Conjugates of cyclodextrins with charged and neutral macrocyclic europium, terbium and gadolinium complexes: sensitised luminescence and relaxometric investigations and an example of supramolecular relaxivity enhancement

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The synthesis and characterisation of lanthanide complexes of mono- and tetra-amide β-cyclodextrin derivatives of 1,4,7,10-tetraazacyclododecane tetraacetate (DOTA) are reported. Luminescence and relaxivity measurements confirm that the Eu, Tb and Gd complexes of the eight-coordinate mono-amide ligand possess one bound water molecule while the tetra-amide complexes are rare examples of $q = 0$ systems in aqueous solution. The relaxivity of the host β-CD Gd complex (8.50 mM^{-1} s^{-1}, 20 MHz, 298 K) is enhanced when non-covalently bound to a second gadolinium complex bearing two phenyl moieties with an enhancement that is limited by the slowness of the water exchange rate ($\tau_m = 0.6$ μs, 298 K). Sensitisation of the terbium luminescence in the mono-amide β-CD complex occurs in the absence of oxygen using various substituted naphthalene derivatives (*e.g.* naphthalene, $K = 1.04 \times 10^4$ M^{-1}, 293 K) and methyl *p-tert*-butylbenzoate. The slowness of the intra-complex energy transfer step severely limits the efficiency of this process and restricts the scope of 'non-covalently triggered luminescence' to a narrow range of guest substrates, as deduced by variable temperature time-resolved luminescence and flash-photolysis studies.

Introduction

The long-lived luminescence of europium and terbium complexes has prompted a large number of studies which seek to take advantage of this aspect in diverse applications of supramolecular photochemistry.[1–10] From the development of complexes as stable luminescence reporters,[11] to the definition of responsive probes for signalling changes in bioactive ion concentration,[12,13] a considerable effort has been made to design efficient systems in which an antenna chromophore has been built into the complex structure, in order to improve the overall efficiency of the photochemical process. Equally attractive are systems in which the sensitising chromophore is non-covalently bound to a suitably designed lanthanide complex. Examples have been promulgated by Nocera in particular,[14] in which a cyclodextrin host serves as a binding site for a size-matched aromatic guest, in conjugation with a neutral[15] or tribasic ligand[16] for binding the lanthanide ion. The binding of the aryl chromophore is signalled by a 'switching-on' of the lanthanide luminescence, following intramolecular energy transfer from the aryl triplet excited state to the bound lanthanide ion. Thus β-cyclodextrin binding of naphthalene and biphenyl has triggered weak terbium emission, but only in the absence of oxygen.[17]

Related cyclodextrin conjugates have also been reported for NMR shift or relaxivity applications. Thus a dysprosium–DTPA complex, apparently despite being separated by six atoms from a mono-amino-β-cyclodextrin, induces measurable chemical shift non-equivalence in the ^{1}H NMR resonances of cyclodextrin-bound racemic guests, such as tryptophan or propranolol.[18] In a quite different application, the addition of β-cyclodextrin to β-benzyl-α-propionic acid substituted [Gd DOTA] and [Gd DTPA]† complexes leads to formation of a non-covalently bound adduct with an enhanced relaxivity[19] as a consequence of the slower tumbling rate of the paramagnetic gadolinium centre in the bound form.

Design features

With this background in mind, we set out to prepare well-defined stable lanthanide complexes of mono- and tetrasubstituted derivatives of DOTA in which there is a link to one and four β-cyclodextrin groups respectively. The monosubstituted ligand **1a** gives rise to kinetically stable charge neutral Ln(III) complexes while the tetra-amide **2** affords a stable triply charged complex. The linking amide carbonyl group in each case also serves as a donor to the Ln(III) ion, thereby bringing the cyclodextrin moiety relatively close to the Ln(III) centre. This is an important feature as any cyclodextrin bound antenna group needs to be as close as possible to the Ln(III) ion, in order to maintain relatively efficient energy transfer. Assuming a Förster mechanism for this energy transfer step (varying as $^1/_{r^6}$), and given that the distance‡ for 50% efficiency in this process is believed to be between 3 and 5.5 Å for common aryl triplet donors and a Tb or Eu(III) acceptor,[20,21] then it is apparent

† DOTA is 1,4,7,10-tetraazacyclododecanetetraacetate; DTPA is diethylenetriaminepentaacetate.

‡ r is the distance between Ln(III) and the chromophore and the efficiency of the energy transfer process, $\eta_{eT} = [1 + (r/R_o^6)]^{-1}$ where R_o is the distance for a 50% efficient process: thus if $r = 7.6$ Å and $R_o = 3.8$ Å, then $\eta_{eT} = 1.5\%$.

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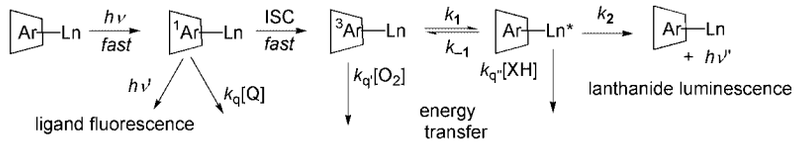
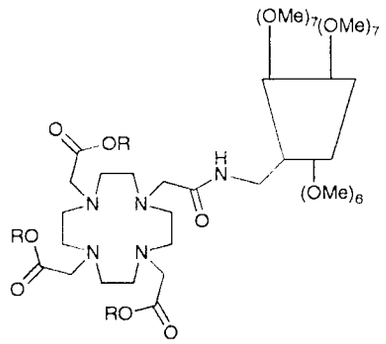
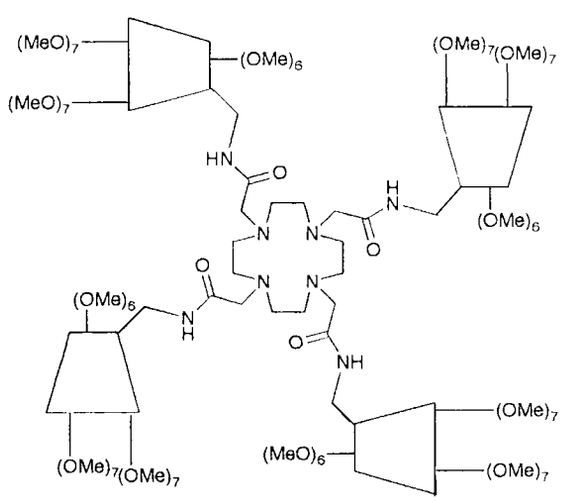


Scheme 1



1 a) R = H
b) R = ^{t}Bu



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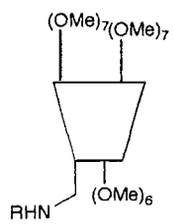
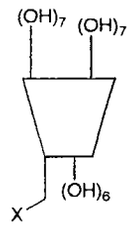
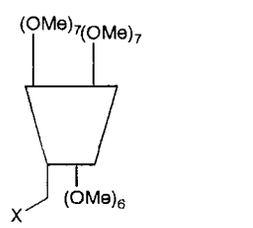


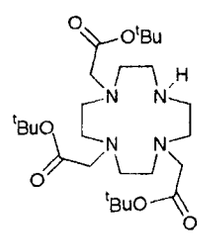
3 a) R =H
b) R = $COCH_2Cl$



4 a) X = OTs
b) X = N_3
c) X = O^{t}BuSIMe$_2$



5 a) X = O^{t}BuSIMe$_2$
b) X = OH
c) X =OMs
d) X = N_3



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that this factor is of paramount importance. Such a mechanism also requires that the aryl triplet energy level be greater than that of the $^5D_4$ terbium excited state (20 400 and 17 200 cm^{-1} for ^5D_o Eu). Again, most efficient transfer occurs when this energy gap is minimised, but this feature is constrained by the practical need to limit thermally activated back-energy transfer which repopulates the triplet (Scheme 1). A compromise is struck with an energy gap of close to 1700 cm^{-1}, although in the absence of competitive triplet quenching (principally by $^3O_2$) then lower energy gaps will obviously be more efficient, *i.e.* in deoxygenated solution.[1,4,12]

The β-cyclodextrin conjugates used in this work were permethylated at each OH group. This was done to enhance the water-solubility of the derived complexes (β-CD is the least soluble of all the cyclodextrins). It has been reported that such per-alkylation does not unduly compromise the stability of certain aryl guests in polar media. Indeed 2-*p*-toluidinonaphthalene-6-sulfonate (2,6-TNS)[22] and 4-methylbenzoic acid[23] both show enhanced binding affinities for per-*O*-methyl β- and α-CD respectively, compared to the parent hosts.

