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Kinetics and Mechanisms of Platinum(II)-Promoted Hydrolysis of Inorganic Polyphosphates

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The hydrolysis reactions of tri- and pyrophosphate ions are accelerated in the presence of *cis*-diaquodiammineplatinum(II) and its corresponding conjugate bases in aqueous perchlorate and nitrate media in the pH range 1–8. The kinetics of hydrolysis of triphosphate in the presence of *cis*-diaquodiammineplatinum(II) exhibit second-order growth of an intermediate, which decays to hydrolyzed products by a first-order process. The rates of both formation and decay of the intermediate increase with increasing pH. The pH-corrected limiting rate constants for the formation and decay of the intermediate were computed to be 0.1 M^{-1} s^{-1} and 1.0×10^{-3} s^{-1}, respectively, at 40 °C in 0.5 M $NaClO_4$. The intermediate has been identified as a triphosphato-bridged diplatinum species on the basis of phosphorus-31 NMR spectra in solution. The hydrolyzed products have been characterized as various ortho- and pyrophosphato complexes of platinum(II) and mixed-valence Pt(II)–Pt(III) species. The hydrolysis of pyrophosphate ion in the presence of *cis*-diaquodiammineplatinum(II) also proceeds through the formation of a pyrophosphato-bridged diplatinum complex to yield orthophosphate ion bound to platinum. The estimated rate constant for the hydrolysis of the intermediate at 55 °C and pH 4.0 is 5×10^{-4} s^{-1}. At pH 8 a pyrophosphato chelate, (dihydrogen pyrophosphato)diammineplatinum(II), was isolated from the reaction between *cis*-dichlorodiammineplatinum(II) and pyrophosphate ion. From the phosphorus-31 NMR spectrum of the complex the two-bond coupling constant, $^2J_{P\text{-}O\text{-}^{195}Pt}$, was found to be 15 Hz. The aquation reaction of this pyrophosphato chelate proceeds through an acid-catalyzed process, and the limiting rate constant at lower pH for the aquation reaction was determined to be 9.0×10^{-6} s^{-1} at 25 °C in 0.1 M $NaClO_4$.

Introduction

Enzymes, in the presence of certain metal ions, catalyze phosphate hydrolysis in biological systems.[2] In this laboratory, the kinetics and mechanisms of phosphate hydrolysis of cobalt(III) complexes of inorganic polyphosphates and nucleotides catalyzed by various amine and macrocyclic complexes of cobalt(III) have been investigated.[3-6] On the basis of the observed pH–rate profile, it has been concluded that a coordinated hydroxide ion acts as the nucleophile for the hydrolysis of the phosphate ligands.[4-6] Similar conclusions have been reached by other workers.[7,8] The hydrolysis of phosphate esters coordinated to cobalt(III) has also been interpreted as being due to the intramolecular base catalysis by the coordinated hydroxide ion.[9] Although cobalt(III) is not a naturally occurring metal ion in biological systems, most of the model systems that have been studied so far have involved cobalt(III) complexes with a variety of phosphate ligands. Cobalt(III) has been the metal ion chosen for these studies because its phosphato complexes are well characterized,[10-12] its solution chemistry is well documented, and its inertness to substitution offers advantages for characterizing the various products of the hydrolysis reaction. Moreover, since all the phosphato complexes of cobalt(III) are low-spin and diamagnetic, phosphorus-31 NMR spectra can be used to characterize the various phosphato products in solution. Other metal ions having octahedral coordination geometry have also been reported to catalyze phosphate hydrolysis.[8,12]

This study extends our investigation of phosphate hydrolysis reactions catalyzed by metal ions to include platinum(II) with its square-planar coordination geometry. Although platinum(II) is also not known to occur naturally in biological systems, its anticancer activity has created great interest in its reaction with biological molecules. The substitution chemistry of platinum(II) is well understood,[14] and the diamagnetism of platinum(II) offers the same advantages for characterization as in the case of cobalt(III) systems. In spite of the interest in the biological activity of platinum complexes and the omnipresence of phosphates in biological systems, few phosphato complexes of platinum have been reported. Stanko[15] reported the X-ray crystal structure of a pyrophosphato complex, $Pt_2(NH_3)_4(P_2O_7)$, in which the pyrophosphate ion acts as a doubly bridging ligand between the two platinum(II) ions. Louie and Bau[16] presented the crystal structure of the dinuclear product $[Pt(en)(5'\text{-CMP})]_2$, where en and 5'-CMP represent ethylenediamine and 5'-cytidine monophosphate. The complex is dinuclear, with each 5'-CMP ligand bridging between the two platinum(II) centers. A given platinum atom is coordinated to the phosphate group of one 5'-CMP molecule and N3 of the other. Phosphato-bridged metal–metal-bonded diplatinum(III) complexes have been reported by Cotton and coworkers,[17] and Appleton et al.[18] have reported the phosphorus-31 and platinum-195 NMR spectra of these phosphato-bridged di-

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platinum complexes. Complexes proposed to be mixed-valence phosphato-bridged platinum species have also been described recently.[19] A few orthophosphato platinum(IV) complexes are described in the literature.[20] Recently we have reported the kinetics of formation of a variety of phosphato complexes of platinum(II) and their phosphorus-31 NMR characterization in solution.[21] Here we describe the synthesis, aquation, and phosphate hydrolysis of platinum(II) complexes of inorganic polyphosphates. We have presented a preliminary communication on the hydrolysis of triphosphate ion promoted by platinum(II).[22]

