respectively) are obtained by the hydrolysis of *N*-ethylidene-threonyl- and *N*-ethylideneallothreonylglycinato complexes (İ), formed in the second phase of the process, by the reaction of acetaldehyde with the N-terminal CH_2 group of the Schiff base of glycylglycinato ligand. The presence of complexes of type İ was proved in the reaction mixture obtained in the reaction of *mer*-[CoCN(glygly)(en)] with acetaldehyde.

When the results obtained in this and in our previous paper are taken into account,[2] the following can be concluded:

(a) The glycylglycinato ligand of all the investigated *mer*-[Co(glygly)L(en)] complexes (L = CN^-, NCS^-, NO_2^-) in the reaction with acetaldehyde gives rise to the corresponding complexes of Schiff bases, as well as a mixture of diastereomeric threonyl- and allothreonylglycinato complexes. The latter are obtained in a much higher yield when the reaction involves the dipeptidato ligand of cyano (~40%) rather than that of the nitro and isothiocyanato (~6%) complexes, respectively; it seems likely that the greater reactivity of the dipeptidato ligand of *mer*-[CoCN(glygly)(en)] is at least partly due to a decreased electron density at the amino nitrogen, which is caused by the trans effect of the cyano ligand. Thus, the first step of the reaction, i.e. deprotonation of the amino group of the dipeptidato ligand, is facilitated.

(b) When the ligand L is changed, the ratio of complexes present in the mixture obtained is altered.

(c) Only one amino group of the ethylenediamine ligand in nitro and isothiocyanato complexes reacts with acetaldehyde, whereby the *N*-ethylidene group in the *mer*-[Co(glygly)NO_2(CH_3CH=en)] complex was found to be in the trans position with respect to the nitro group.[13]

In view of the fact that the position of the *N*-ethylidene group in *mer*-[Co(glygly)NCS(CH_3CH=en)] is not so far determined and that in the reaction of *mer*-[CoCN(glygly)(en)] with acetaldehyde the corresponding condensation product was not obtained, at present, nothing can be said on the effect of the unidentate ligand on the reaction of coordinated ethylenediamine with aldehyde; this will be the subject of our further investigations.

Registry No. I, 18746-17-3; Ia, 93564-67-1; Ib, 93564-68-2; IIa, 93564-70-6; IIb, 93564-72-8; IIIa, 93711-44-5; IIIb, 93711-45-6; IVa, 93564-71-7; IVb, 93564-74-0; Vb, 93564-73-9; *mer*-[CoCN(CH_3CH=threogly)(en)], 93564-69-3; *mer*-[CoCN(CH_3CH=allothreogly)(en)], 93711-43-4; *mer*-[Co(glygly)NO_2(en)], 70738-74-8; acetaldehyde, 75-07-0; glycylglycine, 556-50-3; ethylenediamine, 107-15-3.

Supplementary Material Available: Listings of thermal parameters and observed and calculated structure factor amplitudes (9 pages). Ordering information is given on any current masthead page.

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Nucleoside Complexing. Evidence for Three Metastable Species in the Reaction of Nucleosides with *cis*-Dichlorobis(dimethyl sulfoxide)platinum(II). Use of Restricted Rotation about Pt-N Bonds for Structural Assignments

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The reactions in Me_2SO of *cis*-Pt(Me_2SO)$_2Cl_2$ with 7-methylinosine (7-MeIno), 7-methyl-9-propylhypoxanthine (7-Me-9-PrHX), cytidine (Cyd), and 5-methylcytidine (5-MeCyd) were examined with high-field 360-MHz ^1H NMR spectroscopy as well as limited studies with ^{13}C NMR spectroscopy. In a typical reaction with 7-MeIno at a ratio of one nucleoside per Pt, the products formed in greater abundance at short time periods were found to be *trans*-Pt(7-MeIno)(Me_2SO)Cl_2 and *cis*-[Pt(7-MeIno)$_2$(Me_2SO)Cl]$^+$. With time, these species slowly converted to the stable product, *cis*-Pt(7-MeIno)(Me_2SO)Cl_2. The conversion from a *cis*-dichloro to a *trans*-dichloro species probably proceeds via the complex *cis*-[Pt(7-MeIno)(Me_2SO)$_2$Cl]$^+$, which could be generated by addition of $AgNO_3$ at the initiation of the reaction. When Cyd was employed, direct evidence for the intermediacy of *cis*-[Pt(Cyd)(Me_2SO)$_2$Cl]$^+$ was obtained in the 360-MHz NMR spectrum of the reaction mixture. This species could be made to be the predominant species by addition of $AgNO_3$ as above. The structures were assigned on the basis of the NMR spectrum of complexes since, in certain geometries, the presence of the chiral D-ribose sugars allowed identification of rotamers that are rendered diastereomeric by the chiral sugars. The barrier to rotation about the Pt-N3 bond in Cyd complexes is so large that no evidence for appreciable rotation was observed even at 80 °C, above which temperatures decomposition set in. For the N1-bound 7-MeIno complexes, rotation is somewhat more facile and estimates of rotation barriers were obtained from the partial collapse of the 7-Me ^1H NMR resonances. From $J_{1'2'}$ coupling constants of the complexes, some evidence was found that the N sugar conformer is favored somewhat over the S conformer relative to the distribution of these conformers in the free ligand. This tendency was greater for Cyd and 5-MeCyd complexes than 7-MeIno complexes. This difference was attributed to the sugar and metal being on the same nitrogen heterocyclic ring in the former complexes but on different rings in the latter complexes.

