

# NMR-Studies of the Interaction of Metal Ions with Poly-(1,4-hexuronates). V. Quantitative Separation of Contact and Dipolar Contributions to Lanthanide Induced Shifts in $^1\text{H}$ NMR Spectra of Methyl $\alpha$ -D-Gulo- and Methyl $\beta$ -D-Hamamelopyranosides, 1,6-Anhydro- $\beta$ -D-manno-, 1,6-Anhydro- $\beta$ -D-talo- and 1,6-Anhydro- $\beta$ -D-allo-pyranoses and Epi-inositol

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Some 1:1 complexes between epi-inositol and various lanthanide ions were analysed according to a mirror symmetric structure in solution where an *ax-eq-ax* sequence of three hydroxy groups formed the binding site. It was assumed that in the substrate three of the protons (H6, H2, H4) showed only dipolar lanthanide induced shifts (LIS) in the case of  $\text{Pr}^{3+}$ . The McConnell-Robertson equation was found to account for the dipolar part of the LIS for all Ln ions when compared with data given by Bleaney. The remaining LIS corresponded relatively well with theoretical contact LIS from  $\text{Ce}^{3+}$  to  $\text{Tb}^{3+}$ . Most of the heavier Ln ions gave bad agreement. It was concluded that separation of LIS by using theoretical shift patterns should be limited, at present, to the lighter lanthanides.

Such analysis was carried out for a series of glycopyranosides containing a similar binding site as epi-inositol. The analysis showed that whereas the contact LIS in epi-inositol was large only when bond connecting the proton and the cation formed a planar zig-zag arrangement, the introduction of a ring oxygen altered this pattern leading to a considerable contact interaction also at other protons.

The McConnell-Robertson equation was used in an attempt to locate the Ln-ion bound to methyl  $\alpha$ -D-gulopyranoside.

Numerous attempts have been made recently in applying LIS in NMR spectra for obtaining structural informations of molecules and their metal complexes in solution.<sup>1</sup> One limitation in the method is that two different mechanisms operate in LIS: Fermi contact and dipolar interaction, and that only the dipolar LIS which is geometry dependent, has the potential for quantitative structure determination. Reliable methods are therefore required to dissect the two shift contributions before the technique can become generally applicable. The object of the present paper is to investigate the utility and limitation of such methods for a certain type of compounds containing a high affinity binding site for Ln-ions, consisting of three consecutive hydroxy groups with an *ax-eq-ax* arrangement.

Many recent studies of LIS provide evidence for significant contact interactions at  $^1\text{H}$  nuclei close to the complexing site.<sup>2–4</sup> In epi-inositol, Fig. 1, the contact interaction is found to be stereospecific,<sup>2</sup> i.e. it is greatest when the bonds connecting the proton and the cation form a planar zig-zag arrangement. As pointed out by Angyal,<sup>2</sup> the contact interaction may then be detectable over several intervening bonds. We have used the LIS-data of Angyal<sup>4</sup> for epi-inositol and utilized the mirror symmetry of the complex, and the resulting sym-

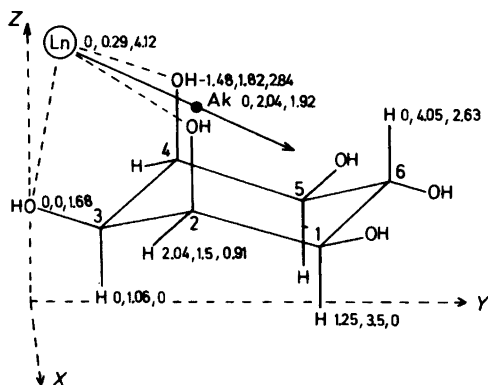


Fig. 1. The proposed Ln-epi-inositol complex located in cartesian coordinates as discussed in the text. The complex has a mirror symmetry through Ln, O3, H3, and H6 in the  $yz$ -plane. The coordinates ( $x, y, z$ ) are given in units of Å.

metry of the LIS, to separate between the contact and dipolar contributions to the shifts. By this quantitative method we are able to confirm the conclusions which Angyal<sup>3</sup> arrived at based upon qualitative arguments. In addition the results suggest that the general structure independent method<sup>5</sup> for separation of contact and dipolar shifts based upon theoretical values<sup>6,7</sup> for different Ln

ions, in the present case should be limited to the lighter Ln-ions.

