Nuclear Magnetic Resonance Studies of Beryllium Complexes in Aqueous Solution

(the late) Dennis F. Evans and Chih Y. Wong*
The Evans Laboratory, Department of Chemistry, Imperial College, London SW7 2AY, UK

Quantitative data for the equilibria $[Be(H_2O)_4]^{2+} + xH_yL \rightleftharpoons BeL_y$, where x and $y = 1$ or 2, HL = 3-hydroxy-2-methylpyridin-4-one, 3-hydroxy-1,2-dimethylpyridin-4-one or 1-ethyl-3-hydroxy-2-methylpyridin-4-one and H_2L = chromotropic acid (4,5-dihydroxynaphthalene-2,7-disulfonic acid), tiron (4,5-dihydroxybenzene-1,3-disulfonic acid), 5-sulfosalicylic acid, 5-nitrosalicylic acid or 3,5-dinitrosalicylic acid have been obtained by ^1H and ^9Be NMR spectroscopy. The shielding effects of the ^9Be nucleus were found to be in accord with the number of ligands and the chelate ring size.

Beryllium is the most toxic non-radioactive element in the Periodic Table.[1] It is also the second lightest metal after lithium and its unique properties are a great asset in todays' nuclear, aerospace and electronic industries. In September 1990 an accident occurred in a nuclear fuel processing plant in Ust-Kamenogorsk of the Soviet Union resulting in tonnes of beryllium being released into the atmosphere.[2] The lack of awareness of the dangers of beryllium poisoning was again highlighted. Coupled with the recent paper by Skilleter,[3] there is now a resurgence of research on the co-ordination chemistry of beryllium(II).[4]

The present treatment for beryllium poisoning involves the formation of non-toxic 'lakes' by chelation with organic substrates such as aurintricarboxylic acid {5-[(3-carboxy-4-hydroxyphenyl)(3-carboxy-4-oxocyclohexa-2,5-dien-1-ylidene)-methyl]-2-hydroxybenzoic acid}.[5] The inherent toxicity of this acid, however, means that there is at present no universally accepted antidote to beryllium poisoning. An alternative approach to the development of beryllium ion inactivation involves the 'chelating process' or 'metal encapsulation'[6] but there is little information in the literature on designer ligands for beryllium encapsulation. The solution chemistry of the hydrated beryllium(II) ion in both acidic and alkaline media has previously been reported.[7] Chelation using 5-sulfosalicylic acid[8] and tiron (4,5-dihydroxybenzene-1,3-disulfonic acid)[9] has also been studied. However, the majority of research has concentrated on *in vivo* and *in vitro* experiments to establish the most effective antidote based on commercially available reagents. The present work establishes quantitatively the reaction of various bidentate ligands with beryllium in aqueous solution. This, we hope, will improve our understanding of the requirements for designing specific chelators of Be^{II}. The reactions were monitored by ^1H and ^9Be NMR spectroscopy in aqueous solution. The results are presented in the form of stepwise equilibrium constants.

Experimental

Safety.—In view of the extreme toxicity of beryllium compounds, it is essential that experiments carried out involving beryllium are monitored with the utmost vigilance. The most common form of intoxication is by inhalation of beryllium salts, especially in the form of a fine powder or dust. In the event of beryllium poisoning, increased oxygen inhalation, judicious use of steroids, and absolute bed rest are necessary.[10] (There is no clear evidence that steroids have cured chronic beryllium poisoning.) Skin poisoning requires surgical removal and, in severe cases, amputation may be required.[10] Note that, although the toxicity of beryllium compounds is well documented,[6,11] no standard treatment is given.

All experiments were carried out in a well ventilated and 'beryllium-exclusive' fumehood. Where possible, the equipment used was of the disposable form. Spillages and especially ones of particulate nature were washed down well with water. Transportation of samples outside the fumehood were minimised and NMR measurements were carried out with extreme caution.