A Pt(II) drug, *cis*-Pt(NH_3)$_2Cl_2$, is currently the most widely sold antitumor agent in the United States.[1] The mechanism of action of this drug probably involves attack on DNA.[2] Reaction of Pt(II) complexes with DNA and DNA constituents has been studied for two additional reasons. First, Pt is a useful heavy-metal label for EM or X-ray studies of nucleic acids.[3] Second, Pt(II) forms inert complexes, and the effects of metal binding on nucleic acid components and the sites of metal binding can be more readily recognized than when labile metal centers are employed.[4]

The Pt antitumor drugs are bifunctional reagents, and the stepwise process that leads to the attachment of Pt to two nucleic

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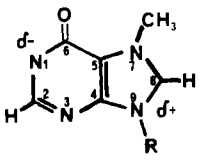
Table I. Selected ^{1}H NMR Spectral Data for 7-MeIno and Some Complexes Derived from *cis*-$Pt(Me_2SO)_2Cl_2$[a]

compd	base[b]			ribose[c]	
	H8	H2	H7(CH_3)	H1′	$^3J_{1'2'}$, Hz
7-MeIno	9.312	8.072	4.096	5.902	4.8
trans-Pt(7-MeIno)(Me_2SO)Cl_2	9.460	8.123	4.095	5.948	2.9
cis-Pt(7-MeIno)(Me_2SO)Cl_2	9.483[d]	8.351	4.110[d]	5.591	2.9
		8.349		5.949	3.2
cis-[Pt(7-MeIno)$_2$(Me_2SO)Cl]$^+$	9.540	8.605	4.080	5.915 (m)[d]	3.5[e]
	9.532	8.600	4.090		3.3[e]
	9.528[d]	8.282	4.053		3.6[e]
		8.277	4.062		3.6[e]
cis-[Pt(7-MeIno)(Me_2SO)$_2$Cl]$^+$	9.563	8.258	4.109[d]	5.986	3.6
	9.557	8.251		5.976	4.0

[a] Me_2SO-d_6, 360-MHz data, chemical shift values in ppm from internal Me_4Si. [b] Singlets. [c] Position of the center of the doublet. [d] Two or more superimposed resonances. [e] Obtained indirectly from H2′ resonances (±0.3 Hz); see supplementary material.

acid bases is not understood. It has been suggested that Pt binding may change the conformation of the ribose sugar, thereby poising the Pt for addition to a second base.[5] Both guanine (G) and adenine (A) bases readily occupy the two coordination sites vacated by chloride when *cis*-$Pt(NH_3)_2Cl_2$ reacts with DNA.[6] However, even the apparently simple situation where *cis*-$Pt(NH_3)_2Cl_2$ reacts with GMP has not been fully resolved.[7,8] Certainly, secondary reactions take place, and although it is widely accepted that N7 of G is the preferred binding site for Pt, subsequent or secondary reaction sites could involve N1 or O6.

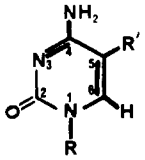
In order to maximize the chance of reaction at N1, we have carried out a series of studies utilizing N7-methylated-6-oxopurines.[9-13]



R = CH_3 7,9DMHX

R = Pr 7Me-9Pr-HX

R = D-Ribose 7MeIno



R = D-Ribose

R′ = H Cyd

R′ = CH_3 5MeCyd

The Pt complex we utilized in the present study, *cis*-Pt-(Me_2SO)$_2Cl_2$, had been investigated earlier with other nucleosides,[14] and we felt that similar chemistry might prevail. However, some unusual coordination chemistry was uncovered, in part by the use of high-field NMR spectroscopy. The nucleoside employed primarily in the study is 7-methylinosine (7-MeIno). However, in view of our findings we also examined cytidine (Cyd) and 5-methylcytidine (5-MeCyd).

Experimental Section

Instrumentation. ^{1}H NMR spectra were obtained on a Nicolet 360NB FT NMR spectrometer operating at 360 MHz and equipped with a VT unit. Conditions: 4–64 transients, quadrature detection, 16K data points, 6.1-μs (45°) pulse, 3.0-s delay, no resolution or signal enhancement. ^{13}C NMR spectra were obtained on a Varian CFT 20 operating at 20 MHz. Conditions employed were described previously.[15] A Beckman Serpass conductivity bridge, Model RCM15B1, was used for conductivity measurements.

Ligands. Nucleosides were obtained from Aldrich, Sigma, or Vega.

7-Methyl-9-propylhypoxanthine (7-Me-9-PrHX) was prepared as follows: the starting material 9-propyladenine was prepared by the method of Takemoto et al.,[16] using NaH and propyl bromide. 9-Propyladenine was converted to the corresponding hypoxanthine by $NaNO_2$ in 10% H_2SO_4. Dimethyl sulfate (1.2 mL) was added to a solution of 2.0 g of 9-propylhypoxanthine in 20 mL of *N*,*N*-dimethylacetamide. The mixture was stirred for 1 h at 120–125 °C and then cooled in an ice–salt bath. The pH was raised to 10–11 with concentrated NH_4OH. The resulting white crystals were collected by filtration, washed with acetone, and air-dried. They were then dissolved in 20 mL of H_2O containing 10 mL of concentrated NH_4OH, and the solution was stirred for several hours before the product was extracted with 5 × 30 mL portions of $CHCl_3$. The solvent was removed, and the residue was recrystallized from CH_3CN to give 7-Me-9-PrHx: 67% yield; mp 212–213 °C. Anal. Calcd for $C_9H_{12}N_4O$: C, 53.72; H, 6.51; N, 27.84. Found: C, 54.12; H, 6.30; N, 28.01.

Complexes. The complexes *cis*-$Pt(Me_2SO)_2Cl_2$ and $K[Pt(Me_2SO)Cl_3]$ were prepared by known methods.[17] Similarly, the unstable *trans*-PtL-(Me_2SO)Cl_2 (L = nucleoside) complexes were prepared by addition of 1 equiv of nucleoside to an aqueous solution of $K[Pt(Me_2SO)Cl_3]$.[14] In a typical preparation, 7-MeIno (282 mg) in 5 mL of H_2O was added to a solution of $K[Pt(Me_2SO)Cl_3]$ (419 mg in 15 mL of H_2O). After the reaction mixture was allowed to stand 10 min at room temperature, the complex was precipitated by addition of a 1:1 mixture of petroleum ether and ethanol. This material contained KCl, but it was not recrystallized to avoid isomerization to the more stable cis isomer.

