Supporting Information for:

**Fluorophore Incorporation Allows Nanomolar Guest Sensing and White-Light Emission in M₄L₆ Cage Complexes**

Prakash P. Neelakandan, Azucena Jiménez, and Jonathan R. Nitschke*

Department of Chemistry, University of Cambridge, Lensfield Road, Cambridge CB2 1EW, United Kingdom

**Experimental Section**

**General experimental techniques:** All reagents and solvents were purchased from commercial sources and used as supplied. All experiments were carried out at room temperature. Spectroscopy grade acetonitrile was used for UV-Vis absorption and fluorescence measurements and was distilled prior to use. Microwave reactions were performed with a CEM Discover microwave reactor. NMR spectra were recorded using either a Bruker DRX-400, a Bruker AVC-500-BB or a Bruker AVC-500-TCI spectrometer. Chemical shifts are reported in parts per million (δ) calibrated to the residual solvent signals: δ_H with [D₃]acetonitrile at 1.940 ppm and δ_C with [D₃]acetonitrile at 118.260 ppm.¹⁹F NMR values are calibrated to the internal standard hexafluorobenzene (−164.90 ppm) or the counter anions triflate (−79.5 ppm) or triflimide (−80.4 ppm).¹ High resolution mass spectrometry was performed on a Waters LCT Premier Mass Spectrometer featuring a Z-spray source with electrospray ionization and modular LockSpray interface. Molecular modelling was done using the modified MM2 force field of CAChe Workspace, WorkSystem Pro Version 7.5.0.85. UV-Vis absorption measurements were performed on a Perkin-Elmer Lambda 750 spectrometer. Fluorescence measurements were performed on a Cary Eclipse fluorescence spectrophotometer. Rhodamine 110 in basic ethanol (Φ_F = 0.92) or quinine sulphate in 1 N H₂SO₄ (Φ_F = 0.55) were used as the standards.² Quantum yields of fluorescence were calculated using the equation 1,

\[
\Phi_u = \frac{A_s F_u n_u^2}{A_u F_s n_s^2} \Phi_s
\]  

(1)

where \( \Phi \) represents the fluorescence quantum yield, \( A \) the absorbance, \( F \) the areas of fluorescence peaks and \( n \) the refractive index of the solvent used. The subscripts “s” and “u” denote these parameters for the standard and unknown compounds, respectively.
**Synthesis**

**Synthesis of bis(aminophenyl)BODIPY 1**

The bis(nitrophenyl)BODIPY 9 (Scheme S1) was synthesized as reported elsewhere. A solution of the bis(nitrophenyl)BODIPY 9 (100 mg, 0.18 mmol) in methanol (20 mL) was stirred at room temperature followed by the addition of 10 % Pd/C (15.0 mg). The reaction mixture was degassed and stirred at room temperature for 5 h under an atmosphere of dihydrogen. The solution was filtered through Celite, concentrated in vacuo, and purified via flash column chromatography over silica gel using dichloromethane as eluent followed by ethyl acetate to yield 1 as a dark green powder (86 mg, 97 %). $^1$H NMR (400 MHz, CD$_3$CN): $\delta$ = 2.12 (s, 6H), 2.38 (s, 3H), 4.27 (s, 4H), 6.61 (d, $J$ = 8 Hz, 4H), 6.70 (s, 2H), 7.06 (s, 2H), 7.32 (d, $J$ = 8.0 Hz, 4H), 8.26 (s, 2H); $^{13}$C NMR (100 MHz, CD$_3$CN): $\delta$ = 20.0, 21.2, 115.5, 122.3, 127.5, 129.0, 130.7, 135.8, 137.0, 137.3, 139.8, 142.5, 145.7, 148.8; $^{19}$F NMR (376 MHz, CD$_3$CN): $\delta$ = –147.8 to –148.1 (m, 2F); HRMS (ESI): $m/z$: calculated for C$_{30}$H$_{27}$N$_4$BF$_2$ [M–H]$^+$: 493.2370, found: 493.2351.

![Synthesis of bis(aminophenyl)BODIPY 1](image)

**Scheme S1.** Synthesis of subcomponent 1. (i) (a) TFA, 5 min; (b) toluene, DDQ, RT, 2 h; (c) DCM, TEA, BF$_3$·OEt$_2$, RT, 30 min, 35 %; (ii) NBS, 1:1 DCM:DMF, 25 °C, 8 h, 80 %; (iii) 4-nitrophenylboronic acid, Pd$_2$(dba)$_3$·CHCl$_3$, tBu$_3$P·HBF$_4$, Cs$_2$CO$_3$, THF : water, 25 °C, 24 h, 65 %; (iv) H$_2$/Pd-C, MeOH, 25 °C, 5 h, 95 %.

