Synthesis and Structures of Five $[Co(Mecyclen)(S\text{-}AlaO)]^{2+}$ Isomers: Use of nOe and COSY ^1H NMR Spectroscopy for Structural Assignment in Solution

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The cyclic quadridentate 1-methyl-1,4,7,10-tetraazacyclododecane (Mecyclen) has been prepared in three simple steps from *p*-tosylaziridine, methylammonium acetate, and tris(*p*-tosyl)diethanolamine. Coordination to Co(III) gives $[Co(Mecyclen)Cl_2]Cl$ in high yield, which on treatment with *S*-alanine (*S*-AlaOH), or its methyl ester (50 °C, pH 7.5−8.0), gives a mixture of six $[Co(Mecyclen)(S\text{-}AlaO)]^{2+}$ isomers. Five of these have been isolated by a combination of cation ion-exchange chromatography and selective crystallization. Each has been structurally assigned using a combination of nOe and COSY ^1H NMR spectroscopies (d_6-DMSO solvent) as, in order of decreasing yield from *S*-AlaOMe, the following: *anti*(Me), *syn*(N), *anti*(O) (isomer **4**); *anti*(Me), *anti*(N), *syn*(O) (isomer **5**); *syn*(Me), *syn*(N), *anti*(O), (isomer **1**); *syn*(Me), *anti*(N), *syn*(O) (isomer **2**); *syn*(Me), *syn*(N), *syn*(O) (isomer **3**). The structures of **1**, **2**, and **4** have been confirmed by single-crystal X-ray analysis: $[Co(Mecyclen)(S/R\text{-}AlaO)](ClO_4)_2{\cdot}H_2O$ (50:50 mixture of isomer **1** containing *S*-AlaO and isomer **4** containing *R*-AlaO), monoclinic, $P2_1/n$, $a = 14.131(3)$ Å, $b = 8.777(2)$ Å, $c = 17.745(4)$ Å, $Z = 4$, $R = 0.0734$; $[Co(Mecyclen)(S\text{-}AlaO)]ZnCl_4{\cdot}2H_2O$ (isomer **2**), orthorhombic $P2_12_12_1$, $a = 8.842(2)$ Å, $b = 14.794(7)$ Å, $c = 17.157(8)$ Å, $Z = 4$, $R = 0.0415$; $[Co(Mecyclen)(S\text{-}AlaO)](ClO_4)_2{\cdot}H_2O$ (isomer **4**); orthorhombic $P2_12_12_1$, $a = 8.757(2)$ Å, $b = 9.684(5)$ Å, $c = 25.291(6)$ Å, $Z = 4$, $R = 0.0380$. Structural assignment by the ^1H NMR method depends critically on the presence of separate N−*H* resonances for the different sites; this can usually be achieved by addition of DCl to a d_6-DMSO solution of the complex.

Introduction

The main purpose of this paper is to show how nOe and COSY ^1H NMR spectroscopy can be used to determine the essential structural features of multidentate amine chelates of Co(III). The study arose out of attempts to prepare such cobalt(III) chelates for use in the stereospecific synthesis of small peptides.[1] Because the nOe is transmitted through space,[2] it is ideally suited to the determination of structure, especially for molecules of fixed or relatively fixed geometry.[3] Although the method does not give the detail of an X-ray study, it does avoid the necessity of obtaining crystals, and the results apply to the solution phase which is generally more appropriate to other investigations.

In recent years 1- and 2-D nOe spectroscopy has been increasingly used for the large-scale structure determination of complex natural products and for the conformational analysis of peptides, proteins, polynucleotides, and oligiosaccharides.[3] However it has not been used extensively for small molecules and, in particular, for metal-coordination complexes. This is surprising since the distances between NMR sensitive atoms (particularly ^1H) attached to different ligands, and especially interchelate $^1H/^1H$ distances in octahedral complexes, are often of the order of 2−3 Å which is ideal for the nOe experiment. An early report on $[Co(EDTA)]^-$ [4] has not been extended in a structural sense, with nOe studies on other Co(III) complexes being restricted to determining the preferred conformations of O-bound formato,[5] formamido,[5] dimethylformamido,[6] acetamido,[6] *N*-methyl- and *N*,*N*-dimethyl acetamido,[6] and a variety of alkyl-substituted imidazole[7] ligands bound to pentaamminecobalt(III). A recent application to the assignment of the absolute configuration of two Pd(II) diastereomers[8] shows how the method can be extended to give information on groups ca. 5 Å apart in a square planar complex.

It is well-known that coordinated secondary or tertiary N centers lead to asymmetric or diastereotopic configurations about an octahedral metal ion.[9] When several such centers are involved, as they are in multidentate amine complexes, many isomers are possible and indeed many are often found in the preparative mixture. This article considers the $[Co(Mecyclen)(S\text{-}AlaO)]^{2+}$ ion containing the cyclic quadridentate 1-methyl-1,4,7,10-tetraazacyclododecane and bidentate (*S*)-alanine. Eight isomers are possible based on the *syn*, *anti* arrangement of the two Me groups and the *syn*, *anti* orientations of the "in-plane" secondary N*H* centers of the macrocycle,[10] Figure 1 .

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(10) Some authors[11,12,16] have used the terms *endo* or *exo* to describe the spatial relationship between an atom or group of one ligand (particularly N−*H* protons) and that belonging to a different ligand in the same molecule. Other authors have used *syn* (toward) and *anti* (away from) in this same sense[13−15] while reserving the terms *endo* (inward) and *exo* (outward) to describe spatial relationships between atoms or groups belonging to the same ligand. In this paper we follow the latter usage (see Figure 2).
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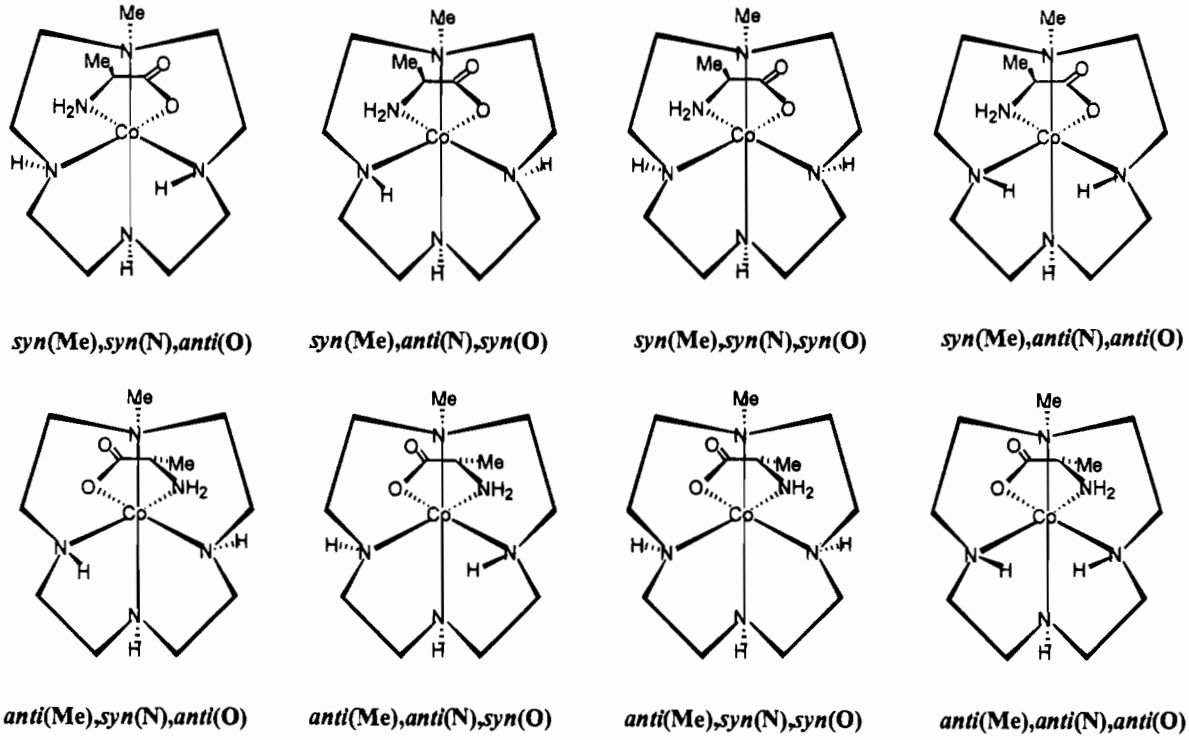

Figure 1. The eight possible configurational isomers of [Co(Mecyclen)(*S*-AlaO)]$^{2+}$: top, *syn*(Me) isomers; bottom, *anti*(Me) isomers.

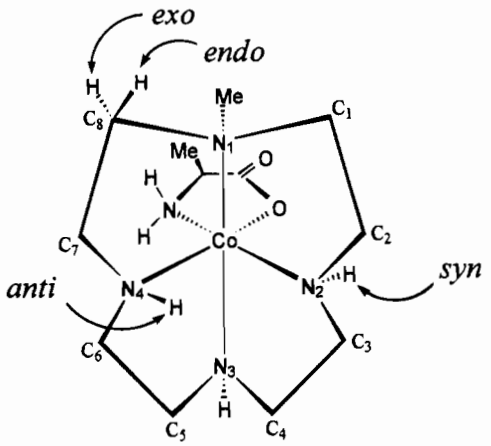

Figure 2. Representation of the *syn*(Me),*anti*(N),*syn*(O)-[Co(Mecyclen)(*S*-AlaO)]$^{2+}$ ion showing the planar *anti*(N), *syn*(O) N−*H* protons and the *exo, endo* C−*H* hydrogens of Mecyclen. The atom-numbering system for C and N is the same for all isomers (cf. Figure 1).

Figure 2 gives the ligand atom numbering system employed in this paper as well as indicating *syn*, *anti*, *exo,* and *endo* protons. Six isomers have been detected in this study, but only five have been isolated. The sixth represents <1% of the reaction mixture, and the other two do not appear to exist as ground state molecules.[17] The visible spectra of all the isolated isomers are very similar as are many of their other properties, but their ^{1}H NMR spectra are different, especially in the N−*H* region. This provides the means of determining their structure by the nOe method. In this paper we describe in some detail how this was achieved for each isolated isomer, but at the conclusion of the study we felt that with a higher field NMR spectrometer and appropriate software[18] this might be achieved in admixture, i.e. before separation of the isomers. The method therefore has considerable promise.

