Electronic Supplementary Information

Stabilisation of a Heptamethine Cyanine Dye by Rotaxane Encapsulation

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Section 1: Experimental procedure for synthesis of $1 \subset \alpha$ -CD·PF₆



Tetramethyl indolium iodide **3** (5.00 g, 16.6 mmol), glutaconaldehyde acetate **2** (1.17 g, 8.36 mmol) and α -cyclodextrin (70.0 g, 72.0 mmol) were suspended in degassed potassium acetate solution (aq. 1.0 M, 100 cm³) and stirred at 80 °C under nitrogen for 48 h. After cooling, water (80 cm³) and dichloromethane (2000 cm³) were added with stirring. The resulting precipitate of α -cyclodextrin-dichloromethane complex was removed by filtration. The filtrate was then transferred to a separating funnel and the aqueous layer was extracted with dichloromethane until the organic layer was colourless.

The aqueous phase was purified by ion exchange chromatography on a Sephadex C-25 column (8 cm × 20 cm), eluting with water, then with 0.10 M aqueous ammonium bicarbonate and collecting the blue-green fraction. (The column was regenerated by a washing cycle of 1.0 M ammonium bicarbonate and water.) The solution was then dried under reduced pressure at 60 °C. The pale blue-green solid obtained was then dissolved in methanol (100 cm³) and filtered to remove some insoluble impurities. The green solution was dried under reduced pressure to yield a dark green solid. The solid was then dissolved in water (200 cm³) and ammonium hexafluorophosphate (1.6 g) was added. Upon cooling to 4 °C for 48 h, $1 \subset \alpha - CD \cdot PF_6$ (1.70 g, 13.3 %) was obtained as a brown-red solid (Found: C, 47.32; H, 6.60; N, 1.63; P, 1.92. $C_{65}H_{93}N_2O_{30}PF_6$ requires C, 47.21; H, 6.52; N, 1.69; P, 1.87); λ_{max} (MeOH) 770 nm; $\varepsilon = 2.82 \times 10^5 \text{ M}^{-1} \text{ cm}^{-1}$; δ_{H} (500 MHz, d_6 -DMSO) 1.59 (s, 6H, $Me_{e1'/e2'}$, 1.72 (s, 3H, Me_{e1}), 1.73 (s, 3H, Me_{e2}), 3.32 (m, 6H, H-2), 3.47 (t, 6H, J = 9.1, H-4), 3.56 (s, 3H, Me_f), 3.58 (m, 6H, H-6), 3.62 (s, 3H, Me_f), 3.64 (m, 6H, H-5), 3.68 (m, 6H, H-6), 3.77 (t, 6H, J = 9.2, H-3), 4.60 (t, 6H, J = 5.2, OH-6), 4.84 (d, 6H, J = 1.7, H-1), 5.49 (s, 6H, OH-3), 5.65 (d, 6H, J = 6.5, OH-2), 6.17 (d, 1H, J = 14.5, H_a), 6.19 (d, 1H, J = 13.6, $H_{a'}$), 6.47 (t, 1H, J = 12.6, $H_{c'}$), 6.69 (t, 1H, J = 12.7, H_{c}), 7.06 (t, 1H, J = 12.8, $H_{d'}$), 7.23–7.57 (m, 8H, aromatic), 7.45 (t, 1H, H_b), 7.63 (t, 1H, J = 13.6, *H_b*[']); δ_C (500 MHz, *d*₆-DMSO): 27.00, 27.37, 27.99, 31.15, 31.20, 48.58, 48.63, 59.77, 72.06, 72.32, 73.64, 81.64, 101.86, 104.41, 104.88, 111.13, 111.19, 121.98, 122.21, 124.71, 124.79, 126.65, 127.29, 128.42, 140.73, 141.18, 142.83, 143.06, 147.53, 147.86, 150.23, 171.21, 171.39; *m/z* (ES+) 1381.70 (100%, M⁺; C₆₅H₉₃N₂O₃₀⁺ requires 1381.58); mp 242–244°C (dec.).

The trifluoroacetate salt $1 \subseteq \alpha$ -CD·CO₂CF₃ was prepared using the above procedure except that after ion exchange chromatography, TFA was added to the aqueous solution of the bicartbonate salt of the rotaxane until it reached pH 4; the solution was then evaporated to dryness. $1 \subseteq \alpha$ -CD·CO₂CF₃ was also prepared from $1 \subseteq \alpha$ -CD·PF₆ by passing an aqueous solution of the rotaxane through a Dowex 1X8 anion exchange column (which had previously been loaded with trifluoroacetate by elution with 0.10 M NaOH followed by 1% aqueous TFA).





