and 21 370 cm^{-1} with continuous absorption commencing at about 21 000 cm^{-1}. The band at 21370 cm^{-1} has been attributed to an f → d Eu(II) transition. The band at 8770 cm^{-1} has been attributed to intervalence transfer between Eu(II) and Eu(III). Assuming Hush theory[14] to apply to Eu_3S_4, then the intervalence transfer band should be related to the half-width at half-height of the band ($E_{1/2}$) by

$$E_{1/2} = \sqrt{16[\ln (2kt)]E_{iT}} \quad (7)$$

For the intervalence transfer band at 8770 cm^{-1} the half-width at half-height should be 4500 cm^{-1}. It is only 3500 cm^{-1}. The measured half-width at half-height implies that the intervalence transfer band should be centered at 5300 cm^{-1} and not at 8700 cm^{-1}. Obviously, Hush theory has broken down for Eu_3S_4. However, this is expected since Eu_3S_4 is a strongly interacting, class II mixed-valence compound and Hush theory was developed for weakly interacting sites.

The thermal barrier and the intervalence transfer band are also related by Hush theory:

$$E_{th} = \tfrac{1}{4}E_{iT} \quad (8)$$

Thus, the thermal barrier obtained from the Mössbauer simulations predicts E_{iT} = 8612 cm^{-1} in excellent agreement with the experimental value. This agreement is fortuitous because we have just concluded from eq 7 that Hush theory does not apply (Eu_3S_4 is a class II compound on the basis of the half-width at half-height for the IT band. Vibronic coupling should lower the thermal barrier yielding a prediction of E_{IT} which is too low. This supports our conclusion that the Mössbauer result is incorrect. The thermal barrier obtained by TDR predicts E_{iT} = 5716 cm^{-1}, and this is the kind of miss expected when Hush theory is used on a class II system.

Finally, using Debye's development of dielectric theory[4] the permanent dipole moment of the sample may be obtained from[15]

$$M = \sqrt{(\epsilon_0 - \epsilon_\infty)3kT/4TN} \quad (9))$$

where N is the unit concentration. Assuming a density correction factor for the TDR data of 2, a value of about 7×10^{-18} esu is obtained at room temperature. The polarizability may be estimated from

$$L = \frac{M^2}{3kT} = \frac{\epsilon_0 - \epsilon_\infty}{4TN} \quad (10)$$

from which a value of 4×10^{-22} cm^3/molecule is obtained. Equations 9 and 10 are crude approximations, but they show that Eu_3S_4 has a very large dipole moment as is expected from the large distance between the charges in this extended lattice.

Conclusions

The use of time domain reflectrometry as a tool to obtain rates of intervalence transfer has been demonstrated. The TDR results have been compared to Mössbauer results obtained from the same sample and have been found to be more accurate. The breakdown of Hush theory applied to this class II sample has also been noted. This article represents the first reported use of TDR to obtain relxation times for intervalence transfer in a mixed-valence species and to determine from those times the thermal barrier for electron transfer. The viability of TDR for the extraction of accurate relaxation times is very significant because reliable values for the relaxation times of mixed-valence species are critical for the theoretical development of models to describe the process of intervalence transfer in diverse systems. Barriers will often be small and differences smaller. Clearly, TDR has the potential to play a significant role in the experimental determination of relaxation times.

Registry No. Eu_3S_4, 12345-98-1; EuS, 12020-65-4; Eu_4S_5, 82190-34-9.

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(15) Since a microscopic field is being used, Onsager's equation should be used instead of the Debye equation. However, for qualitative purposes the mathematically more simple Debye equation should be sufficient.

Syntheses, Kinetics, and Mechanism of Formation of Polynuclear Hydroxo-Bridged Complexes of (*trans*-1,2-Diaminocyclohexane)platinum(II)

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Abstract: The synthesis and ^{195}Pt chemical shifts of the hydroxo-bridged dimer and trimer, isolated from an aquated solution of bis(nitrato)(*trans*-1,2-diaminocyclohexane)platinum (I), at different pH are described. The chemical shifts of these three complexes are widely separated, and ^{195}Pt NMR provides a convenient method for the investigation of dimer formation kinetics from I. Rate constants for the dimerization reaction calculated at different pD, temperature, and concentration agree well with the hypothesis that dimerization occurs with a rate-limiting bimolecular reaction of an intermediate hydroxo species formed by the loss of a proton from the parent complex.

The discovery of cisplatin[1] as an anticancer agent marks the beginning of the upsurge of interest in the use of metal complexes in cancer chemotherapy. The drug has found wide application in the treatment of a variety of cancers.[2] The continuing interest and stimulation has been triggered and guided by two objectives: first, to develop new metal complexes having superior spectra of activities, lower toxicities, better therapeutic indices, and higher solubilities than the presently existing drugs; and second, to un-

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derstand the mechanistic features of the actions of metal complexes in vivo from the model reactions in vitro. To succeed in the former, it is imperative to have a good understanding of the latter. At present, the active antitumor species are believed to be aquated platinum(II) complexes,[3] formed by the aquation of the labile groups, that react in vitro with DNA.[4] Polynuclear complexes are known to form, and X-ray structure studies have been performed on samples isolated from aqueous solutions of bis(nitrato)diammineplatinum(II) complexes at neutral pH.[5] It also has been suggested that the formation of polynuclear "platinum blues", obtained by interaction of diaquodiammineplatinum complexes with pyrimidines, occurs in association with the formation of polynuclear species.[6]

Further, the cytotoxic and anticancer properties of bis(nitrato)(1,2-diaminocyclohexane)platinum(II) (M), bis(μ-hydroxo)-bis[(1,2-diaminocyclohexane)platinum] nitrate (D), and the *cyclo*-tris(μ-hydroxo)tris[(1,2-diaminocyclohexane)platinum] nitrate (T) are quite different from the analogous diammineplatinum complexes (unpublished results). All three complexes, in the case of the former, are active anticancer agents, and the dimer D and the trimer T are less toxic than the monomer M, whereas in the case of latter, all three are marginally active and the polynuclear complexes are much more toxic than the monomer.[7] In order to account for this contrasting behavior, and to get a better perception of reactions with DNA bases, it was of interest to study the kinetics of formation of the polynuclear complexes. The studies on *cis*-bis(nitrato)diammineplatinum[8] were undertaken by Chikuma of this laboratory. The present investigation deals with the syntheses of polynuclear complexes and ^{195}Pt FTNMR investigation of the kinetics of dimerization of bis(nitrato)(trans-1,2-diaminocyclohexane)platinum(II). The effect of changes in concentration, pH, ionic strength, and temperature on the rate of formation of the dimer was studied, and a tentative mechanism is proposed for this dimerization reaction.