Methods. All ^{1}H NMR spectra employed 0.02 M Pt. In a typical experiment, the appropriate amount of nucleoside was added to a 5-mm NMR tube and 0.60 mL of Me_2SO-d_6 was added. Me_4Si (5 μL) was added as an internal standard. The spectrum was recorded, and 1 equiv of *cis*-$Pt(Me_2SO)_2Cl_2$ was added as a solid. The reaction was monitored spectrally with time until it appeared complete, typically 24–36 h. In some cases, more nucleoside was then added to a stoichiometry of two nucleosides to one Pt and the reaction monitored again as above. In experiments designed to maximize the concentration of *cis*-[PtL-(Me_2SO)$_2$Cl]$^+$, the Me_2SO-d_6 was added to a mixture of 1 equiv each of *cis*-$Pt(Me_2SO)_2Cl_2$ and L and 0.9 equiv of $AgNO_3$. For L = Cyd or 5-MeCyd, the formation of *cis*-[$PtL_2(Me_2SO)Cl$]$^+$ was increased by addition of 0.9 equiv of $AgNO_3$ after the other two components had been dissolved in the correct stoichiometry. In the 360-MHz ^{1}H NMR spectrum of *cis*-$Pt(Me_2SO)_2Cl_2$, coordinated Me_2SO was not observed.

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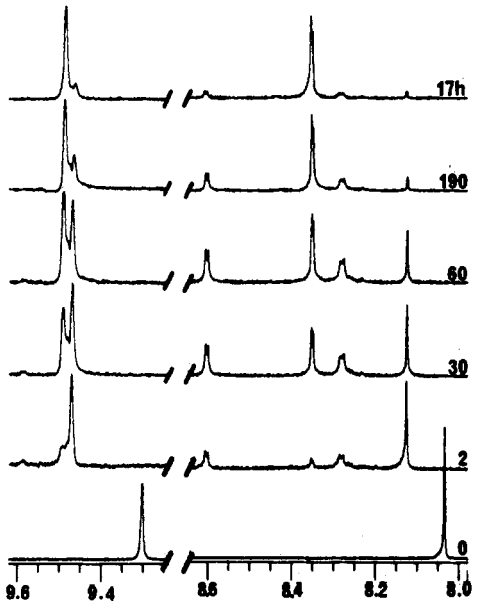


Figure 1. Observed ^1H NMR spectral changes (downfield region) on addition of *cis*-$Pt(Me_2SO)_2Cl_2$ to 7-MeIno: bottom trace, with no Pt, 0.02 M 7-MeIno; upper traces, after the addition of 0.02 M Pt. Spectra were recorded at the times indicated (min, expect top trace).

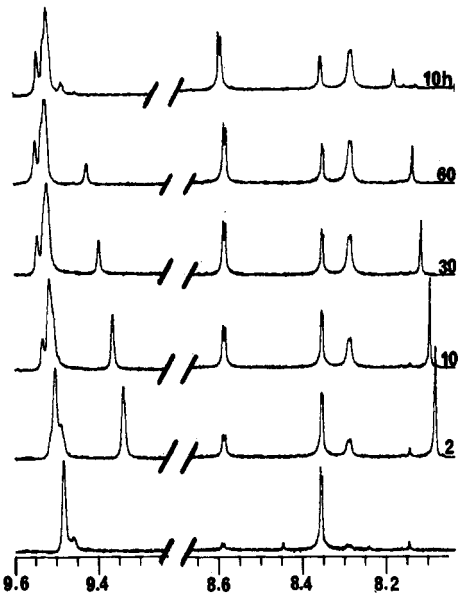


Figure 2. Observed ^1H NMR spectra changes with (downfield region) on addition of a second equivalent of 7-MeIno: bottom trace, with equilibrated 0.02 M 7-MeIno and 0.02 M *cis*-$Pt(Me_2SO)_2Cl_2$; upper traces, After the further addition of 0.02 M 7-MeIno. Spectra were recorded at the times indicated (min, except top trace).

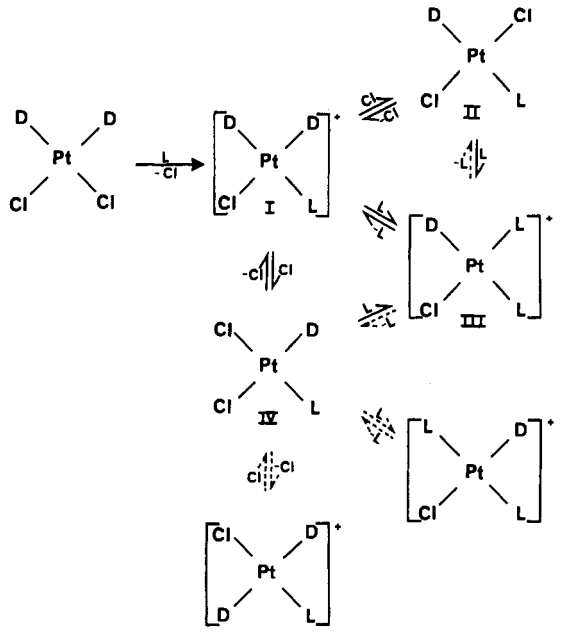
Results

7-MeIno. The spectral changes that accompany the reaction of 7-MeIno with *cis*-$Pt(Me_2SO)_2Cl_2$ are unlike any observed or reported previously for reactions of this complex. The phenomenon is quite complex, and our approach to its description will begin by a detailed description of spectral changes of the nucleoside H2 resonance. Then, before describing the changes in other parts of the spectrum, we will discuss our interpretation of the results. Otherwise, it would be difficult to follow the subsequent presentation of results.

H2 Resonance. H8 of 7-MeIno is readily exchanged for D in D_2O.[18] Addition of D_2O to 7-MeIno in Me_2SO-d_6 establishes that the upfield aromatic signal is H2. When 1 equiv of *cis*-$Pt(Me_2SO)_2Cl_2$ is added to 7-MeIno, the original H2 signal disappears before a spectrum can be recorded. Instead, seven new H2 resonances are observed (Figure 1; Table I). The most upfield resonance (8.123 ppm) is a sharp isolated signal. The other six H2 resonances appear as three pairs of singlets and are shifted further downfield from the free 7-MeIno signal than the single resonance. The signals within each pair are equal in intensity. With time, the upfield singlet decreases essentially to base line. Concomitantly, the most upfield and most downfield pairs increase and then decrease in intensity. All four signals in these two pairs have equal intensity throughout, however. The central pair increases with time to become the dominant signals when spectral changes cease. If at this juncture an additional 1 equiv of 7-MeIno is added, further reaction takes place and the predominant H2 signals are the downfield and upfield pairs (Figure 2). The Me_2SO-d_6 solutions used in the study were unbuffered, and in some cases, a slight downfield shift (~0.05 ppm) was observed for uncoordinated nucleoside. Addition of a base (triethylamine) restores the shifts to the original values.