In our earlier study<sup>8</sup> of methyl  $\alpha$ -D-gulopyranoside (1) the shift ratio for  $\text{Eu}^{3+}$  and  $\text{Pr}^{3+}$  at the H2 proton was found to be significantly different from the nearly equal ratios observed at the other protons. In view of the results for epi-inositol it is reasonable to suggest that the planar zig-zag arrangement of Ln-O2-C2-H2 in 1 may cause a considerable contact shift contribution at proton H2 also in this case. In order to clarify this point we have remeasured LIS for 1 with several of the lighter Ln ions, and used the general structure independent method<sup>5</sup> to dissect the shifts. This method was also applied to recently reported LIS data<sup>9</sup> for methyl,  $\beta$ -D-hamameloside (2), 1,6-anhydro- $\beta$ -D-manno- (3), talo- (4) and allopyranoses (5).

The set of dipolar shifts obtained for the Ln complex of methyl  $\alpha$ -D-gulopyranoside has been used with the computer search program described earlier,<sup>10</sup> to obtain information of the structure of the complex. The results obtained have been compared to those reported previously.<sup>8</sup>

## RESULTS

*Dissection of LIS for complexes with mirror symmetry.* The Ln-epi-inositol complex, Fig. 1, has a mirror symmetry through O3, H3, H6 and the

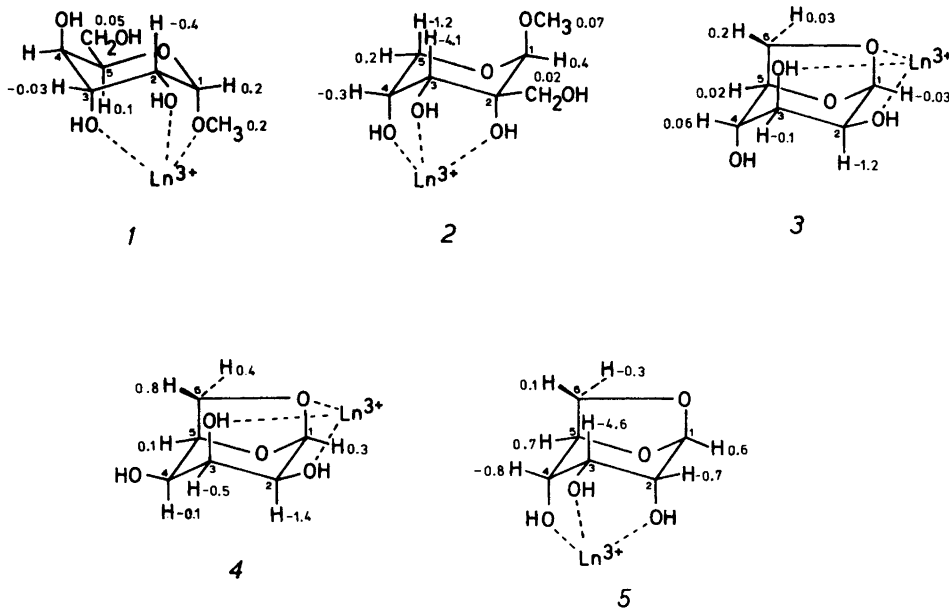


Table 1. Comparison of theoretical and estimated dipolar and contact shifts induced by lanthanides in epi-inositol.<sup>a</sup> Positive values indicate downfield shifts.

Lanthanide	Rel. theor. shifts		Lanthanide induced shifts (ppm) in epi-inositol						H6		
	Bleaney <sup>2</sup>	Goldring <sup>3</sup>	H1,5 $\Delta v_{\text{obs}}^a$	H2,4 $\Delta v_{\text{obs}}^b$	$\Delta v_{\text{cont}}^c$	$\Delta v_{\text{dip}}^b$	$\Delta v_{\text{dip}}^c$	$\Delta v_{\text{cont}}^c$	$\Delta v_{\text{obs}}^c$	$\Delta v_{\text{dip}}^c$	$\Delta v_{\text{cont}}^c$
La <sup>3+</sup>	0	0	0.17	0	0	0	0	0	0.07	0	0
Ce <sup>3+</sup>	-6.3	0.98	-0.4	-0.6	0.03	-0.73	-0.7	-0.8	-1.05	1.05	0
Pr <sup>3+</sup>	-11	3	-1.05	-1.92	0.70	-2.32	-2.05	-2.56	-3.5	4.06	0
Nd <sup>3+</sup>	-4.2	4.5	0.3	-0.93	1.06	-1.12	-1.10	-1.23	-1.65	6.8	0
Sm <sup>3+</sup>	-0.7	-0.06	-0.05	-0.2	-0.02	-0.24	0.15	-0.26	-0.37	0.01	0
Eu <sup>3+</sup>	4	-10.7	-0.35	0.85	-1.37	1.03	1.6	-1.13	1.65	-8.9	0
Tb <sup>3+</sup>	-86	-31.8	-18	-10.2	-7.93	-12.4	-19	-13.6	-19	-22	0
Dy <sup>3+</sup>	-100	-28.5	-21	-14	-7.2	-16.9	-21	-18.6	-26	-5.95	0
Ho <sup>3+</sup>	-39	-22.6	-8	-8.1	-0.07	-9.8	-5	-10.7	-15	-1.85	0
Er <sup>3+</sup>	33	-15.4	0.5	1.96	-1.63	2.36	2.1	2.6	3.7	-6.6	0
Tm <sup>3+</sup>	53	-8.2	-4.5	3.0	-7.7	3.67	-10	4.03	5.7	-20.5	0
Yb <sup>3+</sup>	22	-2.6	-0.25	2.4	-2.82	2.85	-2.05	3.14	4.45	-8.5	0
Lu <sup>3+</sup>	0	0	0.16	0	0	0	0.23	0	0.06	0	0