Experimental Section

1,2-Diaminocyclohexane (dach) was purchased from Strem as a 70:30 isomeric mixture of *trans*- and *cis*-dach, respectively. It was separated into *trans*-dach dihydrochloride and *trans*-dach and *cis*-dach sulfate and *cis*-dach by literature methods.[9] The *trans*(dach)Pt complexes were prepared, starting from potassium tetrachloroplatinate(II) provided by Johnson Matthey, Inc., International Nickel Co., and Engelhard Corp. It was crystallized from water before use. Deuterioxide, sodium deuterioxide, and potassium deuterioxide were obtained from Merck Sharp & Dohme, Ltd., and the latter two were purified by treatment with barium carbonate.[10] The buffers were purchased from Sigma Chemical Co. and were used without further purification.

Preparation of Dichloro(*trans*-1,2-diaminocyclohexane)platinum. To a solution containing 12.45 (0.03 M) of potassium tetrachloroplatinate(II) in 150 mL of water was added 5.55 g (0.03 M) of *trans*-dach dihydrochloride. The resulting solution was heated to 75–80 °C, and a solution containing 2.64 g (0.066 M) of sodium hydroxide in 90 mL of water was added dropwise, with stirring under a N_2 atmosphere. A yellowish precipitate was obtained immediately. After all the sodium hydroxide was added, heating was stopped and the solution stirred for another 20 h. The yellow precipitate was removed by filtration and washed with 0.01 N HCl, cold water, hot water, alcohol, and ether to give a quantitative yield of the product. This was further purified by conversion to a diaquo complex by treatment with silver nitrate in water and precipitation of the dichloro complex with 1 N HCl.

Alternatively, the *trans*-(dach)Pt complex was prepared by stirring together a mixture of *trans*-dach and K_2PtCl_4 in water at room temperature.[11]

Preparation of Bis(nitrato)(*trans*-1,2-diaminocyclohexane)platinum (I). This was prepared and purified by a modification of the procedure described in the literature for bis(nitrato)diammineplatinum(II).[12]

A mixture of Pt(trans-dach)Cl_2 (11.4 g, 0.03 M) and silver nitrate (10.0 g, 0.0588 M) in 210 mL of a solution of 0.1 M sodium nitrate, acidified to pH 1.5 with nitric acid, was stirred for 24 h in a low-actinic glass flask. Silver chloride was removed by filtration, and the pale yellow solution was concentrated on a flash evaporator and allowed to crystallize. This was recrystallized from DMF/ether to give white needle-shaped crystals and finally recrystallized from water acidified with nitric acid. Anal. (Galbraith Lab.). Calcd for $H_{14}C_6N_4O_6Pt$: H, 3.26; C, 16.63; N, 12.93; Pt, 45.03. Found: H, 3.30; C, 16.74; N, 12.80; Pt, 45.14.

The deuterated analogue was prepared by dissolving the bis(nitrato) complex in D_2O and letting it sit at 5 °C for 4 weeks. It was then dried under reduced pressure.

Preparation of Bis(μ-hydroxo)bis[(*trans*-1,2-diaminocyclohexane)-platinum] Nitrate (II). Pt(*trans*-dach)$(NO_3)_2$ (I) (1.0 g) was dissolved in 46 mL of water. The pH of the solution was initially 2.5 and was raised to 5.9 with 1.5 N NaOH. Water was removed on a flash evaporator, and the resulting white product was washed with methanol and acetone and dried to give 90% yield of the product. This was further extracted with a minimum amount of water and freeze-dried. Anal. (Galbraith Lab.). Calcd for $H_{30}C_{12}N_6O_8Pt_2$: H, 3.89; C, 18.53; N, 10.80; O, 16.46; Pt, 50.16. Found: H, 4.02; C, 18.80; N, 10.61; 0, 16.60; Pt, 49.95.

The deuterated dimer of II was obtained by dissolving the deuterated monomer in water and using D_2O and NaOD instead of H_2O and KOH.

Preparation of *cyclo*-Tris(μ-hydroxo)tris[(*trans*-1,2-diaminocyclohexane)platinum] Nitrate (III). Pt(*trans*-dach)$(NO_3)_2$ (6.062 g) was dissolved in water (70 mL). The pH of the solution was initially 2.4 and was raised to 6.45 by dropwise addition of 1.5 N NaOH. The flask was stoppered and allowed to stand at room temperature for 30 min. The volume of the solution was reduced to 35 mL on a flash evaporator at 30 °C, and the solution was allowed to stand at room temperature for 6 days. During this time the trimer crystallized out of the solution (yield 4.0 g, 65%), and the pH of the filtrate was 5.85. The pH of the filtrate was raised to 6.45 again and the above procedure repeated to get 1.0 g more of the trimer. Anal. H, 3.93; C, 18.44; N, 10.65; O, 16.54; Pt, 50.37. Calcd for $H_{45}C_{18}N_9O_{12}Pt_3$: H, 3.89; C, 18.53; N, 10.83; O, 16.46; Pt, 50.16.

The deuterated trimer of III was prepared in satisfactory yields by using deuterated monomer, D_2O, and NaOD.

Preparation of Dihydroxo(*trans*-1,2-diaminocyclohexane)platinum (Ib). Pt(*trans*-dach)$(NO_3)_2$ (1.0 g) was dissolved in 30 mL of water, and the pH of the solution was raised to 10.5 by addition of 2 N sodium hydroxide. This was allowed to stand at room temperature for 2 h and then passed through an anion-exchange resin (OH^- form) to give a very pale yellow solution of the product. The volume of the solution was reduced to 10 mL, and it was allowed to crystallize at 5 °C for a few days. The precipitate of the dihydroxo complex was removed by filtration, washed with acetone, and dried.