Ligand and complex synthesis and characterisation

The synthesis of mono-amino-β-cyclodextrin **3a** was undertaken by two parallel routes. Firstly, using a recent modification[24] of Lincoln's earlier procedure[25] mono-*O*-tosyl-β-CD **4a** was prepared in 40% yield and subsequent reaction with LiN_3 in DMF followed by permethylation (NaH–DMF–MeI) gave the azide **5d** (*via* **4b**). Completion of the final reaction was monitored by electrospray mass spectrometry, monitoring the disappearance of incompletely methylated products.[26] In parallel, a 'one-pot' procedure reported by Bradshaw[27] was followed involving the intermediacy of the silyl ether **4c**, followed by permethylation (NaH–MeI) to give **5a** and deprotection (NH_4F–MeOH) in the final sequence of the procedure to yield the alcohol **5b** in 20–25% overall yield, following chromatographic purification. Conversion to the azide **5d** *via* the mesylate **5c** (MsCl–Et_3N–CH_2Cl_2 then LiN_3–DMF) proceeded in high yield. The second route was higher yielding overall, but more laborious in that repeated chromatographic purification was found necessary. Finally, reduction of the azide **5d** to the amine **3a** proceeded efficiently by catalytic hydrogenation (PtO_2, H_2, EtOH), and reaction with chloroacetyl chloride (Et_3N–CH_2Cl_2) gave the α-chloro amide **3b**. Alkylation of the tri-*tert*-butyl ester **6** (Cs_2CO_3, MeCN) followed by treatment with CF_3CO_2H yielded **1a** as a glassy solid, and complexation with $Ln(NO_3)_3$ (Ln = Eu, Gd, Tb, Yb; pH 4 to 5) in aqueous solution gave the neutral complex which was separated from low molecular weight species by dialysis. In the case of the tetra-amide **2**, reaction of 1,4,7,10-tetraazacyclododecane with 4.2 equivalents of **3b** (DMF, K_2CO_3, 60 °C) was monitored by electrospray mass spectrometry, adding $Y(NO_3)_3$ in MeOH (^{89}Y: 100% abundance) as a means of enhancing the ionisation efficiency, allowing observation of the triply (and doubly MLX) charged species, *in situ*. Purification of the ligand (MW = 5990) from lower mass salts was possible with the aid of dialysis, and subsequent complexation with $Ln(SO_2CF_3)_3$ (Ln = Eu, Tb) in dry MeCN gave the tripositively charged complexes, which were also purified by dialysis, and characterised by positive ion ESMS as their doubly charged species $(ML - H)^{2+}$.

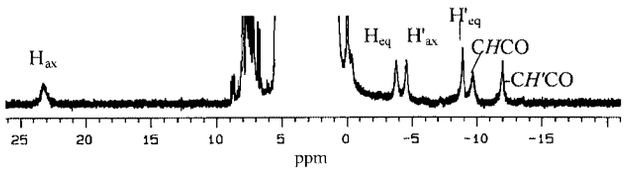


Fig. 1 Partial ^{1}H NMR spectrum of [Eu **2**]$^{3+}$ $(CF_3SO_3)_3$, highlighting the most shifted ring axial and equatorial protons and the diastereotopic NCHH′CO resonances (300 MHz, 293 K, CD_3OD).

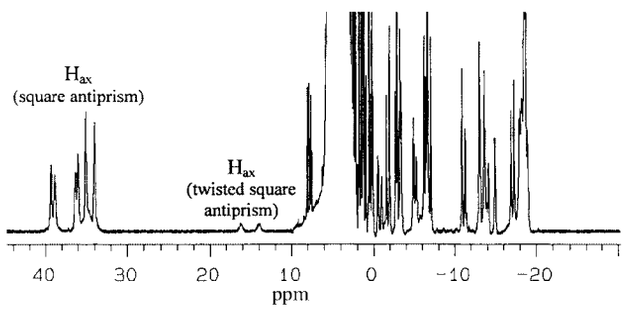
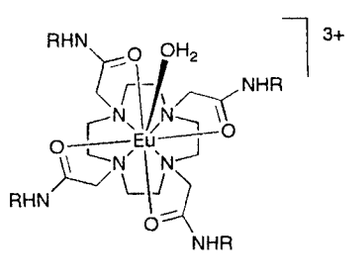


Fig. 2 ^{1}H NMR spectrum of [Eu **1a**] showing the axial ring protons of the two major diastereoisomeric species resonating to higher frequency of a minor twisted square antiprismatic isomer (293 K, 300 MHz, CD_3OD).

The proton NMR spectrum of [Eu **2**]$^{3+}$ in d^4-methanol (Fig. 1) revealed a very similar pattern for the most shifted axial, equatorial and NCH_2CO protons to that defined for related europium complexes such as **7a**.[28] Thus the broad singlet at



7 a) R = (*S*)-CHMePh
b) R = Me

+23.1 ppm corresponds to the closest four axial protons and the observation of single resonances for the other ring protons is consistent with the presence of time-averaged C_4-symmetry in a predominantly square-antiprismatic structure. The absence (≤7%) of a significant amount of the twisted square-antiprismatic complex is inferred from the lack of a resonance around +5 to +10 ppm for H_{ax}; in the complex **7b**, for example, both isomers may be discerned (273 K, CD_3OD) at *ca.* 25 and 5 ppm.[29] In the case of [Eu **2**]$^{3+}$, varying the temperature over the range 233–303 K (CD_3OD) and 275–333 K (D_2O) revealed no significant changes in the multiplicity of signals observed. Thus, one predominant isomeric species is present in solution.

In the case of [Eu **1a**], two sets of non-equivalent axial ring protons occur, as a consequence of the lack of C_4 symmetry. The observation of eight lines for the most shifted protons (Fig. 2; two pairs overlap) is consistent with the presence of two major square antiprismatic isomers (*i.e.* Δ/λλλλ and Λ/δδδδ with respect to the helicity and NCCN five-ring chelate configurations).[30] Evidence for the presence of a small amount of the twisted square antiprismatic isomer is given by the observation of broadened resonances in the region 14–16 ppm. Little change in linewidth and complexity of the ^{1}H spectrum was apparent on warming to 323 K (300 MHz, CD_3OD) so that, notwithstanding the temperature dependence of the paramagnetic shift, no clear evidence for isomer interconversion was obtained. This may be related to the high energy barrier

Table 1 Rate constants[a] for the depopulation of excited states of europium and terbium complexes of **1a** and **2** (293 K, ms^{-1})

Complex	k_{H_2O}	k_{D_2O}	Δk	Δk_{corr}[b]	q[b]
[Eu **2**]$^{3+}$	1.90	1.39	0.51	0	0
[Tb **2**]$^{3+}$	0.65	0.55	0.10	0.04	0.20
[Eu **1a**]	1.49	0.42	1.07	0.75	0.90
[Tb **1a**]	1.14	0.91	0.23	0.17	0.85

[a] Values given are the mean of 3 independent measurements (±10%). [b] $q_{Eu} = 1.2\Delta k_{corr}$ and $q_{Tb} = 5\Delta k_{corr}$ (see ref. 32 for details).

associated with the motion of the coordinated cyclodextrin moiety. Analysis of the ^{1}H NMR spectrum of [Tb **1a**] and [Yb **1a**] (65.6 MHz, CD_3CD) also revealed shift data (see Experimental) consistent with the presence of two major square antiprismatic isomers, by comparison with data reported for analogous complexes of DOTA or tetra-amides derived therefrom.[28,29,31,32] For example, with [Tb **1a**], two sets of shifted axial ring protons were observed around –345 and –365 ppm in the ratio 2:1.

Luminescence behaviour

Measurements of the radiative rate constants for decay of the excited states of the Eu and Tb ions in [Eu **1a**] and [Eu **2**]$^{3+}$ were made in H_2O and D_2O, following direct excitation of the lanthanide ion (*e.g.* Eu: 397 nm) or indirectly *via* a charge transfer band (λ_{exc} 250 nm). In each case mono-exponential decay curves were observed and the data obtained (Table 1) allowed an estimation of the number of coordinated water molecules ('q')—after allowing for the independent quenching effect of exchangeable amide NH protons and of second-sphere waters.[32] The monoamide complexes possess one bound water molecule while both of the tetra-amide complexes do not, providing an unprecedented example of unhydrated cationic complexes of Tb/Eu in aqueous media. Evidently the presence of the numerous methyl groups on the CD surfaces engenders a rather bulky environment which may inhibit the approach of a water molecule to the ninth coordination site. Support for the presence of an eight-coordinate Eu complex was also afforded by comparison of the emission spectra of [Eu **2**] in H_2O with those of [Eu **7a**] in H_2O and MeCN. In the latter case, the complex is mono-hydrated (X-ray and luminescence studies[28]), and the ratio of the $\Delta J = 1 : \Delta J = 2$ manifold is >2 in water and ≤1 in dry MeCN where no water is coordinated. For [Eu **2**], the spectrum closely resembled that found for [Eu **7a**] in dry CH_3CN (the *absence* of a bound water, under these conditions (1 mM complex, 293 K) has been defined[33]). In particular the forms of the hypersensitive $\Delta J = 2$ and $\Delta J = 4$ transitions were very similar and the $\Delta J = 1 : \Delta J = 2$ ratio was <1.