Experimental Section

Reagents and Materials. Solvents were dried over molecular sieves and distilled prior to use. *p*-Tolylsulfonyl chloride, pyridine, triethylamine, and dichloromethane were purified using established procedures.[19] Dean−Stark dried toluene was stored over sodium. Ethanolamine (EA) and diethanolamine (DEA) were purchased from BDH (LR grade), distilled at atmospheric pressure, and stored over molecular sieves. Methylamine was used as purchased from BDH (33% solution in ethanol). Li_2ZnCl_4 was prepared as a 2 M solution.[12a] *N,O,O'*-Tris(*p*-tolylsulfonyl)diethanolamine (Ts_3DEA) was prepared on a 0.5 mol scale using the method of Osvath.[20] *N,O*-Bis(*p*-tolylsulfonyl)ethanolamine (Ts_2EA) was prepared on a 0.8 mol scale by the method of Hope and Horncastle[21] (care was taken to remove residual pyridine by washing with water and/or by crystallizing the product from ethanol[22]). *N*-(*p*-Tolylsulfonyl)-aziridine (TsA) was prepared from EA as described by Lehn *et al.*[23] (care was taken to avoid overheating when removing the solvent, 35 °C maximum). Methylammonium acetate (MeAc) was prepared by neutralizing an ethanolic solution of $MeNH_2$

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with glacial HOAc (below 40 °C), removing the solvent, and crystallizing the product from a concentrated solution in acetone by cooling (−40 °C). The hygroscopic solid was collected and dried under an atmosphere of N_2. Decomposition made it difficult to store MeAc for long periods of time.

Ligand Synthesis. *N*-Methyl-*N'*,*N''*-bis(*p*-tolylsulfonyl)-ethylamine (Ts_2Medien). This material was prepared on a 0.11 mol scale in the absence of light by reacting a stirred solution of TsA (43.3 g) dissolved in dry toluene (100 cm^3) and acetonitrile (50 cm^3) with MeAc (10 g) at 75 °C for 13 h. Dichloromethane (300 cm^3) was then added to the cooled solution which was washed with water (3 × 300 cm^3), dried with Na_2SO_4, and reduced in volume to a viscous oil. The oil was then taken up in dichloromethane (20 cm^3), layered with ether (10 cm^3), and cooled in ice whereupon the product crystallized; further product was obtained by adding additional ether and cooling overnight; mp 113 °C (lit. 113−114 °C).[24] Anal. Calcd for $C_{19}H_{27}N_3O_4S_2$: C, 53.62; H, 5.92; N, 9.88; S, 15.06. Found: C, 53.92; H, 6.18; N, 9.60; S, 15.00. ^{1}H NMR ($CDCl_3$, 25 °C): δ = 7.77 (d, 4H); 7.32 (d, 4H); 2.96 (t, 4H); 2.43 (s, 6H); 2.38 (t, 4H); 1.99 (s, 3H). ^{13}C NMR ($CDCl_3$, 25 °C): δ = 143.3, 136.9, 129.8, 127.3, 56.2, 40.8, 40.4, 21.6. The disodium salt (Na_2Ts_2Medien) was prepared by adding sodium ethoxide in dry ethanol (0.2 mol, 100 cm^3) to refluxing Ts_2Medien (43 g, 0.1 mol in dry ethanol, 200 cm^3) and continuing reflux until all the solid had dissolved. On cooling the solvent was removed (oil pump, azeotrope with benzene) and the solid stored under vacuum.

1-Methyl-4,7,10-tris(*p*-tolylsufonyl)-1,4,7,10-tetraazacyclododecane (Ts_3-Mecyclen). To a stirred solution of Na_2Ts_2-Medien (25.8 g, 55 mmol) in dry DMF (150 cm^3) at 105 °C and under a N_2 atmosphere was added over 120 min Ts_3DEA (31.2 g, 55 mmol) in dry DMF (300 cm^3). Heating was continued for a further 120 min before the volume was reduced (oil pump) to ca. 180 cm^3. Ethanol was then added to the cooled solution and the resulting white solid collected and washed with EtOH (2 × 200 cm^3) and H_2O (2 × 200 cm^3) before drying overnight (50−80 °C). This material was recrystallized by dissolving it in the minimum volume of chloroform (60 °C) and adding 2-propanol (50 °C, 3 volumes) followed by cooling in ice. The recovered product was dried at 50−80 °C: yield 75−80%; mp 204−205 °C. Anal. Calcd for $C_{30}H_{40}N_4O_6S_3$: C, 55.53; H, 6.20; N, 8.64; S, 14.82. Found: C, 55.39; H, 6.27; N, 8.66; S, 14.81. ^{1}H NMR ($CDCl_3$, 25 °C): δ = 7.85 (d, 2H); 7.65 (d, 4H); 7.34 (t, 6H); 3.52 (t, 4H); 3.28 (t, 4H); 3.08 (m, 4H); 2.63 (m, 4H); 2.47 (s, 3H); 2.43 (s, 6H); 2.18 (s, 3H). ^{13}C NMR ($CDCl_3$): δ = 143.6, 143.4, 136.6, 135.0, 129.8, 127.8, 127.5, 59.4, 51.9, 50.2, 48.7, 42.6, 21.6.

1-Methyl-1,4,7,10-tetraazacyclododecane·3HCl (Mecyclen·3HCl). The following procedure is a modification of that described by Richmann and Atkins.[25] A stirred solution of Ts_3-Mecyclen (30 g) in 98% H_2SO_4 (300 cm^3) was heated at 110 °C under a N_2 atmosphere for 45 h. To the cooled solution (0−5 °C) was slowly added ether (500 cm^3) whence an off-white precipitate formed. This was collected, washed with ether (3 × 300 cm^3), and dried *in vacuo* over P_2O_5. The solid was then dissolved in H_2O (400 cm^3), the pH adjusted to ca. 7 by adding solid $BaCO_3$, and the resulting white precipitate of $BaSO_4$ removed. The filtrate was passed through a column of Amberlite IRA-400 anion-exchange resin (Cl^- form) using H_2O as eluant and the total eluate (Ag^+ test for Cl^-) reduced to dryness. The resulting solid was recrystallized from the minimum volume of dilute HCl (80 °C) by adding EtOH to the cooled solution, and the product was dried under vacuum. Anal. Calcd for $C_9H_{22}N_4$·3HCl: C, 26.5; H, 8.01; N, 18.55; Cl, 46.94. Found: C, 26.29; H, 8.23; N, 18.52; Cl, 46.67. ^{1}H NMR (D_2O, 25 °C): δ = 3.18 (m, 16H); 2.62 (s, 3H).

Preparation of Co(III) Complexes. [Co(Mecyclen)Cl_2]Cl was prepared as follows. A vigorous stream of air was passed through a solution of Mecyclen·3HCl (10 g, 34 mmol) in 1.0 M NaOH (68 cm^3) and water (250 cm^3) to which was added dropwise over 10 min a solution of $CoCl_2$·$6H_2O$ (9.0 g, 38 mmol) in water (250 cm^3). Aerial oxidation was continued for a further 90 min during which time the color changed from brown to deep red. HCl (30 cm^3, 12 M) was then added and the purple solution diluted with water (200 cm^3), sorbed on Dowex ion-exchange resin (50 W × 2, H^+ form; 30 × 6 cm column), and eluted with HCl (1−2 M). The three bands which separated (1+, 2+, 3+ species) were combined and reduced to dryness. The resulting purple residue was stirred for 4 h with acetone (600 cm^3), collected, and dried in air; yield ca. 95%. Anal. Calcd for $CoC_9H_{22}N_4Cl_3$: C, 30.74; H, 6.31; N, 15.94. Found: C, 30.79; H, 6.35; N, 15.79.

[Co(Mecyclen)(*S*-AlaO)]Cl_2, Isomer Mixture. The isomer distribution depended on whether alanine methyl ester hydrochloride (*S*-AlaOMe·HCl) or alanine (*S*-Ala) was used in the preparation. [Co(Mecyclen)Cl_2]Cl (5.0 g; 14.3 mmol) and *S*-AlaOMe·HCl (2.5 g, 17.8 mmol), or *S*-Ala (1.6 g, 17.8 mmol), in water (150 cm^3) was adjusted to pH 7.5−8.0 (1 M NaOH) and warmed at 50 °C for 90 min with stirring and with periodic pH adjustment (this was unnecessary when *S*-Ala was used). The red solution was then acidified to pH 2 (dilute HCl), and the complexes were sorbed on to and then eluted from (1 M HCl) Dowex 50 W × 2 ion-exchange resin (H^+ form, 15 × 6 cm column). The combined red 2+ band was collected (some separation of isomers had begun; see below) and reduced to dryness, yield ca. 80−90%.

Isomer Separation and Characterization. Chromatography on Dowex 50W × 2 cation-exchange resin (25 × 6 cm column) of the isomer mixture resulted in the separation of two red bands; the first to elute contained isomers **2**, **3**, and **5**, and the second, isomers **1** and **4** (cf. Figure 3). Isomers **2**, **3**, and **5** showed differing N−*Me* singlets in the ^{1}H NMR spectrum (2.46, 2.41, 2.53 ppm, respectively) as did isomers **1** and **4** (2.39, 2.48 ppm, respectively); isomer purity was established by reference to these absorptions. It was not found possible to separate the isomers further by RP-HPLC (cf. the analogous [Co(cyclen)(*S*-AlaO)]$^{2+}$ isomers[26]). The separate isomers were isolated as crystalline solids as follows.

Isomer 4. The mixture of [Co(Mecyclen)(*S*-AlaO)]$^{2+}$ isomers (ca. 3 g), obtained from the reaction of AlaOMe with [Co(Mecyclen)Cl_2]Cl, was dissolved in the minimum volume of 0.1 M HBr, the solution was filtered, $NaClO_4$·H_2O was added, and the mixture was cooled in ice. The mauve microcrystals were collected and washed with MeOH. The filtrate was then desalted chromatographically (Dowex 50 W × 2 ion-exchange resin, 1−2 M HCl, dryness) and the procedure repeated. This resulted in the isolation of more (but lower purity) **4**. Recrystallization of the combined mauve crystals from 0.1 M HBr by addition of small amounts of $NaClO_4$·H_2O gave pure **4** (yield ca. 0.8 g). ^{1}H NMR (N−*Me*): 2.48 ppm. **Caution: care should be exercised in handling this material since perchlorate salts are potentially explosive (cf.: *J. Chem. Educ.* 1973, *50*, A335; *Chem. Eng. News* 1983 (Dec 5), 4).** Calcd for $CoC_{12}H_{28}N_5O_2$·$2ClO_4$: C, 27.10; H, 5.30; N, 13.16; Cl, 13.32.

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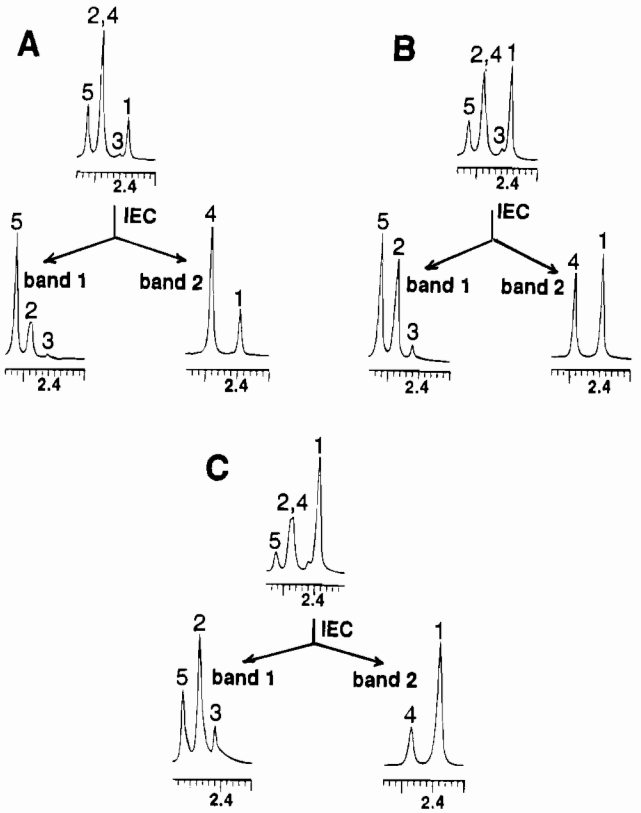


Figure 3. N−*Me* part of the ^{1}H NMR spectrum (0.1 M DCl solvent, 300 MHz) for the preparative mixture starting with (A) (*S*)-AlaOMe·HCl, and (B) (*S*)-AlaOH. (C) gives the isomer distribution following equilibration on activated charcoal at 70 °C. Also shown is the distribution following ion-exchange chromatography (IEC); band 1 (first to elute) contains isomers **2**, **3**, and **5**; band 2 contains isomers **1** and **4**. (Scaling is in 0.02 ppm units.)