The deuterated complex was prepared as before.

NMR Measurements. Solutions of the monomer (I) were freshly prepared in D_2O before use, and their pD was adjusted with carbonate-free NaOD just before each kinetic run. A Radiometer Model H-4 or a Corning Model 7 pH meter was used for measurement of pD, and the correction factor pD = pH + 0.4 was applied.[13] The buffer Mes (2-(*N*-morpholino)ethanesulfonic acid) was used in the pD region between 5 and 7, and Pipes (piperazine-*N*,*N*'-bis(2'-ethanesulfonic acid)) was used above pD 7. A standard kinetic run consisted of 0.05 M of the complex and 0.25 M buffer at pD 6 and at temperature of 25 °C. The ionic strength was adjusted with sodium perchlorate. The ^{195}Pt NMR spectra were obtained on a Bruker WH-180 spectrometer operating at 38.7 MHz, equipped with Fourier transform accessories. NMR tubes (20-mm diameter) and 10 mL of solution were used for each run. Spectra were obtained at different spectral widths by using 2–16K data points. For kinetic runs, optimized parameters, i.e., spectral width of 41667 Hz, pulse width of 50 μs, acquisition time of 24 ms, 2048 accumulated data points, and 17143 transients, were taken over a period of 12 min.[14] In spite of

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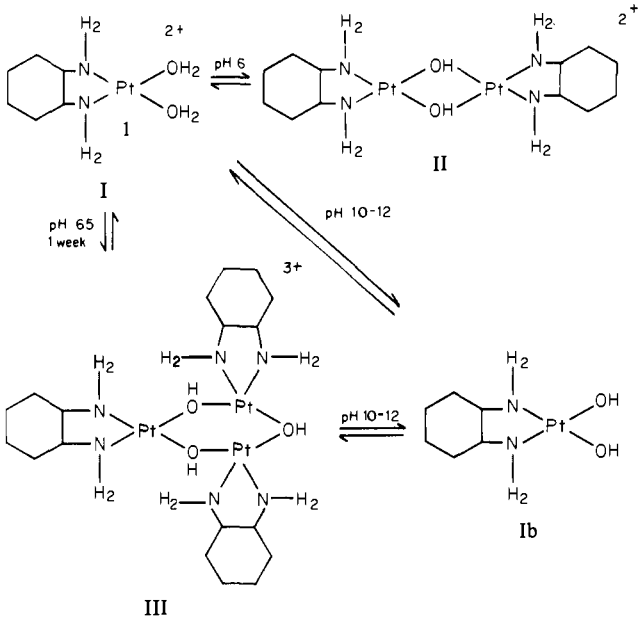
Scheme I



fast pulsing, no saturation effects were observed. Six measurements were taken for each kinetic run. The solutions were maintained at ±1 °C with the help of a WH-180 variable-temperature control. Signal intensities were integrated by a Nicolet data system in the spectrometer. The initial rate of dimer formation (first 15 min.) was obtained from the tangent to the plot of monomer concentration vs. time, and the tabulated results are generally the average of two determinations and are reproducible to ±5%. Na_2PtCl_6 (1 g in 1 mL of D_2O) was used as a reference.[15]

The trans isomer of Pt(dach)$(NO_3)_2$ was chosen for initial kinetic studies for three reasons: first, to simplify the ^{195}Pt NMR spectrum because of partial overlapping of the Pt(*trans*-dach)$(NO_3)_2$ and Pt(*cis*-dach)$(NO_3)_2$ resonances; second, to obtain a better signal to noise ratio; and third, to compare the kinetics of *trans*-dach and *cis*-dach isomers to explain the different cytotoxic properties of the two isomers. The kinetics of Pt(*cis*-dach)$(NO_3)_2$ dimerization are currently being investigated.

pK_a Measurements. Acid equilibrium constant measurements were carried out potentiometrically at 25 ±1 °C under N_2 atmosphere, with carbonate-free potassium hydroxide in 0.5 M potassium nitrate to keep the activity coefficient constant. Similar conditions were followed for the determination of the pK_a values in the case of the deuterated complex in D_2O titrated with KOD. The pK_a values were calculated according to the method given by Martin.[16]

Results

Synthesis. During the investigation of the hydrolytic behavior of Pt(*trans*-dach)$(NO_3)_2$ by ^{195}Pt NMR at different pD's, the presence of three new species was detected. These complexes are now obtained on a preparative scale whose purity is indicated by elemental analysis, and they are characterized by ^{195}Pt NMR. The structures and pH equilibria of their formation are given in Scheme I.

The dimer, II, was obtained by raising the pH of the 0.05 M solution of I to 5.9 and completing the workup in a short time. The extended reaction time and increased concentration produce some formation of the trimer, and a higher pH accelerates the formation of the trimer. At pH >7, a mixture of III and Ib is obtained along with II. The trimer was best prepared at pH 6.45 over a period of 5–7 days, when the trimer crystallizes out. At a pH >10, the dihydroxo complex, Ib, is primarily obtained. The ^{195}Pt NMR chemical shifts of these complexes in D_2O solution are given in Table I. Deuterated complexes were obtained for the interpretation of vibrational spectra, and these results, along with other spectrochemical data, will be reported separately.