Descriptions of the Complexes Formed. Transient Species. The upfield singlet, which appears primarily in the initial phases of the 1:1 reaction, is assigned to the compound *trans*-Pt(7-MeIno)$(Me_2SO)Cl_2$ for the following reasons. See Scheme I, which is generalized, with L = 7-MeIno in this case. First, from symmetry considerations to be described in the Discussion, all uncoupled resonances of species II (L = 7-MeIno) should be singlets.

Scheme I



Second, it is well established that the primary product of the reaction of $K[Pt(Me_2SO)Cl_3]$ in H_2O with nitrogen heterocycles is a *trans*-$PtL(Me_2SO)Cl_2$ complex.[14,17,19] This reaction was carried out with 7-MeIno, and the product precipitated quickly. The precipitate, when freshly dissolved in Me_2SO-d_6, gave the same signals as the transient species. Third, with time the trans compound converted into a species to which we assign the cis geometry and which we believe is the cis isomer (IV). The H2 pair of resonances for this cis isomer (vide infra) is downfield to that of the trans isomer. Similarly, *trans*-Pt(guanosine)-$(Me_2SO)Cl_2$ isomerizes to the cis isomer, and the H8 resonance of the cis complex is downfield to that of the trans isomer.[14] Fourth, this relationship of signals is also found for the isomers of Pt(4-picoline)$(Me_2SO)Cl_2$, and the analogous 2-picoline com-

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Table II. ^{13}C NMR Changes in Shifts (ppm) of 7-Methyl-6-oxopurines on Formation of Pt Complexes[a]

	C2	C4	C5	C6	C8
[Pt(dien)(7,9-Me_2HX)] $(NO_3)_2$[b]	1.6	−1.7	0.7	−1.8	~0.5[d]
[Pt(en)(7,9-Me_2HX)$_2$] $(NO_3)_2$[c]	2.5	−1.7	0.7	−2.1	
cis-Pt(7-MeIno)(Me_2SO)Cl_2	1.9	−1.9	−0.4	−4.8	2.0
cis-[Pt(7-MeIno)$_2$(Me_2SO)Cl]$^+$[e]	2.3	−1.8	0.4	−4.6	2.2
	1.2		−0.3	−5.0	

[a] In Me_2SO-d_6 shifts are upfield. [b] dien = diethylenetriamine. [c] en = ethylenediamine. [d] C8 resonance was broad. [e] Two resonances were not always resolved.

pounds have been studied by X-ray crystallography.[14,17]

Final Product (1:1 Reaction). We believe the final dominant product in the reaction with 1:1 stoichiometry is *cis*-Pt(7-MeIno)(Me_2SO)Cl_2 (IV) for several reasons. First, the relationship to the trans isomer is described above. Second, two closely spaced signals are expected for this compound (see Discussion). Third, conductivity measurements indicate that the product is a nonelectrolyte. Fourth, since this species is the predominant product, we were able to obtain ^{13}C NMR spectra that confirmed only one type of N1-bound nucleoside was coordinated (vide infra).

Final Product (2:1 Reaction). We assign the final product formed under these conditions to *cis*-[Pt(7-MeIno)$_2$(Me_2SO)Cl]$^+$ (III) for the following reasons. First, the ^1H NMR and ^{13}C NMR spectra establish that there are two nucleosides in different environments in the complex. Second, almost all the nucleoside has been coordinated, establishing a 2:1 stoichiometry. Third, the molar conductivity of 16.5 $cm^2\ \Omega^{-1}\ mol^{-1}$ for a Me_2SO solution is in the range of a 1:1 electrolyte (see supplementary material). Fourth, we expect two sets of closely spaced pairs of signals for this geometry (see Discussion).

^{13}C NMR Spectra of the Final Products of the 1:1 and 2:1 Reactions. We have shown that in the related molecule 7,9-dimethylhypoxanthine (7,9-Me_2HX), the N1 binds directly to Cu(II)[13] and Pt(II).[9-12] The coordination chemistry of nucleosides and of alkylated bases in which the sugar is replaced by an alkyl group appears to be very similar, as regards metal-to-base binding sites.[20] Therefore, 7-MeIno should bind via N1. To verify this expectation, we have evaluated the effect of Pt(II) coordination in the ^{13}C NMR spectrum of 7,9-Me_2HX (Table II) and compared the results with those observed for 7-MeIno in Table II. For 7,9-Me_2HX, there is an ~2 ppm downfield shift of the C2 resonance and a 1.7 ppm upfield shift of the C4 resonance. For 7-MeIno, these values are ~1.2-2.3 ppm downfield and ~1.8 ppm upfield, respectively. Similarities are also apparent in the pattern of the effects of Pt coordination on the C6 and C8 resonances of the two ligands although no pattern is evident for the C5 resonance. Since the inert Pt(II) complexes of 7,9-Me_2HX have been shown to have N1 binding in the solid, there is little doubt that the complexes formed between *cis*-Pt(Me_2SO)$_2Cl_2$ and 7-MeIno in this study involve N1 binding.

Additional Resonances. In the 1:1 reaction, four new H8 resonances are observed downfield (<0.25 ppm) from the free ligand shift. This range of shifts is less than the 0.5 ppm found for the H2 resonances and is consistent with N1–Pt binding. From the time dependence of the intensity of the shifts the most upfield H8 resonance corresponds to the *trans*-Pt(7-MeIno)(Me_2SO)Cl_2 complex. The next resonance to lower field belongs to the cis isomer and is not resolved from the resonance of one cis bis rotamer. The two downfield resonances belong to the other *cis*-[Pt(7-MeIno)$_2$(Me_2SO)Cl]$^+$ rotamer. These latter three resonances predominate at the end of the 2:1 reaction. Note that, in contrast to the H2 resonances, the resonances usually do not appear in pairs and there are five resonances corresponding to the three complexes whereas there were seven H2 resonances.

