5$ ns) was observed for Hg^{2+} (Figure 2).

To observe the **2**−Hg^{2+} complex stability as a function of pH, we recorded absorption and luminescence spectra of a solution containing **2** and Hg^{2+} in equimolar amounts (2.5×10^{-5} M) at varied pH values. The plot reporting absorbance (Figure 1) or luminescence intensity as a function of pH can be conveniently interpreted in analogy to observations made with the **1**−Mg^{2+} complex,[6] i.e., the formation of stable quinolate complexes with particular metal ions shifts the two deprotonation processes to lower pH. In the system containing **2** and Hg^{2+}, the complexation process with the concomitant deprotonation of both phenolic groups occurred at pH below 3. The appearance of a new band at 265 nm in the absorption spectrum was very similar to that observed for other quinolate complexes and gave further evidence of deprotonation of the phenolic groups.

From the changes observed in the absorption and luminescence spectra, we were able to determine association constants of **2** in methanol−water (1:1 vol:vol) with Mg^{2+} (1.2×10^5 M^{-1}) and Ni^{2+} (3.6×10^5 M^{-1}), while with Zn^{2+}, Cu^{2+}, Cd^{2+}, and Hg^{2+} only a lower limit (1×10^8 M^{-1}) could be measured. However, competition experiments performed at neutral pH with **2** in the presence of 1 equiv of Zn^{2+} and Cd^{2+} showed that Hg^{2+} ions could replace these ions with high efficiency, leading to the formation of the luminescent mercury complex (Figure 3). This result indicated that the association constant with the latter ion was likely at least 2 orders of magnitude higher than those with zinc and cadmium. In contrast, only a partial recovery of the luminescent band was obtained when 1 equiv of Hg^{2+} was added to the **2**−Cu^{2+} complex suggesting that **2** has similar affinities for copper and mercury. Nevertheless, interference due to the presence of copper ions, which do not form a luminescent complex with **2**, can be observed only when saturation of **2** occurs; otherwise, all of the mercury is complexed, giving its characteristic luminescence. Finally, no interference was observed while performing titrations with Hg^{2+} in a complex matrix containing alkali, and earth alkali metal ions present in 100-fold excess.

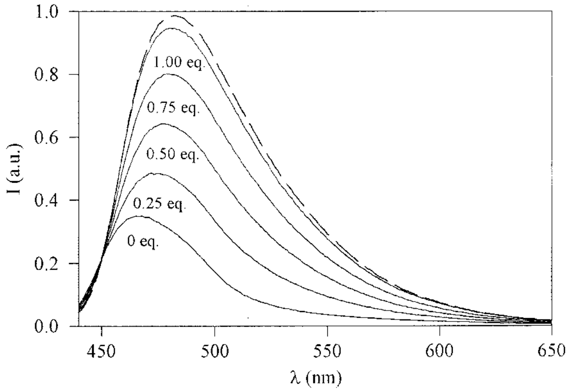

Figure 3. Fluorescence spectra of **2** (2.5×10^{-5} M) in methanol−water (1:1 v/v, pH 7.0) in the presence of 1 equiv of Cd^{2+} and Zn^{2+} upon addition of increasing amounts of Hg^{2+} (dashed line corresponds to the complex (**2**−Hg^{2+}) without other metal ions).

The selective response of **2** to Hg^{2+} is not completely unexpected. In an unrelated system, Winkler et al.[13] showed that a compound containing a 5-nitro-8-hydroxyquinoline group displayed a strong luminescent response to Hg^{2+}. However, these experiments were performed in an aprotic, noncompetitive solvent where high affinity would be expected. In **2**, the macrocyclic ring presumably provides a degree of ion affinity that results in stable complex formation in a protic medium. While other potential chemosensors for transition metal ions have been reported,[2,7,8,14] to our knowledge, no other demonstrates positive fluorescent response and the degree of selectivity for Hg^{2+} over other transition metal ions exhibited by **2**.

Ligand **2** displays the criteria necessary for a functional, fluorescent chemosensor for Hg^{2+}. The ligand displays good sensitivity, affinity, and selectivity for Hg^{2+} in a very large pH window (>2 pH units). Consequently, ligand **2** in solution or immobilized in a solid support may find applications for the determination of Hg^{2+} concentrations in waste effluent streams and drinking water. Further studies designed to increase the fluorescent intensity and the selectivity are in progress.

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Supporting Information Available: Experimental details, plots of derivatives of the titration curves in Figure 1, and results from titrations of **2** with Mg^{2+} and Ni^{2+} (PDF). This material is available free of charge via the Internet at http://pubs.acs.org.

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[11] The solubility of **2** in water is ca. 1×10^{-5} M. The behavior of **2** in the presence of metal cations was measured in methanol−water (1:1 v/v) to avoid working near the solubility limit and possible aggregation concentration of **2**. Nevertheless, the ion selectivities of **2** are expected to be the same in water as they are in the mixed aqueous solvent system (for an example of ion selectivities of macrocycles in methanol−water mixtures compared to selectivities in water see: Zhang, X. X.; Izatt, R. M.; Krakowiak, K. E.; Bradshaw, J. S. *Inorg. Chim. Acta* **1997**, *254*, 43).

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