The inclusion of aromatic guests by the appended cyclodextrins in the Eu and Tb complexes of **1a** and **2** affords a means by which lanthanide luminescence may be sensitised, provided that the energy transfer step is sufficiently efficient (Scheme 1). Several potential aryl guests were screened possessing a triplet energy of greater than 244 kJ mol^{-1} (the $^5D_4$ level of Tb(III)). For example, addition of tryptophan to [Tb **2**]$^{3+}$ (pH 5 to 8; 0.1–1 mM host; 293 K; D_2O) with excitation at 250 or 280 nm led to a diminution in terbium emission intensity and a similar reduction was observed with 2-acetylnaphthalene, naphthalene and quinoline (in MeOH–H_2O 90:10), both in aerated and degassed solution. Tryptophan has previously been shown to act as a sensitiser for Tb emission in modified proteins[34] and each of these substrates is reported to bind to β-cyclodextrin with affinities in the range 300–800 M^{-1}.[35,36] In the case of [Eu **2**]$^{3+}$, 2-*p*-toluidinonaphthalene-6-sulfonate (2,6-TNS) was added—as it was known to bind to per-*O*-methyl-β-cyclodextrin with an affinity of 2×10^3 M^{-1}.[37] A large enhancement of the guest fluorescence was observed, consistent

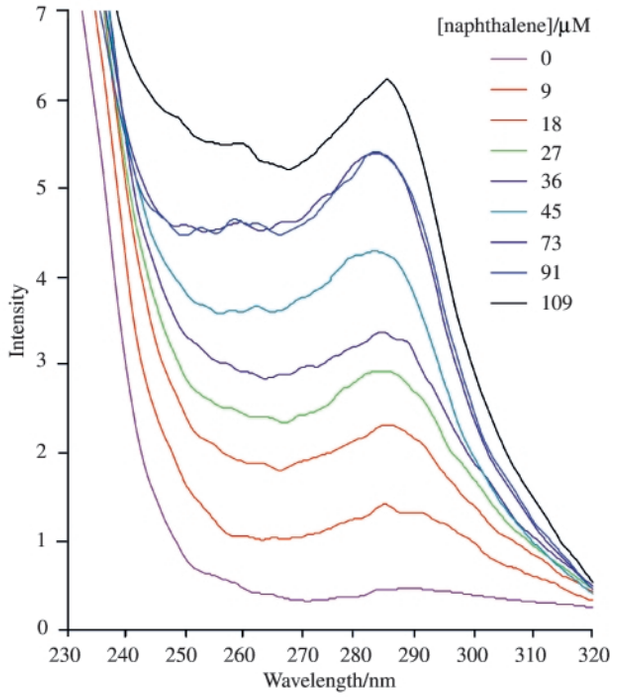


Fig. 3 Increases in terbium emission intensity as a function of added naphthalene concentration (μM) (λ_{exc} 275 nm, [Tb **1a**] = 0.14 mM; 293 K; 1 : 1 MeOH–H_2O).

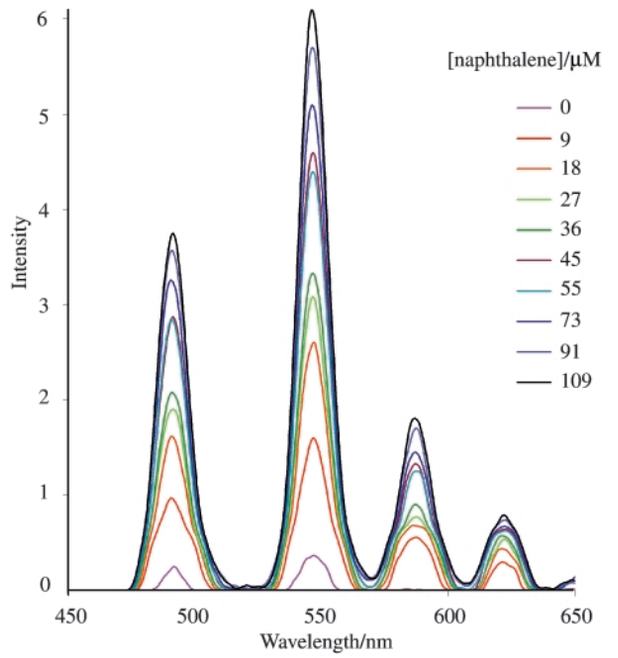


Fig. 4 Excitation spectrum of [Tb **1a**] as a function of added naphthalene concentration (μM) (λ_{em} 547 nm; [Tb **1a**] = 1.4×10^{-4} M; 50% MeOH–H_2O, 293 K).

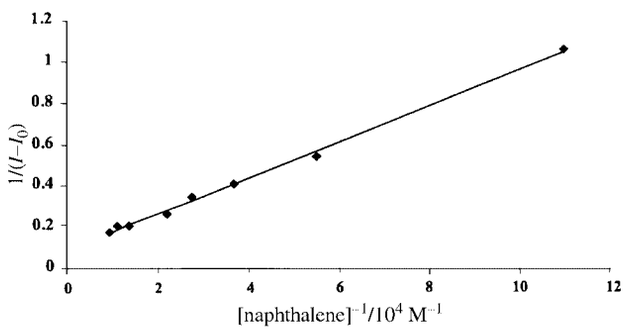


Fig. 5 Benesi–Hildebrand analysis of naphthalene complexation by [Tb **1a**] (λ_{exc} 275 nm, λ_{em} 547 nm; [Tb **1a**] = 1.4×10^{-4} M; 50% MeOH–H_2O, 293 K).

with guest binding,[38] but no enhancement of Eu emission was found for a range of host–guest concentrations (0.1 to 0.5 mM host; 0.1 to 1 mM guest). A similar degree of fluorescence enhancement was noted when 2,6-TNS was added to per-*O*-methyl-β-cyclodextrin under equivalent conditions, suggesting that inter-system crossing or, more likely that energy transfer is inefficient in this and the other studied cases with terbium.

The addition of naphthalene (φ_{isc} is *ca.* 0.8 in polar media) to [Tb **1a**] in 1 : 1 aqueous methanol, following excitation at 275 nm, also did not result in enhancement of terbium emission in aerated solution. However, in deoxygenated solution, the terbium emission increased by a factor of over 20 as naphthalene was added to [Tb **1a**] (0.14 mM, 293 K) (Fig. 3). The excitation spectrum revealed a parallel increase in intensity corresponding to changes in the naphthyl chromophore (Fig. 4), confirming that the terbium emission arose from energy transfer from the naphthyl triplet. Analysis of the binding isotherm, using a modified Benesi–Hildebrand analysis (Fig. 5), gave a value for the binding constant of 1.02 (± 0.1) × 10^4 M^{-1}, which is more than an order of magnitude greater than the association constant (760 M^{-1}) for complexation of naphthalene in water by β-cyclodextrin itself.[39] The enhanced binding affinity of cyclodextrins in which a more shielded hydrophobic environment is created by monofunctionalising the primary face has been reported in a number of cases involving diverse substituents such as polyamine or pyridyl groups.[40,41*a*]

Increases in sensitised terbium emission§ were also observed upon incremental addition of 1- or 2-methylnaphthalene, 2-bromonaphthalene or methyl *p*-*tert*-butylbenzoate in degassed 1 : 1 MeOH–H_2O solution, but changes were smaller than those observed for naphthalene itself and binding was not quantified. In contrast, addition of 1-bromonaphthalene, quinoline, or D-tryptophan led to a reduction in metal-emission intensity, although simultaneous observation of their fluorescence emission spectra, showing enhanced emission, demonstrated that binding to the cyclodextrin was occurring. That a 2-substituted naphthalene binds more strongly than the corresponding 1-substituted analogue has been observed previously; for example with the naphthalenesulfonates, the 2-isomer binds three times more strongly to β-CD than the 1-isomer.[41*b*] Evidently there is a fine balance struck between the rate of energy transfer (determined by the spectral overlap integral, and the donor–acceptor separation) and competitive deactivation of the intermediate aryl triplet state, highlighted by the sensitivity to triplet oxygen quenching.

Time resolved and variable temperature studies

Emission and excitation spectra for [Tb **1a**] and [Eu **1a**] (0.09 mM) in the presence of excess naphthalene (3 mM) and 1-methylnaphthalene (3 mM) were recorded over the range 293–240 K in degassed 50 : 50 MeOH–H_2O. As the temperature was lowered, an increase in metal emission intensity was observed. The lifetime for Tb emission increased from 0.5 ms at 295 K to 2.0 ms at 240 K in the presence of naphthalene and from 0.78 to 3.0 ms (240 K) for 1-methylnaphthalene. For [Eu **1a**] in the presence of 1-methylnaphthalene, the lifetime increased by 80%. For both [Eu **1a**] and [Tb **1a**], no significant changes in lifetime occurred in the absence of added guest. The observed lifetime increases for the Tb complexes are most likely to be primarily due to the reduction in the rate of thermally

§ No sensitisation of Yb emission from [Yb **1a**] was observed in degassed MeOH–H_2O (1 : 1) in the presence of one equivalent of naphthalene or phenanthridine, although both chromophores are known to sensitise Yb emission,[42] possibly *via* an electron transfer process.[43]

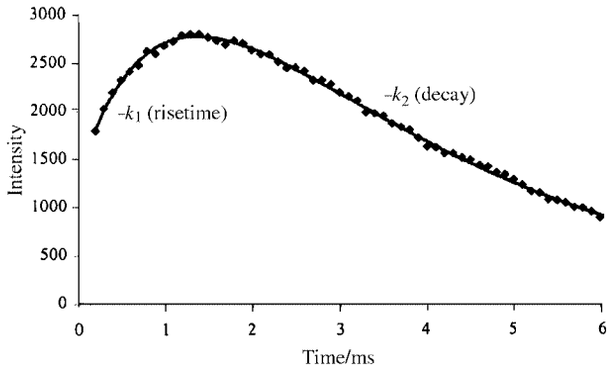
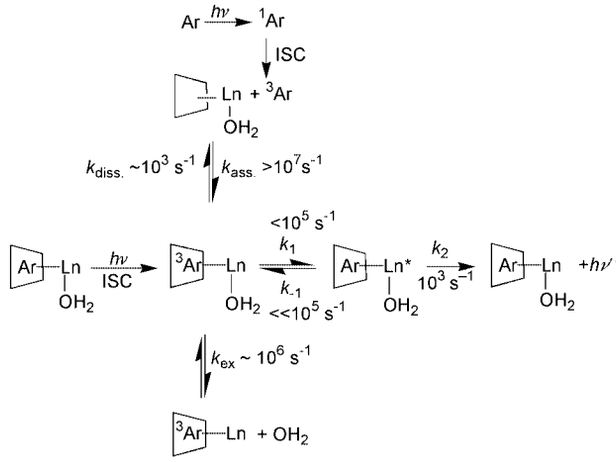


Fig. 6 Luminescent emission profile of [Tb **1a**] (0.09 mM) and naphthalene (3 mM) showing distinctive 'grow-in' and decay components (240 K; 50% MeOH–H_2O).