**Synthesis of 5-(pyren-1-yl)picolinaldehyde (2)**

![Synthesis of 5-(pyren-1-yl)picolinaldehyde (2)](image)

**Scheme S2.** Synthesis of subcomponent 2. (i) Pd(PPh$_3$)$_4$, aq. NaOH (1 M), toluene, 100 °C, 30 min, 200 W microwave, 65 %.
As shown in Scheme S2, the coupling between pyren-1-ylboronic acid (10) and 5-bromopicolinaldehyde (11) under Suzuki conditions yielded the pyrene appended 2-formylpyridine (2). In a vial suitable for microwave reactions, pyrene-1-boronic acid (10, 160 mg, 0.65 mmol), 5-bromo-2-formylpyridine (11, 100 mg, 0.54 mmol), and tetrakis(triphenylphosphine)palladium(0) (6.2 mg, 0.0054 mmol) were suspended in a mixture of toluene (8 mL) and 1 M aqueous NaOH solution (4 mL). The vial was flushed with nitrogen, capped, and heated in the microwave reactor at 100 °C for 30 minutes at 200 W. The reaction mixture was cooled down to room temperature, washed with water (3 × 20 mL), dried over MgSO4 and concentrated in vacuo to yield a dark yellow residue. The product was isolated by column chromatography over silica gel using dichloromethane as eluent to yield 2 as yellow crystals (108 mg, 65%).

1H NMR (400 MHz, CD3CN): δ = 8.20 (m, 11H), 9.06 (s, 1H), 10.16 (s, 1H); 13C NMR (100 MHz, CD3CN): δ = 122.2, 125.0, 125.2, 125.5, 125.9, 126.4, 126.8, 127.6, 128.3, 128.7, 129.1, 129.38, 129.44, 131.7, 132.3, 132.5, 133.5, 139.9, 141.9, 152.4, 152.6, 194.4; HRMS (ESI): m/z: calculated for C22H13NO [M–H]+ 308.1070, found: 308.1073.

**General procedure for the preparation of cages:** In a Teflon-capped J-Young NMR tube, 1 (0.0203 mmol), either 2 or 3 (0.0406 mmol), and either FeII(OTf)2 (0.0136 mmol) or ZnII(NTf2)2 (0.0136 mmol) were dissolved in CD3CN (0.5 mL). The solution was degassed by three evacuation/nitrogen-fill cycles and heated at 70 °C overnight. After cooling down to room temperature, the solvent was removed and the residue was washed with diethyl ether to yield the self-assembled structures.

4: 1H NMR (400 MHz, CD3CN): δ = 2.14 to 2.19 (36H, m), 2.41 to 2.47 (18H, m), 5.27 to 5.33 (24H, m), 7.00 to 7.38 (36H, m), 7.46 to 7.55 (24H, m), 7.69 to 7.73 (12H, m), 8.32 to 8.47 (24H, m), 8.57 to 8.64 (12H, m), 8.71 to 8.78 (12H, m); 13C NMR (100 MHz, CD3CN): δ = 19.8 to 19.9, 20.8 to 21.1, 122.3 to 122.6, 125.1 to 125.5, 126.8 to 127.4, 128.9 to 129.1, 129.8 to 129.9, 130.2 to 130.3, 131.5 to 131.7, 133.0 to 133.8, 136.8 to 137.1, 140.0 to 140.2, 143.2 to 143.4, 143.8 to 144.1, 144.5, 148.3, 149.6 to 150.0, 156.4, 158.9, 175.1 to 175.4; 19F NMR (376 MHz, CD3CN): δ = –79.5 (OTf), –143.2 to –144.1, –144.3 to –144.8, –145.3 to –145.5, –146.4 to –146.9, –147.9 to –148.5.

5: 1H NMR (400 MHz, CD3CN): δ = 2.22 to 2.30 (36H, m), 2.40 to 2.45 (18H, m), 5.71 to 5.76 (24H, m), 7.03 to 8.48 (180H, m), 8.72 to 8.90 (24H, m), 9.05 to 9.13 (12H); 13C NMR (100 MHz, CD3CN): δ = 20.1 to 20.3, 21.1 to 21.2, 122.8 to 123.6, 124.6, 125.1, 125.8, 126.3 to 126.5, 127.1 to 127.3, 127.5, 128.0, 129.2 to 129.3, 129.6, 130.1, 130.8, 131.9, 132.8, 133.5, 137.1 to 137.5, 140.4, 141.5 to 141.7, 142.8, 143.6, 148.7, 157.2 to 157.3, 157.6 to 157.8, 175.4 to 175.7; 19F NMR (376 MHz, CD3CN): δ = –79.5 (OTf), –142.7 to –144.3, –145.1 to –145.3, –146.0 to –146.5, –147.4 to –148.0.

6: 1H NMR (400 MHz, CD3CN): δ = 2.08 to 2.19 (36H, m), 2.35 to 2.46 (18H, m), 6.06 to 6.27 (24H, m), 6.97 to 7.20 (24H, m), 7.50 to 7.57 (24H, m), 7.82 to 7.98 (24H, m), 8.07 to
8.21 (12H, m), 8.39 to 8.60 (36H, m); $^{19}$F NMR (376 MHz, CD$_3$CN): $\delta = -80.5$ (NTf$_2$), –143.4 to –144.8, –145.1 to –146.1, –146.7 to –147.5.

