

A gadolinium cryptate with two coordinated water molecules

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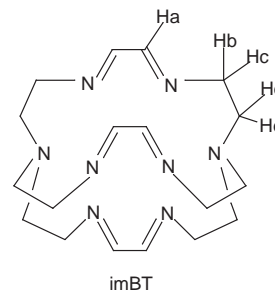
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An X-ray crystallographic structure determination of the nitrate salt of the gadolinium imBT cryptate shows coordination of two water molecules; NMRD relaxometry of this cryptate reveals a relaxivity of $5.8 \text{ mM}^{-1} \text{ s}^{-1}$ (at 10 MHz, 298 K and pH 6), which is pH sensitive over the range 4–9.

Complexes of lanthanide cations are in general characterised by high lability in aqueous solution; therefore specially designed hosts are needed to achieve the kinetic stability required for biomedical applications. Polydentate chelates such as diethylenetriaminepentaacetic acid (DTPAH₅)¹ and 1,4,7,10-tetraazacyclododecane-1,4,7,10-tetraacetic acid (DOTAH₄)² have proved valuable in preventing the release of toxic gadolinium(III) aqua ions under physiological conditions when used as hosts for gadolinium as contrast agents in magnetic resonance imaging (MRI).³ Magneto-pharmaceutical products developed for MRI so far are mostly anionic or neutral Gd(III) complexes *e.g.* Gd(DTPA)²⁻, trade-named MAGNEVIST; Gd(DOTA)⁻, DOTAREM; or the related compounds Gd(DTPA-BMA) {DTPA-BMA = HO₂CCH₂N[(CH₂)₂N(CH₂CO₂H)(CH₂CONHMe)₂], OMNISCAN; and Gd(HPDO3A) [HPDO3A = 10-(2-hydroxypropyl)-1,4,7,10-tetraazacyclododecane-1,4,7-triacetic acid], PROHANCE. Cationic complexes exist which are known to have relaxivity greater than [Gd(H₂O)₉]³⁺ (10 MHz, 298 K); these are macrocyclic complexes such as⁴⁻⁶ [Gd(porphyrin)]⁺, [Gd(texaphyrin)]²⁺, and [Gd(N6-tetraimine)]³⁺. In these cases, the Gd³⁺ ion is above the macrocyclic cavity and exposed to coordination with either water molecules or anions. In cryptates, however, the metal ion is present within the cavity and the water exchange process will differ from that in macrocyclic or chelate complexes. Given the significance of symmetry in relation to the mechanism of electronic relaxation⁷ it is important to examine the relaxivity of any gadolinium cryptates which can be synthesised. To date the strategy of using the *cryptate effect*⁸ to achieve the desired kinetic stability for biomedical applications of lanthanides has been barely exploited. Lehn's polyether cryptand hosts have insufficient coordinating power to compete with solvent O-donors, and while N-donors prove unexpectedly good donors for lanthanide cations,⁹ many existing azacryptand ligands lack the appropriate geometry to efficiently coordinate these relatively large cations. However, the [2 + 3] Schiff-base condensation route to azacryptands^{10,11} offers a range of new hosts including some with good potential for coordination of lanthanide cations.

The small iminocryptand generated by this method¹⁰ using tris(2-aminoethyl)amine and the 2-carbon dialdehyde, glyoxal, (imBT) provides a sufficiently large cavity to ensure good fit of the heavier main group¹² or lanthanide cations. In earlier studies of the hexaimino cryptand imBT we observed unusual kinetic stability^{12,13} for transition and main group ions encapsulated in mononuclear fashion within the hexamine cage; this appears to protect both cation and ligand in that the ligand also seems to be stabilised against the metal-assisted hydrolysis of C=N bonds which normally affects Schiff-base complexes of Lewis-acid cations. Complexation of lanthanide cations is

easily achieved by treating a solution of the ligand, imBT, with the metal salt,[†] under anhydrous conditions. As for other imBT cryptates so far isolated, apart from disilver¹⁴ or dicopper salts,¹⁵ the lanthanide cryptates are obtained as mononuclear complexes. The lanthanide cryptates La[imBT][ClO₄]₃ **1** and [Gd(imBT)(H₂O)₂][NO₃]₃·MeCN **2** need to be made in dried MeCN. Once formed, however, they do not appear to be susceptible to hydrolysis, as they can be recrystallised without deterioration, and treatment with basic aqueous solution (≈1 M NaOH) fails to cause precipitation on standing over a period of weeks, as does the addition of phosphate, even at acidic pH (4.5). Such chemically robust behaviour can, like the observation of satellites in the NMR spectra of the heavy metal cryptates,¹² be attributed to kinetic stability against decomplexation, an unusual and potentially valuable property in Gd(III) complexes.¹⁶



