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Biphenyl Appended BODIPY Derivatives Showing Combined Environmental Polarity and Heavy Metal Cation Sensing Functionality

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BODIPY derivatives showing a strong variation in photoluminescence quantum yield in solvents of different polarity were prepared. Further elaboration of these compounds with dithiacrown ether substituents provides additional Hg^{2+} and Pb^{2+} sensing functionality.

Dipyrrins, in particular, BODIPY derivatives have attracted significant attention due to their high stability, ease of preparation and interesting optical properties such as high photoluminescence quantum yields, narrow emission bands and large extinction coefficients. These properties have led to the application of BODIPY derivatives to a wide variety of applications including organic light emitting diodes, energy transfer cassettes, non-linear optics, biological labeling and chemical sensors with various modes of operation.¹

Development of efficient chemical sensors is of great importance in environmental, chemical process, pharmaceutical, biomedical and clinical monitoring applications.² Chemical sensors and chemodosimeters usually operate by reacting either reversibly (e.g. through guest-host interaction at a suitable binding site) or irreversibly with the analyte in such a way as to cause a change in the optical properties of the sensing units. For example, interaction with an analyte may cause a significant increase or decrease in the fluorescence properties of the sensing molecule. In general, chemical sensors responding to an analyte with an increase in fluorescence or a 'switch-on' mode of operation are more desirable. The fluorescence properties of BODIPY derivatives can often be strongly influenced when interaction with an analyte causes a perturbation of intramolecular charge transfer processes in the BODIPY derivative. In particular, the photoinduced electron transfer (PeT) process can act as a strong photoluminescence quenching mechanism in certain BODIPY compounds.³ If this process is then perturbed by interaction with an analyte, this can lead to a large increase in the photoluminescence quantum yield of the BODIPY derivative and a sensing event with a photoluminescence 'switch-on' mode of operation is realized. This basic principle has been employed in BODIPY derivatives to create sensors responding to stimuli including pH, environmental polarity and a wide variety of ionic species.4 ⁵ Of particular interest is the detection of heavy metal ions including Hg^{2+} , Pb^{2+} and Cd^{2+} , which are often highly toxic environmental pollutants.⁶ In this paper we present our studies into a series of BODIPY compounds, which show a strong fluorescence

response to changes in solvent polarity. This behavior, analogous to that shown in previously reported BODIPY derivatives is a result of the PeT-dependent fluorescence quenching mechanism, which is favoured in highly polar media.⁵ Based on these initial studies, the compounds were further modified to incorporate dithiacrown ether moieties, which are capable of binding heavy metal cations such as Hg^{2+} .⁷ We show that such compounds can act as highly sensitive switch-on fluorescence sensors for toxic heavy metal ions including Hg^{2+} and Pb^{2+} .



Scheme 1 Environmental polarity sensors 1-4 and Hg^{2+}/Pb^{2+} sensor 5.

Our initial investigations focused on the synthesis and photo-optical properties of compounds 1-4 (Scheme 1), which were prepared in moderate to high yields using a modified Suzuki-Miyaura coupling between 2,6-dibromo-BODIPY and the appropriate biphenyl boronic acid or acid esters as shown for compound 5 in Scheme 2. Synthesis of the biphenyl boronic acids and acid esters is shown in Scheme S1.⁸ All four compounds exhibit very similar UV/vis absorption spectra, which do not change drastically in a variety of solvents. The UV/vis absorption and fluorescence spectra for 10⁻⁵ M solutions of compounds 1-5 in CHCl₃ are shown in Figure S1. A maximum absorption of around $\lambda_{max} = 578$ nm is observed for each of the compounds with an extinction coefficient of ϵ = 4.6 \times $10^4~M^{-1}~cm^{-1}$ (compound 1). Compounds 1-5 are soluble in a wide variety of organic solvents across a broad range of polarity and although not water soluble, 5 can form stable solutions in up to 40% v/v water in acetonitrile. In solvents of low polarity, the compounds are highly fluorescent with quantum yields as high as $\Phi = 0.70$ for compounds 2 and 4 in *n*-hexane. Peak emission wavelengths of between 624 and 627 nm were observed. Whereas the UV/vis absorption properties show very little variation in solvents of different polarity, all of the

compounds show a dramatic decrease in fluorescence quantum yields in solvents with increasing dielectric constant such that in highly polar media like DMSO and MeCN, fluorescence is almost completely quenched in the case of compounds 1 and 2 and reduced to around $\Phi = 0.30$ for compounds 3 and 4. This behaviour is analogous to that observed in a series of BODIPY-based environmental polarity sensors designed by Nagano and co-workers and is due to a PeT quenching mechanism.⁵ Here, photoelectron transfer occurs from the dialkoxybenzene moiety attached to the *ortho* positions of the benzene rings on the 2,6-dibenzoBODIPY chromophore. The PeT process becomes less favoured with decreasing solvent polarity due to an increase in the oxidation potential of the dialkoxybenzene moiety in the lower polarity solvent. Thus in low polarity solvents, the main quenching process is inhibited and a fluorescence switch-on effect occurs.⁵



Scheme 2 Synthesis of **5** through a modified Suzuki-Miyaura coupling reaction.