<sup>a</sup>  $\Delta v_{\text{obs}} = \Delta v_{\text{obs}}(\text{La}^{3+}) + \Delta v_{\text{dip}} + \Delta v_{\text{cont}}$ . <sup>b</sup>  $\Delta v_{\text{dip}}$  was calculated according to  $\Delta v = C(3\cos^2\theta - 1)/r^3$  by use of appropriate coordinates given in Fig. 1. The constant  $C$  was adjusted to give only dipolar shift at proton H6. <sup>c</sup>  $\Delta v_{\text{cont}}$  was assumed to be zero at H2,4 for Pr<sup>3+</sup>, in order to determine the location of the symmetry axis.

Ln<sup>3+</sup> ion, which is demonstrated by equally large LIS observed at protons H2,4 and H1,5, as shown in Table 1. This means that the principal magnetic symmetry axis must be located in the mirror plane, corresponding to the  $yz$ -plane in Fig. 1. The Ln<sup>3+</sup>-oxygen distance used,  $\sim 2.46$  Å, was estimated as a mean value of reported solid state crystal values.<sup>11-14</sup> Space coordinates for the hydrogens of epi-inositol were obtained by using conformational angles and bond lengths given in a crystal structure<sup>15</sup> and assuming a C-H distance of 1.09 Å. As pointed out by Angyal,<sup>3</sup> the observed LIS for protons H6 and H2,4 can be accounted for by an approximately pure dipolar mechanism. This is demonstrated in Fig. 2 where the measured LIS at H6 is shown to match the theoretical dipolar LIS<sup>6</sup> through the Ln series quite closely. The same finding prevails for measured LIS at H2,4 except for Tm<sup>3+</sup> and Yb<sup>3+</sup>. Since the contact interaction caused by Pr<sup>3+</sup> is small,<sup>7</sup> it may therefore be a good approximation to use the total Pr<sup>3+</sup> induced shifts at protons H6 and H2,4 in determining the orientation of the principal magnetic symmetry axis in the presumed axially symmetric complex by using the space coordinates defined in Fig. 1 and the McConnell-Robertson equation<sup>16</sup>

$$\Delta v/v = C(3\cos^2\theta - 1)/r^3 \quad (1)$$

Here  $r$  is the length of the vector joining the Ln ion and the proton and  $\theta$  is the angle between this vector and the symmetry axis.

Calculations on this rigid substrate molecule is thus simple. The axis was found to go through the point Ak as indicated in Fig. 1. By assuming the same structure for the homologous Ln complexes, the relative dipolar LIS for all protons are given by eqn. (1). The absolute dipolar LIS values were found by adjusting the constant  $C$  in eqn. (1) according to pure dipolar shift at proton H6 throughout the Ln series. The relationship between the LIS contributions is given by

$$\Delta v_{\text{obs}} = \Delta v_{\text{obs}}(\text{La}^{3+}) + \Delta v_{\text{dip}} + \Delta v_{\text{cont}} \quad (2)$$

Hence, the numerical values for the contact LIS,  $\Delta v_{\text{cont}}$ , are found as the difference between the observed LIS,  $\Delta v_{\text{obs}}$ , corrected for La-induced diamagnetic shifts,  $\Delta v_{\text{obs}}(\text{La}^{3+})$ , and the calculated dipolar LIS,  $\Delta v_{\text{dip}}$ . The resulting dipolar and contact LIS values are listed in Table 1. As shown in Fig. 2 the estimated relative contact LIS at H3 and H1,2