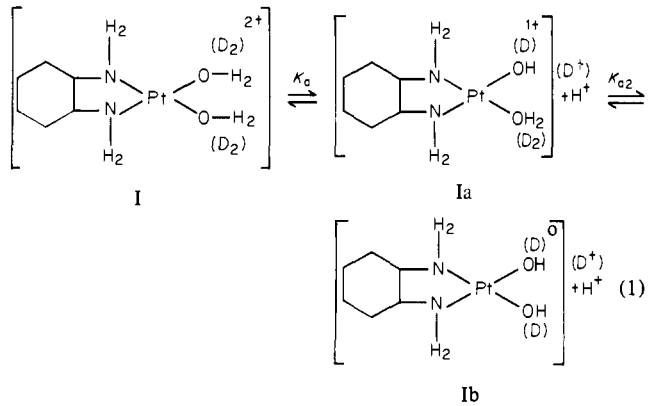
Table I. ^{195}Pt Chemical Shifts, Coupling Constants, and Line Widths for the (*trans*-1,2-Diaminocyclohexane)platinum Complexes

complex	concentration, M	pD	buffer	−δ	$J_{^{195}Pt-^{14}N}$, Hz	$\nu_{1/2}$, Hz
I	0.05–1.5	2.5–3.2	nil	1898	285	629
I	0.05	2.5–3.2	Mes	1898		629
I	0.05	5.1	Mes	1894		
I	0.05	5.6	Mes	1885		
I	0.05	6.0	Mes	1880		
I	0.05	6.1	Mes	1878		
I	0.05	6.5	Mes	1870		
I	0.05	7.0	Mes	1862		
I	0.05	7.3	Mes	1855		
I	0.05	7.45	Pipes	1847		
I	0.05	8.0	Hepes	1825		
I	0.05	8.3	Hepes	1820		
I	0.05	8.4	Pipes	1800		
I	0.05	10–12	nil	1796		
I-B	0.004	6.1–6.8	Mes	1780		
	0.004	7–8	Hepes	1769		
	0.004	8.4	Pipes	1763		
II	0.05	5.4	nil	1462	142	320
II	0.05	5–7.4	Mes	1462		320
III	0.025	7.5	nil	1757	140	264
Ib	0.05	>9	nil	1796		325

Table II. Acid-Base Equilibria for Diaquo(*trans*-1,2-diaminocyclohexane)platinum(II)

solvent	temperature	pK_{a_1}	pK_{a_2}
H_2O	25	6.14	7.56
D_2O	25	6.48	8.08

Scheme II



Acid–Base Equilibria. A study of the solute equilibria (Scheme II) was required in order to interpret kinetic data for the dimerization reactions. Dissolution of complex I in D_2O releases coordinated nitrate readily to form the dideuterated complex, whose acid–base equilibria were determined by KOD titration. Similarly, the acid-base equilibria of I in water were determined by dissolving it in water and titrating against KOH. The values of pK_{a1} and pK_{a2} are reported in Table II, and the increase in pK values of 0.38–0.48 in D_2O compared to H_2O is in agreement, within experimental error, with those reported by Glasoe for weak acids and is tentatively explained in terms of Hauggard's theory of the glass electrode.[13] From these data, the percentages of I, Ia, and Ib of the total platinum concentration at different pD's were calculated and are shown in Figure 1. It can be seen that formation of Ia is a maximum at a neutral pD.

Kinetics Studies. pD Dependence. The kinetics of formation of the dimer was followed by the disappearance of the monomer at different pD's. The chemical shifts of complexes I, II, III, and Ib are separated by large values (Table I, Figure 2). However, in an equilibrium mixture, the three monomers I, Ia, and Ib could

(14) The total acquisition time is longer than the acquisition time for one transient multiplied by the total number of transients because of processor time required to implement the PAPS sequence.

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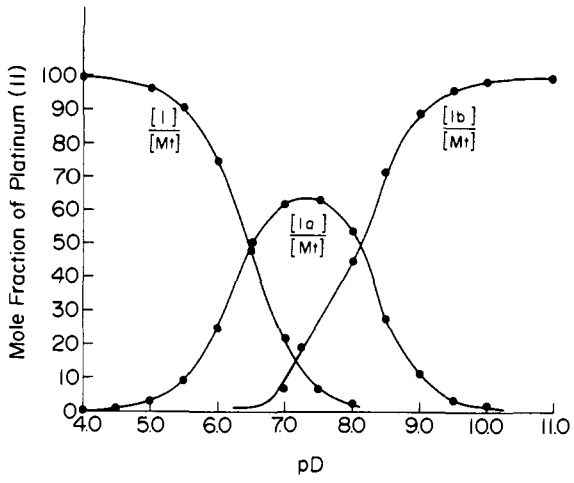

Figure 1. The equilibrium distribution of the monomeric Pt(II)–dach complexes I, Ia, and Ib as a fraction of total concentration Mt. The initial concentration of I is 0.05 M. The percentage is calculated from pK_{a_1} and pK_{a_2}.

not be seen separately due to rapid D^+ exchange among these species. Complex I (0.05 M) is stable to hydrolysis and dimerization up to a pD of 3.1 indefinitely. No formation of any other species was observed by NMR. At pD's 4 and 5.2, a conversion of 20% and 44%, respectively, to the dimer was observed over a period of 20 h. At pD's 9.5–13, only Ib was observed after a period of 0.5 h. The plot 1/[Mt] vs. time (Figure 3) gives a straight line, indicating second-order dependence on the concentration of [Mt]. The slope of the curve increases with increases in the pD. At higher pD's, a considerable scatter in the data was observed due to formation of III and Ib. From the kinetic results, the half-life time of Mt at different pD's was calculated and is shown in Figure 4, along with the percent fraction of [Ia] of the total concentration [Mt]. Again, the half-life time decreases with increases in pD and is a minimum at a pD of 7.4. Further increases in pD cause an increase in half-life time. The initial rates are given in Table IIIA.

Concentration Dependence. The concentration was varied from 0.05 to 0.15 M. The concentration range is rather limited because of solubility problems at higher concentrations and poor reproducibility or greater times required for the acquisition of spectra for kinetic runs at low concentrations. Initial rates of change in molar concentration of Mt are given in Table IIIB, and a plot of initial rates vs. the square of [Mt] gives a straight line in Figure 5, indicating second-order dependence.

Ionic Concentration. Ionic concentration was varied from 0.3 to 0.75 M by addition of sodium perchlorate. The plot of 1/[Mt] vs. time is shown in Figure 6, and initial rates are given in Table IIIC. The rate increases by 23% on addition of 0.3 M sodium perchlorate.