The structure of a single crystal of [Gd(imBT)(H₂O)₂][NO₃]₃·2H₂O **2'** obtained by recrystallisation from MeCN–MeOH[‡] has been solved by X-ray crystallography, and is seen to consist of discrete cations (Fig. 1), nitrate anions and two solvent water molecules. The metal ion is bonded to six nitrogen atoms of the macrobicycle and two water molecules; the nitrate counter ions are uncoordinated. The Gd–N_{imino} distances are somewhat longer (2.60 vs. 2.57 Å) on the strands adjacent to coordinated waters. The Gd–O_{water} distances lie toward the short end of the normal range;^{11,17} Fig. 2 illustrates the siting of the water ligands within the cryptand host. The distances from the metal to the bridgehead nitrogens are too long to represent more than very weak interactions. The demonstrated 8-coordination of the gadolinium(III) cation might be expected to ensure an associative¹⁸ water-exchange process, more rapid than the dissociative exchange normally encountered.

In comparison with the free ligand system where axial and equatorial signals are frozen out in the methylene ¹H NMR spectrum at ambient temperatures and below, that of La[imBT]³⁺ in CD₃CN indicates a fully mobile conformation consisting of a pair of triplets illustrating equivalence of the H_B/H_C and H_D/H_E pairs due to rapid interconversion of configurations on the ¹H NMR time scale throughout the fluid range of the deuterioacetonitrile solvent. The gadolinium cryptate **2** fails, as expected, to exhibit any ligand spectrum; the only signals to be seen are those of solvate molecules. Preliminary investigation of relaxivity of gadolinium cryptates¹⁹ at 500

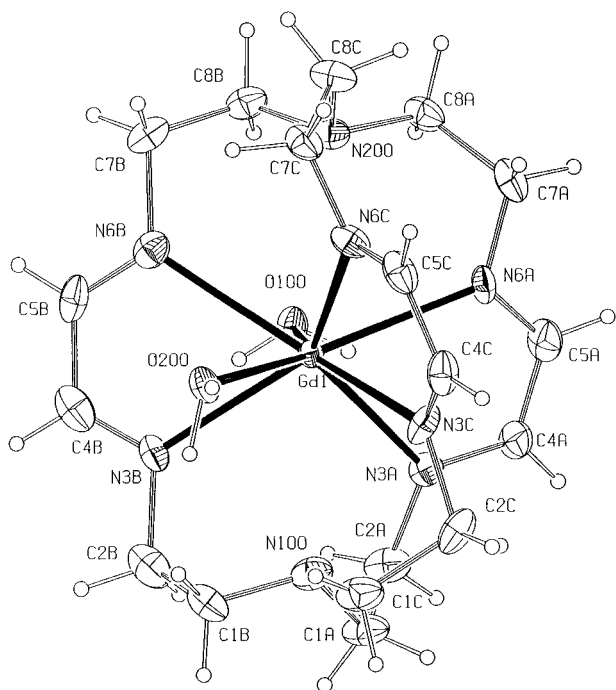


Fig. 1 Crystal structure of the cation in **2'** together with atomic numbering scheme; ellipsoids at 30% probability. Selected distances (Å) Gd–N(6C,3A,3C) 2.563(8)–2.569(9); Gd–N(6A,6B,3B) 2.600(11)–2.608(8); Gd–O 2.434(6), 2.466(7) Å, Gd⋯N(100), Gd⋯N(200) 2.973(9), 2.982(8).