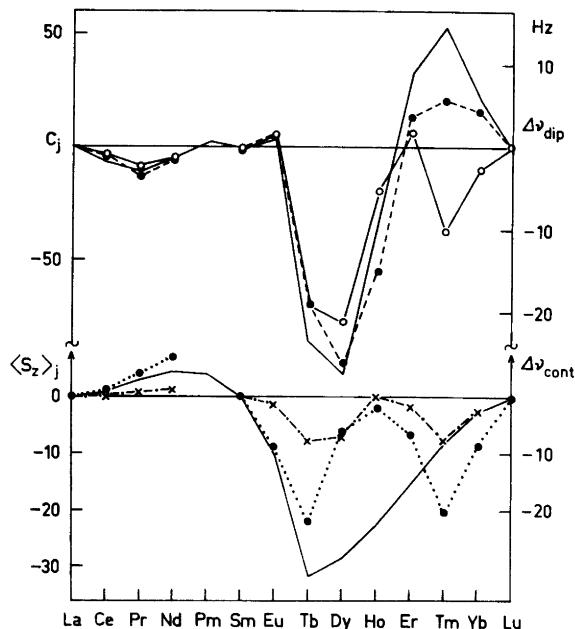


Fig. 2. Variation of the theoretical relative dipolar,  $C_j$ , and contact,  $\langle S_z \rangle_j$ , LIS (—) through the lanthanide series. Measured LIS: H6 (●---●), H2,4 (○—○). Estimated contact LIS: H1,2 (×-×), H3 (●··●). (All protons of epi-inositol).

both agree with theory down to  $Tb^{3+}$ . However, a substantial discrepancy occurs for the heavier lanthanides.

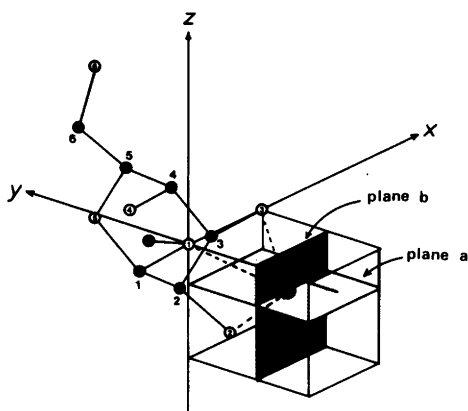


Fig. 3. Location of the cube used for calculation of the  $Ln^{3+}$  - methyl  $\alpha$ -D-gulopyranoside complex. The coordinates for the lanthanide ion were found to be  $x=1.1 \text{ \AA}$ ,  $y=-1.5 \text{ \AA}$  and  $z=-0.8 \text{ \AA}$ . The magnetic axis has been omitted from the figure in sake of clarity.

*Dissection of LIS by the general structure-independent method.*<sup>5</sup> The methyl  $\alpha$ -D-gulopyranoside (1), Fig. 3, is a neutral sugar derivative containing an ax-eq-ax sequence of two hydroxy groups and one  $OCH_3$  group, which most probably constitute the  $Ln^{3+}$  binding site as reported recently. Due to the ring oxygen and the  $OCH_3$  group, this substrate lacks the symmetry found in epi-inositol which enabled us to deduce the structure of its  $Ln$  complex in a straightforward manner. In the case of (1) we used a structure-independent method, outlined and tested by Reilly *et al.*,<sup>5</sup> where advantage is taken of the fact that dipolar and contact LIS vary in a characteristic but different way with  $Ln$  ions. It is essential for using this method that the structure of the complexes stay constant throughout the lanthanide series employed, and that they have axial symmetry around their  $Ln$  cations. Our results for epi-inositol support this assumption in case of the lighter lanthanides. The contact hyperfine coupling constant and the ligand field should then remain invariant with changing the  $Ln$  ion. The contact shift is proportional to the expectation value of the  $z$  component of the total electron spin,  $\langle S_z \rangle$ , whereas

Table 2. Measured LIS values<sup>a</sup> (ppm) at 0.24 M substrate and 0.24 M lanthanide,  $\Delta v_{\text{obs}}$ , for methyl  $\alpha$ -D-gulopyranoside and the corresponding computed values for contact,  $\Delta v_{\text{cont}}$ , and dipolar,  $\Delta v_{\text{dip}}$ , contributions.