Found: C, 26.77; H, 5.65; N, 13.17; Cl, 13.90. Visible−UV spectrum: ϵ_{515}, 298; ϵ_{354}, 223.

Isomer 1. The filtrate from the above isolation of isomer **4** was desalted chromatographically (see above) and the eluate taken to dryness. The residue was dissolved in the minimum volume of 0.1 M HCl and 2 M Li_2ZnCl_4 added dropwise to the filtered solution. Scratching and cooling resulted in the crystallization of a red solid which was collected, washed with MeOH, and dried in air. Recrystallization using this same procedure was repeated until the product was isomerically pure (N−*Me:* 2.39 ppm). Anal. Calcd for $CoC_{12}H_{28}N_5O_2{\cdot}ZnCl_4{\cdot}1.5\ H_2O$: C, 25.39; H, 5.51; N, 12.34; Cl, 24.98. Found: C, 25.47; H, 5.42; N, 12.16; Cl, 25.83. Visible−UV spectrum: ϵ_{509}, 265; ϵ_{353}, 213. Alternatively, isomer **1** was isolated in larger amounts from the initial preparative mixture as follows. [Co(Mecyclen)-(*S*-AlaO)]Cl_2 (1.0 g) in water (30 cm^3) was adjusted to pH 4−6 and equilibrated by stirring with Norite activated animal charcoal (0.4 g) at 70 °C for 45 min (longer times or higher temperatures resulted in N-demethylation of the macrocycle[27] and/or hydrolysis of AlaO from the complex). The solution was filtered (Celite) and the isomer mixture sorbed onto ion-exchange resin (40 × 2 cm column) as a fine red band. Elution with 1 M HCl resulted in the separation of two red bands, the second of which (major) contained isomers **1** and **4** (**1**:**4** ≃ 2:1). This second band was taken to dryness and 2 M Li_2ZnCl_4 added dropwise to the residue dissolved in the minimum volume of 0.1 M HCl (filtered). The impure sample of **1** (containing some **4**) was purified by recrystallization from 0.1 M HCl by addition of Li_2ZnCl_4.[12a] The product was then washed with MeOH and dried in air. Isomerically pure **1** as its $ZnCl_4^{2-}$ salt was used to seed all subsequent preparations, resulting in fewer steps.

Isomer 2. An aqueous solution of isomer **1** (1.0 g) in water (30 cm^3) was adjusted to pH 7 (0.01 M NaOH) and the equilibrated mixture sorbed onto ion-exchange resin (40 × 2 cm column) as a fine red band. Elution with 1 M HCl resulted in the separation of two red bands (ratio 2:7) the first of which contained pure isomer **2**. This eluate was reduced to dryness, the residue dissolved in a small volume of 0.1 M HCl, and 2 M Li_2ZnCl_4 added dropwise to the filtered solution. Red crystals of [Co(Mecyclen)(*S*-AlaO)]$ZnCl_4{\cdot}2H_2O$ (**2**) crystallized on scratching and cooling in ice. ^{1}H NMR (N−*Me*): 2.48 ppm). Anal. Calcd for $CoC_{12}H_{28}N_5O_2{\cdot}ZnCl_4{\cdot}2H_2O$: C, 24.99; H, 5.59; N, 12.15; Cl, 24.60. Found: C, 24.65; H, 5.55; N, 11.83; Cl, 24.73. Visible−UV spectrum: ϵ_{515}, 236; ϵ_{354}, 184.

Isomer 3. Isomer **2** (1 g) was dissolved in a formate buffer (30 cm^3, 0.2 M, pH 4.6) and left at ambient temperature (ca. 18 °C) for 15 min before quenching with 1 M HCl to pH ~ 2. The mixture was desalted chromatographically (see above) and the residue taken to dryness. The solid was then sorbed as a fine band on a long column of ion-exchange resin (40 × 2 cm) and eluted with 1 M HCl. The major first band (a small amount of **1**, which is in the second band to elute, may be present if the complex was left too long in the buffer solution[26]) was rather broad, and the tail of this band was found to be enriched in isomer **3** (ca. 80% **3** (N−*Me*: 2.41 ppm): 20% **2** (N−*Me*: 2.48 ppm)). This tail was collected and taken to dryness. Further enriched samples from other preparations were pooled and the combined material rechromatographed on a long ion-exchange column (1 m × 2 cm). The tail end of the major diffuse red band (~10%) was collected and taken to dryness. This was shown to be ca. 95% isomer **3** and was used (as the chloride salt) without further purification.

Isomer 5. A solution of isomer **4** (0.3 g) in water (50 cm^3) was adjusted to pH ~ 7 (0.01 M NaOH) and the mixture sorbed and eluted (1 M HCl) from a column of ion-exchange resin (40 × 2 cm). The first band to elute (ratio of two bands ca. 1:2) was taken to dryness and dissolved in the minimum volume of 0.1 M HBr, and solid $ZnBr_2$ was added. Scratching and cooling in ice resulted in purple crystals, which were collected, washed with MeOH, and dried in air (N−*Me*: 2.53 ppm). Anal. Calcd for $CoC_{12}H_{28}N_5O_2{\cdot}ZnBr_4$: C, 20.01; H, 3.93; N, 9.75; Br, 44.5. Found: C, 20.00; H, 4.80; N, 9.70; Br, 44.5. Visible−UV spectrum: ϵ_{519}, 291; ϵ_{355}, 204. Subsequent preparations involved crystallization as the $ZnCl_4^{2-}$ salt.

Isomer Equilibration. This was achieved on Norite A activated charcoal at 70 °C as described above for the (alternate) preparation of isomer **1**. This material was purified (for NMR determination of isomer distribution) by ion-exchange chromatography (1 M HCl eluent) and taken to dryness.

NMR Spectroscopy. Standard ^{1}H and ^{13}C NMR spectra (D_2O, d_6-DMSO, $CDCl_3$) were obtained using a Varian VXR 300 spectrometer. Difference nOe ^{1}H spectra (d_6-DMSO) were recorded on a Brucker AC 300 spectrometer using a standard Brucker NOEMULT pulse program. Acquisition times were typically 2 h per irradiated peak with sets of 6−8 experiments interleaved sequentially with the offset resonance frequency in multiples of 32 FIDS. No special precautions (such as the exclusion of dissolved oxygen) were used in sample preparation. In a typical experiment the irradiated peak frequency (or multiple of frequencies if the peak was a multiplet) was presaturated for ca. 3 s (T_1 values for various N−*H* and C−H protons were shown to be 0.1−0.3 s), a 90° pulse applied, and the FID acquired with a 3 T_1 delay. Spectra were acquired across a 15

(27) Searle, G. H.; Keene, F. R.; Lincoln, S. F. *Inorg. Chem.* **1978**, *17*, 3262.

Table 1. Crystallographic Data for **1/4b**, **2**, and **4a**

	1/4b	2	4a
chem formula	$C_{12}H_{30}N_5O_{11}C_{12}Co{\cdot}H_2O$	$C_{12}H_{32}N_5O_4Cl_4CoZn{\cdot}2H_2O$	$C_{12}H_{30}N_5O_{11}C_{12}Co{\cdot}H_2O$
a, Å	14.131	8.845(2)	8.757(2)
b, Å	8.777(2)	14.794(7)	9.684(5)
c, Å	17.745(4)	17.157(8)	25.291(6)
α, deg	90	90	90
β, deg	107.80(3)	90	90
γ, deg	90	90	90
V, Å^3	2095.5(8)	2245(2)	2144.7(13)
Z	4	4	4
fw	550.24	576.54	550.24
space group	$P2_1/n$ (No. 14)	$P2_12_12_1$ (No. 19)	$P2_12_12_1$ (No. 19)
T, °C	193(2)	193(2)	193(2)
λ, Å	0.710 73	0.710 73	0.710 73
ϱ_{calcd}, g cm^{-3}	1.738	1.694	1.698
μ(Mo Kα), cm^{-1}	11.41	23.10	11.15
transm coeff	0.801 (max), 0.628 (min)	0.914 (max), 0.837 (min)	0.763 (max), 0.713 (min)
R^a	0.0734	0.0415	0.0380
$wR_2(F_o^2)^a$	0.1426	0.0883	0.0980

^{a} $R = [\sum||F_o| - |F_c||/\sum|F_o|)$ $[F > 2\sigma(F)]$. $wR_2(F_o) = [\sum w(F_o^2 - F_c^2)^2/\sum wF_o^4]^{1/2}$ (all data); $w = (1/(\sigma^2F_o^2 + 0.0222P)^2) + 17.25P)$ for **1/4b**, $w = (1/(\sigma^2F_o^2 + (0.0378P)^2) + 2.77P)$ for **2**, $w = (1/\sigma^2F_o^2 + (0.0535P)^2) + 1.83P)$ for **4a**; $P = (\max(F_o^2,0) + 2F_c^2)/3$.

ppm window and were digitized using 16 K data points. Sufficient power was used to saturate the chosen peak to ca. 50% its normal height, but in some cases, where peak centers were too close to allow selective saturation, a lower peak power was used and/or the side of the peak in question was irradiated. Difference spectra were generated by subtracting the off resonance FID from the irradiated resonance FID of interest, and the difference was transformed using a line broadening of 0.5−1.0 Hz.

For these small molecules (M_r ca. 300 Da) the 1D NOE experiment is superior to the 2D NOSEY or ROSEY experiment.[28] The small number of well-separated N−*H* absorptions allows for selective irradiation, and because of better digital resolution, the enhancements of overlapping peaks in the C−H region are better distinguished. The set-up is also far simpler (i.e. mixing times).

X-ray Data Collection, Reduction, and Structure Determination. Diffraction data were collected on mauve crystals of **1/4b**, deep red crystals of **2**, and mauve crystals of **4a** using a Nicolet R3M diffractometer at 193(2) K and graphite-monochromated Mo Kα radiation. The data were corrected for Lorentz and polarization effects, and empirical absorption corrections were applied using SHELXTL.[29] Analysis of systematic absences was consistent with the space groups $P2_1/n$ for **1/4b** and $P2_12_12_1$ for **2** and **4a**.[30] Details of the crystals, data collections, and refinements are summarized in Table 1.