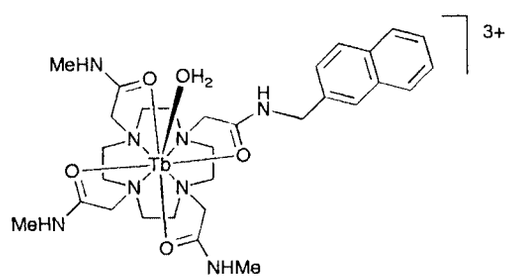
activated back energy transfer from the excited metal to the naphthyl triplet state. Similar behaviour has been defined previously with the related terbium complex **8**, where the energy gap between the naphthyl triplet and the emissive terbium level is sufficiently small to give rise to T and pO_2 dependent Tb emission.[4]

The lanthanide emission profile for each of these systems showed two components, corresponding to a 'grow-in' (or rise-time; Fig. 6) of the lanthanide luminescence followed by a slower decay. The apparent grow-in of the Ln luminescence was observed to increase with decreasing temperature for [Tb **1a**] with both naphthalene and 1-Me-naphthalene and for [Eu **1a**] with 1-Me-naphthalene [degassed solution; 0.09 mM complex, 3 mM guest]. The measured lifetime grew rapidly at temperatures below 260 K, in each case (from *ca.* 200 μs to ~1 ms). The observation of a 'grow-in' component to the emission profile also provides evidence for an inefficient energy transfer step, either as a result of its slow forward rate or of a significant back transfer process.

A flash-photolysis study for [Tb **1a**] (0.32 mM) in the presence of 1-methylnaphthalene (0.17 mM; with $K = 10^4$ M^{-1} this is equivalent to 70% bound) was undertaken, in an attempt to more closely define the salient kinetic processes. A xenon chloride excimer laser (308 nm) was used to excite the sample. The duration of each laser pulse was 20 ns, during which time the triplet level of the naphthyl group is populated to a significant extent, allowing the triplet state to be probed by transient absorption spectroscopy. The transient absorption was investigated over a range of wavelengths (360–430 nm) and the maximum found to be 410 nm. This is close to the literature value for λ_{max}^{T-T} of naphthalene of 415 nm.[44] Neither ground state naphthalene nor terbium absorbs at this wavelength, and hence the rate of triplet deactivation can be probed by monitoring the decay of this absorption band, following the excitation pulse. A small modification of the instrumental set-up also allowed the decay of the terbium luminescence (490, 545 and 590 nm) to be monitored on the same timescale. Decay of the 1-Me-naphthyl triplet excited state fitted a single exponential with good residuals and gave a lifetime of 43 μs at 295 K and 180 μs at 240 K. These values may be compared to 92 and 915 μs respectively, measured for 1-methylnaphthalene, under identical conditions in the absence of the Tb complex, and contrast with the shorter value of 4 μs observed[4] for the terbium complex **8**—where energy transfer to Tb is presumably more efficient. The terbium emission profile was fitted to a double exponential scheme, with one component defining the grow-in and the other the decay. A grow-in lifetime of 55(5) μs and a decay of 1.1(0.5) ms was estimated, at 240 K. The 'grow-in' lifetime of terbium emission does not match very well with that measured for triplet decay [180(20) μs] and consideration therefore needs to be given to competing processes and the relative rates of the key steps involved (Scheme 2). Thus, if the rates of energy transfer and



Scheme 2



8

lanthanide emission are similar, then they will not match well to those estimated from analysis of the 'grown-in' or decay profiles. Secondly, the rate of water exchange at such neutral terbium centres is likely to be around 10^6 s^{-1}.[45] Furthermore dissociation of naphthalene from the cyclodextrin cavity prevents energy transfer to the metal and this rate is estimated to be in the range of 10^4 to 10^3 s^{-1} (*i.e.* $K = 10^4$ and assuming a forward association rate of 10^8 to 10^7 s^{-1}).[46] Evidently there are likely to be several competing reactions occurring which determine the important bound aryl triplet lifetime and the rate of terbium decay, so that a rigorous kinetic analysis is not feasible.

Relaxivity behaviour of [Gd 1a]

Measurement of the relaxivity of a gadolinium complex as a function of magnetic field strength generates a nuclear magnetic resonance dispersion profile (NMRD). Over the past ten years in particular, a very large number of structurally related complexes have been analysed[45,47] and their NMRD profiles have been modelled to provide reliable estimates of τ_m (the water exchange rate), τ_r (the reorientational correlation time) and τ_s (the electronic spin relaxation time). For [Gd **1a**], profiles were obtained at 298 and 310 K (Fig. 7) and each was fitted assuming $q = 1$ (as determined by independent luminescence measurements, Table 1). A value for τ_m of 0.58 μs (298 K) was obtained which is very similar to those obtained for related neutral mono-amide–DOTA complexes.[45] The rotational correlation time was estimated to be 200 ps.

Previous work has established that β-cyclodextrin forms inclusion complexes with aryl-substituted gadolinium complexes, such as $[Gd{\cdot}BOPTA]^{2-}$ and the related cyclic complex **9**.[19] Water proton relaxation rates (R_1) were observed to increase as a function of added cyclodextrin concentration, consistent with an increase in the effective τ_r and associated with formation of a more slowly tumbling, higher molecular weight complex [$K \approx 200$ M^{-1}, 298 K]. Addition of a 0.01 mM solution of [Gd **1a**]—bearing a 'free' β-cyclodextrin—to aqueous solutions of the Gd complex **9**, varying in concentration

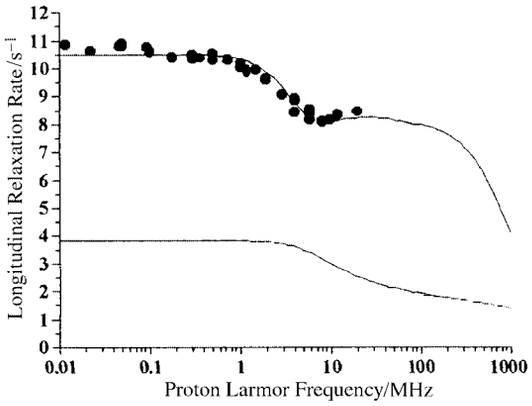
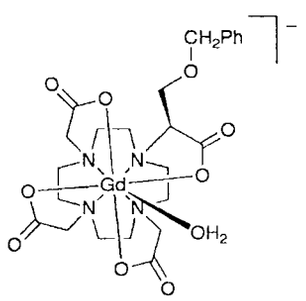


Fig. 7 NMRD Profile for [Gd **1a** OH_2] at 298 K (H_2O, 1 mM complex); the fitting parameters used gave $\tau_r = 200$ ps, $\tau_m = 0.58$ μs, $r = 3.0$ Å, $a = 3.8$ Å and $D = 2.24 \times 10^{-5}$ cm^2 s^{-1} [line; dots represent experimental points].