	LIS	Pr <sup>3+</sup>	Nd <sup>3+</sup>	Eu <sup>3+</sup>	Tb <sup>3+</sup>	$G_i$	$F_i$	AF
H1	$\Delta v_{\text{obs}}$	-0.60	-0.32	0.35	-3.16			
	$\Delta v_{\text{dip}}$	-0.51	-0.20	0.19	-4.02	0.047	-0.019	0.082
	$\Delta v_{\text{cont}}$	-0.06	-0.08	0.20	0.60			
H2	$\Delta v_{\text{obs}}$	0.20	0.35	-0.44	-0.40			
	$\Delta v_{\text{dip}}$	0.11	0.04	-0.04	0.88	-0.010	0.041	0.190
	$\Delta v_{\text{cont}}$	0.12	0.18	-0.44	-1.31			
H3	$\Delta v_{\text{obs}}$	0.41	0.20	-0.15	2.80			
	$\Delta v_{\text{dip}}$	0.39	0.15	-0.14	3.03	-0.035	0.003	0.051
	$\Delta v_{\text{cont}}$	0.01	0.01	-0.03	-0.10			
H5	$\Delta v_{\text{obs}}$	-0.60	-0.32	0.28	-3.40			
	$\Delta v_{\text{dip}}$	-0.53	-0.20	0.19	-4.13	0.048	-0.013	0.094
	$\Delta v_{\text{cont}}$	-0.04	-0.06	0.14	0.41			
H6	$\Delta v_{\text{obs}}$	-0.16	-0.08	0.07	-0.60			
	$\Delta v_{\text{dip}}$	-0.12	-0.05	0.04	-0.93	0.011	-0.004	0.311
	$\Delta v_{\text{cont}}$	-0.01	-0.02	0.05	0.14			
OCH <sub>3</sub>	$\Delta v_{\text{obs}}$	-0.61	-0.32	0.28	-3.24			
	$\Delta v_{\text{dip}}$	-0.52	-0.20	0.19	-4.08	0.047	-0.014	0.124
	$\Delta v_{\text{cont}}$	-0.04	-0.06	0.15	0.44			

<sup>a</sup>NMR spectra were measured with a Varian XL-100-12 NMR spectrometer at 100 MHz in D<sub>2</sub>O solutions at 30 °C. Positive values indicate downfield shifts. The diamagnetic La<sup>3+</sup> induced LIS could not be measured and  $\Delta v_{\text{par}}$  was approximated by  $\Delta v_{\text{obs}}$  in the calculations.

the dipolar shift is proportional to the constant  $C$  in eqn. (1). These terms are dependent only upon the Ln ion employed. Under these conditions the sum of the two paramagnetic LIS contributions,  $\Delta v_{\text{par}}$ , at proton  $i$  induced by the Ln ion  $j$  may be written

$$\Delta v_{\text{par},ij} = \Delta v_{\text{cont},ij} + \Delta v_{\text{dip},ij} = F_i \langle S_z \rangle_j + G_i C_j \quad (3)$$

where  $F_i$  and  $G_i$  are related to the structure and depend only upon the proton  $i$  being observed. The factor  $F_i$  reflects the effectivity of contact interaction at proton  $i$  while the  $G_i$  value includes the geometric factor of that proton in eqn. (1). By measuring the LIS for Pr<sup>3+</sup>, Nd<sup>3+</sup>, Eu<sup>3+</sup> and Tb<sup>3+</sup> (varying indices  $j$ ), Table 2, an appropriate number of simultaneous equations was set up, eqn. (3), using theoretical values for  $\langle S_z \rangle_j$  and  $C_j$  given by Golding<sup>7</sup> and Bleaney,<sup>6</sup> respectively. In order to obtain the weighted average least squares values for  $F_i$  and  $G_i$  in Table 2, the observed predominant shift for

Tb<sup>3+</sup> and the values of  $\langle S_z \rangle$  and  $C$  for this ion were scaled down by a factor of 8 before solving the equations. In the calculated agreement factors

$$AF = \left[ \frac{\sum_i (\Delta v_{\text{par},i}^{(\text{obs})} - \Delta v_{\text{par},i}^{(\text{cal})})^2}{\sum_i (\Delta v_{\text{par},i}^{(\text{obs})})^2} \right]^{1/2} \quad (4)$$

also listed in Table 2, the full observed paramagnetic LIS,  $\Delta v_{\text{par}}^{(\text{obs})}$ , and calculated LIS,  $\Delta v_{\text{par}}^{(\text{cal})}$ , were used. The resulting dipolar and contact shift values are listed in Table 2. Here  $\Delta v_{\text{par}}$ , approximated by  $\Delta v_{\text{obs}}$ , is not perfectly equal to the sum of  $\Delta v_{\text{dip}}$  and  $\Delta v_{\text{cont}}$  because these values found via a least square approach deviated from a perfect fit *i.e.* the agreement factor  $AF > 0$ .