All three structures were solved using SHELXS-86[31] including any additional non-hydrogen atoms found in least-squares refinement and Fourier synthesis cycles. Weighted full-matrix refinement of the structures on F^2 was performed using SHELXL-93,[32] with all non-hydrogen atoms assigned anisotropic temperature factors. Hydrogen atoms were included in calculated positions. A difference Fourier following the location of all non-hydrogen atoms revealed the presence of peaks that could be sensibly assigned to the oxygen atoms of single water molecules of crystallization for **1/4b** and **4a** and to two such molecules for **2**. Inclusion of these atoms in the refinement led to a significant improvement in the residuals. Hydrogen atoms were not included for the solvent water molecules. For **1/4b** evidence was found in the difference Fourier map of an alternative location for the methine C atom of the alaninato ligand. This disorder was initially resolved by refining the two atom positions with the sum of the occupancy factors tied to unity. The separate occupancy factors converged to 0.50(3) consistent with the presence of equal proportions of the *R* and *S* enantiomers in the asymmetric unit and with the centrosymmetric space group. In final refinement cycles the occupancy factors were fixed at 0.5 for C(11) and C(11′) and the positions of the H atoms on C(11), C(11′), and the adjacent C(12) and N(5) calculated independently for the two orientations. For the noncentrosymmetric structures **2** and **4a** calculation of the Flack absolute structure parameter[33] gave −0.01(3) for **2** and −0.05(3) for **4a** in the final cycle, confirming that the chosen coordinates represented the correct absolute configurations of the chiral C atoms. Final difference Fourier maps were essentially flat for all three structures with the highest peak at 0.44 e Å^{-3} for **1/4b**, 0.77 e Å^{-3} for **2**, and 0.43 e Å^{-3} for **4a**. Final positional and equiv thermal parameters are listed in Tables 2−4. Tables of bond length and angle data, anisotropic thermal parameters, and hydrogen positional and thermal parameters are available as supporting information.

Results and Discussion

Preparations. Ciampolini has previously synthesized the related *trans*-dimethylcyclen ligand (Me$_2$cyclen·4HBr) by selective N-methylation of diethylenetriamine and subsequent condensation with bis(2-chloroethyl)methylamine.[24] Adaptation of this procedure to the synthesis of Mecyclen·3HCl gave only a ca. 15% yield in our hands, and a more convenient method requiring fewer steps and giving a higher overall yield is that shown in Scheme 1. This involves treating methylammonium acetate with 2 mol equiv of tosylaziridine and condensing the product (as its Na^+ salt) with the protected diethanolamine **3** to give Ts$_3$-Mecyclen, **4**. Deprotection using concentrated H_2SO_4 followed by anion-exchange chromatography gives **5** in 30% overall yield based on ethanolamine. The mixture of [Co(Mecyclen)(*S*-AlaO)]$^{2+}$ isomers was prepared from [Co-

(28) Reference 3, Chapter 8, p 253.
(29) Sheldrick, G. M. SHELXTL, An integrated system for solving, refining, and displaying crystal structures from diffraction data. University of Göttingen, 1981.
(30) *International Tables for X-ray Crystallography*; Kynoch Press, Birmingham, U.K., 1966; Vol. 1.
(31) Sheldrick, G. M. SHELXS-86, A program for the solution of crystal structures from diffraction data. University of Göttingen, 1986.
(32) Sheldrick, G. M. SHELXL-93; FORTRAN-77 program for the refinement of crystal structures from diffraction data. University of Göttingen, 1993. Sheldrick, G. M. *J. Appl. Crystallogr.*, in press.
(33) Flack, H. D. *Acta Crystallogr.* **1983**, *A39*, 876.

Table 2. Atomic Coordinates ($\times 10^4$) and Equivalent Isotropic Displacement Parameters (Å^2 $\times$ 10^3) for **1/4b**[a]

	x	*y*	*z*	*U*(eq)
Co(1)	4616(1)	2348(1)	7409(1)	15(1)
N(1)	4961(6)	871(9)	6654(4)	21(2)
C(9)	4092(8)	91(12)	6085(6)	40(3)
C(1)	5645(8)	−326(11)	7150(6)	32(3)
C(2)	5510(8)	−384(11)	7969(5)	27(3)
N(2)	5548(6)	1166(8)	8244(4)	18(2)
C(3)	5284(7)	1437(11)	8971(5)	22(2)
C(4)	5227(7)	3090(11)	9074(5)	26(3)
N(3)	4686(5)	3804(8)	8284(4)	16(2)
C(5)	5215(7)	5168(10)	8140(6)	21(3)
C(6)	6042(7)	4668(11)	7823(5)	20(2)
N(4)	5552(5)	3688(8)	7143(4)	14(2)
C(7)	6200(7)	2835(11)	6767(5)	24(2)
C(8)	5506(7)	1738(11)	6204(5)	27(3)
O(1)	3619(5)	1125(7)	7604(3)	19(2)
O(2)	2001(5)	677(9)	7302(4)	46(2)
C(10)	2711(9)	1397(14)	7213(6)	40(3)
C(11)	2506(15)	2990(26)	6788(12)	18(5)
C(12)	1626(7)	2999(13)	6136(7)	46(3)
C(11′)	2574(14)	2250(24)	6460(12)	12(5)
N(5)	3461(5)	3373(8)	6649(4)	18(2)
Cl(1)	8304(2)	1827(3)	9274(1)	24(1)
O(11)	7783(5)	1352(9)	8485(4)	48(2)
O(12)	8709(6)	3282(9)	9243(5)	55(2)
O(13)	7647(5)	1851(10)	9740(4)	47(2)
O(14)	9090(5)	792(9)	9610(4)	50(2)
Cl(2)	8052(2)	2280(3)	4906(1)	30(1)
O(21)	8979(5)	3002(8)	5305(4)	44(2)
O(23)	7577(6)	3048(11)	4189(5)	78(3)
O(22)	8251(6)	778(10)	4722(6)	80(3)
O(24)	7422(7)	2223(13)	5383(5)	83(3)
O(3)	4423(5)	−3876(8)	6170(4)	36(2)

[a] *U*(eq) is defined as one-third of the trace of the orthogonalized $\mathbf{U}_{ij}$ tensor.

Table 3. Atomic Coordinates ($\times 10^4$) and Equivalent Isotropic Displacement Parameters (Å^2 $\times$ 10^3) for **2**[a]

	x	*y*	*z*	*U*(eq)
Co(1)	3700(1)	4894(1)	9238(1)	16(1)
N(1)	2738(7)	5966(4)	9771(4)	23(2)
C(9)	3637(10)	6816(5)	9788(5)	35(2)
C(1)	2414(10)	5682(6)	10583(5)	34(2)
C(2)	1853(9)	4705(5)	10575(4)	29(2)
N(2)	2998(7)	4209(4)	10120(3)	22(2)
C(3)	2706(10)	3264(5)	9899(4)	28(2)
C(4)	4034(9)	3000(5)	9381(5)	32(2)
N(3)	4128(7)	3707(4)	8761(4)	25(2)
C(5)	3004(9)	3519(5)	8107(4)	31(2)
C(6)	2157(9)	4369(5)	7901(4)	29(2)
N(4)	1847(6)	4834(4)	8634(3)	21(1)
C(7)	1259(10)	5770(5)	8574(4)	26(2)
C(8)	1239(9)	6167(5)	9366(4)	29(2)
O(1)	5527(5)	5010(4)	9818(3)	21(1)
C(10)	6616(8)	5472(5)	9500(4)	22(2)
O(2)	7756(6)	5685(4)	9860(3)	33(2)
C(11)	6447(8)	5694(5)	8642(4)	17(2)
C(12)	7002(9)	6622(5)	8413(5)	33(2)
N(5)	4827(6)	5537(4)	8412(4)	22(1)
Zn(1)	8691(1)	2636(1)	7941(1)	21(1)
Cl(1)	10084(2)	1444(1)	8365(1)	36(1)
Cl(2)	9362(3)	3175(1)	6768(1)	35(1)
Cl(3)	6243(2)	2151(1)	7828(1)	34(1)
Cl(4)	8738(3)	3707(1)	8894(1)	27(1)
O(11)	617(7)	654(4)	2905(3)	52(2)
O(12)	1797(7)	9467(5)	3920(3)	53(2)

[a] See footnote *a* of Table 2.

(Mecyclen)Cl_2]Cl using either (*S*)-alanine or its methyl ester in aqueous solution at ca. 50 °C. These two reactions almost certainly occur via chloro−aqua intermediates since the solutions rapidly turn crimson (from purple) and then much more slowly

Table 4. Atomic Coordinates ($\times 10^4$) and Equivalent Isotropic Displacement Parameters (Å^2 $\times$ 10^3) for **4a**[a]

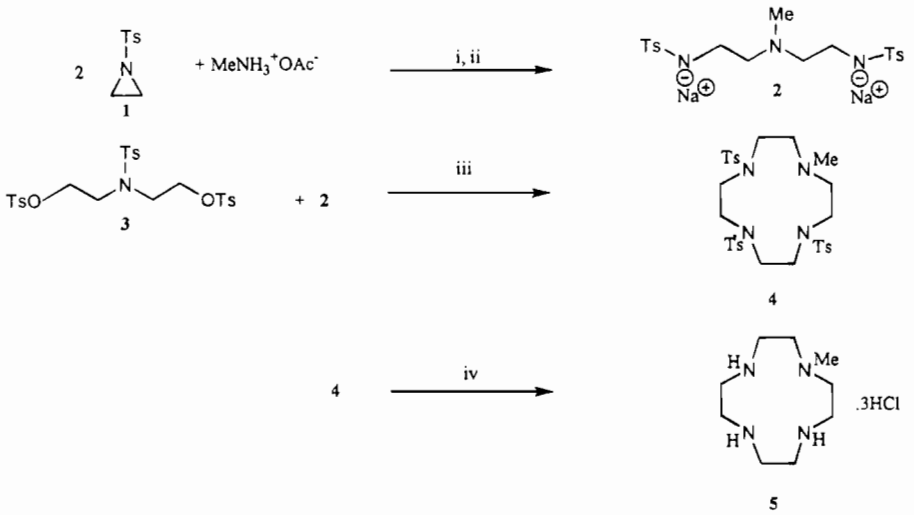
	x	*y*	*z*	*U*(eq)
Co(1)	2348(1)	5917(1)	1092(1)	17(1)
N(1)	754(5)	6365(5)	1657(2)	26(1)
C(9)	−205(7)	7629(7)	1577(2)	39(2)
C(1)	1617(7)	6520(6)	2166(2)	31(1)
C(2)	2872(6)	5447(6)	2187(2)	26(1)
N(2)	3725(4)	5636(4)	1679(2)	19(1)
C(3)	4911(6)	4589(6)	1545(2)	23(1)
C(4)	5381(5)	4935(6)	987(2)	24(1)
N(3)	3954(5)	5000(5)	659(2)	21(1)
C(5)	3447(6)	3576(6)	500(2)	28(1)
C(6)	1740(6)	3448(7)	568(2)	31(1)
N(4)	1402(4)	4097(5)	1086(2)	22(1)
C(7)	−246(5)	4307(6)	1202(2)	31(1)
C(8)	−299(6)	5141(7)	1699(2)	33(1)
O(1)	1081(4)	6332(4)	503(1)	22(1)
C(10)	1136(6)	7572(5)	307(2)	22(1)
O(2)	371(5)	7938(4)	−79(1)	33(1)
C(11)	2111(6)	8614(5)	605(2)	23(1)
C(12)	2932(6)	9637(6)	246(2)	28(1)
N(5)	3172(5)	7815(4)	957(2)	21(1)
Cl(1)	7609(2)	656(1)	994(1)	34(1)
O(11)	9104(6)	600(7)	787(2)	86(2)
O(12)	6793(8)	−537(7)	859(3)	120(3)
O(13)	7705(8)	696(9)	1546(2)	114(3)
O(14)	6871(6)	1825(6)	778(2)	70(2)
Cl(2)	2717(2)	1116(1)	1989(1)	29(1)
O(21)	1532(6)	2127(5)	2051(2)	55(1)
O(22)	2236(9)	−149(5)	2200(3)	109(3)
O(23)	4026(7)	1610(7)	2257(2)	87(2)
O(24)	3059(6)	985(7)	1448(2)	69(2)
O(3)	4407(6)	3039(5)	3202(2)	63(2)