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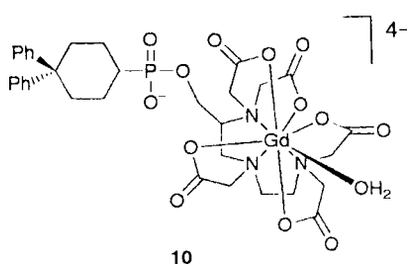


10

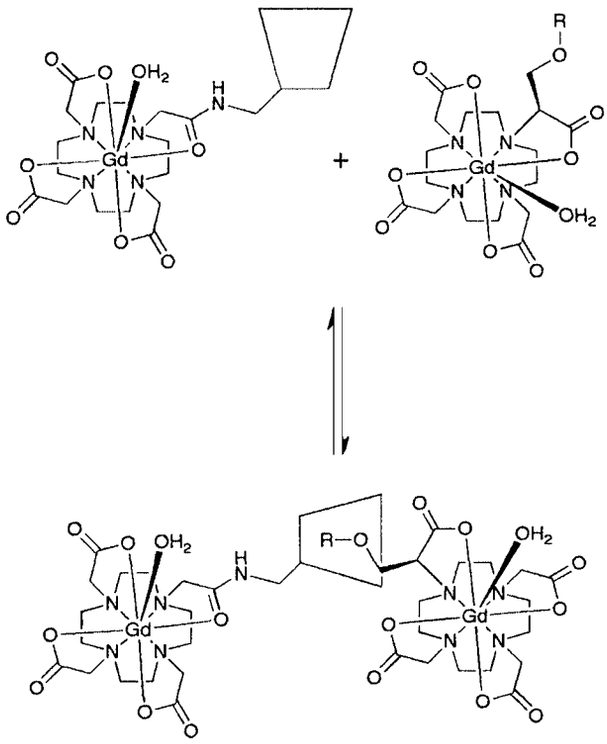
from 0.1 to 10 mM, gave measured relaxivities that were within 4% of the sum of the individual complexes. Alternative guest Gd complexes were then considered (Scheme 3) possessing a sub-unit that binds more strongly to a cyclodextrin host. An example is provided by **10**, (MS-325, AngioMARK™, EPIX/Mallinckrodt).[48] This complex has been shown to bind to β-cyclodextrin with an affinity of the order of 8×10^4 M^{-1}.[49] A 0.09 mM solution of [Gd **1a**] and a 0.18 mM solution of **10** gave an observed relaxation rate of 2.83 s^{-1} (298 K, 20 MHz), which is higher than the value expected (2.56 s^{-1}) if the complexes do not interact. An enhancement factor, ε, of 1.3 was estimated using eqn. (1), where c represents the complex concentration of

$$\varepsilon = R_1^{obs} - R_1^{d} - c^{[Gd\ 10]} r_{1p}^{[Gd\ 10]} / r_{1p}^{[Gd\ 1a]} c^{[Gd\ 1a]} \quad (1)$$

the indicated complexes, r_{1p} is the relaxivity value, R_1^{d} is the relaxation rate of water in the absence of added complex and R_1 is the observed or calculated relaxation rate. Using a similar approach with a related Gd complex bearing a cholic acid group, an enhancement factor of 7.2 has been found.[49]

Conclusion

In the terbium and europium complexes of the monocyclodextrin **1a**, non-covalent binding of a limited range of aryl chromophores leads to sensitisation of the proximate lanthanide luminescence. The overall scope and efficiency of this



Scheme 3

process are primarily limited by the energy transfer step, which is slow as a result of the rather long donor–acceptor distance. The slowness of the energy transfer step subjects the aryl triplet to competing deactivation processes, principally involving quenching by triplet oxygen. The sensitivity of the process to aryl chromophore structure and triplet energy is intriguing: thus naphthalene (E_T 255 kJ mol^{-1}),[50] 1- and 2-Me naphthalene (254 kJ mol^{-1}) and 2-Br naphthalene (252 kJ mol^{-1}) **do** act as sensitisers for Tb ($^5D_4 \sim 244$ kJ mol^{-1}) emission in degassed solution, whereas 1-Br naphthalene (247 kJ mol^{-1}) and quinoline (261 kJ mol^{-1})[50] do not. Such a finely poised situation presumably reflects the interplay between competing rates of forward and reverse energy transfer, dissociation from the cyclodextrin, the slightly different distance of the bound guest in the CD complex and the relative rates of other non-radiative quenching processes.

In the case of [Gd **1a**], the first example of relaxivity enhancement is presented involving non-covalent binding of a host gadolinium complex [Gd **1a**] with a different guest gadolinium complex. The synergicity obtained in the relaxivity of the supramolecular complex augurs well for the future application of such an approach, involving the docking of a migrating guest complex with a higher molecular weight, less mobile (or indeed immobilised) host complex.

Experimental

Reactions requiring an inert atmosphere or anhydrous conditions were carried out under a dynamic atmosphere of dry, oxygen-free argon using standard Schlenk-line techniques. Solvents were dried by distillation from the appropriate drying agent with the exception of *N*,*N*-dimethylformamide and dimethyl sulfoxide which were used directly from "sure-seal" bottles. Water was purified by the $PURITE_{STILL}$ plus system.

Thin layer chromatography was carried out using aluminium backed silica plates with or without Merck Art 5554, a fluorescent indicator. Plates containing non-fluorescent cyclodextrin compounds were developed by immersion in aqueous sulfuric acid solution (50% v/v) and heating (400 °C), the compound(s) appearing as charred black spot(s). Preparative column chrom-

atography was carried out using silica (Merck silica gel 60, 230–400 mesh). Benzoylated dialysis tubing (Sigma, D-2272 or D-7884) with a molecular weight cut off of 1200–2000 amu was used in water as supplied and sealed by tying at both ends. Dialysis was also carried out using a Spectrum Spectra/Por® MacroDialyzer using CEA (Cellulose Ester Asymmetric) membranes with a cut-off of 1000, 2000 or 5000 amu. Membranes were washed with water before use, and the chamber operated in a flow dialysis mode using distilled water as a reservoir solvent.

^{1}H NMR spectra were recorded at 65.26 MHz on a 1.53 T magnet connected to a Varian VXR400 console, at 199.99 MHz on Varian Mercury-200, Gemini-200, and VXR200 spectrometers, at 299.91 MHz on a Varian Unity-300, at 399.96 MHz on a Varian VXR400 and at 499.79 MHz on a Varian Unity Inova-500 spectrometer. ^{13}C NMR spectra were recorded on the Varian Gemini-200 and Mercury-200 spectrometers operating at 50.29 MHz, the Varian Unity-300 operating at 75.41 MHz, the Varian VXR400 operating at 100.58 MHz and the Varian Unity Inova-500 operating at 125.67 MHz. Infra-red spectra were recorded on a Perkin-Elmer 1600 FT-IR spectrometer using a 'Golden Gate' accessory and absorbance maxima are quoted in wavenumbers. Ultraviolet spectra were recorded on a Unicam UV2 spectrometer.

Electrospray ionisation mass spectra were obtained on a VG II platform (Fisons Instruments) with methanol as carrier solvent. VG 7070E and Micromass Autospec spectrometers were also used, operating in EI ionisation mode. MALDI-TOF spectra were recorded on a Kratos Kompact 4 spectrometer, operated in linear or reflection detection mode to generate positively charged ions. 2,5-Dihydroxybenzoic acid was applied as a matrix. Accurate masses were obtained from the EPSRC National Mass Spectrometry Service Centre, University of Wales, Swansea.

Elemental analyses were determined on an Exeter Analytical CE440 Elemental Analyser. Metal and halogen concentrations were determined on a Dionex DX500 Ion Chromatograph. Melting points were determined on a Reichert Köfler block melting point apparatus and are uncorrected.

NMRD profiles were obtained at the Università degli Studi di Torino, Italy. The profiles were acquired between 0.01–10 MHz on a Stelar Field Cycling Relaxometer, at 20 MHz on a Stelar Spinmaster and at 90 MHz on a Jeol EX 90.

Fluorescence and phosphorescence spectra were acquired using a Perkin-Elmer LS 50B Luminescence Spectrometer controlled through Perkin-Elmer Fluorescence Data Manager Instrument Control Version 3.00 on a PC, or using an Instruments SA Fluorolog^{-3} controlled through DataMax for Windows Version 2.1 on a PC. Samples were held in a degassing cell equipped with a 10 mm pathlength square cuvette and a degassing bulb. Degassing was achieved using the 'freeze–pump–thaw' method.

Lifetime measurements were acquired on the same spectrometer using a program designed and written by Dr Andrew Beeby (University of Durham) on a PC. Decays were fitted using Microsoft Excel to single or double exponential functions of the form $I(t) = A_o + A_1e^{-k_1t} + A_2e^{-k_2t}$. Low temperature data were acquired using cells held in an optical cryostat (Oxford Instruments DN-704). Temperatures could be maintained at 77–330 ± 0.1 K using an Oxford Instruments ITC-6 temperature controller. The sample was contained in a degassing cell equipped with a degassing bulb and a cell made from square section fused silica tubing with a 10 mm nominal pathlength.

Laser flash photolysis was carried out with a 90° geometry for pump and probe beams. Excitation was carried out using the 4th harmonic of a Q-switched Nd-YAG laser (Spectra Physics GCR-150–10) emitting at a wavelength of 266 nm. The pulse energy delivered was less than 1 mJ per pulse at the sample. The probe beam was supplied by a continuous wave W lamp. Transmitted light was passed through a monochromator (Spex Triax 320) and the intensity recorded using a photomultiplier (Hamamatsu R928) operating in DC-mode. The signal was recorded and averaged using a digital oscilloscope (Tektronix TDS-320) and the transient transferred to a PC for analysis. Ytterbium complexes were excited at 266 nm in an identical manner as for flash photolysis experiments. Ytterbium emission was detected in the range 980–1000 nm using a liquid nitrogen cooled germanium diode (North Coast EO817P).