This method was also used to analyse reported LIS data<sup>9</sup> for the complexes 2, 3, 4 and 5. The dipolar and contact LIS contributions are given in Table 3. The contact shifts (ppm) for Eu<sup>3+</sup> in these complexes are indicated in the figures. The LIS data for 1–5 were measured at about the same

Table 3. Measured LIS values,  $\Delta v_{\text{par}} = \Delta v_{\text{obs}} - \Delta v(\text{La}^{3+})$  at 0.25 M substrate and 0.25 equiv. of lanthanide, in  $^1\text{H}$  NMR spectra, and the corresponding computed values for contact,  $\Delta v_{\text{cont}}$ , and dipolar,  $\Delta v_{\text{dip}}$ , contributions. Positive values indicate downfield shifts.

Cation	$\Delta v_{\text{par}}$	$\Delta v_{\text{dip}}$	$\Delta v_{\text{cont}}$	$\Delta v_{\text{par}}$	$\Delta v_{\text{dip}}$	$\Delta v_{\text{cont}}$	$\Delta v_{\text{par}}$	$\Delta v_{\text{dip}}$	$\Delta v_{\text{cont}}$	$\Delta v_{\text{par}}$	$\Delta v_{\text{dip}}$	$\Delta v_{\text{cont}}$	$\Delta v_{\text{par}}$	$\Delta v_{\text{dip}}$	$\Delta v_{\text{cont}}$	$\Delta v_{\text{par}}$	$\Delta v_{\text{dip}}$	$\Delta v_{\text{cont}}$
1,6-Anhydromannose																		
$\text{Pr}^{3+}$	-1.67	-1.68	0.01	0.87	0.62	0.35	0.96	0.91	0.04	H2	H3	H4	H5	H6 <i>endo</i>	H6 <i>exo</i>			
$\text{Nd}^{3+}$	-0.65	-0.64	0.01	1.12	0.24	0.51	0.34	0.35	0.06									
$\text{Eu}^{3+}$	0.58	0.62	-0.03	-1.33	-0.23	-1.23	-0.49	-0.33	-0.14									
1,6-Anhydrotalose																		
$\text{Pr}^{3+}$	-1.62	-1.55	-0.08	0.52	0.12	0.40	0.77	0.63	0.14									
$\text{Eu}^{3+}$	0.83	0.56	0.27	-1.47	-0.04	-1.43	-0.72	-0.23	-0.49									
1,6-Anhydroallose																		
$\text{Pr}^{3+}$	-1.88	-1.71	-0.17	-0.38	-0.57	0.19	1.05	-0.25	1.30									
$\text{Eu}^{3+}$	1.24	0.62	0.62	-0.48	0.21	-0.69	-4.55	0.09	-4.64									
Methyl $\beta$ -D-hamamelopyranoside																		
$\text{Pr}^{3+}$	-1.60	-1.48	-0.12	1.31	0.15	1.16	1.38	1.29	0.09	H3	H4	H5 <sub>eq</sub>	H5 <sub>ax</sub>	CH <sub>2</sub>	Me			
$\text{Eu}^{3+}$	0.98	0.54	0.44	-4.19	-0.06	-4.14	-0.79	-0.47	-0.32									

conditions and allow an approximate comparison of the LIS values in these compounds. The LIS values for epi-inositol are reported as limiting shifts and are not directly comparable as far as absolute shifts are concerned.

*Evaluation of the Ln ion binding site.* For methyl  $\alpha$ -D-gulopyranoside the existence of a single, best-fit site in the vicinity of the three oxygen functions O1, O2 and O3 was investigated by computer calculation of an agreement factor for space points equally spaced within a cube. The cube was located with one corner in O1, one corner in O3, and with O2 lying in the plane of the side O1–O3 as shown in Fig. 3. The edge of the cube is thus equal to the distance O1–O3 (2.6 Å). No other atoms in the carbohydrate structure will be located inside this cube. The agreement factor is given by a formula similar to eqn. (7) in which the dipolar LIS is used, and where  $\Delta v_{\text{dip},i}^{(\text{obs})}$  was taken as the value relative to the dipolar shift for H1. The resulting value was divided by six to give the average square-root deviation per proton signal observed.