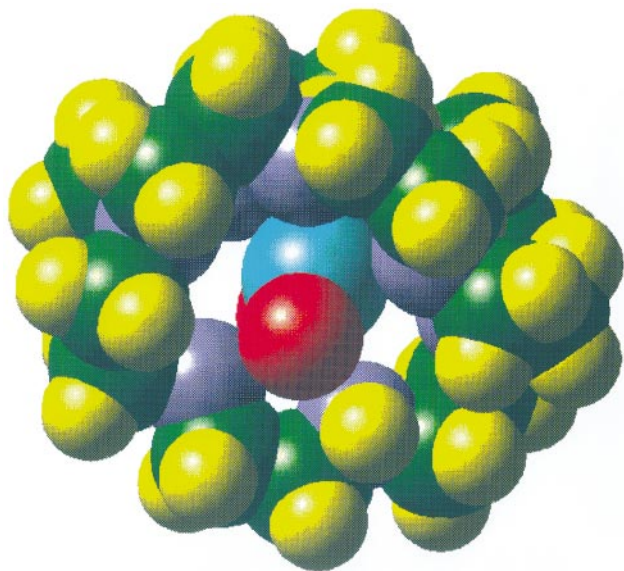


Fig. 2 Space-filling model of **2** illustrating siting of coordinated water: colours; gadolinium light blue, nitrogen blue, oxygen red, carbon green, hydrogen yellow.

MHz indicates that it is faster than 2 mM s^{-1} at this frequency. In order to quantify relaxivity at the low frequencies normally quoted, we have examined the relaxation rates of the water solvent molecules by an NMRD study§ over the frequency range 0.1–100 MHz.

At 10 MHz, pH 6 and 298 K, the proton relaxivity of **2** obtained from a plot of relaxation rate against complex concentration, is $5.8 \text{ mM}^{-1} \text{ s}^{-1}$. The relaxivity is sensitive to pH change and it reduces to 3.5 at pH 9. The magnetic field dependence of the proton relaxivities (Fig. 3) was fitted to the modified Solomon–Bloembergen–Morgan (SBM) theory²⁰ by assuming that parameters established in our X-ray study apply in solution: *i.e.*, hydration number, $q=2$, and distance of the metal from first coordination sphere protons, $r=3 \text{ Å}$. By extrapolation, the closest distance to the water protons in the second

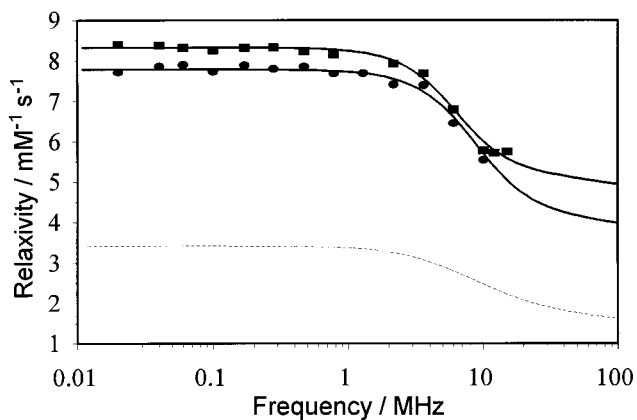


Fig. 3 NMRD profiles, experimental and theoretically fitted, of the $[\text{Gd}(\text{imBT})(\text{H}_2\text{O})]^{3+}$ complex at 298 K (■) and at 310 K (●); calculated outer-sphere contribution to the NMRD profile at 298 (-----).

coordination sphere was estimated as $d=4 \text{ Å}$, and this, along with the parameters τ_v (correlation time for electron relaxation) set at 18 ps, diffusion coefficient, D_{diff} , of $2.5 \times 10^{-5} \text{ cm}^2 \text{ s}^{-1}$, and static zero field splitting $D(\text{ZFS})=0$, was fitted in the final fitting procedure.

The values of Δ (the average quadratic transient zero field splitting), τ_m (chemical exchange time) and τ_r (rotational correlation time) obtained from the NMRD profile are 0.033 cm^{-1} , $4.18 \mu\text{s}$ and 71 ps respectively and they are typical values of low molecular mass Gd(III) complexes. The rate of water exchange obtained from the SBM fitting calls for comment. As the inner-sphere exchange mechanism is generally considered to dominate the relaxation process, it might be expected that the presence of two coordinated water molecules would give rise to enhanced relaxivity. However, the value of $4.18 \mu\text{s}$ obtained for the rate of water exchange is slower than for comparable negatively charged or neutral complexes; a trend noted earlier²¹ by Parker and co-workers. It seems likely that as in this earlier case,²¹ exchange of coordinated water molecules is not the major relaxation mechanism; instead, prototropic exchange arising from the relatively high acidity of water coordinated to the Gd(III) cation is the dominant mechanism. Potentiometric titration shows²² a $\text{p}K_a$ of around 8, associated with deprotonation of the coordinated water. Above this pH, the complex is in the hydroxo form where inner-sphere water exchange is no longer possible; the fall off in relaxivity with increasing pH supports this hypothesis. We plan to carry out variable temperature ¹⁷O NMR transverse relaxation measurements as a function of pH to further investigate this behaviour.