¹H NMR spectra of compounds 1 and 2 in chloroform show an unexpected splitting of a number of the proton resonances. The mesityl aromatic proton and the mesityl 2-methyl proton resonances are split into three peaks with integrals of 1:2:1 ratio. Also, the BODIPY β proton resonances are split slightly but only resolved sufficiently to reveal two peaks. The splitting is not observed in compounds 3 and 4 that do not have a methyl group attached to the dimethoxybenzene substituents. It is likely that this methyl group restricts the rotation of the dimethoxybenzene substituent in compounds 1 and 2 and that in turn leads to an energy barrier between different conformers (atropisomers) that cannot be easily overcome on NMR timescales at lower temperatures. VT NMR studies using compound 1 in 1,1,2,2-tetrachloroethane show that the split resonances quickly coalesce with increasing temperature as the conformational energy barrier is overcome. From the VT NMR measurements, the energy barrier was estimated to be 87 kJ.mol⁻¹ in CDCl₃ which is reasonably consistent with energy barriers measured previously for 2,2'-disubstituted biphenyl derivatives.⁹ Compounds 3 and 4, which do not have the methyl group are less conformationally restricted and so show no splitting of the ¹H NMR resonances. This is exemplified in Figure 1, which shows the crystal structures of compounds 3 and 4. Although structurally very similar, the two compounds adopt very different conformations in the crystalline state, which illustrates that these compounds are relatively unrestricted conformationally. Compound 3 has a P-1 space group whearas compound 4 shows a more symmetrical C2/c space group. A significant difference is noted in the dihedral angles formed between the BODIPY pyrrole units and the appended phenyl groups. Compound 3 shows a dihedral angle of 37.5° between carbons C18-C17 and C19-C24 (Figure S4a) and 49.5° between carbons C14-C13 and C31-C36. A much larger dihedral angle of 135.6° between carbons C2-C11 and C10-C9 (Figure S4b) is observed for compound 4. Unfortunately, attempts to prepare crystals of compounds 1 and 2 suitable for single-crystal X-ray

diffraction analysis proved unsuccessful. Again, this may be a result of the higher conformational barriers in these compounds.

Based on our observations for compounds 1-4, we aimed to synthesise analogous compounds with a suitable cation-binding site on the photoinduced electron donating substituent. A cation binding event at this site is expected to further increase the oxidation potential of the substituent which should then cause a 'switch on' of the BODIPY fluorescence in solvents of appropriate polarity. In the case of compound 1, the two methoxy substituents can be replaced with an appropriate crown ether unit for recognition of a variety of metal cations.



Fig. 1 Crystal structures of (a) compound **3** and (b) compound **4** illustrating the very different conformations adopted by these structurally quite similar compounds.

Here we utilised a benzo-18-crown-6 ether unit with two of the oxygen atoms replaced with sulphur atoms, as this macrocycle is known to act as a highly selective host for Hg^{2+} cations.⁷ The final Suzuki-Miyaura coupling reaction for the preparation of compound **5** was found to be much slower than in the case of compounds **1-4**, possibly due to steric hindrance from the crown ether unit. A longer reaction time at an elevated temperature of 50 °C was necessary to give an isolated yield of 9%.

As expected, compound 5 was found to exhibit a similar solventdependent fluorescence quantum yield to that observed in compounds 1-4. The Hg^{2+} binding properties of compound 5 were investigated by monitoring fluorescence and UV/vis changes upon addition of Hg^{2+} to solutions of 5 in a variety of solvents. In highly polar media such as MeCN, the fluorescence of 5 is almost completely quenched ($\Phi = 0.01$) due to PeT from the benzodithacrown-ether moiety to the BODIPY fluorophore. Addition of Hg²⁺ ions leads to a very slight increase in fluorescence ($\Phi = 0.05$) although a significant 'switch-on' effect is not observed, even in the presence of a large excess of Hg²⁺. Although the dithacrown ether macrocycle does recognise Hg^{2+} ion in this solvent, the high polarity of the media results in a very low oxidation potential of the dithiabenzocrown-ether substituent and the Hg²⁺ binding event is likely not sufficient to raise the oxidation potential enough to prevent PeT to the BODIPY fluorophore. In the case of a less polar solvent, such as dichloromethane, PeT is already somewhat inhibited due to the higher oxidation potential of the benzo-dithacrown-ether substituent. So even in the absence of Hg^{2+} ion, significant fluorescence ($\Phi = 0.30$) is observed. This fluorescence is increased upon addition of Hg^{2+} ion ($\Phi = 0.40$), but as the compound was already significantly fluorescent (i.e. the PeT was already significantly inhibited in the low polarity media), a large overall