Temperature Dependence. The rate of dimer formation was measured at four different temperatures, and the initial rates are given in Table IIID. The rates increase with increase in temperature.

Table III. Initial Rates of Monomer Conversion and Rate Constants for Dimer Formation

(A) pD Dependent[a]

pD	initl rate × 10^4, mol min^{-1}	k_{Mt} × 10^3, mol^{-1} s^{-1}	k_{Ia} × 10^3, mol^{-1} s^{-1}
5.1	1.17	1.04	25.98
5.5	3.66	2.5	26.38
6.0	8.33	7.4	29.8
6.5	12.48	13.3	26.35
6.7	12.99	14.2	23.35
7.1	16.66	22.2	29.8
7.4	16.66	22.2	29.53
8.0	12.18	12.8	23.83
8.4	8.41	7.5	23.75

(B) Concentration Dependent[b]

concn, M	initl rate × 10^4, mol min^{-1}	k_{Mt} × 10^3, mol^{-1} s^{-1}	k_{Ia} × 10^3, mol^{-1} s^{-1}
0.05	8.33	7.4	29.81
0.07	13.0	7.8	31.42
0.10	24.98	6.66	26.83
0.15	47.34	6.66	26.83

(C) Ionic Dependent[c]

$NaClO_4$, M	initl rate × 10^4, mol min^{-1}	k_{Mt} × 10^3, mol^{-1} s^{-1}	k_{Ia} × 10^3, mol^{-1} s^{-1}
0.0	8.33	7.4	29.81
0.3	9.73	9.16	36.89
0.5	11.20	11.25	45.32
0.75	11.70	12.12	48.74

(D) Temperature Dependent[d]

temp, K	initl rate × 10^4, mol min^{-1}	k_{Mt} × 10^3, mol^{-1} s^{-1}	k_{Ia} × 10^3, mol^{-1} s^{-1}
298	8.33	7.40	29.81
303	13.17	14.52	58.49
312	20.32	34.72	139.86
317	23.73	54.95	221.35

[a] 0.05 M; temperature = 25 °C. [b] pD = 6; temperature = 25 °C. [c] pD = 6; 0.05 M; temperature = 25 °C. [d] pD = 6; 0.05 M.

Discussion

Syntheses and ^{195}Pt NMR. The common intermediate in the preparation of these polynuclear complexes is assumed to be the monohydroxo complex Ia formed in an equilibrium reaction shown in Scheme II. Under different conditions of concentration of I, pH of solution, temperature, and time, the complexes II and III are formed, presumably by the oligomerization of the intermediate Ia. At pD > 10, Ib is formed by deprotonation of Ia, pK_{a2} being 7.56. Although there are various other possible routes for the formation of these complexes, the one involving the monohydroxo intermediate is considered to be the predominant pathway. Other possibilities for the formation of dimer are discussed later. In general, the formation of trimer (III) is favored at higher concentrations of I, neutral pH, and longer reaction time. The selective preparation is based upon the solubility difference in water and controlled reaction conditions. The trimer, which is less

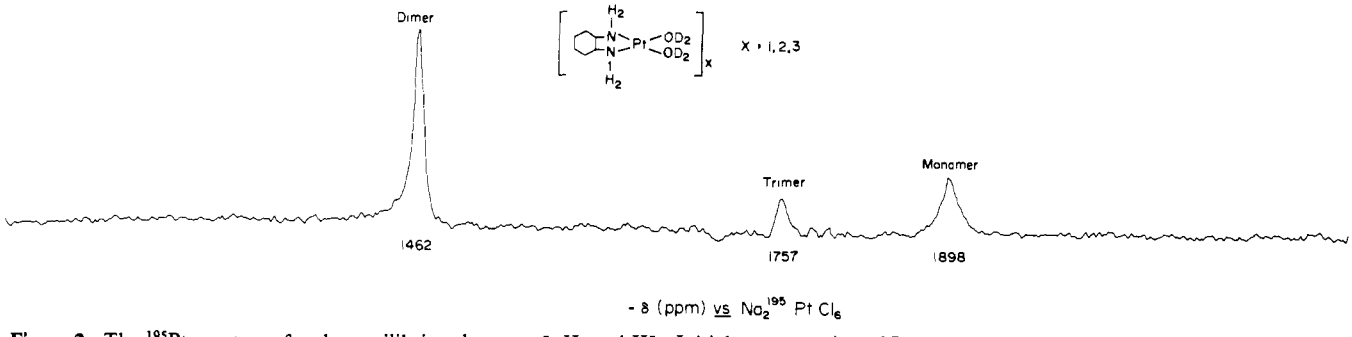

Figure 2. The ^{195}Pt spectrum for the equilibrium between I, II, and III. Initial concentration of I, 0.05 M; Mes, 0.25 M; temperature 25 °C; Data collection time 12 min. Chemical shifts are high field of Na_2PtCl_6.

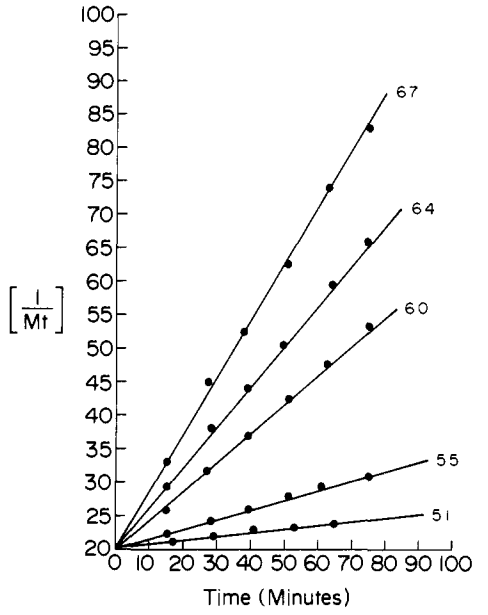

Figure 3. Plot of the reciprocal of [Mt] vs. time at different pD.