Figure S1. Part of the ¹H/¹H ROESY spectrum of $1 \subset \alpha$ -CD·CF₃CO₂ (500 MHz, d_6 -DMSO) showing correlations between the methyl groups Me_{e1/e2} and Me_{e1'/e2'} to the dye backbone and the cyclodextrin unit (Me_{e1/e2} to H3, OH3 and OH2; Me_{e1'/e2'} to H6, H6' and OH6).



Figure S2. Part of the ¹H/¹H ROESY spectrum of $1 \subset \alpha$ -CD·CF₃CO₂ (500 MHz, d_6 -DMSO) showing correlations of the polymethine chain to cyclodextrin resonances (H_c, H_d and H_c⁻ to H5; H_a, H_b and H_c to H3).



Figure S3. Part of the ¹H/¹H ROESY spectrum of $1 \subset \alpha$ -CD·CF₃CO₂ (500 MHz, d_6 -DMSO) showing correlations within the dye backbone.

Section 3: X-Ray Crystallographic Analysis

Single crystals of $1 \subset \alpha$ -CD·PF₆ were grown by slow cooling of a saturate aqueous solution (40° to 5°C). The crystals produced promising X-ray diffraction patterns, on both laboratory and synchrotron sources, but the structure was extremly difficult to solve and refine by conventional methods because of the large size of the unit cell and asymmetric unit, the weak scattering of the atoms involved and the extensive disorder of solvating water molecules and counterions, which challenge the boundaries of small-molecule single crystal analysis.

A single crystal was mounted on a two-stage fiber using Fomblin Y-1800 oil. The two-stage fiber was constructed from a 10–20 µm glass wool fiber glue mounted to a 0.1 mm borosilicate capillary, to reduce background scatter and enable selection of small crystals. The sample was cooled to 120 K using an Oxford Cryosystem Cobra+.¹ The data collection was undertaken on a Bruker-Nonius APEXII diffractometer using a Bruker Nonius APEXII² CCD area detector, controlled by BIS 2.0.1.9³ and APEX 2.1 on Station 9.8, SRS, Daresbury UK.⁴ A "full-sphere" data collection strategy using three 180° ω scans at three φ settings 120° apart was used. Saint-7.34a⁵ was used to integrate the data and SADABS 2004⁶ used to scale the resulting merged raw file; λ /2 correction was not applied; no beam decay correction was applied. The optimum data set (reported here) was collected at 0.68140 Å.⁴ Variable temperature data collections ranging from 85 to 180 K were attempted, however the ambient temperature of 120 K gave the optimal diffraction pattern.

The structure was solved by direct methods using the program SIR2004.⁷ The refinement (on *F*) and graphical calculations were performed using the CRYSTALS program suite⁸ and using SHELX.⁹ The disorder in the solvent molecules does not affect the main part of each of the the rotaxane molecules, which are very well behaved; it should not therefore impact on the discussions of the intra- and intermolecular distances and angles in the main components.

Due to the poor quality of the data, refinement of the Flack parameter resulted in an ambiguous value due to its large esd. Instead, an alternative model where the Friedel pairs were merged before the refinement (and as such, were the Flack parameter was not refined) is reported here. Due to the low number of observed data, anisotropic atomic displacement parameters were only included in the model for the rotaxane core. There is a large number of disordered water molecules in the asymmetric unit. Hydrogen atoms attached to oxygen could not be identified by examination of a difference Fourier map. Hydrogen atoms on carbon atoms were placed geometrically after each cycle. All PF_6 counterions are disordered and were idealised. Phenyl rings in the rotaxane thread severely disordered refinement with similarity restraints

Cambridge Crystallographic Data Centre deposition number: CCDC 661299; formula: C260 H298 F24 N8 O134 P4; unit cell parameters: a = 24.8064(4) Å, b = 20.5004(3) Å, c = 30.7944(5) Å, beta = 94.643(1)°, space group P2₁.

Section 4: Photobleaching Kinetics



Figure S4. Photodegradation of $1 \subset \alpha$ -CD·CF₃CO₂ and $1 \cdot$ CF₃CO₂ in water during irradiation with a continuous-wave Ti:sapphire laser (Coherent Mira; *ca.* 300 mW) at 752 nm. Both samples were at the same concentration, and had the same absorbance at 752 nm. Data are fitted to first-order decay curves with rate constants of $7.5 \times 10^{-4} \text{ s}^{-1}$ and $2.9 \times 10^{-3} \text{ s}^{-1}$ for the $1 \subset \alpha$ -CD and 1 respectively.

Section 5: Electrochemistry

Electrochemistry experiments were performed using an Autolab PGSTAT12 potentiostat, giving the cyclic voltammograms plotted in Figure 4. Compounds were dissolved in DMSO containing Bu_4NPF_6 (0.10 M), under argon. A 3 mm glassy carbon working electrode was used with a Pt wire counter electrode and a Ag/AgNO₃ pseudo-reference electrode. The redox potentials were measured using square-wave voltammetry (step potential 0.002 V; amplitude 0.02 V, frequency 8 Hz), and referenced to internal ferrocinium/ferrocene at 0 V.