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change in fluorescence is not visually observed. In order to observe the most significant fluorescence 'switch-on' effect upon cation binding, it is necessary to select a solvent of intermediate polarity. Here, we chose a 1:5 MeCN:DCM solvent mixture in which compound **5** is quite weakly fluorescent ($\Phi = 0.04$). A 10^{-5} M solution of **5** in 1:5 MeCN:DCM was prepared and changes in the UV/vis absorption and fluorescence were monitored upon addition of aliquots of 10^{-3} M mercury (II) perchlorate in the same solvent system. Figure 2 shows the changes in UV/vis absorption and fluorescence upon Hg²⁺ addition. The UV/vis spectrum shows an initial λ_{max} of 577 nm, which undergoes an 11 nm bathochromic shift to 588 nm upon addition of 4 equivalents of Hg²⁺. Changes in fluorescence spectra were more dramatic with an initial λ_{max} of 626 nm and intensity of 590 (a.u) shifting slightly to a λ_{max} of 629 nm but with a much higher intensity of 4293 (a.u) corresponding to a change

in fluorescence quantum yield from $\Phi = 0.04$ to $\Phi = 0.24$. In order to check the selectivity for the Hg^{2+} cation, 4 equivalents of a variety of cations were added to solutions of 5 and the change in fluorescence observed. Results are shown in Figure 3. As well as responding to ${\rm Hg}^{2^+}$ cations, a somewhat lower response is also observed in the case of ${\rm Ag}^+,~{\rm Pb}^{2^+}$ and ${\rm Sr}^{2^+}$ cations. Although a response to Ag⁺ was expected given a similar response reported with a fluorescence sensor using an identical dithiacrown ether cation binding unit, the response to Pb²⁺ and Sr²⁺ was unexpected as these cations did not show a response with the previously reported sensor.⁷ The response to Pb^{2+} and Sr^{2+} cations may be due to secondary interactions with the BODIPY BF2 unit. Further additions of each of the cations up to 20 equivalents showed no change in the fluorescence response to Hg^{2+} and Ag^+ suggesting that cation binding is fully saturated upon addition of 4 equivalents. However, in the case of \mbox{Pb}^{2+} and $\mbox{Sr}^{2+},$ a further increase in fluorescence response is observed, such that in the case of Pb²⁺ ion, the saturated fluorescence intensity is somewhat higher ($\Phi = 0.36$) than that observed in the case of Hg^{2+} ion.



Fig. 2 (a) UV/Vis changes of 10^{-5} M **5** in 5:1 DCM:MeCN upon titration with Hg(ClO₄)₂. Inset shows the binding isotherm at 588 nm (blue diamonds) and the calculated 1:2 binding curve (solid line). (b) PL changes of 10^{-5} M **5** in 5:1 DCM:MeCN upon titration with Hg(ClO₄)₂.

From the UV/vis spectral changes, a titration curve was obtained (Figure 2 (a) inset) and a nonlinear least-squares regression analysis

supported a 1:2 5:Hg²⁺ stoichiometry with association constants $\log K_1 = 7.7 \pm 0.6$ and $\log K_2 = 5.7 \pm 0.1$.¹⁰ The lower value of K₂ is probably a result of electrostatic repulsion.¹¹ The 1:2 ligand to Hg²⁺ binding mode was confirmed by Job's plot analysis (Figure S2 ESI). The fluorescence spectral changes upon Hg²⁺ titration show that despite the high value for the first binding constant ($\log K_1 = 7.7$) there is very little increase in fluorescence until at least 1 equivalent of Hg²⁺ is added. This is to be expected given that binding to one dithiabenzorown ether substituent still leaves an unbound dithiabenzorown ether substituent from which PeT quenching can easily occur.



Fig. 3 Fluorescence changes $(I-I_0)$ at 626 nm for 10^{-5} M solutions of 5 in 1:5 MeCN:DCM upon addition of 4 and 20 equivalents of various metal cations.

Binding of a second Hg^{2+} cation leads to an increase in oxidation potential of both dithiabenzocrown ether substituents and the PeT quenching mechanism is thus inhibited leading to a large increase in fluorescence. Neither Hg^{2+} nor Pb^{2+} sensing properties were significantly affected by the presence of other cations, as confirmed by addition of an excess of a variety of competing cations (Figure S3 ESI).

In conclusion, a series of BODIPY derivatives has been synthesised whose fluorescence properties can be strongly influenced by environmental polarity. Further elaboration of these derivatives with dithiacrown ether units at appropriate positions gives compounds whose fluorescent properties are not only affected by environmental polarity, but also through binding of appropriate cations including the highly toxic heavy metals Hg^{2+} and Pb^{2+} . We anticipate that these, or similar compounds might find application as a tool for the detection of environmental polarity changes in biological samples as well as for monitoring Hg^{2+} and Pb^{2+} contamination in such samples.

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