[a] See footnote *a* of Table 2.

deep red as the product is formed. Both reactions gave excellent yields (80−90%). Six out of the eight possible isomers (cf. Figure 1) were detected in the preparative mixtures by ^{1}H NMR, and five have been isolated by a combination of ion-exchange chromatography and selective crystallization. The N−*Me* resonances (^{1}H NMR) were used as a means of determining isomer purity and distribution. The latter depends on the starting material used, with less **4** and **5** being formed with alanine than with its methyl ester. Figure 3 shows this before and after chromatography. Neither preparation gives the thermodynamic distribution, which is also shown. A sixth isomer (N−*Me*: 2.44 ppm) was detected as a very minor component of band **1** in the preparation starting with AlaOMe, but it was never isolated in pure form. Its behavior suggests that it is an *anti*(Me) isomer, possibly the *syn,syn* intermediate between isomers **4** and **5**. Different isomer distributions have been noted previously in similar reactions and have been attributed to initial coordination by carboxylate-O with the amino acid and by amino-N with the ester.[34] However the nonequilibrium *syn*(Me)/*anti*(Me) distributions found here must be decided by subsequent events. Intramolecular hydrolysis of the dangling ester function by coordinated H_2O or OH^- is likely in the reaction of the ester,[35] whereas direct substitution at the metal by the amino group is likely with the monodentate O-bound amino acid. Such processes apparently involve different steric requirements as far as the Me groups are concerned, and neither duplicates the equilibrium conditions found in the final chelate. Further consideration of these aspects will be left to a subsequent publication.[26] It is also to be noted that ion-exchange chromatography separates isomers on the basis of the orientation of the planar N−*H* proton adjacent to carboxylate-O rather than

(34) Marzilli, L. G.; Buckingham, D. A. *Inorg. Chem.* **1967**, *6*, 1042.
(35) Sutton, P. A.; Buckingham, D. A. *Acc. Chem. Res.* **1987**, *20*, 357.

Scheme 1. Reactions for Preparation of the Mecyclen Ligand[a]



[a] Key: i, CH_3CN, toluene, 18 h; ii, 2NaOEt; iii, DMF, 100 °C, 2 h; iv, H_2SO_4, 100 °C, 50 h, HCl.

syn(Me), *anti*(Me) isomers on the basis of the different stereochemistries of the two Me groups. This suggests hydrogen bonding to the resin by the *sec*-N−*H* centers plays an important role in their chromatography.

Following chromatography, isomer isolation was based on the selective crystallization of isomers **1** and **4**. Isomerization at pH ~ 5 then led to **2** and **5**, respectively. Isomer **3** was formed in larger than thermodynamic amounts using the kinetic relationships **2** → **3** → **1**[26] and by quenching at an appropriate time. It was found that isomers **1**−**3** could be readily interconverted at pH ~ 5, as could isomers **4** and **5**, but that the two sets remained distinct under these conditions. Complete equilibration was only possible on activated charcoal. The kinetics and thermodynamics of the transformations **3** ↔ 1 + 2 and **5** ↔ 4 will be taken up in a subsequent publication.[26]

^{1}H and ^{13}C Spectra. Figure 4a gives ^{1}H NMR spectra for each of the five isomers in acidified D_2O. Comparisons of the C−*H* (2.5−3.5 ppm) and N−*H* (4.5−7.5 ppm) regions show similarities between **1** and **4** and between **2** and **5**. All resonances downfield of 4.5 ppm are absent in spectra run at pD ~ 7 and are therefore assigned to exchangeable N−*H* protons. The 4.0 ppm multiplet (1 H) is coupled to the Me doublet of AlaO at 1.5 ppm and is therefore assigned to α-C−*H*. Separate resonances occur for each of the five N−*H* hydrogens in isomers **2** and **5**, but not for isomers **1** and **4** where two resonances overlap at 6.7 ppm; with **3** one N−*H* absorption is obscured by the HOD signal. The N−*H* and C−*H* chemical shifts are essentially independent of acid concentration (DCl, D_2SO_4) and addition of other electrolytes; this differs from spectra run in d_6-DMSO (see below). ^{13}C NMR data for spectra run in acidified D_2O are given in Table 1. The 18.5 and ~186 ppm absorptions are assigned to C−*Me* and $-CO_2^-$ carbons, respectively. A DEPT experiment assigned the N−*Me* and α-*C*−H carbons. Remaining *C*H_2 carbons of the macrocycle were assigned to specific sites using the method of Jackson.[11] This required partial N−*H*/*D* exchange and a knowledge of the relative exchange rates.[36] Such assignments are given in Table 5, and refer to the numbering scheme given in Figure 2.

The following general observations can be made concerning the chemical shifts of the Mecyclen carbons (Table 5). C_1 and C_8 adjacent to *tert*-*N*Me occur at lowest field (>63 ppm), and C_4 and C_5 adjacent to apical *sec*-*N*H come at the next lowest field (53−59 ppm) while carbons adjacent to the two planar *sec*-*N*H centers occur at highest field (<51 ppm). For the latter it appears that those adjacent to *syn*-*N*H occur at higher field (ca. 47 ppm) than those adjacent to *anti*-*N*H (48−50.4 ppm). Such generalizations add to those previously made by Searle *et al.*[37] for related Co(III) complexes containing dien and Medien ligands.

^{1}H nOe and COSY Spectra and Structure Assignment. 1H spectra run in d_6-DMSO are given in Figure 4b. These show "sharper" N−*H* absorptions than in Figure 4a, and some of the chemical shifts are now sensitive to the addition of electrolytes, such as DCl and LiCl. Such observations are not entirely new,[38,39] but they are of use in the nOe experiment where the separation of the N−*H* absorptions is necessary for selective irradiation. With the present complexes, protons **g** and **h** were found to be particularly sensitive to the addition of DCl or LiCl; signals **d** and **e** were relatively insensitive.

Table 6 contains all the essential information. Labels **a**−**h** refer to irradiated absorptions as given right to left in Figure 4b (high to low field). Absorptions **a**−**c** refer to C−*Me*, N−*Me*, and α-C−*H*, respectively, and **d**−**h** to the five N−*H* resonances. The latter do not always represent the same protons in the different isomers. Table 6 also contains chemical shift data for CH_2 hydrogens enhanced in the nOe experiment. COSY correlated C−*H* hydrogens are designated by an asterisk.

As indicated above, it is important in the nOe experiment that the irradiated absorptions be distinct, and this is so for the N−*H* hydrogens in d_6-DMSO solvent. It is also true of the *Me* groups (cf. Figure 3), and their presence in these complexes is therefore useful but not essential.[40] However the various CH_2 hydrogens of the ethylene bridges are strongly coupled, being spread over the 2.5−3.5 ppm range. Selective irradiation of these is therefore difficult, and no structural information was gained from this source. This property holds for other ethylenediamine or propylenediamine chelates of Co(III) where strong *J*−*J* couplings, and broadenings resulting from rapid

(36) Details of these assignments are given in the following: Rogers, A. J. Ph.D. Thesis, University of Otago, 1995.

(37) Searle, G. H.; Lincoln, S. F.; Teague, S. G.; Rowe, D. G. *Aust. J. Chem.* **1979**, *32*, 519.

(38) Nakazawa, H.; Sakaguchi, U.; Yoneda, H.; Morimoto, Y. *Inorg. Chem.* **1981**, *20*, 973.

(39) Brasch, N. E.; Buckingham, D. A.; Clark, C. R.; Finnie, K. S. *Inorg. Chem.* **1989**, *28*, 3386.

(40) Buckingham, D. A.; Rogers, A. J. Other results (to be published) will show that nOe structure determination does not require the presence of Me groups.