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Table III. Selected ^1H NMR Spectral Data for 7-Me-9-PrHX and Some Complexes Derived from *cis*-Pt(Me_2SO)$_2Cl_2$[a]

	base[b]		
compd	H8	H2	H7(CH_3)
7-Me-9-PrHX	9.170	8.061	4.081
trans-Pt(7-Me-9-PrHX)(Me_2SO)Cl_2	9.360	8.105	4.079
cis-Pt(7-Me-9-PrHX)(Me_2SO)$_2Cl_2$	9.375	8.327	4.099
cis-[Pt(7-Me-9-PrHX)$_2$(Me_2SO)Cl]$^+$	9.426[c]	8.589	4.061
		8.265	4.030

[a] Me_2SO-d_6, 360-MHz data, chemical shift values in ppm from internal Me_4Si. [b] Singlets. [c] Two superimposed resonances.

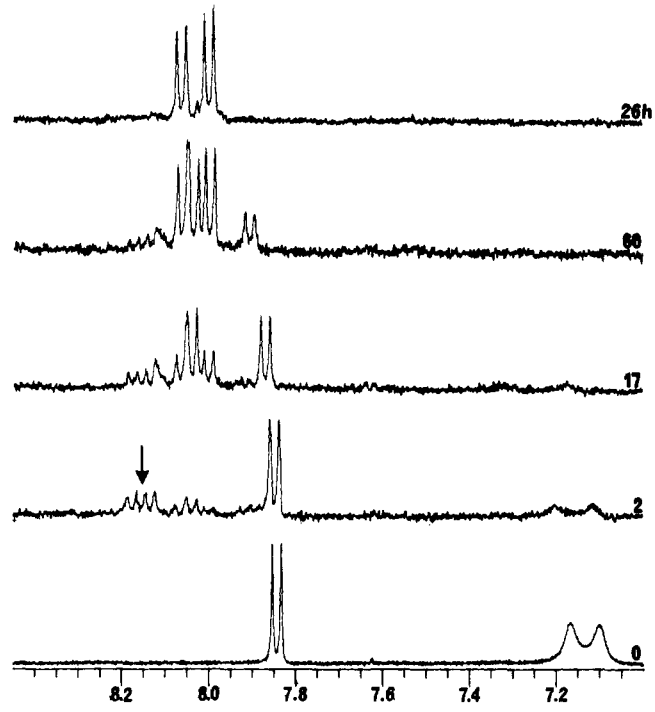

Figure 3. Observed ^1H NMR spectral changes (downfield region) on addition of *cis*-Pt(Me_2SO)$_2Cl_2$ to Cyd: bottom trace, with no Pt, 0.01 M Cyd; upper traces, after the addition of 0.02 M Pt. Spectra were recorded at the times indicated. Note that the marked signals in the 2-min spectrum are due to species I and are not the small downfield signals seen in later spectra, which arise from species III.

In the 1:1 reaction, six new N7–CH_3 resonances are found. The most downfield is assigned to *cis*-Pt(7-MeIno)(Me_2SO)Cl_2 and the next most downfield to the trans isomer. The four upfield resonances correspond to the *cis*-[Pt(7-MeIno)$_2$(Me_2SO)Cl]$^+$ species, and these predominate in the 2:1 reaction. Note in this case that only this latter species has resonances in pairs. The sugar resonances are complex because of spin–spin coupling. These will be considered in the Discussion.

7-Me-9-PrHX. Many of the same experiments described above for 7-MeIno have been conducted with 7-Me-9-PrHX, which differs from 7-MeIno in that it lacks an asymmetric sugar and has a normal propyl group at the 9-position. The same species were identified, but there were no evidence for pairs of signals (Table III).

Cytidine. The H6 resonance of Cyd is doublet split by H5. Cyd is less reactive than 7-MeIno, and consequently the reaction was carried out at a 0.5:1 Cyd:Pt ratio (Figure 3; Table IV). In contrast to the 7-MeIno reaction, at rather short time periods after mixing (~2 min), the predominant species in solution was not *trans*-Pt(Cyd)(Me_2SO)Cl_2 but a rather downfield set of two doublets (Figure 3). After 17 min, this species had essentially disappeared and the predominant complex was *trans*-Pt(Cyd)(Me_2SO)Cl_2. However, some trace amount of *cis*-[Pt(Cyd)$_2$(Me_2SO)Cl]$^+$ was now evident and an approximately equal

Table IV. Selected ^1H NMR Spectral Data for Cyd and Some Complexes Derived from *cis*-$Pt(Me_2SO)_2Cl_2$[a]

compd	base[b]				ribose[b]	
	H6	H5	$^3J_{56}$, Hz	H4(NH_2)[c]	H1′	$^3J_{1'2'}$, Hz
Cyd	7.832	5.690	7.4	7.17	5.754	3.6
				7.10		
trans-Pt(Cyd)(Me_2SO)Cl_2	8.042	5.928	7.6	8.695		
				8.115	5.757	3.3
cis-Pt(Cyd)(Me_2SO)Cl_2	8.065	5.910	7.6	8.751	5.799	3.6
				8.740		
	8.001	5.897	7.6	8.483	5.752	2.6
				8.468		
cis-[Pt(Cyd)$_2$(Me_2SO)Cl]$^+$	8.227	6.074	7.6	9.210	5.812	
	8.195	6.058	7.6	9.165		3.3
	8.147	6.041	7.6	9.128[d]	5.802	3.8
	8.125	6.003	7.6			
					5.740	2.8
					5.712	2.0
cis-[Pt(Cyd)(Me_2SO)$_2$Cl]$^+$	8.180	5.994	7.6	8.977	5.769	2.8
	8.138	6.003	7.6	8.708	5.724	2.7

[a] Me_2SO-d_6, 360-MHz data, chemical shift values in ppm from internal Me_4Si. [b] Position of the center of the doublet. [c] Singlet. [d] Two superimposed resonances.