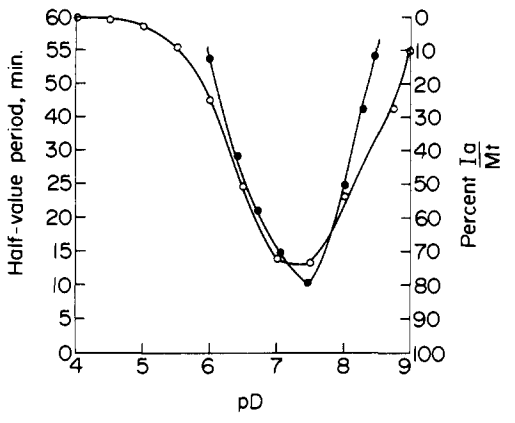

Figure 4. The effect of pD on the half-life of Mt and on the percentage of Ia/Mt (●) half-value period and (○) percent Ia/Mt.

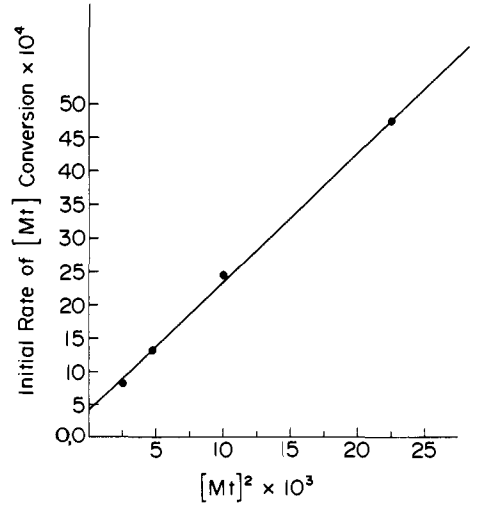

Figure 5. Plot of initial rates of change in molar concentration of Mt as function of [Mt].[2]

soluble, precipitates out of solution over a period of 5–7 days. This is in contrast to the dimer and trimer of diammineplatinum, where the dimer is less soluble than the trimer.[5] The reasons for this behavior are not clear at this time. The complexes have been assigned the dimeric and trimeric structures on the basis of their ^{195}Pt NMR spectra in conjunction with the known structures and

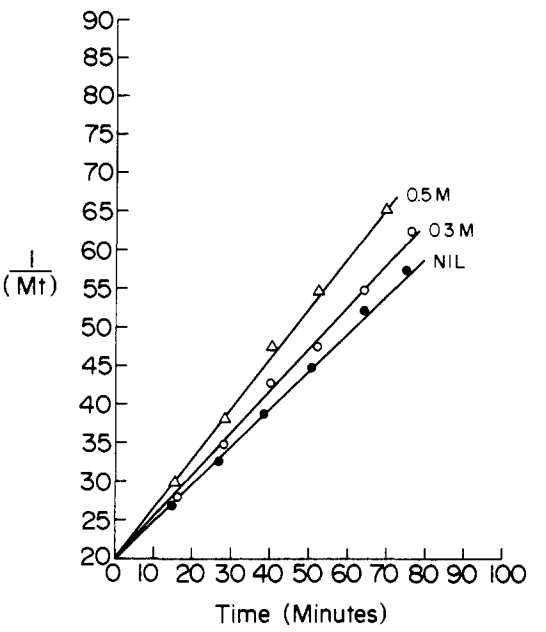

Figure 6. Plot of reciprocal of [Mt] vs. time at different ionic concentrations. Ionic concentration was adjusted with sodium perchlorate: (●) no perchlorate; (○) 0.3 M $NaClO_4$; (△) 0.5 M $NaClO_4$.

^{195}Pt NMR of the dimer and trimer of diammineplatinum[5] and a preliminary crystal structure determination of III.[17] The ^{195}Pt NMR of a mixture of I, II, and III is shown in Figure 2. Upon addition of base to I (^{195}Pt −1898 ppm) in D_2O, first the formation of II (−1462 ppm) was observed, and over an extended period or at a higher pD, another peak due to III was observed at −1757 ppm. A similar trend in chemical shifts for the dimer and trimer of the diammineplatinum was observed.[5e] These values are in agreement with the expected dominance of σ_p in determining the chemical shift.

The main contribution to the screening constant of a heavy metal nucleus such as platinum is comprised of two terms ($\sigma = \sigma_p + \sigma_d$), where σ_p and σ_d are the paramagnetic and diamagnetic contributions to the shielding, respectively.[18a] Of the two, σ_p is the dominant contribution and is defined by Ramsay[18b] and evaluated for ^{195}Pt by Dean and Green:[18c]

$$\sigma_p = \frac{-e^2h^2}{2m^2c^2}\langle r^{-3}\rangle C_{a_{1g}}{}^2\left[8C_{a_{2g}}{}^2\Delta E_A{}^{-1} + 4C_{e_g}{}^2\Delta E_E{}^{-1}\right] \quad (2)$$

where $\langle r^{-3}\rangle$ is an average over the radial 5^d functions used as a basis set; C_{eg}, $C_{a_{1g}}$, and $C_{a_{2g}}$ are the coefficients of the corresponding d orbitals in the molecular orbitals of platinum; and ΔE terms are differences in energies between highest occupied and lowest unoccupied orbitals accessible for one-electron transitions.[18] The ^{195}Pt chemical shift for monomers I and Ib differs considerably ($\Delta(\delta I - \delta Ib) = 102$ ppm). The high-field chemical shift of I may be explained in terms of greater covalency of the Pt–OD_2 bond compared to the Pt–OD bond, D_2O being more polarizable than OD^-. Increase in covalency may result from a decrease in $C_{a_{1g}}{}^2$ and, hence, increase in the δPt. The chemical shift of the intermediate complex, Ia, could not be determined because it cannot be isolated. In solution it exists in equilibrium with I or Ib or with both, depending upon the pD of the solution. Thus, due to fast D^+ exchange between three monomeric species I, Ia, and Ib, only an average value of the chemical shift was obtained, which increased monotonically with an increase in pD of the solution because of the varying proportions of these species at different pD. Qualitatively similar changes in chemical shifts were observed in the case of the $Pt(NH_2–CH_2–CH_2–NH_2)(D_2O)_2{}^{2+}$ complex

(17) Personal communication from Dr. C. J. L. Lock.