Mono-(6^A-*O*-tolylsulfonyl-6^A-deoxy)-β-cyclodextrin, 4a

The synthesis was adapted from a procedure described by Zhong *et al.*[24] β-Cyclodextrin (11 g, 10.4 mmol) and toluene-*p*-sulfonyl chloride (5.2 g, 27.3 mmol, recrystallised from 60 : 80 petroleum ether) were stirred as a suspension in water (250 cm^3). After 3 h, aqueous sodium hydroxide (60 cm^3, 2 M) was added and the solution stirred for a further 30 min. Ammonium chloride (20 g) was added until the pH of the solution reached 8. The solution was chilled for 18 h and the resulting precipitate collected by vacuum filtration to afford a white solid (5.19 g, 4.03 mmol, 39%), mp: 168–170 °C dec. (lit. 168–170 °C dec.).[25] R_f = 0.53 (5 : 4 : 3 t-BuOH–EtOH–H_2O, silica). *m*/*z* (ES+): 1306 $[M + NH_4]^+$, 1312 $[M + Na]^+$.

Mono-(6^A-azido-6^A-deoxy)-β-cyclodextrin, 4b

Mono-6-*O*-tolylsulfonyl-6-deoxy-β-cyclodextrin, **4a**, (1.94 g, 1.5 mmol) was dried under vacuum for 18 h (overnight) then dissolved in dry DMF (30 cm^3). Lithium azide (0.82 g,15 mmol) was added and the solution was stirred at 100 °C for 5 h. The solvent was removed under reduced pressure (20 mmHg, 40 °C), the residual solid was dissolved in warm water (40 cm^3) and added to chilled acetone (250 cm^3). The resulting precipitate was collected by vacuum filtration to afford a cream solid (1.62 g, 1.4 mmol, 93%), mp: 215–220 °C dec. (lit. 210–220 °C dec.).[25] R_f = 0.48 (5 : 4 : 3 t-BuOH–EtOH–H_2O; silica). *m*/*z* (ES+): 1177 $[M + NH_4]^+$.

6^A-Hydroxyheptakis(2,3-di-*O*-methyl)-6^B,6^C,6^D,6^E,6^F,6^G-hexa-*O*-methyl-β-cyclodextrin, 5b

β-Cyclodextrin **(1)** (4.54 g, 4.0 mmol) and imidazole (0.61 g, 9.0 mmol) were dried under vacuum for 18 h, then dissolved in dry DMF (100 cm^3) under argon. *tert*-Butyldimethylsilyl chloride (1.24 g, 8.2 mmol) dissolved in dry DMF (20 cm^3) was added dropwise over 2 h. After a further 2 h the solution was cooled to 0 °C and sodium hydride (6.4 g, 260 mmol) added slowly over 5 min. The solution was then stirred at 0 °C for 30 min and at room temperature for a further hour. The solution was then cooled to 0 °C and methyl iodide (77 g, 545 mmol) added dropwise over 1 h and the mixture stirred at room temperature for 3 days. The solution was then cooled to 0 °C, excess hydride quenched by the addition of methanol (15 cm^3) and the mixture added to chilled water (400 cm^3). The solution was extracted into chloroform (4 × 100 cm^3), the combined organic extracts were washed successively with aqueous sodium thiosulfate solution (75 cm^3, 3% w/v) and water (3 × 75 cm^3), and dried (K_2CO_3), filtered and solvent removed under reduced pressure to yield the crude silyl ether **5a** as a cream solid (6.35 g). This cream solid was dissolved in methanol (300 cm^3) and ammonium fluoride (2.15 g, 58.1 mmol) added. The solution was boiled under reflux for 30 h then solvents removed under reduced pressure. The residual solid was redissolved in ethyl acetate (100 cm^3), filtered and solvent removed under reduced pressure to yield a yellow solid (6.06 g). Purification by column chromatography (1% MeOH–$CHCl_3$, silica) gave a yellow solid (1.15 g, 0.81 mmol, 20%). R_f = 0.52 (10 : 1 $CHCl_3$–MeOH; silica). *m*/*z* (ES+): 1432 $[M + NH_4]^+$. δ_H($CDCl_3$): 5.30–4.95 (m, 7H, C(1)-*H*), 3.95–2.95 (m, 103H), of which 3.38 (s), 3.50 (s) and 3.64 (s) are for OCH_3 groups.

6^{A}-*O*-Methylsulfonyl-6^{A}-deoxyheptakis(2,3-di-*O*-methyl)-6^{B},6^{C},6^{D},6^{E},6^{F},6^{G}-hexa-*O*-methyl-β-cyclodextrin, 5c

Mono-6-hydroxy-per-*O*-methyl-β-cyclodextrin **5b** (1.015 g, 0.72 mmol) and triethylamine (0.100 g, 0.99 mmol) were dissolved in dry THF (10 cm^3) and chilled to −15 °C. Methanesulfonyl chloride (0.250 g, 2.2 mmol) was added dropwise and the solution stirred at −15 °C for 1 h, and at room temperature for a further 2 h. The solvent was removed under reduced pressure and the resulting solid dissolved in water (30 cm^3) and extracted into chloroform (4 × 50 cm^3). The combined organic extracts were dried (K_2CO_3), filtered and solvent removed under reduced pressure to yield mono-6-*O*-methylsulfonyl-6-deoxy-per-*O*-methyl-β-cyclodextrin as a colourless glassy solid (1.06 g, 0.71 mmol, 99%). R_f = 0.70 (10 : 1 $CHCl_3$–MeOH; silica). *m*/*z* (ES+): 1510 $[M + NH_4]^+$. mp: 74–76 °C. $\delta_H(CD_3OD)$: 5.30–5.10 (m, 7H, C(1)-*H*), 3.95–2.95 (m, 105H), of which 3.34 (s), 3.48 (s) and 3.61 (s) are for OCH_3 groups and 3.11 (s) is for one SO_2CH_3 group. $\delta_C(CD_3OD)$: 98.5–97.6 (m, inc 98.4, 98.1, 98.0, 97.9, 97.8), 82.1–81.2 (m, inc 81.9, 81.7, 81.6, 81.4), 79.8–78.6 (m, inc 79.7, 79.3, 79.2, 78.9, 78.7), 71.3–70.7 (m, inc 71.2, 71.1, 71.0, 70.8), 69.7, 69.5, 60.5–60.1 (m, inc 60.4, 60.3), 58.0–57.2 (m, inc 57.9, 57.8, 57.6, 57.5, 57.4), 36.0, 31.2. ν_{max}: 2926, 2830, 1466, 1366, 1140, 1106, 1034, 976, 862, 754, 704, 528 cm^{-1}. Found: C: 50.4%, H: 7.20%, S: 2.14%. $C_{63}H_{111}O_{37}S$ requires C: 50.7%, H: 7.56%, S: 2.15%.

6^{A}-Azido-6^{A}-deoxyheptakis(2,3-di-*O*-methyl)-6^{B},6^{C},6^{D},6^{E},6^{F},6^{G}-hexa-*O*-methyl-β-cyclodextrin, 5d

Method 1. Mono-6-azido-6-deoxy-β-cyclodextrin **4b** (0.40 g, 0.34 mmol) and 18–crown-6 (0.004 g) were dried under vacuum for 18 h then dissolved in dry DMF (20 cm^3). Sodium hydride (0.5 g, 21 mmol) was added and after stirring the solution for 1 h, methyl iodide (2 cm^3, 32 mmol) was added over a period of 1 h. The mixture was stirred at 50 °C for 7 days. Further additions of sodium hydride (0.15 g, 6 mmol) and methyl iodide (1 cm^3, 16 mmol) were subsequently made on the 2nd, 3rd and 4th days. After 7 days the solution was analysed by electrospray mass spectrometry which showed the presence of mono-6-azido-6-deoxy-per-*O*-methyl-β-cyclodextrin and no detectable mono-6-azido-6-deoxy-β-cyclodextrin or per-*O*-methyl-β-cyclodextrin. The solution was then quenched by the addition of methanol (10 cm^3) and filtered, the solid being washed with further methanol (30 cm^3). The solvents were removed under reduced pressure (20 mmHg, 30 °C) to give a pale orange solid which was treated with dichloromethane (100 cm^3) and water (100 cm^3) and the organic phase separated and washed with water (3 × 30 cm^3). Sodium chloride (20 g) was then dissolved in the combined aqueous phases and the aqueous solution washed with dichloromethane (50 cm^3). The combined organic phases were dried (K_2CO_3), filtered and solvent removed under reduced pressure to yield a deep yellow oil. TLC Analysis (10 : 1 $CHCl_3$–MeOH, silica) of the oil revealed one major component (R_f = 0.70) and one minor component (R_f = 0.50) and purification by column chromatography (1% MeOH–$CHCl_3$, silica) gave a yellow oil (0.11 g, 0.076 mmol, 22%), which slowly crystallised on standing.

Method 2. Mono-6-*O*-methylsulfonyl-6-deoxy-per-*O*-methyl-β-cyclodextrin **5c** (0.88 g, 0.59 mmol) was dried under vacuum for 18 h then dissolved in dry DMF (30 cm^3) and lithium azide (0.31 g, 6.3 mmol) added. The solution was heated to 100 °C for 18 h and solvent removed under reduced pressure to yield a brown solid. The brown solid was dissolved in water (50 cm^3) and extracted into chloroform (3 × 60 cm^3). The combined organic phases were dried (K_2CO_3), filtered and solvent removed under reduced pressure to yield a yellow solid (0.53 g, 0.365 mmol, 62%), mp: 77–79 °C (lit. 87–90 °C).[27] R_f = 0.70 (10 : 1 $CHCl_3$–MeOH; silica). *m*/*z* (ES+): 1458 $[M + NH_4]^+$. $\delta_H(CD_3OD)$: 5.15–4.95 (m, 7H, C(1)-*H*), 3.95–2.95 (m, 102H), of which 3.28 (s), 3.41 (s) and 3.54 (s) are for OCH_3 groups.