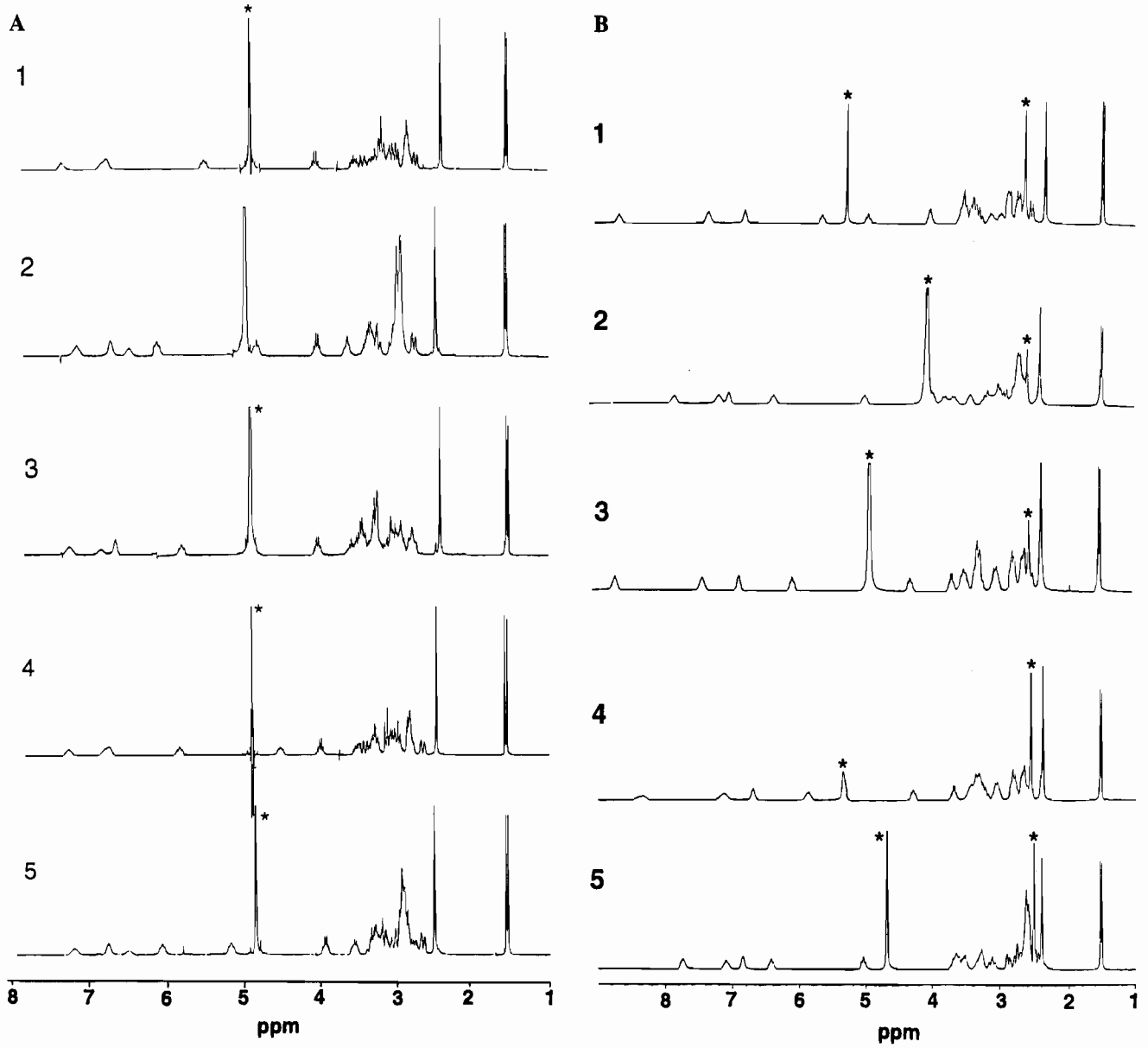


Figure 4. ^{1}H NMR spectra (300 MHz) for isomers **1**−**5**: (A) in 0.1 M DCl (asterisk represents HOD signal); (B) in d_6-DMSO containing sufficient DCl to differentiate the five N−*H* protons (asterisk represents residual HOD and d_5-DMSO impurity). Absorptions downfield of 4.1 ppm in (A) represent N−*H* protons (absent from spectra recorded in neutral D_2O). Spectra similar to (B) were used in the nOe and COSY experiments.

Table 5. ^{13}C NMR Assignments for Isomers **1**−**5**[a]

isomer	CM*e*	NM*e*	α-*C*H	C_1	C_2	C_3	C_4	C_5	C_6	C_7	C_8
1	18.8	47.8	54.6	65.3	49.6	48.6	55.2	56.9	47.6	47.3	66.2
2	18.5	48.0	53.2	67.7	47.0[b]	47.1[b]	57.4	53.5	50.4	50.4	63.3
3	18.9	50.3	54.8	69.2	50.3[b]	50.3[b]	58.8	57.6	49.5[b]	51.2[b]	68.0
4	18.5	48.8	53.9	66.7	47.2	47.7	56.4	55.2	49.1	49.4	65.4
5	18.2	49.4	53.3	63.6	50.2	50.8	53.1	56.9	47.4	47.4	67.6

[a] ppm from Bu^{t}OH reference; acidified D_2O solvent, 25 °C. [b] Assignment is equivocal due to insufficient resolution of the β-isotopic shift from *ap*-N*H*/*D*.

conformational equilibria, make individual C−*H* assignments difficult or impossible.[41]

A complete analysis of the nOe and COSY data is given for isomer **1** in what follows. For isomers **2**−**5** the essential information is given in Table 6 and supporting nOe and COSY spectra are available on request (cf. supporting information).

Isomer 1 (Table 6; Figures 5 and 6). Figure 5 shows the results of irradiating absorptions **a**-**h**. Irradiation of **a** (C−*Me*) shows a strong nOe to **c** (α-C−*H*), a weak nOe to **b** (N−*Me*), and a weak nOe to one N−*H* proton, **d**. Irradiation of **c**, **b**, and **d** gives the reverse enhancements to **a** suggesting each is closely related in space to **a**. The **a** ↔ **c** association is to be expected since they are also *J*−*J* coupled, but the **a** ↔ **b** association suggests a *syn*(Me) isomer. The **a** ↔ **d** and **b** ↔ **d** associations suggest that **d** is *syn* to both Me groups, and consideration of a molecular model suggests **d** to be the N−*H* hydrogen of the amino group of alanine *cis* to C−*Me* (i.e. N*H* (*cis* C−H)). Irradiation of **g** gives nOes to **e**, and possibly to

(41) Hawkins, C. J. In *Absolute Configurations of Metal Complexes*; Wiley-Interscience: New York, 1971; Chapter 6.

Table 6. nOe Enhancement Data for Isomers **1**–**5** and Assignment of N–*H* and C–*H* Hydrogens

irradiated peak (ppm)[a]	enhancement, %[b] **a**	**b**	**c**	**d**	**e**	**f**	**g**	**h**	CH_2, ppm	assgn (**a**–**h**)
					Isomer **1**					
a (1.50)		1	3	1						C–*Me*
b (2.34)	1			2					2.80, 3.20	N–*Me*
c (3.96)	4				1	1				α-C–*H*
d (4.91)	3	4	4		9					N*H*(*cis*-C–Me)
e (5.45)			15	13		1	3			N*H*(*cis*-C–H)
f (6.70)			5						2.45, 3.4[c]	*ap*-N–*H*
g (6.96)				1	2				2.6[c]	N*H*(*syn*(N))
h (8.05)									2.7,[c] 3.45	N*H*(*anti*(O))
					Isomer **2**					
a (1.50)		2	+	2		+				C–*Me*
b (2.40)	1			2					3.05	N–*Me*
c (3.95)	+				+	+				α-C–*H*
d (4.95)	6	6			13					N*H*(*cis*-C–Me)
e (6.24)			+	13		+			2.70, 3.00, 3.40	N*H*(*cis*-C–H)
f (6.90)			+						2.7–2.85[c]	*ap*-N–*H*
g (7.15)									2.70[c]	N*H*(*syn*(O))
h (7.40)									2.75, 3.7, 3.8	N*H*(*anti*(N))
					Isomer **3**					
d (5.13)	5	4	2		9		1	3.7		N*H*(*cis*-C–Me)
e (6.15)			12	14			4	3.9		N*H*(*cis*-C–H)
f (7.16)			5						2.9, 3.1, 3.9	*ap*-N–*H*
g (7.28)									2.8, 3.1	N*H*(*syn*(O))
h (7.81)					2				2.7, 3.9	N*H*(*syn*(N))
					Isomer **4**					
a (1.50)			2	1						C–*Me*
b (2.36)			3		1				2.80, 3.05, 3.2–3.3	N–*Me*
c (3.66)	5	4			3					α-C–*H*
d (4.27)	5				16	3	2			N*H*(*cis*-C–Me)
e (6.05)	*d*	*d*	9	13			2			N*H*(*cis*-C–H)
f (6.83)		+		1					3.30[c]	*ap*-N–*H*
g (7.38)				+	2				2.55,[c] 3.3	N*H*(*syn*(N))
h (8.70)									2.6,[c] 3.30, 3.5	N*H*(*anti*(O))
					Isomer 5					
a (1.50)			2	1						C–*Me*
b (2.37)			3		1				2.92	N–*Me*
c (3.55)	2	1			+			+	2.65	α-C–*H*
d (4.96)	4				11	3			2.7	N*H*(*cis*-C–Me)
e (6.27)	2	1	+	14					2.7, 3.3	N*H*(*cis*-*C*–H)
f (6.72)				3					2.75, 3.3[c]	*ap*-N–*H*
g (7.15)									2.65	N*H*(*syn*(O))
h (7.25)									2.67,[c] 3.55	N*H*(*anti*(N))

[a] Chemical shifts relative to C*Me* doublet (1.50 ppm). [b] + indicates calculation of % enhancements for the irradiated or enhanced peak not possible due to overlapping signal. [c] Enhanced signal also coupled to irradiated peak. [d] Poor subtraction does not allow calculation of % enhancement.

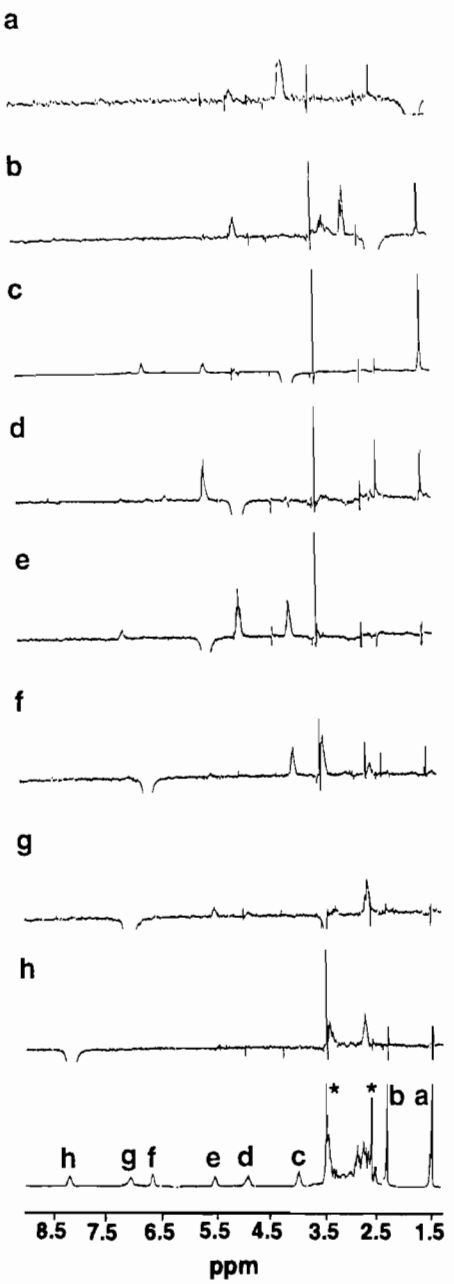


Figure 5. nOe difference spectra for isomer **1** showing the results of irradiating absorptions **a**–**h**.

d, and the reverse enhancement of **g** on irradiation of **e** is very apparent. Thus **g** must be the planar N–*H* proton of Mecyclen *syn* to the alanine amino group, N*H*(*syn*(N)).