Starting at the far end of the cube with respect to the oxygen atoms, the agreement factor was calculated for all points constituting a cube-grid of 0.2 Å. The calculation is equivalent to placing the Ln ion in a grid position, calculating the theoretical shifts and the agreement factor and minimizing the latter with respect to the direction of the magnetic axis through a search procedure. The minimum value for the agreement factor is thus obtained for all the grid points. A smaller part of the cube was also investigated using a grid size of 0.1 Å. These calculations gave a contour plot for the AF values consistent with only one minimum within the cube, thus demonstrating that there exists only one best-fit site for the Ln ion within the part of space around the sugar molecule which is physically and chemically reasonable. Fig. 4 shows contour plots in two perpendicular planes a and b through the best-fit position. In three dimensional space the inner contours form a cigarshaped cavity. The equidistance corresponds to 0.0003, and the minimum value of the agreement factor is 0.0024 (grid size 0.1 Å, angular accuracy for magnetic axis 0.1°). The distances from the three oxygen functions O1, O2, and O3 are 2.0, 2.2 and 2.3 Å, respectively. The direction of the magnetic axis is defined by the angle  $\alpha = 62.8^\circ$  with the x-axis and  $\beta = 62.8^\circ$  with the z-axis (intersection with the xz-plane in 0.20, 0.00, 0.10).

The evaluated binding site was checked by using

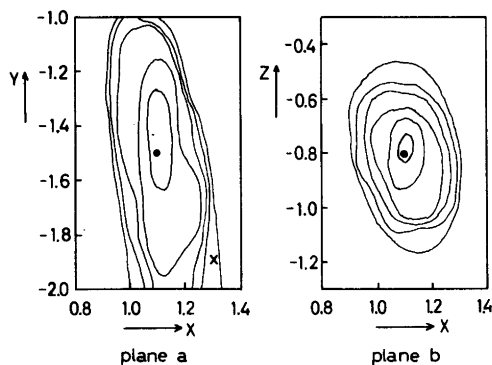


Fig. 4. Contour plot of two perpendicular planes through the best-fit position according to the agreement factor between observed and calculated dipolar LIS for methyl  $\alpha$ -D-gulopyranoside. The average position from crystal structures lies in plane a, and is marked with a cross.

relaxation reagent. After addition of  $\text{Pr}(\text{NO}_3)_3$  to separate overlapping resonances,  $\text{Gd}(\text{NO}_3)_3$  was added and its relaxation effects on the separated resonances were measured. Appropriate corrections for outer sphere effects were made by subtracting the observed relaxation rate for internal dioxane. The proton relaxivity in dioxane induced by  $\text{Gd}^{3+}$  was very low indicating that dioxane did not bind significantly to  $\text{Ln}^{3+}$  ions in  $\text{D}_2\text{O}$  solutions. The net effects of the  $\text{Gd}^{3+}$  in the binding site were of the order:  $\text{H1} \approx \text{H2} \approx \text{H3} \approx \text{H5} \approx \text{OMe} > \text{H4} > \text{H6}$ . This is in accordance with an  $r^{-6}$  dependence valid for a dipolar paramagnetic relaxation mechanism where  $r$  is the distance from the substrate proton to the evaluated Ln binding site.

## DISCUSSION

Here we have dealt with complexes consisting of six-membered rings, cyclitols and neutral sugar derivatives, possessing three neighbouring hydroxy groups in an *ax-eq-ax* sequence which are known to form an active site of rather high affinity for calcium and lanthanide ions.<sup>3</sup> Our estimates of contact, and dipolar shift contributions to proton LIS data clearly demonstrate some geometrical relationships which strongly influence the effects of contact interaction in these lanthanide complexes.

The contact shifts are caused by either or both of two mechanisms: (a) Direct delocalization of

unpaired electron spin density from the paramagnetic cation leading to<sup>17</sup> upfield shifts for all protons with  $\text{Eu}^{3+}$  and (b) spin polarization giving alternating shifts in pairs along the bonds.<sup>18</sup> The fact that alternating upfield and downfield contact shifts occur indicates clearly that spin polarization plays an important role in these systems. Evidently the total effect of contact interaction is increased by a planar zig-zag path which is most clearly demonstrated by the LIS data for epi-inositol,<sup>4</sup> Table 1. Here the upfield contact shift for  $\text{Eu}^{3+}$  at proton H3 is surprisingly large,  $\sim 8.9$  ppm. The bonds connecting H1,5 to the cation are in a plane, those of H2,4 are not. These two proton pairs show contact shift in opposite direction which is about a factor of 4 greater at H1,5 even though these protons are one bond further away from the cation. A similar type of stereospecificity of contact interaction is found in all the investigated complexes. The pyranose rings show a more complex LIS pattern than the cyclohexane ring.