Materials.—3-Hydroxy-2-methylpyridin-4-one[12] (Hhmpyo), 3-hydroxy-1,2-dimethylpyridin-4-one[13] (Hhdmpyo) and 1-ethyl-3-hydroxy-2-methylpyridin-4-one[13] (Hehmpyo) were prepared by literature methods. The remaining ligands were pure commercial samples.

Physical Measurements.—Proton and ^9Be NMR spectra were recorded on a JEOL EX270 FT spectrometer at 270 and 38.0 MHz respectively. All measurements were made in aqueous solution using a coaxial inner tube containing D_2O to provide a lock signal. The digital resolution was 0.33 and 0.48 Hz per point for ^1H and ^9Be respectively. The acquired free induction decay (f.i.d.) was multiplied by an appropriate line-broadening window function before transformation. In all cases $\pi/4$ pulses were used. Hydrogen-1 measurements were made using pre-saturation of the water resonance before acquisition (to reduce the magnitude of the solvent signal). The recycle times (acquisition and pulse delay) for ^1H and ^9Be were 5.0 and 2.0 s respectively. Previous experiments on these systems indicated this repetition time gave acceptable integration of the proton spectra. The integral accuracy for ^9Be was considered to be acceptable as all the lines were broad ($\nu_{\frac{1}{2}}$ 5–24 Hz). Integration was carried out using the standard JEOL software: accuracy is in the range 5–10%. Chemical shifts are quoted relative to internal 3-(trimethylsilyl)propane-1-sulfonic acid sodium salt for the ^1H nucleus and $[Be(H_2O)_4]^{2+}$ for the ^9Be nucleus. pH Measurements were made with a micro combination electrode with a PHA 230 pH meter (Whatman Labsales).

Equilibrium Studies.—A stock solution of Be^{2+} (0.02 mol dm^{-3}) was prepared by dissolution of $BeSO_4 \cdot 4H_2O$ (88.6 mg) in distilled water (25.00 cm^3). Stock solutions of each ligand (0.06 mol dm^{-3}) were also prepared. In some cases, the less-soluble ligands were completely dissolved by the addition of a minimum volume of saturated NaOH.

Samples were prepared by transferring equal volumes (350 μl) of the Be^{2+} stock solution and the ligand solution to a small sample tube. Thus, when mixed, the solution contains 0.01 mol dm^{-3} Be^{2+} ions and 0.03 mol dm^{-3} of ligand. The sample was

Table 1 Ionisation constants and abbreviations of ligands studied

Ligand	Abbreviation	pK_{1L}[a]	pK_{2L}[b]	Ref.
Chromotropic acid[c]	H_4dndsa	15.6	5.36	14
3,5-Dinitrosalicylic acid	H_2dnsa	7.40	2.25	15
5-Nitrosalicylic acid	H_2nsa	10.34	2.12	16
5-Sulfosalicylic acid	H_3ssa	11.74	2.49	8
3-Hydroxy-2-methylpyridin-4-one	Hhmpyo	9.80	3.65	17
1-Ethyl-3-hydroxy-2-methylpyridin-4-one	Hehmpyo	9.81	3.64	17
3-Hydroxy-1,2-dimethylpyridin-4-one	Hhdmpyo	9.86	3.70	17
Tiron[d]	H_4dbdsa	12.48	7.57	14

[a] $K_{1L} = HL^- \rightleftharpoons L^{2-} + H^+$ except for Hhmpyo, Hhdmpyo and Hehmpyo where $K_{1L} = HL \rightleftharpoons L^- + H^+$. [b] $K_{2L} = H_2L \rightleftharpoons HL^- + H^+$ except for Hhmpyo, Hhdmpyo and Hehmpyo where $K_{2L} = H_2L^+ \rightleftharpoons HL + H^+$. [c] 4,5-Dihydroxynaphthalene-2,7-disulfonic acid. [d] 4,5-Dihydroxybenzene-1,3-disulfonic acid.

adjusted to the required pH using minimum volumes of either concentrated HCl or saturated NaOH. Equilibrium is established quickly: spectra may be acquired as soon as a stable pH is obtained and the sample transferred to the NMR tube. Although no particular attempts were made to maintain a constant temperature, spectra were recorded at temperatures in the range 18–22 °C.