6^{A}-Amino-6^{A}-deoxyheptakis(2,3-di-*O*-methyl)-6^{B},6^{C},6^{D},6^{E},6^{F},6^{G}-hexa-*O*-methyl-β-cyclodextrin, 3a

Mono-6-azido-6-deoxy-per-*O*-methyl-β-cyclodextrin **(5d)** (0.55 g, 0.38 mmol) was dissolved in ethanol (10 cm^3) in the presence of platinum oxide (0.02 g). The solution was then shaken under hydrogen (50 psi) at room temperature. After 4 days analysis by electrospray mass spectrometry indicated that complete conversion to the desired compound had occurred. The solution was filtered and solvent removed under reduced pressure to yield a colourless crystalline solid (0.52 g, 0.37 mmol, 96%), mp: 91–92 °C (lit. 91–93 °C).[27] R_f = 0.20 (10 : 1 $CHCl_3$–MeOH; silica). *m*/*z* (ES+): 1414 $[M + H]^+$. $\delta_H(CDCl_3)$: 5.12 (d, *J* = 3.4, 7H, C(1)-*H*), 3.95–2.95 (m, 104H), of which 3.38 (s), 3.50 (s) and 3.64 (s) are for OCH_3 groups.

6^{A}-(Chloromethylcarbamoyl)-6^{A}-deoxyheptakis(2,3-di-*O*-methyl)-6^{B},6^{C},6^{D},6^{E},6^{F},6^{G}-hexa-*O*-methyl-β-cyclodextrin, 3b

Mono-6-amino-6-deoxy-per-*O*-methyl-β-cyclodextrin **3a** (3.2 g, 2.26 mmol) and triethylamine (0.34 g, 3.36 mmol) were dissolved in ether (100 cm^3) and cooled to −30 °C. Chloroacetyl chloride (0.38 g, 3.36 mmol) was added and the solution stirred at −30 °C for 1 h and at room temperature for a further hour. The solvent was removed under reduced pressure to yield an orange solid which was purified by column chromatography (CH_2Cl_2, then 5% MeOH–CH_2Cl_2, silica) to yield a glassy yellow solid (2.93 g, 1.97 mmol, 87%), mp: 85–87 °C. Found: C: 51.3%, H: 7.60%, N: 0.71%, Cl: 2.69%. $C_{64}H_{112}O_{35}NCl$ requires C: 51.6%, H: 7.57%, N: 0.94%, Cl: 2.38%. R_f = 0.73 (10 : 1 $CHCl_3$–MeOH; silica). *m*/*z* (ES−): 1524 $[M + Cl\ (2 \times {}^{35}Cl)]^-$, 1525, 1526, 1527, 1528, 1529. $\delta_H(CD_3OD)$: 5.12 (d, *J* = 3.4, 7H, C(1)-*H*), 4.09 (s, 2H, CH_2Cl), 4.00–2.95 (m, 102H), of which 3.34 (s), 3.48 (s) and 3.61 (s) are for OCH_3 groups. $\delta_C(CD_3OD)$: 99.5 (*C*(1)), 83.5, 83.2, 80.8, 72.8, 72.3, 61.8 (O-*C*H_3), 59.4 (O-*C*H_3), 59.0 (O-*C*H_3): ^{13}C NMR partial analysis is consistent with a lack of C_7 symmetry within the molecule. ν_{max}: 2930, 2830, 1690 (C=O), 1156, 1030, 968, cm^{-1}.

1,4,7,10-Tetrakis[6^{A}-deoxyheptakis(2,3-di-*O*-methyl)-6^{B},6^{C},6^{D},6^{E},6^{F},6^{G}-hexa-*O*-methyl-β-cyclodextrin-6^{A}-acetamido]-1,4,7,10-tetraazacyclododecane, 2

1,4,7,10-Tetraazacyclododecane (0.026 g, 0.15 mmol), potassium carbonate (0.10 g, 0.72 mmol) and potassium iodide (1 crystal, cat.) were dissolved in anhydrous DMF (1 cm^3) and heated to 60 °C. Mono-6-(chloromethylcarbamoyl)-6-deoxy-per-*O*-methyl-β-cyclodextrin **3b** (1.00 g, 0.67 mmol), dissolved in further DMF (1 cm^3), was added and the solution stirred at 60 °C for 5 days. The solvent was removed under reduced pressure and the solid residue dissolved in water (10 cm^3) and placed in benzoylated dialysis tubing. The sealed tubing was placed in water (2000 cm^3) and allowed to stand. After 2 days the solution within the tubing was removed and solvent removed under reduced pressure. Purification by column chromatography (98% CH_2Cl_2, 1.8% MeOH, 0.2% NH_3, rising to 95% CH_2Cl_2, 4.5% MeOH, 0.5% NH_3, silica) gave the tetra-amide as a glassy yellow solid (0.16 g, 0.027 mmol, 18% yield), mp: 132–136 °C. Found: C: 50.0%, H: 7.64%, N: 1.95%. $C_{264}H_{464}O_{140}N_8 \cdot 18H_2O$ requires C: 50.2%, H: 7.98%, N: 1.77%. R_f = 0.20 (90% CH_2Cl_2, 9% MeOH, 1% NH_3; silica). *m*/*z* (ES+): 2024 $[M + Y]^{3+}$: yttrium nitrate was added *in situ* to improve ionisation; (MALDI-TOF): 5987 $[M + H]^+$: $C_{264}H_{464}O_{140}N_8$ requires 5984 (M^+). $\delta_H(CDCl_3)$: 5.35–5.00 (m, 28H, C(1)-*H*), 4.05–2.50 (m, 436H), of which 3.37 (s), 3.48 (s) and 3.62 (s) are for OCH_3 groups. $\delta_C(CD_3OD)$: 100–97 (*C*(1)), including 99.0, 82.5–79, including 83.0, 82.6, 80.4, 79.7, 74–70, including 72.2, 71.8, 63–57, including 61.3, 60.9, 58.9,

58.5. The ^{13}C spectrum is consistent with a lack of C_7 symmetry within the molecule.

Lanthanide complexes of ligand 2

[Eu·2]$^{3+}$. Ligand **2** (0.052 g, 8.7 mmol) was dissolved in anhydrous, freshly distilled, acetonitrile (1 cm^3). Europium trifluoromethanesulfonate (0.008 g, 12.6 mmol) was added and the solution stirred at 70 °C for 24 h. Solvent was removed under reduced pressure, the solid residue was taken up in water (3 cm^3) and purified through benzoylated dialysis tubing for 3 days. Solvent was removed under reduced pressure. Column chromatography using a 0.5 cm depth of silica (100% CH_2Cl_2, then 90% CH_2Cl_2, 9% MeOH, 1% NH_3, silica) gave the complex as a white solid (0.022 g, 3.3 mmol, 39%). Analysis by UV absorption showed the presence of unknown chromophores at 260 and 340 nm. The complex was dissolved in an aqueous solution of tetraethylammonium perchlorate (4×10^{-3} M, 5 cm^3) and placed in benzoylated dialysis tubing. The sealed tubing was placed in an aqueous solution of tetraethylammonium perchlorate (4×10^{-3} M, 1000 cm^3) and stirred for 3 days. The solution within the tubing was then removed and solvent removed under reduced pressure. Analysis by UV absorption showed the presence of no significant chromophore within the range 250–600 nm. *m*/*z* (ES+): 3070 $[M{\cdot}Eu+H]^{2+}$, 3146 $[M{\cdot}Eu+SO_3CF_3]^{2+}$. mp: 130–133 °C. $\delta_H(CD_3OD)$: 23.1 (s, 4H, H_{ax}), 9.0–1.0 (m, 438H), 0.0 (s, 2H, C(6^A)H_2), −3.8 (s, 4H, H_{eq}), −4.2 (s, 4H, H′$_{ax}$), −8.8 (s, 4H, H′$_{eq}$), −9.3 (s, 4H, C*H*CO), −12.0 (s, 4H, C*H*′CO). The ^1H spectrum is consistent with a lack of C_7 symmetry within the complex.

[Tb·2]$^{3+}$. The terbium complex was prepared and isolated in a similar manner to that described for the europium complex, from the ligand **2** (0.050 g, 8.3 mmol) and terbium trifluoromethanesulfonate (0.008 g, 13.2 mmol). (Yield = 0.045 g, 82%) mp: 149–151 °C. *m*/*z* (ES+): 3074 $[M{\cdot}Eu+H]^{2+}$.