Although the tendency of positively charged gadolinium complexes to localise in negatively charged *e.g.* bone or membrane tissue²³ means that they are rarely utilised as contrast agents, the relaxivities reported here are promising. Together with the unusually high kinetic stability deriving from the cryptate effect, they warrant a full investigation of the relaxation behaviour of these cryptates to allow comparison with other gadolinium complexes currently in use as contrast agents. The possibilities for substitution on the cryptand skeleton to generate *e.g.* charge-neutral derivatives or biological conjugates makes these systems additionally attractive.

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Notes and references

† The lanthanide cryptates **1**, **2** were prepared as follows: to a solution of 1 mmole of imBT prepared as described^{24,25} elsewhere in 20 cm^3 of

dry acetonitrile and enough CHCl_3 to aid dissolution of the ligand, was added 1.2 mmole of the appropriate lanthanide salt in 20 cm^3 dry CH_3CN under N_2 atmosphere. Upon removal of the solvent (rotary evaporation or standing in air) a white microcrystalline solid was obtained. This could be recrystallised from acetonitrile–methanol; crystals of **2'** used for crystal structure determination, were obtained in this way. **1** FAB-MS: m/z 695(100); 596(65); % C,H,N (calc. values in parentheses): 27.2(27.9); 3.8(3.8); 13.7(14.1). **2** FAB-MS: m/z 640(14); 658(5); 577(46); 595(35); % C,H,N (calc. values in parentheses): 30.8(30.8); 4.8(4.8); 21.6(21.6). Yields of recrystallised samples: **1**, 20%; **2'**, 30%. **Safety note:** although all perchlorates must be treated as potentially explosive, and the quantities indicated in the syntheses described should not be exceeded, we experienced no problems in working with complex **1** in the manner described.

‡ Crystal data: $\text{C}_{18}\text{H}_{38}\text{GdN}_{11}\text{O}_{13}$, $M = 773.84$, orthorhombic, space group $Pn2_1a$; $a = 15.798(12)$, $b = 14.100(12)$, $c = 13.736(12)$ Å; $U = 3060(4)$ Å³, $Z = 4$, $D_m = 1.680$ Mg m^{-3} , $F(000) = 1564$, $\mu = 2.244$ mm^{-1} , $T = 293$ K, largest differential peak and hole 2.131, -1.582 $\text{e} \text{Å}^{-3}$ situated close to the Gd atom. Data were collected on a Marresearch Image Plate system using Mo-K α radiation. 95 frames were collected using 2 min per frame and 8254 reflections were collected up to 2θ of 50° of which 4545 were unique, $R_{\text{int}} = 0.0372$. The structure was refined to an $R1$ of 0.0422, $wR2 = 0.1299$ for 3911 data with $I > 2\sigma(I)$. All calculations were carried out on a Silicon Graphics R4000 Workstation at the University of Reading. CCDC reference number 186/1198. See <http://www.rsc.org/suppdata/dt/1998/3711/> for crystallographic files in .cif format.

§ Relaxation measurements of **2** were carried out on samples in the concentration range 0–25 mM at pH 6, 298 K. pH studies were carried out in tris(hydroxymethyl)aminoethane (0.3 M) solution and titrated with aqueous HCl (6 M). All NMRD profiles were acquired on the Stelar FFC relaxometer from 0.00024 to 0.36 T (0.01–15 MHz). The profiles are theoretically fitted using a computer program which calculates the paramagnetic enhancements due to inner and outer sphere contributions, and takes into account both dipolar and contact relaxation, as well as zero-field splitting, g anisotropies and hyperfine coupling, if any.²⁶ The paramagnetic contribution of the water relaxivity of the Gd(III) cryptate has been calculated by subtracting the values of water proton relaxation rates, $0.38 \text{ mM}^{-1} \text{ s}^{-1}$.

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