**Figure S1.** (A, B) UV-Vis absorption and (C, D) fluorescence spectra of the subcomponents 1 (3.5 µM) and 2 (5 µM) and the metallo-supramolecular cages 4 (65 nM), 5 (110 µM), and 6 (75 nm) in acetonitrile, $\lambda_{ex} =$ 326 nm. Insets show the fluorescence of 4, 6, 2 and 5 in acetonitrile illuminated with a hand held UV lamp.
Figure S2. UV-Vis absorption (red), fluorescence emission (blue) and fluorescence excitation (black) spectra of the subcomponent 2 (4 to 7 µM) in various solvents, $\lambda_{ex} = 357$ nm. Excitation spectra were collected at the respective emission maxima.

Figure S3. Temperature-dependent changes in the fluorescence emission and excitation spectra of the subcomponent 2 (10 µM) in acetonitrile, $\lambda_{ex} = 357$ nm. Excitation spectra were collected at the respective emission maxima.
**Figure S4.** Partial $^1$H NMR spectra of the subcomponents (A) 1 and (B) 2 and the cages (C) 4, (D) 5, (E) and 6 in CD$_3$CN.
**Figure S5.** Partial $^{19}$F NMR spectrum of the subcomponent (A) 1 and the cages (B) 4, (C) 5, and (D) 6 in CD$_3$CN.

**Figure S6.** High-resolution mass spectrum of cage 4 along with the simulated and experimental isotopic distributions as inset.
**Figure S7.** High-resolution mass spectrum of cage 5 along with the simulated and experimental isotopic distributions as inset.

**Figure S8.** High-resolution mass spectrum of cage 6 along with the simulated and experimental isotopic distributions as inset.
Figure S9. (A) DOSY, (B) $^1$H-$^1$H COSY, and (C) NOESY spectra of cage 4 in CD$_3$CN.
Figure S10. (A) Partial $^1$H NMR spectrum of cage 4 (1 mM), and in the presence of 2 equivalents of tetrabutylammonium salts of (B) acetate and (C) azide in CD$_3$CN.

Figure S11. Partial $^{19}$F NMR spectrum of the cage 4 (1 mM) (A) in the absence and (B) presence of 2 equivalents of tetrabutylammonium acetate in CD$_3$CN. Insets show the region between $-140$ to $-150$ ppm.
Figure S12. Changes in the (A) UV-Vis absorption and (B) fluorescence spectra ($\lambda_{\text{ex}} = 590$ nm) of 5 (110 nM) with the addition of tetrabutylammonium acetate (0→5.77 µM) in acetonitrile. Inset of (B) shows the corresponding fluorescence spectra excited at 333 nm.

Figure S13. UV-Vis absorption spectra of cages (A) 4, (B) 5, and (C) 6 in the absence and presence of tetrabutylammonium acetate in acetonitrile.

Figure S14. Changes in the fluorescence spectrum of cage 6 (75 nM) with the addition of tetrabutylammonium acetate (0→5.77 µM) in acetonitrile.
Figure S15. Relative changes in the fluorescence intensity of cages 4 (67 nm), 5 (110 nm), and 6 (75 nM) at 653 nm with the successive addition of tetrabutylammonium acetate (0-5.77 μM), $\lambda_{ex} = 590$ nm.

Figure S16. Changes in the UV-Vis absorption spectrum of 1 (3.5 μM) following the addition of (A) tetrabutylammonium acetate (0→5.77 μM) in acetonitrile and (B) L-cysteine (0-49.5 μM) in 50% acetonitrile-water mixture.

Figure S17. Time-dependent changes in the (A) UV-Vis absorption and (B) fluorescence spectra ($\lambda_{ex} = 580$ nm) of 4 (150 nM) in the absence and presence of EDTA in acetonitrile. As EDTA is not highly soluble in acetonitrile, it was added as solid to a solution of 4. The resulting solution was filtered and used for optical measurements. For comparison, an absorption spectrum of 1 in acetonitrile is also shown in (A).
**Figure S18.** Changes in the (A) UV-Vis absorption and (B) fluorescence spectra of 5 (110 nM) following the addition of perylene (0→12.9 µM) in acetonitrile, $\lambda_{ex} = 590$ nm.Inset of (B) shows the corresponding spectra when excited at 333 nm.

**Figure S19.** Relative changes in the fluorescence intensity of 5 (110 nM) at 653 nm with the addition of various polycyclic aromatic hydrocarbons (PAHs) in acetonitrile.

**Figure S20.** Absorption (red) and fluorescence spectra (blue) of a mixture of 1 (5.5 µM), 2 (11 µM) and perylene (16.6 µM) in acetonitrile, $\lambda_{ex} = 365$ nm.
Figure S21. Relative changes in the fluorescence intensity of cage 4 (52 nM) at 653 nm with the addition of amino acids in 50% acetonitrile-water mixture.

References