The remaining N–*H* proton at lowest field, **h**, does not give nOe enhancements to the other N–*H* centers so that no direct structural information can be gained from this source. However **h** is the only unassigned N–*H* proton and so must be associated with the planar N–*H* center *cis* to carboxylate-O; this is in keeping with its lack of nOes to the other N–*H* centers. This proton may be either *syn*(O) or *anti*(O), and a decision may be arrived at by considering its nOe to the CH_2 region of the spectrum. Interpretation depends on the fact that we might expect an *anti* N–*H* proton to show nOes to *endo* C–*H* hydrogens on both sides of the Mecyclen macrocycle, that is, to hydrogens coupled to *both* planar *sec*-N–*H* centers. On the other hand a *syn* N–*H* proton should show nOe to only adjacent *exo* N–*H* hydrogens to which it is coupled. Put another way, if a planar N–*H* hydrogen shows nOe to CH_2 hydrogens coupled to a remote N–*H* center of the same ligand, then that proton must be *endo* and occupy an *anti* site. Figure 6 shows the *sec*-N–*H* part of the H/H 2D correlation spectrum for isomer **1** (protons **f**, **g**, **h**). **g** is coupled to CH_2 hydrogens centered at 2.6 and 3.45 ppm; likewise **h** is coupled to CH_2 hydrogens centered at 2.7 and 2.85–3.15 ppm. Irradiation of **h** (Figure 5) shows nOe enhancements to CH_2 hydrogens centered at 2.7 and 3.45 ppm, but the latter absorption is also coupled to **g**; it must therefore represent *endo* hydrogens adjacent to the N*H*(*syn*(N)) center, and **h** must be an *anti*(O) proton. Thus isomer **1** has the *syn*(Me), *syn*(N), *anti*(O) configuration. Other CH_2 hydrogens can be assigned similarly. Those at 3.34 ppm are both coupled to **f** (Figure 6) and enchanced in the nOe experiment (Figure 5). They are assigned to *exo* Mecyclen hydrogens adjacent to *ap* N–*H*. Those at 2.8 ppm and coupled to **f** but not enhanced in the nOe experiment (Figure 5) must be the corresponding *endo* hydrogens. The **g** coupled hydrogens at 2.6 ppm (Figure 6) are assigned to adjacent *exo* hydrogens,

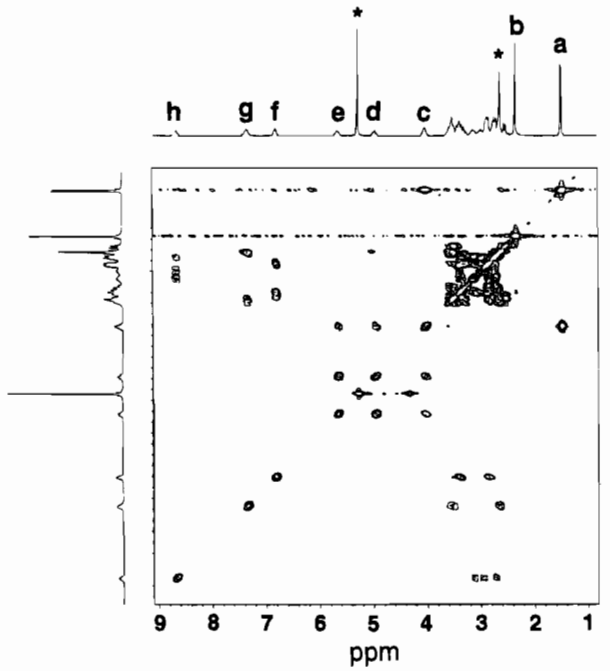


Figure 6. COSY ^{1}H spectrum for isomer **1**. This sample differs from that used for the nOe experiment (Figure 5).

and this is confirmed by the spectrum given in Figure 5. The **h** coupled hydrogens at 2.7 ppm are also nOe enhanced and are therefore assigned to the adjacent *endo* hydrogens, whereas the nonenhanced, but coupled, hydrogens at 2.85−3.15 ppm are assigned to *exo* hydrogens *syn* to carboxylate-*O*.

The *syn*(Me), *syn*(N), *anti*(O) assignment for this isomer is confirmed by the crystal structure shown in Figure 7.

Isomer 2 (Table 6). Many of the nOe enchancements for this isomer parallel those for isomer **1**. Thus **a**, **b**, and **d** give positive nOes to each other confirming **2** as a *syn*(Me) isomer with **d** being the alanine N−*H* proton *cis* to asymmetric C−*Me* (i.e. N*H*(*cis*C−*Me*)). Likewise the **d** ↔ **e**, **c** ↔ **f**, and **c** ↔ **e** associations confirm **e** as being *cis* to **c** (i.e. N*H*(*cis*-CH)) and **f** as being the *ap* N−*H* proton. The major difference with this isomer is the total lack of nOes to and from **g** and **h** to other N−*H* hydrogens. This suggests an *anti*(N), *syn*(O) configuration since an *anti*(N), *anti*(O) arrangement would be expected to give **g** ↔ **h** nOes as well as cross-correlations with the COSY spectrum. A comparison of the *sec*-N−*H* region of the COSY spectrum with the corresponding nOe enhancements to the CH_2 region (supporting information D2/D3) shows **h** giving nOes to two distinct hydrogens (3.7, 3.8 ppm) which are also coupled to **g**. These hydrogens must be *endo*, and **h** is therefore the N*H*(*anti*(N)) proton. Isomer **2** is identified as *syn*(Me), *anti*(N), *syn*(O) (cf. Figure 1). This is confirmed by the crystal structure, Figure 8.

Isomer 3 (Table 6). As for isomers **1** and **2**, isomer **3** shows nOes (supporting information D3/D5) for N−*H* protons **a**−**c** characterizing it as a *syn*(Me) isomer. Absorptions **d**−**f** belong to the alanine NH_2 group (**d**, **e**) and *ap* N−*H* (**f**) as before, and **c** is the α-C−*H* hydrogen *cis* to **e**. Irradiation of **e** also gives enhancement to **h**, and the reverse enhancement from **h** to **e** is also seen, characterizing **h** as the *syn*(N) proton. **g** does not show nOes to other N−*H* protons, nor to CH_2 protons of Mecyclen which are coupled to **h**. It must therefore be the *syn*(O) proton, and this isomer is therefore assigned the *syn*(Me)-, *syn*(N), *syn*(O) configuration.

Isomer 4 (Table 6). Irradiations of **a** and **b** show no enhancements to **b** and **a**, respectively, making this a probable *anti*(Me) isomer (supporting information D6). This is confirmed

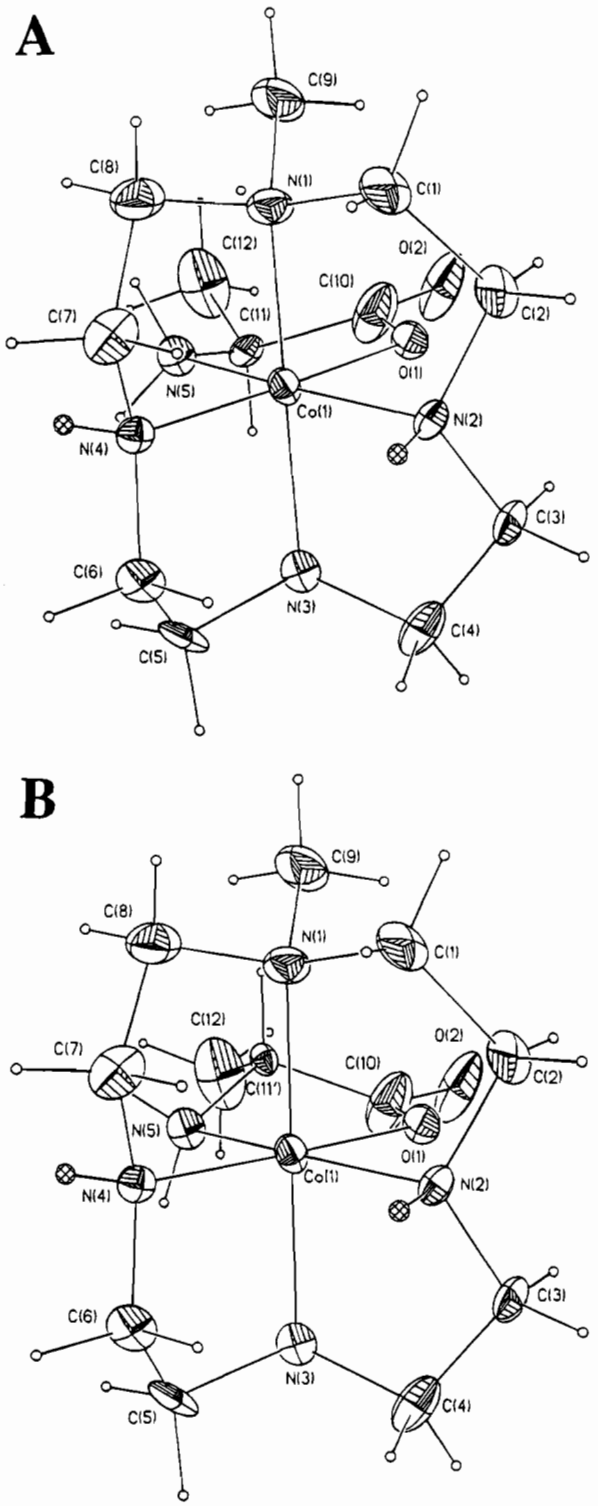


Figure 7. Molecular structure of cations **1** and **4b** showing the atom-numbering scheme: (A) **1**; (B) **4b**.

by the positive **b** ↔ **c** association. Proton **f** is again assigned to the *ap*-N*H* proton, and it shows a definite **f** ↔ **d** association. The **d** ↔ **e**, **c** ↔ **e**, and **d** ↔ **a** enhancements require **d** and **e** to be the two NH_2 protons of alanine, with **d** being *cis* to C−*Me* and **e** *cis* to α-C−*H* respectively. The **g** ↔ **d** and **g** ↔ **e** enhancements require **g** to be a *syn*(N) proton, and the lack of enhancement from **h** to other N−*H* protons confirms **h** as being the N−*H* proton of the planar N center adjacent to carboxylate-*O*. Once again by examining nOes to the CH_2 region of the spectrum it is found that **h** gives enhancements to hydrogens coupled to **g** making **h** an *anti*(O) proton (supporting information

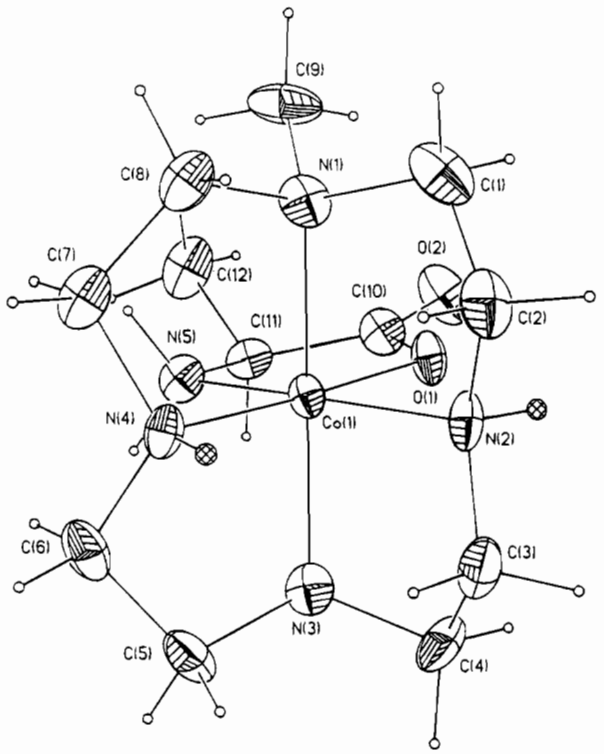


Figure 8. Molecular structure of cation **2** showing the atom-numbering scheme.