Table V. Selected ^1H NMR Spectral Data for 5-MeCyd and Some Complexes Derived from *cis*-$Pt(Me_2SO)_2Cl_2$[a]

compd	base[b]			ribose[c]	
	H6	H5(CH_3)	H4(NH_2)	H1′	$^3J_{1'2'}$, Hz
5-MeCyd	7.686	1.825	7.289	5.758	3.6
			6.812		
trans-Pt(5-MeCyd)(Me_2SO)Cl_2	7.943	1.925	8.215	5.749	3.3
			8.161		
cis-Pt(5-MeCyd)(Me_2SO)Cl_2	7.966	1.918[d]	8.606	5.801	4.0
			8.592		
			8.306		
	7.886		8.295	5.746	1.4
			9.112		
cis-[Pt(5-MeCyd)$_2$(Me_2SO)Cl]$^+$	8.155	1.920[d]	9.019	5.840	2.9
			8.895		
	8.099		8.575	5.814	3.6
			8.742		
	8.056		8.715	5.760	3.4
	8.037			5.730	1.8
cis-[Pt(5-MeCyd)(Me_2SO)$_2$Cl]$^+$	8.099	1.935[d]	8.360[d]	5.771	3.7
	8.045		8.782[d]	5.720	2.3

[a] Me_2SO-d_6, 360-MHz data, chemical shift values in ppm from internal Me_4Si. [b] Singlets. [c] Position of center of doublet. [d] Two or more superimposed resonances.

amount of *cis*-Pt(Cyd)(Me_2SO)Cl_2. After 1 h, most of the Cyd was in this latter complex with ~25% in the trans isomer. After 3 h, the only significant species left was the cis mono compound. In contrast to 7-MeIno, free Cyd was still evident at 1 h.

Thus, it is clear that Cyd follows the same chemistry as 7-MeIno although complex formation is much less favorable and rather low for Cyd. For example, even with 2 equiv of Cyd, the bis complex is not formed appreciably. To obtain appreciable percentages of this compound, we had to add $AgNO_3$ to remove Cl.

The NH_2 resonance of Cyd in Me_2SO-d_6 is readily observable and, due to restricted rotation about the C4–N bond, appears as two signals (Table IV). These resonances shift downfield ~ 1.5–2.0 ppm on complex formation, with the greatest shifts occurring when cationic complexes are formed.

5-Methylcytidine. This nucleoside, 5-MeCyd, behaved similarly to Cyd except that the H6 resonance is no longer a doublet since H5 has been replaced with a methyl group (Table V).

Nature of the Transient Species in the Cyd Reaction. In the Cyd reaction with 2 equiv of Pt, at early stages of the reaction a species predominates that corresponds to none of the products observed in the 7-MeIno reaction. In Scheme I we describe the likely pathways for the formation of the species we have observed. It is possible that *cis*-$Pt(Me_2SO)_2Cl_2$ directly forms *trans*-Pt(L)(Me_2SO)Cl_2, but this seems unlikely. We suspected, therefore, that the first species observed in the Cyd reaction was *cis*-[Pt(L)(Me_2SO)$_2$Cl]$^+$ (I). Indeed, when $AgNO_3$ was added to a 1:1 mixture as described in Methods, this species was formed almost exclusively (Figure 4). There is no free Cyd present, and the complex must have a cis geometry because of the presence of a pair of H6 doublets.

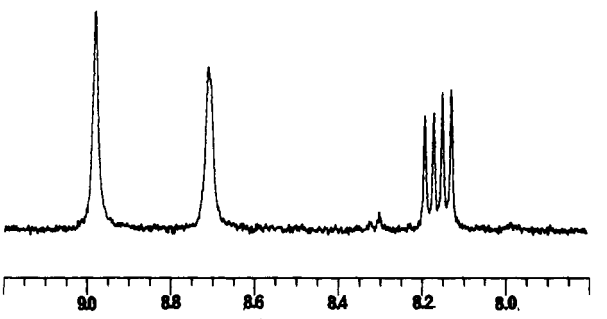

Figure 4. ^1H NMR spectrum (downfield region) of *cis*-[Pt(Cyd)(Me_2SO)$_2$Cl]$^+$ prepared as described in Methods.

Observation of a New 7-MeIno Species with $AgNO_3$. Although the corresponding early transient species, I, was not observed with 7-MeIno, we found that on adding $AgNO_3$ we were able to see an appreciable percentage of this species (Table I). On addition of Cl^- to I, the trans compound, II, was formed. On addition of 7-MeIno, the cis bis product, III, was formed.

Table VI. Number of Resonances Expected for Each Type of Observable Nucleus in Several Types of Square-Planar Pt(II) Compounds[a]

complex	rotation: fast	rotation: slow[b]
trans-[PtLDCl$_2$]	1	1
trans-[PtL*DCl$_2$]	1	1
cis-[PtLDCl$_2$]	1	1
cis-[PtL*DCl$_2$]	1	2
trans-[PtLD$_2$Cl]$^+$	1	1
trans-[PtL*D$_2$Cl]$^+$	1	1
cis-[PtLD$_2$Cl]$^+$	1	1
cis-[PtL*D$_2$Cl]$^+$	1	2
trans-[PtL$_2$DCl]$^+$	1	1 + 1
trans-[PtL*$_2$DCl]$^+$	1	2 + 2
cis-[PtL$_2$DCl]$^+$	2	2 + 2
cis-[PtL*$_2$DCl]$^+$	2	4 + 4
trans-[PtL$_2$Cl$_2$]	1	1 + 1
trans-[PtL*$_2$Cl$_2$]	1	2 + 2
cis-[PtL$_2$Cl$_2$]	1	1 + 1
cis-[PtL*$_2$Cl$_2$]	1	2 + 2

[a] L = achiral, L* = chiral. [b] For L_2 compounds the two numbers are for head-to-tail and head-to-head conformers.

Discussion

Possible species with zero or unipositive charge and one or two L were shown in Scheme I. Probable important reaction pathways are indicated by full arrows whereas broken arrows indicate pathways that have not been observed or are unlikely. Species I–IV have been observed whereas the other two reaction products, *trans*-[PtL(Me$_2$SO)$_2$Cl]$^+$ and *trans*-[PtL$_2$(Me$_2$SO)Cl]$^+$, have not been identified and may not be formed under the conditions employed.