Section 6: Chemical Oxidation

<u>Generation of radical dication of $1 \subseteq \alpha$ -CD</u>: A solution of ceric ammonium nitrate (CAN, Aldrich) in dry DMSO (dried by reduced pressure distillation from CaH₂) was added in portions to a solution of $1 \subseteq \alpha$ -CD·PF₆, also in DMSO, under nitrogen until the UV-vis-NIR spectrum (recorded in 1 cm cuvettes using a Varian Cary 5E spectrometer) showed the absorption from $1 \subseteq \alpha$ -CD·PF₆ had largely disappeared. The resulting spectrum (Figure 5c of paper) is assigned to the radical dication due to its similarity to that of previously reported cyanine radical dications¹⁰ and to TD-DFT spectra (vide infra), and based on ESR spectroscopy (vide infra). Over two hours, the radical dication absorptions slowly decayed to give a spectrum similar to that of $1 \subseteq \alpha$ -CD (Figures S5 and S6) with an additional feature at ca. 400 nm. MALDI mass spectrometry and GPC traces are identical before and after the oxidation / decomposition experiment, suggesting that $1 \subseteq \alpha$ -CD is indeed regenerated. Presumably, the dication is reduced slowly by the solvent (or by impurities in the solvent), with the side products of this redox reaction absorbing around 400 nm.



Figure S5. Decay of the one-electron oxidation product of $1 \subset \alpha$ -CD·PF₆ to give a cyanine-like product.



Figure S6. Absorption spectra of $1 \subset \alpha$ -CD·PF₆ before oxidation, and after oxidation and decay of the oxidized species.

Figure S7 shows spectra recorded during the addition of increasing amounts of CAN to the free dye, $1 \cdot PF_6$. While the absorption at ca. 550 nm seen in some of the spectra is similar to that of the dication formed from the rotaxane, this species has a lifetime of a few seconds; the final spectrum is very different from those seen for oxidized $1 \subset \alpha - CD \cdot PF_6$ and from those of other cyanine radical cations. Moreover, the product cannot be converted back to 1, indicating that oxidation leads to rapid irreversible decomposition of the free dye.



Figure S7. Absorption of $1 \cdot PF_6$ in DMSO (1), and with the addition of increasing amounts of CAN in DMSO (2–5).

Section 7: ESR Spectroscopy

Room-temperature ESR spectra were acquired using a Bruker EMX spectrometer. Solutions were generated by addition of < 0.1 eq. CAN in dry DMSO to $1 \subset \alpha$ -CD·PF₆ in dry DMSO in 2 mm Suprasil ESR tubes. Figure 5b compares the experimental spectrum with that simulated using values from DFT calculations (vide infra). The DFT coupling constants are consistently slightly overestimated and so are scaled by a factor of 0.9.

Section 8: DFT calculations

The geometry optimizations of the cation and radical dication, and the calculations of the hyperfine splittings (isotropic Fermi contact couplings) for the latter system, were performed at the Density Functional Theory (DFT) level. The excitation energies and transition dipole moments of the low-lying excited states of both species were calculated at the time-dependent (TD) DFT level. All DFT and TD-DFT calculations were performed at the spin-unrestricted level using the B3LYP functional and the 6-31G** basis set as implemented in the Gaussian-03 program.¹¹

The optimized geometry of the radical dication and the atom numbering are shown in Figure S8. The isotropic Fermi coupling constants obtained from the DFT simulations are given in Table S1.

The TD-DFT calculations predict the first optical transition $(D_0 \rightarrow D_1)$ of radical dication $\mathbf{1}^{2^+}$ at 1.3 eV with an oscillator strength f = 0.2. This transition primarily involves the HOMO (β) \rightarrow LUMO (β) excitation. A second optical transition ($D_0 \rightarrow D_5$), dominated by the HOMO (α) \rightarrow LUMO (α) excitation, is predicted at 2.1 eV with an oscillator strength of f = 1.7 (the corresponding spin up (α) and spin down (β) molecular orbitals are shown in Figure S9).



Figure S8. B3LYP/6-31G^{**} optimized geometry of the radical dication, and atom numbering used in the simulation of the ESR spectrum.



Figure S9. Pictorial representation of the spin unrestricted UB3LYP/6-31G**-derived molecular orbitals of 1^{2+} .