Results and Discussion

Theory.—The abbreviations and ionisation/protonation constants for the ligands studied are given in Table 1. In the derivations below the assumption is made that 'BeL' type complexes exist in solution. The actual formulation may be hydrous but this does not affect the equilibrium data and for simplicity the 1:1 ligand:metal species is referred to as 'BeL' throughout this paper. There are two sets of equilibria corresponding to the two types of ligands used: monobasic (HL) and dibasic (H_2L). The exact charges of the ionised ligands are not necessarily mono- or di-negative, *e.g.* tiron is actually a 4− anion due to the two additional SO_3^- ionisations. As these do not play a role in the chelation, the ligands have been grouped according to the ionisations required for complexation, L^- or L^{2-}.

For the dibasic ligands the formation constants are given by equations (1) and (2), and similarly for the monobasic ligands the formation constants are given by equations (3) and (4). If we

$$Be^{2+} + L^{2-} \overset{K_1}{\rightleftharpoons} [BeL] \quad (1)$$

$$[BeL] + L^{2-} \overset{K_2}{\rightleftharpoons} [BeL_2]^{2-} \quad (2)$$

$$Be^{2+} + L^- \overset{K_1}{\rightleftharpoons} [BeL]^+ \quad (3)$$

$$[BeL]^+ + L^- \overset{K_2}{\rightleftharpoons} [BeL_2] \quad (4)$$

consider equations (1)–(4) and formulate K_1 and K_2 in terms of mole fractions, *i.e.* let c_M, c_L, x and y be respectively the total concentration of Be^{2+} (0.01 mol dm^{-3}), the total concentration of ligand (0.03 mol dm^{-3}), the total concentration of 1:1 ligand:metal complex and 2:1 ligand:metal complex, we obtain equations (5)–(8), where A^{-1} represents the fraction of

$$[Be^{2+}] = c_M - x - y \quad (5)$$

$$[L^{n-}] = (c_L - x - 2y)/A \quad (6)$$

$$[BeL^{(2-2n)-}] = x \quad (7)$$

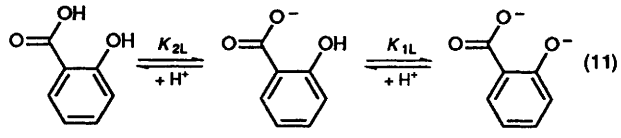
$$[BeL_2^{(2n-2)}] = y \quad (8)$$

ligand in the form of L^{2-} or L^- at the particular pH. Substituting for K_1 and K_2 gives equations (9) and (10).

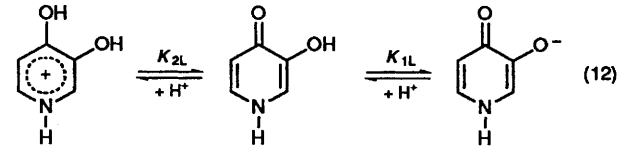
$$K_1 = \frac{xA}{(c_M - x - y)(c_L - x - 2y)} \quad (9)$$

$$K_2 = \frac{yA}{(c_L - x - 2y)x} \quad (10)$$

In order to calculate A the relevant ionisation/protonation processes are considered. For example, the ionisations of a salicylate moiety (H_2L) and a 3-hydroxypyridin-4-one moiety (HL) are represented by equations (11) and (12) respectively.