Some of the evidence we used to assign the structures was detailed in the Results. A powerful NMR aid in structural assignment arises from the consequences of restricted rotation about the Pt–N bonds for the nucleoside complexes used in this study. Well-documented cases of restricted rotation about Pt–N(nucleoside) bonds were first presented by Cramer[21] and were later investigated in detail by Reedijk's group.[5]

In Table VI, we summarize the number of signals we anticipate for each H in the species outlined in Scheme I. First, it should be noted that the plane of the heterocyclic bases of coordinated ligands lies roughly perpendicular to the coordination plane.[4,22] For L ligands lacking two mirror planes both in and perpendicular to the heterocyclic ring, two orientations are possible with respect to the coordination plane in *cis*-L$_2$ complexes. These are the so-called head-to-head orientations where the L are related by a mirror plane (or pseudo mirror plane) or the head-to-tail orientation where the L are related by a C_2 axis (or pseudo C_2 axis). In the trans L_2 compounds, head-to-head L are related by a mirror plane or pseudo mirror plane perpendicular to the coordination plane and including the metal and the two ligating atoms not in L whereas the head-to-tail L are related by an inversion center or pseudo inversion center.

For chiral L*, the number of NMR signals expected for L*$_2$ compounds can double with respect to L_2 compounds, if rotation about the Pt–N bond is slow enough. This is also true for complexes with only one L* if both ligands cis to L* are not identical. The possibilities are summarized in Table VI. In this table, all possible mono and bis complexes of L or L* are summarized. It is clear that for an L*$_2$ compound, the only geometry that permits four resonances for each nucleus is a *cis*-[PtL*$_2$(Me$_2$SO)Cl]$^+$ arrangement in slow rotation. If a compound exists as a mixture of head-to-head and head-to-tail isomers, then one does not expect an equal intensity for all resonances.

Therefore, as mentioned in the results, the final major product in the 2:1 reaction with 7-MeIno is *cis*-[Pt(7-MeIno)$_2$(Me$_2$SO)Cl]$^+$. This is the only bis species that would give four resonances for each type of ligand proton. The corresponding complex with the achiral ligand 7-Me-9-PrHX gives only two resonances per nucleus, as expected.

The ^{13}C NMR spectrum of *cis*[Pt(7-MeIno)$_2$(Me$_2$SO)Cl]$^+$ reveals just one or two resonances for each C. In theory, we might also expect four resonances. However, the differences in environment between the rotational isomers are small, and at the field strength employed, the resonances are not resolved. Nevertheless, this finding adds support to the proposed structure since it is consistent with two nucleosides in different coordination environments as required by structure III. In addition, some of the H resonances other than H2 are not resolved into the expected number of resonances. For example, there are only three H8 resonances for *cis*-[Pt(7-MeIno)$_2$(Me$_2$SO)Cl]$^+$ where four are expected. The difference in environment is not as great as for H2, and consequently, one resonance pair is not resolved even at 360 MHz. The N7–CH$_3$ ^{1}H NMR signal is resolved.

Similar considerations apply to the other 7-MeIno complexes in Table I. Thus, we are very confident of our assignments. In addition, although we did not carry out as extensive a study, the arguments put forth for 7-MeIno also apply to Cyd and 5-MeCyd.

These arguments rest on the idea that there is restricted rotation in these compounds. To our knowledge, well-documented cases of restriction rotation about a Pt–nucleoside bond have involved bulky ligands coordinated to Pt such as *N,N,N',N'*-tetramethylethylenediamine.[5,21] There has been one report of two NMR-observable species that may be the result of restricted rotations.[8] This report involves GMP but is not convincing.

In retrospect, it is not unexpected that 6-oxopurine or 6-aminopurine nucleosides rotate rapidly about the Pt–N7 bond. We have extensively evaluated the nonbonded repulsions associated with purine and pyrimidine exocyclic groups.[23] Coordination of a metal at N3 of pyrimidines or N1 of purines will result in substituents that are ortho to the metal position. Such a substitution pattern will produce greater hindrance to rotation than a substituent further away on the nonmetalated purine ring.[24]

In the complexes studied here, the exocyclic group is ortho to the metalation site. In the cyd species, there are two ortho substituents. Indeed, warming solutions of *cis*-Pt(Cyd)(Me$_2$SO)Cl$_2$ to 80 °C did not give evidence of rapid rotation. At this temperature, the complex began to decompose. On the other hand, investigation of the temperature dependence of the ^{1}H NMR spectrum of *cis*-[Pt(7-MeIno)$_2$(Me$_2$SO)Cl]$^+$ over the temperature range 25–65 °C (above this temperature, decomposition began to occur) led to partial coalescence of the signals. Using the treatment of Dimitrov[25] for an equal-population two-site exchange process led to a lifetime, τ, of ~0.3 s at 25 °C and an activation barrier of 6 ± 2 kcal/mol for rotation about the Pt–N1 bond when the N7–CH$_3$ resonances were used. These values are necessarily approximate because of the low temperature at which decomposition sets in. However, it seems likely that rotation is more facile about the Pt–N1 bond in 7-MeIno complexes than the Pt–N3 bond in Cyd complexes and that this is a consequence of the presence of two ortho substituents in the latter.

The Ribose Sugar. The ^{1}H coupling constants in nucleosides and nucleotides can be used to assess the nature of the ring conformation. In general, two conformations predominate, C3′ENDO(N) or C2′ENDO(S).[26] These conformations typically exist in relatively similar proportions and are in rapid intercon-

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(24) Other observations on restricted rotation in nucleoside complexes are consistent with this analysis: Haring, R. K.; Martin, R. B. *Inorg. Chim. Acta* **1983**, *78*, 259. Marcelis, A. T. M.; Korte, H.-J.; Krebs, B.; Reedijk, J. *Inorg. Chem.* **1982**, *21*, 4059.
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version on the NMR time scale. Methods have been developed that allow reasonably close estimates of the nature of the ring conformation from ^1H coupling constants in aqueous solution.[26] In general, a complete analysis of the spectrum is most useful, but it has been found that $J_{1'2'}$ alone gives a good indication of ring conformation. The typically complex nature of the spectra we observed, due to the closely related rotamers, precluded a complete analysis and assignment of coupling constants. In most cases, we were able to observe $J_{1'2'}$, and since a small value indicates a preference for the N conformer, we believe that a substantial decrease in $J_{1'2'}$ from the free 7-MIno value of 4.8 Hz is indicative of an increased preference for the N conformer. For 7-MeIno complexes $J_{1'2'}$ ranges from 2.9 to 3.6 Hz, suggestive of a small increase of the percentage of N conformer on complex formation. All four H2′ resonances of *cis*-$[Pt(7\text{-MeIno})_2(Me_2SO)Cl]^+$ can be resolved. There seems to be little difference in the conformations of the rings as judged by $J_{1'2'}$ values of 3.5, 3.3, 3.6, and 3.6 Hz.