1-[6^A-Deoxyheptakis(2,3-di-*O*-methyl)-6^B,6^C,6^D,6^E,6^F,6^G-hexa-*O*-methyl-β-cyclodextrin-6^A-acetamido]-4,7,10-tris(*tert*-butoxycarbonylmethyl)-1,4,7,10-tetraazacyclododecane, 1b

1,4,7-Tris(*tert*-butoxycarbonylmethyl)-1,4,7,10-tetraazacyclododecane (0.54 g, 1.0 mmol) and caesium carbonate (0.66 g, 2.0 mmol) were stirred under argon in freshly distilled acetonitrile (20 cm^3) for 5 min. Mono-6-(chloromethylcarbamoyl)-6-deoxy-per-*O*-methyl-β-cyclodextrin **3b** (1.55 g, 1.0 mmol) was added and the solution stirred at 50 °C for 3 days. Solvent was removed under reduced pressure and the resulting solid purified by column chromatography (CH_2Cl_2 to 4% MeOH–CH_2Cl_2, silica) to give an off-white solid (0.914 g, 0.46 mmol, 45%), mp: 118-120 °C. Found: C: 50.2%, H: 7.83%, N: 2.86%. $C_{90}H_{161}N_5O_{41}{\cdot}3CH_2Cl_2$ requires C: 50.2%, H: 7.57%, N: 3.15%. $R_f = 0.18$ (10% MeOH–CH_2Cl_2; silica). *m*/*z* (ES+): 1004 $[M + 2NH_4]^{2+}$; (MALDI-TOF+): 2007 $[M + K]^+$. $\delta_H(CDCl_3)$: 5.1–5.0 (m, 7H, C(1)-*H*), 4.0–3.0 (m, 127H, of which 3.60, 3.58, 3.47, 3.46 and 3.35 are for OCH_3 groups), 1.43 (s, 18H, C(CH_3)$_3$), 1.41 (s, 9H, C(CH_3)$_3$). $\delta_C(CDCl_3)$: 172.3 (*C*(O)O^{t}Bu), 171.8 (*C*(O)O^{t}Bu), 99.0–97.9 (*C*(1), including 99.0, 98.9, 98.8, 98.6, 98.0), 82.4–79.6 (including 82.3, 82.1, 82.0, 81.9, 81.7, 81.5, 81.4, 80.6, 80.3, 80.0, 79.9, 79.6), 71.3–70.8 (including 71.3, 71.2, 71.0, 70.9, 70.8), 61.5–61.0 (including 61.5, 61.4, 61.3, 61.2, 61.1), 59.8, 59.0–58.1 (including 59.0, 58.9, 58.8, 58.5, 58.3, 58.2, 58.1), 55.7, 28.0, 27.8. ν_{max}: 2906, 2832, 1725, 1154, 1093, 1011 cm^{-1}.

1-[6^A-Deoxyheptakis(2,3-di-*O*-methyl)-6^B,6^C,6^D,6^E,6^F,6^G-hexa-*O*-methyl-β-cyclodextrin-6^A-acetamido]-4,7,10-tris(carboxymethyl)-1,4,7,10-tetraazacyclododecane, 1a

1-[6^A-Deoxyheptakis(2,3-di-*O*-methyl)-6^B,6^C,6^D,6^E,6^F,6^G-hexa-*O*-methyl-β-cyclodextrin-6^A-acetamido]-4,7,10-tris(*tert*-butoxycarbonylmethyl)-1,4,7,10-tetraazacyclododecane **1b** (0.89 g, 0.45 mmol) was dissolved in trifluoroacetic acid (10 cm^3) and stirred at room temperature for 24 h. Solvent was removed under reduced pressure and the solid residue dried under high vacuum to yield an off-white solid (0.81 g, 0.45 mmol, 100%), mp: 89–94 °C. $R_f = 0.00$ (10% MeOH–CH_2Cl_2; silica). *m*/*z* (MALDI-TOF+): 1801 $[M + H]^+$, 1823 $[M + Na]^+$, 1839 $[M + K]^+$. $\delta_H(CDCl_3)$: 4.90–5.70 (m, 7H, C(1)-*H*), 2.90–4.30 (m, 130H) of which 3.64 (s), 3.50 (s) and 3.32 (s) are for OCH_3 groups. ν_{max}: 2934, 2042, 1736, 1676 (C=O), 1466, 1364, 1151, 1090, 1030, 970, 916, 796 cm^{-1}.

Lanthanide complexes of ligand 1a

[Tb·1a]. 1-[6^A-Deoxyheptakis(2,3-di-*O*-methyl)-6^B,6^C,6^D,6^E,6^F,6^G-hexa-*O*-methyl-β-cyclodextrin-6^A-acetamido]-4,7,10-tris(carboxymethyl)-1,4,7,10-tetraazacyclododecane **1a** (0.33 g, 0.18 mmol) was dissolved in Purite water (10 cm^3) and the pH adjusted to 4.2 with the addition of 0.1 M aqueous potassium hydroxide solution. Terbium nitrate pentahydrate (0.25 g, 0.59 mmol) was added and the solution readjusted to a pH of 4.8. The solution was stirred at 60 °C for 18 h, poured into methanol (20 cm^3) and extracted into dichloromethane (2 × 50 cm^3). Solvent was removed under reduced pressure to yield a pale yellow solid (0.29 g, 0.15 mmol, 79%), mp: 180–182 °C. *m*/*z* (MALDI-TOF+): 1980 $[M + Na]^+$. $\delta_H(CD_3OD,\ 20\ °C,\ 65\ MHz)$: 310–210 (br m, 4H, NC*H*CO), 170–120 (br m, 4H, H′$_{ax}$), 70–40 (br m, 4H, NC*H*′CO), 50 to −20 (m, 110H), −40 to −160 (m, 8H, H_{eq} and H′$_{eq}$), −370 to −440 (br m, 4H, H_{ax}).

[Eu·1a]. Ligand **1a** (0.15 g, 0.08 mmol) was dissolved in Purite water (8 cm^3) and the pH adjusted to 4.2 with the addition of 0.1 M aqueous potassium hydroxide solution. Europium nitrate pentahydrate (0.12 g, 0.28 mmol) was added and the solution pH readjusted to 5.3. The solution was stirred at 60 °C for 18 h, poured into methanol (20 cm^3) and extracted into dichloromethane (2 × 50 cm^3). Solvent was removed to yield a white crystalline solid (0.09 g, 0.04 mmol, 55%), mp: 164–166 °C. *m*/*z* (MALDI-TOF+): 1973 $[M + Na]^+$, 1989 $[M + K]^+$. $\delta_H(CD_3OD,\ 20\ °C)$: 39.2 (s, 0.5H, H_{ax}), 38.8 (s, 0.5H, H_{ax}), 36.2 (s, 0.5H, H_{ax}), 36.0 (s, 0.5H, H_{ax}), 35.0 (s, 1H, H_{ax}), 34.0 (s, 1H, H_{ax}), 10.0–0.0 (m, 110H), 0 to −8 (m, 8H, H_{eq} and H′$_{eq}$), −10.9 (s, 0.5H, C*H*CO), −11.3 (s, 0.5H, C*H*CO), −12.5 to −15.5 (m, 4H, H′$_{ax}$), −17.0 (s, 0.5, C*H*′CO), −17.3 (s, 0.5, C*H*′CO), −17.5 to −19.1 (m, 6H, C*H*CO and C*H*′CO).

[Gd·1a]. Ligand **1a** (0.37 g, 0.20 mmol) was dissolved in Purite water (10 cm^3) and the pH adjusted to 4.5 with the addition of 0.1 M aqueous potassium hydroxide solution. Gadolinium nitrate hexahydrate (0.24 g, 0.53 mmol) was added and the solution readjusted to a pH of 5.0. The solution was stirred at 60 °C for 18 h, filtered and partitioned between water and dichloromethane. The aqueous fraction was removed and solvent removed under reduced pressure. The residual solid was taken up in minimum water and purified by dialysis (2000 MWCO filter) for 2 days. Solvent was removed under reduced pressure to yield a white glassy solid (0.175 g, 0.09 mmol, 44%), mp: 242 °C (dec.). *m*/*z* (ES+, H_2O): 978 $[M + 2H]^{2+}$, 985 $[M + H + NH_4]^{2+}$.

[Yb·1a]. Ligand **1a** (0.36 g, 0.20 mmol) was dissolved in Purite water (5 cm^3) and the pH adjusted to 4.2 with the addition of 0.1 M aqueous potassium hydroxide solution. Ytterbium nitrate pentahydrate (0.50 g, 1.11 mmol) was added and the solution readjusted to a pH of 4.8. The solution was stirred at 60 °C for 18 h, filtered and partitioned between water and dichloromethane. The aqueous fraction was separated and solvent removed under reduced pressure. The residual solid was taken up in minimum water and purified by dialysis (1000 MWCO filter) for 2 days. Solvent was removed under reduced pressure to yield a white glassy solid (0.318 g, 0.16 mmol, 81%), mp: 210 °C (dec.). *m*/*z* (ES+, H_2O): 1998 $[M + NH_4]$, 994

$[M + H + NH_4]^{2+}$. δ_H(CD_3OD, 20 °C, 65 MHz): 140–120 (br m, 4H, H_{ax}), 36 to −16 (m, 118H, of which 4.88 (s), 3.46 (s) and 3.30 (s) are for OCH_3), −22 to −60 (br m, H'_{ax}, 4H), −60 to −95 (br m, 8H, CH_2CO).

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