(11)



(12)

The concentration of L^{2-} or L^- can be derived by an analogous mole fraction method, thus for both H_2L and HL ligands we obtain equations (13) and (14) where $r = [HL^-]$ or [HL],

$$K_{2L} = \frac{r[H^+]}{U - r - s} \quad (13)$$

$$K_{1L} = s[H^+]/r \quad (14)$$

$s = [L^{2-}]$ or $[L^-]$ and U = total *uncomplexed ligand* concentration = $c_L - x - 2y$. Combining equations (6), (13) and (14) gives (15).

$$A = 1 + \frac{[H^+]}{K_{1L}} + \frac{[H^+]^2}{K_{1L}K_{2L}} \quad (15)$$

Equilibrium Studies.—The rate of exchange between free ligand, 1:1 and 2:1 species is slow on the NMR time-scale and if the separation in chemical shift is sufficient then distinct resonances due to the individual species may be identified. Beryllium complexes are tetrahedral and therefore no geometrical isomerism is expected. In all cases, the $[Be(H_2O)_4]^{2+}$ ion, 1:1 and 2:1 species can be unambiguously identified in the ^9Be NMR spectra. This is clearly illustrated by a stack plot of ^9Be NMR spectra over a range of pH for the Be(hdmpyo)$_x$

Table 2 Results of NMR integral analysis of the $Be(ssa)_x$ system with pH; 1:1 and 2:1 represent $[Be(ssa)]^-$ and $[Be(ssa)_2]^{4-}$ complexes respectively. Equilibrium constants calculated using equations (9) and (10)

	^1H NMR results				^9Be NMR results			
pH	1:1 mmol dm^{-3}	2:1 mmol dm^{-3}	$\log_{10} K_1$	$\log_{10} K_2$	1:1 mmol dm^{-3}	2:1 mmol dm^{-3}	$\log_{10} K_1$	$\log_{10} K_2$
1.72	0.0	0.0			0.0	0.0		
2.69	6.8	0.0	11.22		6.9	0.0	11.19	
3.21	9.3	0.0	11.41		8.8	0.0	11.15	
3.61	9.8	0.0	11.55		9.6	0.0	11.23	
4.22	8.2	1.8		8.61	9.1	0.9		8.24
4.87	5.5	4.5		8.59	6.4	3.6		8.41
5.50	2.7	7.3		8.57	3.5	6.5		8.38
5.99	1.2	8.8		8.57	Overlap			
7.32	0.0	10			0.0	10		
10.91	0.0	10			0.0	10		
			Average $\log_{10} K_1$ = 11.20				Average $\log_{10} K_1$ = 11.19	
			Average $\log_{10} K_2$ = 8.59				Average $\log_{10} K_2$ = 8.34	
			$\log_{10} \beta_2$ = 19.79				$\log_{10} \beta_2$ = 19.53	

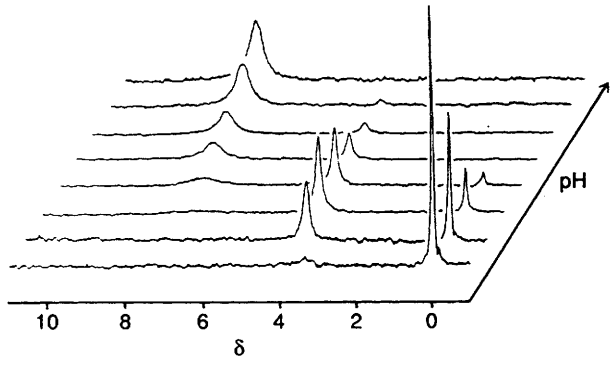


Fig. 1 Stack plot of ^9Be NMR spectra for the $Be(hdmpyo)_x$ system in aqueous solution over a pH range between 2 and 8. The $[Be(H_2O)_4]^{2+}$ ion is referenced at δ 0, $[Be(hdmpyo)]^+$ occurs at δ *ca.* 4 and $[Be(hdmpyo)_2]$ occurs at δ *ca.* 7.5 (38.0 MHz, water with D_2O lock)