The validity of our analysis is strongly supported by the internal consistency of the results obtained for epi-inositol bound to lighter lanthanides. In dissecting the LIS data for these complexes, no assumptions concerning values of theoretical shifts were utilized. We stress that for lanthanides lighter than  $\text{Tb}^{3+}$ , both the calculated dipolar and contact LIS show a remarkably good overall agreement with theoretical values given by Bleaney<sup>6</sup> and Golding,<sup>7</sup> respectively. These values have earlier been reported to be consistent with experimental data.<sup>19</sup> Possible errors in our analysis of epi-inositol arising from structure faults are probably small and play a minor role because no protons possess position close to the magic angle ( $54.7^\circ$ ) position in eqn. (1). The reason why the contact LIS for the heavier lanthanides showed bad agreement may probably be due to some alterations in the metal-substrate bond and/or small conformational changes. Reported works<sup>20</sup> indicate that although the bonding may be predominantly of pure electrostatic nature in all Ln complexes, the possibility of  $f\pi-p\pi$  bonding may be important in complexes of the heavier Ln ions. The ionic radius decreases slightly along the Ln series, being largest for  $\text{Ce}^{3+}$  (1.034 Å), and this may cause some structural changes for the heavier lanthanides. The hydration number of the Ln ions has also been reported to decrease from 9 for  $\text{La}^{3+}$  to 8 for  $\text{Yb}^{3+}$  somewhere in the middle of the Ln series.<sup>21</sup>

Except for methyl  $\alpha$ -D-gulopyranoside, the LIS

data have been corrected for the small downfield diamagnetic shift contributions caused by the charge of the cation as measured by adding  $\text{La}^{3+}$ . For methyl  $\alpha$ -D-gulopyranoside (1) the NMR spectral lines were so heavily overlapping that no  $\text{La}^{3+}$  induced shifts could be measured. This is also the reason why the shift values at proton H4 are absent in Table 2. Only small shifts comparable to diamagnetic shifts were observed at this proton.

We have used the dipolar shifts in Table 2 to determine the Ln binding site on 1. The consistent results for epi-inositol suggest that the dipolar shifts in this tridentate system can be accounted for by use of the McConnell-Robertson equation. This equation requires the complex to have an axial symmetry. Provided this is the case, the calculations strongly suggest that there exists only one site within a reasonable distance from the three oxygen functions O1, O2 and O3. It should be mentioned here that it is very important to use a sufficiently small angular step (less than  $2^\circ$ ) in the search procedure for the axis direction giving the minimum  $AF$ -value, otherwise apparent local minima may develop.

The site defined by the minimum in  $AF$  has Ln—O distances of 2.0, 2.2 and 2.3 Å to O1, O2 and O3, respectively. The shortest Ln—O distance in our "best fit" site, 2.0 Å is somewhat outside the range of Ln—O distances in crystals  $2.46 \pm 0.3$  Å.<sup>11-14</sup> The error in our calculated position depends mainly upon uncertainties in dissecting the LIS which in turn relies on relative theoretical values given by Bleaney<sup>6</sup> and Golding.<sup>7</sup> We have carried out Monte-Carlo calculations to study the effect of a randomly distributed error inside  $\pm 10\%$  in all the shifts, and compared calculated  $AF$  values for our "best fit" site and the "average" site from crystal data letting the computer in all cases find the minimum in  $AF$  by varying the orientation of the magnetic axis. Our "best fit" site yielded  $AF$  values from 0.8 to  $5.2 \times 10^{-3}$ . For the "average" site the  $AF$  values ranged from 1.0 to  $6.5 \times 10^{-3}$ . Therefore, if we assume an error of  $\pm 10\%$  in our dipolar LIS data, the heavy overlap between the two sets of  $AF$ -values shows that we are not able to discriminate between these sites which are *ca.* 0.5 Å apart.

However, even though substantial uncertainties in this type of structure elucidation still exist, important conclusions concerning the localization of ions in complexes may still be drawn, particularly when one has the choice between different sites as is often the case in many large or macromolecular substrates.

In a previous paper<sup>8</sup> we demonstrated that the complex formed between  $\text{Eu}^{3+}$  and methyl  $\alpha$ -D-gulopyranoside had a 1:1 stoichiometry. An attempt was made to explain the LIS as being due to a complex of rhombic symmetry. The site arrived at after applying a correction for the asymmetry gave the following distances between the Ln ion and the oxygen atoms O1, O2, and O3, respectively: 1.7, 1.85, and 3.0 Å. The distance to O3 was larger than expected for a complex where this oxygen plays a significant role. The too short distances to the other two oxygens, and the resulting unsymmetric position of the cation relative to the binding oxygens also seemed questionable.

The more reasonable position found in this paper indicates that the LIS for methyl  $\alpha$ -D-gulopyranoside is due to both contact and dipolar shift mechanisms, and that these contributions may be effectively separated provided measurements have been carried out with a set of Ln ions.

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