The changes in coupling constants for Cyd are much more varied and somewhat more pronounced. The values of $J_{1'2'}$ vary from 2.0 to 3.9 Hz compared to a free ligand value of 3.6 Hz. For example, for *cis*-$[Pt(Cyd)_2(Me_2SO)Cl]^+$, the $J_{1'2'}$ for the four H1′ resonances in the order of increasing upfield shift are as follows: 5.821 ppm, 3.3 Hz; 5.802 ppm, 3.9 Hz; 5.740 ppm, 2.8 Hz; 5.712 ppm, 2.0 Hz. These values suggest a rather wide range of conformational preference for the ribose moieties in the two rotamers. In one case, the conformer equilibrium seems to be shifted toward the S conformer, but the effect, if real, is small.

More dramatic differences are observed for 5-MeCyd. The $J_{1'2'}$ value for free ligand is essentially identical to Cyd's value of 3.6 Hz. For the *trans*-Pt(5-MeCyd)$(Me_2SO)Cl_2$ compound, the value decreases slightly to 3.3 Hz. For the cis isomer, one rotamer has a value of 4.0 Hz whereas the other has a value of only 1.4 Hz. Thus, for one, S is slightly more favored whereas for the other the N conformation is considerably favored. Likewise, in the *cis*-$[Pt(5\text{-MeCyd})_2(Me_2SO)Cl]^+$ compound, the four $J_{1'2'}$ values vary from 1.8 to 3.6 Hz.

The factors influencing these conformational preferences are not clear. The *trans*-$PtL(Me_2SO)Cl_2$ compounds present an environment where the cis ligand is Cl. There is a slightly greater preference for the N conformer compared to the free ligand value. For the *cis*-$PtL(Me_2SO)Cl_2$ compound, the sugar of one rotamer will be closer to Cl and that of the other rotamer closer to Me_2SO. For L = 7-MeIno, there is very little difference between the rotamers. One rotamer has a ribose ring in a conformational state similar to that of the trans isomer, as judged by $J_{1'2'}$. For L = Cyd, one $J_{1'2'}$ is lower than that for the trans compound and one is very close to the free-ligand value. Thus, it is not possible to assign which rotamer may be experiencing the largest sugar conformational change. However, it is clear that the Cyd and 5-MeCyd compounds contain sugars with a greater degree of conformational diversity, and this may result from the position of the sugar on the metalated heterocyclic ring. In comparison, the sugar of 7-MeIno is on the nonmetalated ring and is further removed from the metal.

Finally, in the light of our results, the conclusions and results in earlier studies need to be reexamined. Kong, Iyamuremye, and Rochon[14] concluded that the initial step in the reaction of nucleosides with *cis*-$Pt(Me_2SO)_2Cl_2$ was displacement of Me_2SO not Cl and therefore this complex species was not a good model for *cis*-$Pt(NH_3)_2Cl_2$ where the Cl is displaced. We do not wish to argue that *cis*-$Pt(Me_2SO)_2Cl_2$ is a good model for the antitumor agent, but our results *are* most consistent with initial Cl displacement. Second, these workers observed two closely spaced H8 resonances for inosine and xanthosine complexes. They attributed this finding to particular conformations of the Me_2SO ligand whereas the two resonances probably arise from restricted rotation. In all of these studies, no evidence for a bis species was obtained although excess nucleoside or 4-picoline was added.[14] The pK_a of 4-picoline is 6.[27] The pK_a of 7-MeIno has been reported to be slightly greater than 6.[18] The preference of 7-MeIno to form bis complexes cannot be exclusively attributed to its basicity. Although less well understood or exploited than trans effects, cis effects are known in Pt chemistry[28] and certainly cis effects may be more relevant than trans effects to difunctional adduct formation when *cis*-$Pt(NH_3)Cl_2$ reacts with DNA. However, it is premature to draw any conclusions concerning cis effects in DNA–Pt chemistry since the systems studied here are not close analogues of *cis*-$Pt(NH_3)_2Cl_2$.

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Registry No. I, 93683-35-3; II, 93779-91-0; III, 93683-40-0; IV, 93683-36-4; 7-MeIno, 20245-33-4; 7-Me-9-PrHX, 93646-08-3; Cyd, 65-46-3; 5-MeCyd, 2140-61-6; *trans*-Pt(7-Me-9-PrHX)$(Me_2SO)Cl_2$, 93683-38-6; *cis*-Pt(7-Me-9-PrHX)$(Me_2SO)Cl_2$, 93779-92-1; *cis*-$[Pt(7\text{-Me-9-PrHX})_2(Me_2SO)Cl]^+$, 93714-38-6; *trans*-Pt(Cyd)$(Me_2SO)Cl_2$, 65150-38-1; *cis*-Pt(Cyd)$(Me_2SO)Cl_2$, 93779-93-2; *cis*-$[Pt(Cyd)_2(Me_2SO)Cl]^+$, 93683-37-5; *cis*-$[Pt(Cyd)(Me_2SO)_2Cl]^+$, 93683-41-1; *trans*-Pt(5-MeCyd)$(Me_2SO)Cl_2$, 93683-39-7; *cis*-Pt(5-MeCyd)$(Me_2SO)Cl_2$, 93779-94-3; *cis*-$[Pt(5\text{-MeCyd})_2(Me_2SO)Cl]^+$, 93714-39-7; *cis*-$[Pt(5\text{-MeCyd})(Me_2SO)_2Cl]^+$, 93683-42-2; *cis*-$Pt(Me_2SO)_2Cl_2$, 22840-91-1; $K[Pt(Me_2SO)Cl_3]$, 31168-86-2; Me_2SO, 67-68-5; Cl_2, 7782-50-5; 9-propyladenine, 707-98-2; 9-propylhypoxanthine, 6972-38-9.

Supplementary Material Available: Tables of additional ribose ^1H NMR resonances for the nucleosides and their complexes, ^{13}C NMR data on 7,9-Me_2HX and 7-MeIno and some Pt complexes, and conductivity data (6 pages). Ordering information is given on any current masthead page.

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