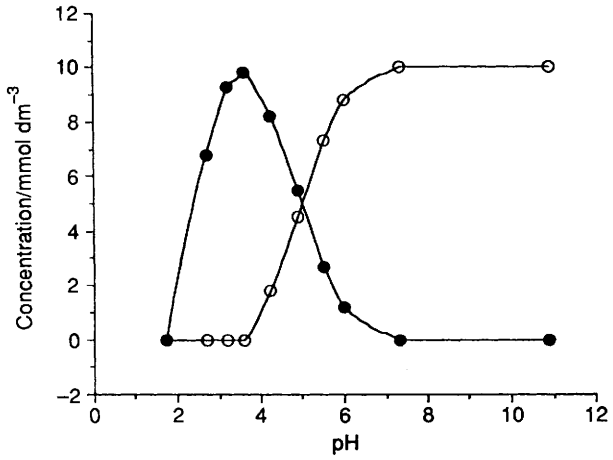


Fig. 2 A plot of concentration *versus* pH for the stepwise formation of 1:1 (●) and 2:1 (○) ligand:metal complexes in the $Be(ssa)_x$ system

(x = 1 or 2) system (Fig. 1). Despite the higher sensitivity of the ^1H nucleus, there was more overlap observed in the corresponding ^1H NMR spectra. In some systems, as is observed for the $Be(ssa)_x$ equilibria, results for both nuclei gave satisfactory analysis. Table 2 shows the concentrations of the 1:1 and 2:1 species at various pH values calculated by intensity measurements on the NMR spectra for the $Be(ssa)_x$ system. These concentrations are then used to calculate the stepwise formation constants from equations (9) and (10). The results can be represented graphically by a plot of concentration *versus* pH (Fig. 2). The plot shows typical stepwise equilibria behaviour with the BeL_2 species being stable to high pH. The other systems all gave similar results and the averaged formation constants are presented in Table 3. A close correlation of K_1 and K_2 with literature values is observed for ligands previously reported. The results indicate that the best chelator for beryllium in the systems studied is chromotropic acid. This ligand has one of the 'hardest' oxygens of all donors described and it binds to form a six-membered chelate ring. The latter feature may be significant since tiron which also has two 'hard' phenolic oxygens but a five-membered chelate ring gives much lower formation constants.

The electronic differences between 5-nitro- and 5-sulfosalicylic acid are small and this is reflected in the formation constants. It also illustrates the similarity of the complexes despite the difference in charge caused by the sulfonate group. The increased electronegativity of 3,5-dinitrosalicylic acid over 5-nitrosalicylic acid decreases the ionisation constant of the ligand. As a consequence, the formation constants of the $Be(dnsa)_x$ system are also lowered compared with the $Be(nsa)_x$ system. This reflects the similarity between the proton and the Be^{2+} cation. That is, the affinity of the Be^{2+} cation for a chelator parallels its acidity. Thus, there is a balance between having a chelator with strong O–H bonding for strong O–Be bonding and yet weak enough O–H bonding for easy displacement. For increased complexation the chelate effect must therefore dominate.

The complexes derived from hmpyo, hdmpyo and ehmpyo gave rather low formation constants. These ligands are now well investigated for other 'hard' metal ions. The effect of changing the alkyl group on the nitrogen appears to give negligible difference chemically. The only apparent effect appears to be the effect on solubility. The ligands formed the first examples of relatively water-soluble neutral 2:1 complexes of Be^{II}.

^9Be *NMR Spectroscopy*.—The observed ^9Be NMR chemical shifts and linewidths are given in Table 4. These values are essentially the same throughout the pH range studied. In all cases the chemical shift, δ, is recorded relative to the $[Be(H_2O)_4]^{2+}$ cation. Two distinct ranges are observed for both 1:1 and 2:1 complexes. The δ range depends on the chelate ring size, such that five-membered chelate rings give 1:1 complexes at *ca.* 4.0 and 2:1 complexes at *ca.* 7.5. Six-membered chelate rings give 1:1 complexes at *ca.* δ 1.7 and 2:1 complexes at *ca.* δ 3.2. The relationship between the size of the chelate ring and δ has been previously reported for tris(bidentate ligand) octahedral complexes derived from ions such as Al^{III} (ref. 18) and Si^{IV}.[19,20] In all cases, six-membered chelate rings were shifted