Table S1. Isotropic Fermi contact couplings, A_{iso} (in Gauss), obtained at the B3LYP/6-31G** DFT level.

atom no.	atom type	Aiso
30	N(14)	3.79347
31	N(14)	3.79343
32	H(1)	-0.11691
33	H(1)	0.08164
34	H(1)	-0.07634
35	H(1)	-1.19035
36	H(1)	0.66082
37	H(1)	0.33306
38	H(1)	-2.05773
39	H(1)	-5.89678
40	H(1)	5.40558
41	H(1)	5.39171
42	H(1)	0.16128
43	H(1)	-0.11685
44	H(1)	0.08161
45	H(1)	-0.07634
46	H(1)	-0.11667
47	H(1)	-0.07641
48	H(1)	0.08159
49	H(1)	3.11074
50	H(1)	-6.69462
51	H(1)	3.76550
52	H(1)	-6.69462
53	H(1)	3.11074
54	H(1)	-5.89676
55	H(1)	0.66079
56	H(1)	-0.07641
57	H(1)	-0.11660
58	H(1)	0.08155
59	H(1)	-1.19034
60	H(1)	-2.05769
61	H(1)	0.33305
62	H(1)	5.38619
63	H(1)	0.16131
64	H(1)	5.41097

Section 9: References

- 1) Cobra+: Non-liquid Nitrogen Cryo-cooling Device (80-500 K); Oxford Cryosystems, Oxford, 2005.
- 2) APEXII 2.1: Crystallography suite; Bruker AXS Inc.: Madison, WI, 2006.
- 3) BIS 2.0.1.9: Bruker Instrument Service; Bruker AXS Inc.: Madison, WI, 2006.
- 4) R. J. Cernik, W. Clegg, C. R. A. Catlow, G. Bushnell-Wye, J. V. Flaherty, N. Greaves, I. Burrows, D. J. Taylor and M Hamichi, *J. Synchrotron Rad.*, 1997, **4**, 279–286.
- 5) SAINT: Area-Detector Integration Software.; Bruker AXS Inc.: Madison, WI, 1995.
- 6) SADABS: Area-Detector Absorption Correction; Sheldrick, G.M., Bruker AXS Inc.: Madison, WI, 1996.
- a) A. Altomare, G. Cascarano, C. Giacovazzo and A. Guagliardi, *J. Appl. Cryst.*, 1993, 26, 343. b)
 M. C. Burla, R. Caliandro, M. Camalli, B. Carrozzini, G.L. Cascarano, L. De Caro, C. Giacovazzo,
 G. Polidori and R. Spagna. SIR2004, http://www.ic.cnr.it/
- 8) a) P. W. Betteridge, J. R. Carruthers, R. I. Cooper, C. K. Prout and D. J. Watkin, *J. Appl. Cryst.*, 2003, **36**, 1487. b) D. J. Watkin, C. K. Prout and L. J. Pearce, 'CAMERON', 1996.
- 9) G. M. Sheldrick, Acta Cryst., 2008, A64, 112–122.

- 10) J. R. Lenhard and A. D. Cameron, J. Phys. Chem., 1993, 97, 4916–4925.
- 11) Gaussian03, Revision B.05. Frisch, M. J.; Trucks, G. W.; Schlegel, H. B.; Scuseria, G. E.; Robb, M. A.; Cheeseman, J. R.; Montgomery, J., J. A.; Vreven, T.; Kudin, K. N.; Burant, J. C.; Millam, J. M.; Iyengar, S. S.; Tomasi, J.; Barone, V.; Mennucci, B.; Cossi, M.; Scalmani, G.; Rega, N.; Petersson, G. A.; Nakatsuji, H.; Hada, M.; Ehara, M.; Toyota, K.; Fukuda, R.; Hasegawa, J.; Ishida, M.; Nakajima, T.; Honda, Y.; Kitao, O.; Nakai, H.; Klene, M.; Li, X.; Knox, J. E.; Hratchian, H. P.; Cross, J. B.; Bakken, V.; Adamo, C.; Jaramillo, J.; Gomperts, R.; Stratmann, R. E.; Yazyev, O.; Austin, A. J.; Cammi, R.; Pomelli, C.; Ochterski, J. W.; Ayala, P. Y.; Morokuma, K.; Voth, G. A.; Salvador, P.; Dannenberg, J. J.; Zakrzewski, V. G.; Dapprich, S.; Daniels, A. D.; Strain, M. C.; Farkas, O.; Malick, D. K.; Rabuck, A. D.; Raghavachari, K.; Foresman, J. B.; Ortiz, J. V.; Cui, Q.; Baboul, A. G.; Clifford, S.; Cioslowski, J.; Stefanov, B. B.; Liu, G.; Liashenko, A.; Piskorz, P.; Komaromi, I.; Martin, R. L.; Fox, D. J.; Keith, T.; Al-Laham, M. A.; Peng, C. Y.; Nanayakkara, A.; Challacombe, M.; Gill, P. M. W.; Johnson, B.; Chen, W.; Wong, M. W.; Gonzalez, C.; Pople, J. A.; Gaussian, Incorporated: Wallingford, CT, 2004.