(18) (a) T. G. Appleton, H. C. Clark, and L. E. Manzer, *Coord. Chem. Rev.*, **10**, 335 (1973); (b) N. F. Ramsey, *Phys. Rev.*, **78**, 699 (1950); (c) R. R. Dean and J. C. Green, *J. Chem. Soc.* A, 3047 (1968); (d) J. J. Pesek and W. R. Mason, *J. Magn. Reson.*, **25**, 519 (1977).

at different pD's.[19] Covalency may be invoked for the downfield chemical shift of II and III as the bridged OD^- ligands are liable to be less covalently bonded in relation to terminal OD^- and D_2O ligands in I and Ib. However, the very large downfield shift of II is surprising, and no satisfactory explanation can be given at this time. Similar trends in chemical shifts of dimers and trimers were observed in ethylenediamineplatinum[19] and diammineplatinum complexes;[5,20] in the latter case, the downfield shift of the dimer has been explained in terms of ring strain at the metal center.[20] The bridged monohydroxo complex, which may be an intermediate in the formation of II, was not observed, presumably because of fast conversion to II on the NMR time scale. The line width $\nu_{1/2}$ of I at different pD is large, broadened by the scalar coupling to the quadrupole nuclei N and O and also by exchange processes. However, the line widths of II, III, and Ib are relatively narrower; the number of quadrupole nuclei per platinum atom is less in II and III, and there is no exchange among these species. The coupling constants ($^1J_{Pt-^{14}N}$) (obtained under best instrumental conditions and optimum parameters) of II and III are about half the magnitude of I. The change in coupling constants is related to trans effect and trans influence.[18a,21] Since the trans effects of D_2O and OD^- ligands are comparable,[22] it is the number of ligands per platinum that govern the coupling constants. In I, there are two D_2O ligands per platinum, whereas in II and III there is only one OD^- ligand per platinum atom.

Kinetics and Mechanism. A number of possible pathways may be envisaged for the formation of II from I. However, on the basis of the second-order dependence on [Mt], the possibilities shown in eq 3–5 are considered most pertinent.

$$2\mathrm{I} \underset{k_{-1}}{\overset{k_1}{\rightleftharpoons}} \mathrm{II} + 2D_2O + 2D^+ \tag{3}$$

$$2\mathrm{Ib} \underset{k_{-2}}{\overset{k_2}{\rightleftharpoons}} \mathrm{II} + 2OD^- \tag{4}$$

$$2\mathrm{Ia} \underset{k_{-3}}{\overset{k_3}{\rightleftharpoons}} \mathrm{II} + 2D_2O \tag{5}$$

In addition, there is an initial formation of a buffer complex (I–B) and the formation of the trimer, III, at higher pD. A number of assumptions are made to try for some understanding of the mechanism of the formation of the dimer: (1) the concentrations of the buffer complex and the trimer are negligible compared to the concentrations of monomer and dimer; (2) the rates for maintaining equilibria between three monomeric aquated species of eq 1 are rapid compared to the rate of dimerization; and (3) the rate of dimer breakup is slow compared to the rate of formation. These are not unreasonable assumptions, because initial rates were used for calculation of the rate data; therefore, reverse reactions can be neglected. The stability of a 0.05 M solution of II in D_2O was monitored over a period of 2 h, and the decomposition to I was less than 5%. However, a 0.05 M solution of II in 0.05 N $HClO_4$ decomposed completely to Pt(*trans*-dach)$(OD_2)_2^{2+}$ over a period of 6 h. The initial formation of I–B and the trimer at pD 7 is <5% and is neglected for the calculation of the rate data. Of the three possibilities for the formation of II, eq 3 is ignored, because at a pD where I is predominant, no formation of II was observed over a period of 1 month; eq 4 is unimportant because kinetics were run at pD's where the formation of Ib is negligible. Moreover, the formation of II from Ib was not observed over a period of 2 h. Thus, the formation of II is considered to take place from Ia, and reaction 5 is considered to be the predominant pathway for the formation of II. The overall reaction kinetics can be expressed as

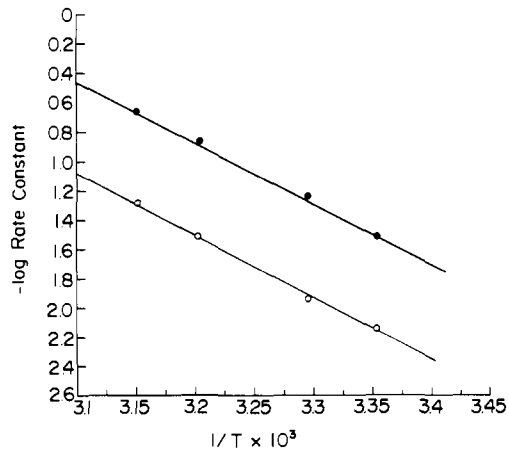

Figure 7. Plot of negative logarithm of rate constants k_{M_1} and k_{1a} vs. the reciprocal of the temperature. (●) $-\log k_{M_1}$; (○) $-\log k_{1a}$

$$\frac{d[Mt]}{dt} = \frac{-2d[II]}{dt} \tag{6}$$

or

$$\frac{d[II]}{dt} = k_{M_1}[Mt]^2 \tag{7}$$

There is a linear relationship between $1/[Mt]$ and time (Figure 3), indicating a second-order dependence on [Mt]. This is further supported by the data of Figure 5. However, the rate equation can be best expressed in terms of Ia, as discussed before, and under the conditions of the kinetic runs, Ia reaches its maximum value (Figure 1).