Table 3 Formation constants of beryllium complexes in aqueous solution determined by NMR spectroscopy (0.01 mol dm^{-3} Be^{2+}, 0.03 mol dm^{-3} ligand, 18–22 °C)

Ligand	$\log_{10} K_1$	$\log_{10} K_2$	$\log_{10} \beta_2$
dndsa[a]	16.2	12.0	28.2
dbdsa[b]	12.2	9.3	21.5
ssa[c]	11.2	8.5	19.7
nsa	10.1	8.0	18.1
hdmpyo	8.7	7.4	16.1
ehmpyo	8.5	7.3	15.8
hmpyo	8.4	7.2	15.6
dnsa	7.8	5.5	13.3

[a] Literature values are 16.34 and 11.85 respectively.[9] [b] Literature values are 12.88 and 9.37 respectively.[9] [c] Literature values are 11.50 and 8.84 respectively.[8]

Table 4 ^{9}Be NMR chemical shifts and linewidths of 1:1 and 2:1 ligand:metal complexes; chemical shifts are relative to the $[Be(H_2O)_4]^{2+}$ cation

Complex system	Chelate ring size	1:1		2:1	
		δ	$\nu_{\frac{1}{2}}$/Hz	δ	$\nu_{\frac{1}{2}}$/Hz
Be(dndsa)	6	1.63	11.4	2.53	11.8
Be(dnsa)	6	1.68	4.2	3.19	12.0
Be(nsa)	6	1.80	5.3	3.18	12.7
Be(ssa)	6	1.85	6.6	3.25	15.3
Be(hmpyo)	5	3.92	6.6	7.51	15.7
Be(ehmpyo)	5	3.94	6.8	7.46	23.1
Be(hdmpyo)	5	3.96	5.6	7.47	18.1
Be(dbdsa)	5	4.09	7.1	7.41	24.2

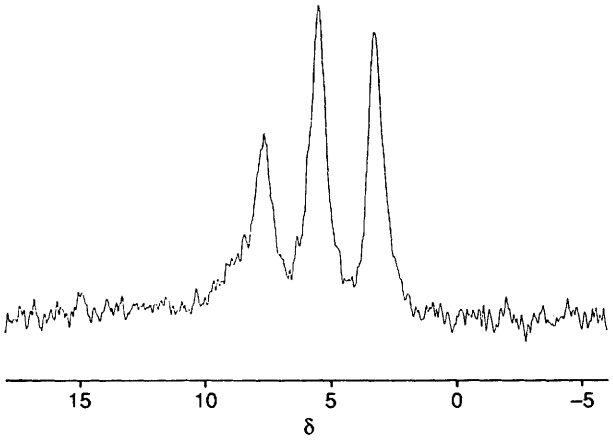


Fig. 3 ^{9}Be NMR spectrum of a mixture of ssa and dbdsa (0.03 mol dm^{-3}) and Be^{2+} (0.01 mol dm^{-3}) at pH 7.4, showing the presence of $[Be(ssa)_2]^{4-}$ at δ *ca.* 3.3, $[Be(dbdsa)_2]^{6-}$ at δ *ca.* 7.6 and $[Be(ssa)(dbdsa)]^{5-}$ at δ *ca.* 5.5 (38.0 MHz, water with D_2O lock)

to higher field compared to the five-membered chelate ring analogues. This correlates well with the data observed in the work described.

In most cases the 2:1 complexes have an appreciably larger linewidth, $\nu_{\frac{1}{2}}$, than the corresponding 1:1 complexes: this may be a consequence of relaxation times or exchange processes. The notable exception to the observation is the $Be(dndsa)_x$ system where the 2:1 complex has a much lower δ ($\Delta\delta \approx 0.7$) and the 1:1 complex has a much larger $\nu_{\frac{1}{2}}$ (≈two-fold). This anomaly has been attributed to the high rigidity enforced by the naphthalene ring in the dndsa ligand.