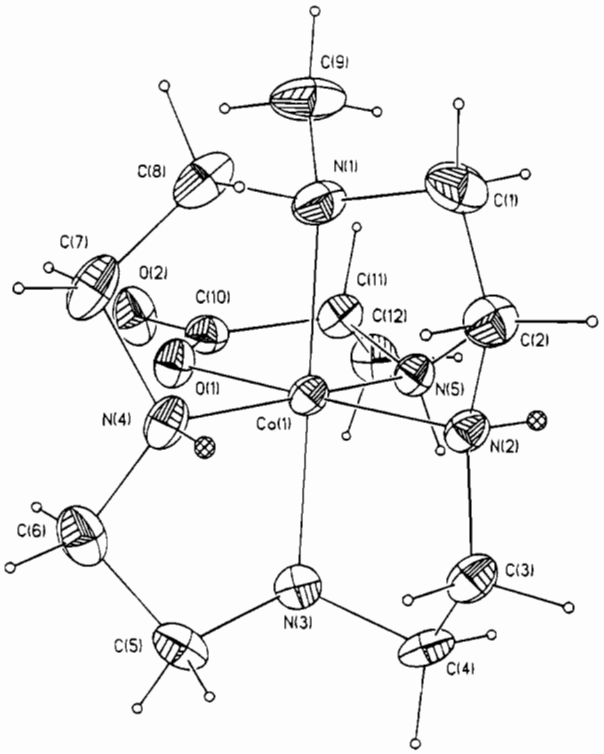


Figure 9. Molecular structure of cation **4a** showing the atom-numbering scheme.

D7). Isomer **4** thus has the configuration *anti*(Me), *syn*(N), *anti*(O). This is confirmed by the crystal structure, Figure 9.

Isomer 5 (Table 6). Once again the lack of **a** ↔ **b** associations suggests an *anti*(Me) structure, and this is confirmed by the **b** ↔ **c** association (supporting information D8/D9). The **d** ↔ **f** and **d** ↔ **a** enchancements confirm **d** as being the NH_2 proton *syn* to *ap*N−*H* and *cis* to C−*Me*. The lack of any **g** or **h** associations with other N−*H* protons again suggests an *anti*(N), *anti*(O) or an *anti*(N), *syn*(O) configuration for the two planar N*H* centers, and the latter configuration was again confirmed by nOe/COSY comparisons; proton **h** gives nOes to CH_2 hydrogens coupled to **g** making **h** an *anti* proton, while **g** gives nOes to CH_2 hydrogens none of which are coupled to **h**. Thus this isomer has the *anti*(Me), *anti*(N), *syn*(O) configuration.

Crystal Structures. Crystal structures of isomers **1**, **2**, and **4** were carried out to confirm the nOe assignments. Crystals of **1** were grown from a solution which contained some **4** (cf. Experimental Section), and it is apparent that the two diastereomers, **1** + **4b**, cocrystallized as the least soluble ClO_4^- salt. Furthermore the centrosymmetric space group $P2_1/n$ requires each diastereomer to be present in enantiomeric pairs so that at least some racemization of (*S*)-alanine has occurred during the synthesis of this sample.[42] Figure 7 gives Ortep diagrams of cations **1** (containing (*S*)-AlaO) and **4b** (containing (*R*)-AlaO) and these confirm the *syn*(Me), *syn*(N), *anti*(O) and *anti*(Me), *syn*(N), *anti*(O) designations, respectively. Likewise Figure 8 shows cation **2** containing the *syn*(Me), *anti*(N), *syn*(O) configuration, and Figure 9 shows cation **4a** with another representation of the *anti*(Me), *syn*(N), *anti*(O) structure. Both the latter structures exist in asymmetric space groups, and the absolute (*S*)-configuration of alanine is confirmed in each case. All structures consist of independent $[Co(Mecyclen)(S\text{-}AlaO)]^{2+}$ cations, water molecules, and ClO_4^- (**1** + **4b**, **4a**) or $ZnCl_4^{2-}$ (**2**) anions, linked by extensive networks of hydrogen bonds.

Bond lengths Co(1)-N(2) and Co(1)-N(4) (mean 1.94(1) Å) are significantly shorter than the corresponding Co(1)-N(1) and Co(1)-N(3) vectors (mean 2.01(2) Å), and this can be attributed to minimization of angular strain in the folded macrocycle. A clearer indication of strain is given by the N(1)−Co(1)−N(3) bond angle (mean 164.5(7)°), and this has the effect of bending N(1), N(3) away from the alaninate chelate with concomitant decreases in the N(1,3)−Co(1)−N(2,4) bond angles. Such deformations have been observed previously in crystal structures of cyclen complexes[43,44] but are significantly less than those found in the related, but sterically less demanding, β-trien chelates.[45] The δ and λ conformations of the two chelate rings spanning the N−*Me* group N(1)−C(1)−C(2)−N(2) and N(1)−C(8)−C(7)−N(4) in **2** and **4a** are enantiomeric to those below the square plane N(2)−C(3)−C(4)−N(3) and N(4)−C(6)−C(5)−N(3) containing the alaninato chelate. A similar situation is found with $[Co(cyclen)(NO_2)_2]Cl$.[43] Bond distances within the alaninato chelate compare well with those previously found for alaninato chelates of Co(III)[44] so that no additional strain is apparent as a result of a strained Mecyclen quadridentate or of close nonbonded contacts with it. As expected the C−O bonds involving coordinated O atoms, C(10)−O(1) (mean 1.29(1) Å), are significantly longer than those involving the carbonyl oxygen, C(10)−O(2) (mean 1.232(7) Å). The Co(1)−N(5) (mean 1.99(1) Å) and Co(1)−O(1) vectors (mean 1.89(1) Å) also corresponds well with those of other amino acidate complexes. The angle subtended at Co(1) by the five-membered ring, O(1)−Co(1)−N(5) (mean 83.6(7)°), is also normal.

Other Considerations. (1) Alaninate Ring Conformation. The **e** ↔ **g** (isomers **1** and **4**) and **e** ↔ **h** (isomer **3**) enhancements are much larger than the corresponding **d** ↔ **g** (**1** and **4**) and **d**

(42) We are uncertain as to how this occurred since inversion at α-C−*H* only occurs under alkaline conditions in chelated alaninate complexes (pH > 11), and we were very careful not to allow these conditions to apply. However, the crystals did wait some 8 months before analysis.
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Table 7. Nonbonded H/H Distances in Crystal Structures **1/4b**, **2**, and **4a**

struct	nonbonded atoms[a]	dist, Å	nOe[c]
1[b]	**a**, **b**	3.026, 3.670	+
	d, **b**	2.135, 3.195	+
	g, **d**	2.607	+
	g, **e**	2.254	+
	f, **c**	2.541	+
	f, **e**	2.699	-
	h, *endo* C−*H*	2.316, 2.470	+
2	**a**, **b**	2.457, 2.928	+
	d, **b**	2.177, 3.121	+
	f, **c**	2.883	+
	f, **e**	2.446	+
	h, *endo* C−*H*	2.452, 2.303	+
4a (**4b**)[b]	**a**, **f**	3.681 (3.956)	-
	b, **c**	2.119, 2.687 (1.739, 2.288)	+
	b, **e**	2.586 (2.558)	+
	d, **f**	2.431 (2.399)	+
	g, **e**	2.245 (2.218)	+
	g, **d**	2.634 (2.705)	+
	h, *endo* C−*H*	2.385, 2.411 (2.316, 2.470)	+

[a] **a** and **b** represent C−*Me* and N−*Me* groups. Distances in column 3 represent distances to nearest H atoms. [b] Since structures **1** and **4b** were obtained by averaging electron densities, some of the distances in column 3 have large standard deviations. [c] Observed (+), not observed (−) (cf. Table 6).

↔ **h** (**3**) enhancements (Table 6) suggesting that **e** is closer than **d** to *syn*-N−*H*. Furthermore, enhancements are observed between **d** and **b** but not between **e** and **f** in the *syn*(Me) isomers (**1**−**3**) and between **d** and **f** but not between **e** and **b** in the *anti*(Me) isomers (**4** and **5**). These results suggest that the C−*Me* group adopts an equatorial orientation, forcing **e** to be more equatorial than **d** and pushing the α-C−*H* proton close to *ap*-N−*H* in the *syn*(Me) isomers and close to N−*Me* in the *anti*(Me) isomers. This equatorial conformation is confirmed by the crystal structures; Table 7 lists appropriate nonbonded distances. Table 7 also shows that at 300 MHz nOe enhancements are observed up to 2.6 Å for C−*H*/ N−*H* separations and beyond 3.0 Å when a methyl group is involved. The above equatorial conformation contrasts with [Co(Mecyclen)(GlyO)]$^{2+}$ where no C−*Me* group is involved. In this complex similar nOe enhancements are seen from **d** and **e** to **g** and from the two CH_2 hydrogens of glycine (analogous to **c**) to N−*Me* and *ap*-N−*H*.[46] This suggests that the glycinate ring is reasonably planar, at least on average, in this structure.

(2) N*H* Chemical Shifts. As suggested elsewhere,[47] these usually follow the order $NH_3 > RNH_2 > R_1R_2NH$ in Co(III) complexes, but in the present examples where a fair degree of stereochemical rigidity exists it is found that the N−*H* chemical shifts are particularly sensitive to stereochemical location and to ion-pairing effects. Thus the two NH_2 protons of the alanine chelate are well separated (≥1.0 ppm) with that *cis* to α-C−*H* always being at lowest field. Also, the "planar" N−*H* proton of the *sec*-N−*H* group is usually found at lower field than the corresponding "apical" *sec*-N−*H* proton,[47] but in the present examples there is not much in it with the N*H*(*syn*(O)) and *ap*-N−*H* protons having similar (sometimes overlapping) chemical shifts (cf. Figure 4a, isomers **2** and **3**). However, of the two, the planar *syn*(O) proton is the more sensitive to ion-pairing effects. Also when present (isomers **1**, **2**, **4**, and **5**) the *anti* N−*H* proton (*endo* to Mecyclen) is always found at lowest field (**h**, 7.5−8.2 ppm) and is also the most sensitive to the addition of electrolytes. We ascribe this sensitivity to ion-pairing effects which, in related complexes,[48] appears to be more significant on the "back-face" of the molecule distant from anionic ligands.

Acknowledgment. We thank Associate Professor A. Wilkins and Dr. R. Thompson, University of Waikato, for assistance with the nOe experiments and Professor W. T. Robinson, University of Canterbury, for collecting the X-ray data.

Supporting Information Available: Tables giving bond lengths and angles (D1), anisotropic displacement parameters (D2), hydrogen coordinates and isotropic displacement parameters (D3) for crystal structures **1/4b**, similar tables for structure **2** (D5−D7) and structure **4a** (D9−D11), and figures of ^{1}H-decoupled ^{13}C spectra for isomers **1**−**5** (D1), nOe and 1D COSY spectra for isomers **2** (D2, D3), **3** (D4, D5), **4** (D6, D7), and **5** (D8, D9) (22 pages). Ordering information is given on any current masthead page.

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