Mt is related to Ia, by eq 8

$$[Mt] = [Ia]\left[1 + \frac{[D^+]}{K_{a1}} + \frac{K_{a2}}{[D^+]}\right] \tag{8}$$

or

$$\frac{1}{[Ia]} = k_{1a}t + \frac{1}{[I_0]} \tag{9}$$

or

$$k_{1a} = k_{M_1}\left(1 + \frac{[D^+]}{K_{a1}} + \frac{K_{a2}}{[D^+]}\right) \tag{10}$$

The values of rate constants k_{1a} (26.58×10^{-3} M^{-1} s^{-1} ± 12%) are more consistent than k_{M_1} (11.62×10^{-3} M^{-1} S^{-1} ± 91%) (Table IIIA–C). It is, therefore, justifiable to consider the formation of the dimer as a second-order reaction with respect to Ia, which is in agreement with the suggestion of Martin[4] for dimerization of Pt(en)$(OH_2)_2^{2+}$. The small variations in the values of k_{1a} at different pD's may be attributed to assumption 1 in the calculation of rate data and ±5% uncertainty in the kinetic runs. The activation energy of dimerization was calculated from the plot of $-\log k_{1a}$ vs. reciprocal temperature (Figure 7), and a value of 82 kJ mol^{-1} was obtained. In conformity with earlier studies for the formation of binuclear bridged complexes,[8,23] a concerted mechanism is tentatively proposed for the dimerization reaction.

Biologically, the formation and anticancer activities of the polynuclear (dach)Pt complexes are interesting. The formation of these complexes is favored in the pH range close to the biological pH. Cleare and Hoeschele have correlated the antitumor activity of platinum complexes to charge neutrality and ligands of moderate lability. The charged complexes or complexes having ligands with fast leaving rates were shown to be inactive and toxic.[24]

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(22) F. A. Cotton and G. Wilkinson, "Advanced Inorganic Chemistry", Interscience, New York, 1972, p 668.
(23) (a) J. Chatt and L. M. Venanzi, *J. Chem. Soc.*, 3858 (1955); (b) R. G. Pearson and M. M. Muir, *J. Am. Chem. Soc.*, **88**, 2163 (1966); (c) L. I. Elding and L. F. Olson, *Inorg. Chem.*, **16**, 2789 (1977).
(24) M. J. Cleare and J. D. Hoeschele, *Bioinorg. Chem.*, **2**, 187 (1973).

However, later reports by Schwartz, confirmed in our laboratory, have shown that complex I, which is charged and has fast leaving groups, is very active.[25] The anomolous activity of this complex may be attributed to the formation in vivo of II and III, which have superior activities and are less toxic than I. The hydroxo-bridged complexes have been shown to be substitutionally inert[4] and may not be easily sequestered to an ineffective and possibly toxic form in extracellular reactions. However, in intracellular reactions, the polynuclear complexes may undergo a slow equilibrium to the monomeric form. This may be promoted by ease of reaction of the monomer with DNA bases. This may provide the basis for a slow conversion of the polynuclear complexes to a monomeric aquated form that is responsible for the anticancer activity in intracellular reactions.[5e] Hence, it is tantamount to a slow release of the drug in its active form within the cell. Slow infusion or divided doses in the case of cisplatin produce less toxicity to the patient than the same dose given as a rapid push.[5e]

(25) P. Schwartz, S. J. Meischen, G. R. Gale, L. M. Atkins, A. B. Smith, and E. M. Walker, Jr., *Cancer Treat. Rep.*, **61**, 1519 (1977).

Acknowledgment. This work was supported in part by fellowship funds from Engelhard Corp. and International Nickel Co.

Registry No. I, 81473-15-6; Ib, 82373-56-6; II, 82398-34-3; III, 82338-62-3; Pt(*trans*-dach)Cl_2, 38780-40-4; K_2PtCl_4, 10025-99-7.

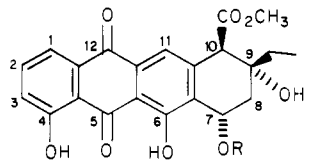
An Efficient Total Synthesis of (±)-Aklavinone[1]

Robert K. Boeckman, Jr.,* and F.-W. Sum

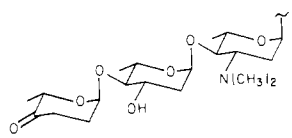
Contribution from the Department of Chemistry, University of Rochester, Rochester, New York 14627. *Received December 4, 1981*

Abstract: An 11-step total synthesis of (±)-aklavinone from 1,3-cyclohexadione is reported. The key steps include a Diels–Alder condensation of **3** and **4** and the stereoselective aldol condensation of **21** to **22**. The overall yield is ~13%.

Among the most promising of the new generation of anthracyclinone antitumor agents currently under clinical evaluation is aclacinomycin A (**1**).[2] Toxicity problems associated with



1, R =

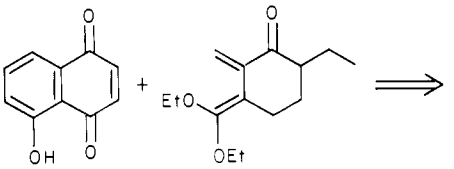


2, R = H

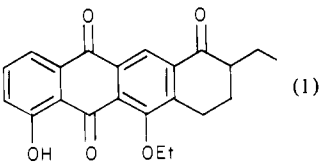
adriamycin appear to be reduced in the case of **1**.[2] Since microbiological methods permit transformation of the aglycon, (+)-aklavinone (**2**), to **1**,[3] there has been a significant effort directed torward the development of synthetic routes to **2**. Recently these efforts have culminated in four total syntheses of **2**.[4-6]

We also have been engaged in studies directed toward synthesis of **2** and report the results of our efforts, which have resulted in a short efficient total synthesis of **2**.

Our strategy for construction of the tetracyclic nucleus of **2** is based upon the combination of the two major synthons **3** and **4** by means of a Diels–Alder cycloaddition, as shown in eq 1.[7,8]



3 4



(1)

Preparation of cyclobutene **5**, the anticipated precursor of **4**, was initiated by a one-pot successive alkylation and acylation of the dianion of 1,3-cyclohexanedione, as indicated in Scheme I. The resulting mixture of enol pivalates is readily separated by preparative chromatography (20-g scale) and the minor isomer recycled to afford 72% of **6** after one recycle. Photolysis of **6** (1 equiv) and ketene diethyl acetal (6 equiv)[9] in ether at room

(1) Preliminary stages of this investigation were carried out at Wayne State University, Detroit, MI.

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