The higher δ and $\nu_{\frac{1}{2}}$ observed for 2:1 over 1:1 complexes indicate progressive deshielding and distortion caused by chelation to the beryllium nucleus. The difference caused by five- and six-membered chelate rings can be further illustrated by the observation of a mixed species containing one five- and one six-membered chelating ligand. Thus, at pH 7.4 a mixture of ssa and dbdsa in $[Be(H_2O)_4]^{2+}$ solution shows the presence of $[Be(ssa)_2]^{4-}$ at δ 3.3, $[Be(dbdsa)_2]^{6-}$ at δ 7.6 and $[Be(ssa)(dbdsa)]^{5-}$ at the intermediate δ 5.5 (Fig. 3). This technique was unable to resolve mixed complexes containing two different ligands with the same chelate ring size. At lower pH values the presence of 1:1 complexes causes complicated overlapping of resonances.

Acknowledgements

C. Y. W. would like to thank Dr. J. Derek Woollins and Mr. Richard N. Sheppard for their advice, and the SERC for a grant.

References

1 *The Guinness Book of Records 1992*, ed. D. McFarlan, Guinness Publishing, London, 1991, p. 107.
2 V. Rich, *New Sci.*, 1990, **128** (1737), 19; 1990, **128** (1743), 15.
3 D. N. Skilleter, *Chem. Br.*, 1990, **26**, 26.
4 H. Schmidbaur, O. Kumberger and J. Riede, *Inorg. Chem.*, 1991, **30**, 3101; M. Inamo, K. Ishihara, S. Funahashi, Y. Ducommun, A. E. Merbach and M. Tanaka, *Inorg. Chem.*, 1991, **30**, 1580.
5 S. Seidel, in *Gmelin Handbook of Inorganic Chemistry*, 8th edn., Springer, Berlin, 1986, Supplement vol. A1, p. 300 and refs. therein.
6 L. B. Tepper, H. L. Hardy and R. I. Chamberlin, *Toxicity of Beryllium Compounds*, Elsevier, Amsterdam, 1961, p. 139.
7 D. A. Everest, *The Chemistry of Beryllium*, Elsevier, Amsterdam, 1964, p. 7.
8 C. V. Banks and R. S. Singh, *J. Am. Chem. Soc.*, 1959, **81**, 6159; *J. Inorg. Nucl. Chem.*, 1960, **15**, 125.
9 M. Bartusek and J. Zelinka, *Collect. Czech. Chem. Commun.*, 1967, **32**, 992.
10 *Encyclopaedia of Occupational Health and Safety*, 3rd edn., ed. L. Parmeggiani, International Labour Office, Geneva, 1983, vol. 1; P. Cooper, *Poisoning by Drugs and Chemicals, Plants and Animals*, 3rd edn., Alchemist Publications, London, 1974.
11 *Beryllium, Biomedical and Environmental Aspects*, eds. M. D. Rossman, O. P. Preuss and M. B. Powers, Williams and Wilkins, Baltimore, 1991.
12 R. L. N. Harris, *Aust. J. Chem.*, 1976, **29**, 1329.
13 G. J. Kontoghiorghes and L. Sheppard, *Inorg. Chim. Acta*, 1987, **136**, L11.
14 G. A. L'Heureux and A. E. Martell, *J. Inorg. Nucl. Chem.*, 1966, **28**, 481.
15 M. Bartusek, *Collect. Czech. Chem. Commun.*, 1967, **32**, 116.
16 Z. L. Ernst and J. Menashi, *Trans. Faraday Soc.*, 1963, **59**, 2838.
17 D. J. Clevette, D. M. Lyster, W. O. Nelson, T. Rihela, G. A. Webb and C. Orvig, *Inorg. Chem.*, 1990, **29**, 667.
18 J. W. Akitt, *Prog. Nucl. Magn. Reson. Spectrosc.*, 1989, **21**, 123.
19 J. A. Cella, J. D. Cargioli and E. A. Williams, *J. Organomet. Chem.*, 1980, **186**, 13.
20 D. F. Evans and C. Y. Wong, *Polyhedron*, 1991, **10**, 1131.

Received 20*th January* 1992; *Paper* 2/00312K