Experimental Section

Reagents. The platinum substrate, *cis*-dichlorodiammineplatinum(II), was prepared from hexachloroplatinate(IV) by following a literature procedure.[23] The corresponding diaquo species *cis*-diaquodiammineplatinum(II) nitrate was synthesized by the procedure of Mann.[24] In many cases, *cis*-$[Pt(NH_3)_2(H_2O)_2]^{2+}$ was prepared in situ by adding a stoichiometric amount of $AgNO_3$ while the acidity was maintained at 0.01 M by HNO_3 in order to avoid formation of the hydroxo-bridged dimer and higher oligomers.[25] Potassium triphosphate (Baker) was used without further purification. All other reagents were of analytical reagent grade and were used without further purification. Elemental analyses were performed by Galbraith Laboratories, Knoxville, TN. In some instances, platinum was analyzed gravimetrically.[26] Phosphate analyses were performed spectrophotometrically as phosphomolybdate following the procedure of Chen et. al.[27] modified to include vanadium(II) reduction for analysis of coordinated phosphate.[4]

Physical Measurements. The proton-decoupled phosphorus-31 NMR spectra were recorded on a JEOL FX-90Q spectrometer operating at 36.3 MHz, and the chemical shifts are reported with respect to an external reference of 85% phosphoric acid. Typical experimental conditions were as follows: spectral width, 2000 Hz; data points, 16K; pulse width, 10 μs (45°); repetition time, 2.0 s. UV–visible spectra were recorded on a Cary 219 spectrophotometer and infrared spectra on a Perkin-Elmer 1330 spectrophotometer. Potentiometric titrations were performed with an Orion Model 701 pH meter using a motor-driven syringe interfaced with an Apple II Plus computer.[28] The pK values were evaluated by using a literature method.[29]

Synthesis of (Dihydrogen pyrophosphato)diammineplatinum(II). A sample of *cis*-dichlorodiammineplatinum(II) (0.1 g) was dissolved in water (250 mL), and tetrasodium pyrophosphate decahydrate (0.4 g) was added. The pH of the solution was adjusted to 8.0 by the addition of 0.1 M HNO_3. The solution was incubated at 40 °C for 15 h and passed through an anion-exchange column (60 × 0.5 cm; Bio-Rad AG-1 X2, 100–200 mesh) pretreated with 0.1 M $NaNO_3$. The unreacted platinum complex passed directly through the column, leaving behind a brown band 1-cm wide. The column was washed with 1 mM HNO_3, which moved a faint yellow component from the brown band, leaving a small brownish black band on the column. The pale yellow eluate was adjusted to pH 1.0 with 0.1 M HNO_3 and cooled in an ice bath. A faint yellow precipitate, which formed immediately, was filtered and washed with ethanol (3 × 5 mL). Yield: 0.09 g (55%). Anal. Calcd for $Pt(NH_3)_2H_2P_2O_7 \cdot H_2O$: N, 6.61; H, 2.36; P, 14.64; Pt, 46.11; PO_4^{3-}, 44.4. Found: N, 6.62; H, 2.32; P, 14.47; Pt, 46.32; PO_4^{3-}, 43.8. IR (KBr pellet): 3550 sh, 3280 br, 1580 br, 1355 sp, 1250 br, 1180 br, 1090 sp, 855 br, 728 sp, 715 sp, 585 sp, 555 sp, 495 sp, m, 430 sp, s, 295 sp, m, 210 w cm^{-1}. Attempts to move the brownish black band from the column with solutions of HNO_3 as concentrated as 1.0 M were unsuccessful, giving a brownish black precipitate within the column.

Reaction of Triphosphate Ion with *cis*-Diaquodiammineplatinum(II) Nitrate. A solution of *cis*-$[Pt(NH_3)_2(H_2O)_2]^{2+}$ (5.0 mM) and a series of solutions of eight different triphosphate concentrations in the range 1.0–50.0 mM were prepared at 0.5 M ionic strength ($NaClO_4$) and pH 3.5. Equal volumes of the platinum solution and each of the triphosphate solutions were mixed and incubated at 40 °C for 24 h. The solution turned blue during the reaction. The final pH of each of the blue solutions depended upon the extent of buffering by triphosphate ion. The pH of the sample solution containing the lowest triphosphate concentration (0.5 mM) decreased to 1.2 from its initial value of 3.5. The pH of the solution containing 25.0 mM triphosphate, on the other hand, was lowered only to 3.3. The pH values of the other solutions with intermediate triphosphate concentrations were in the range 1.5–3.2. It was generally observed that when the concentration of triphosphate ion was 10-fold or more in excess over the platinum concentration, as in the kinetic experiments, then the pH of the reaction mixture changed by 0.2 or less.

The phosphate content of each of the sample solutions was analyzed spectrophotometrically.[4,27] A blank experiment using the same triphosphate concentration without the platinum substrate was also carried out. The amount of orthophosphate obtained in the blank experiment was subtracted from the amount found in the sample containing the platinum complex. The ratio of the concentration of orthophosphate released to the initial concentration of platinum substrate used was found to be 0.55 ± 0.05 at pH 3.5. At pH 5.0 this stoichiometric ratio was found to be 0.3 ± 0.1. Formation of the blue color was also observed at this pH. A yellow precipitate was obtained at pH 8.0, and no blue color developed at this pH. The yellow precipitate was filtered, washed with ethanol (3 × 5 mL), and dried in vacuo over phosphorus pentoxide. Anal. Found: Pt, 62.0 ± 1.0; PO_4^{3-}, 29.3 ± 0.2; ratio on formula basis, 1.03 ± 0.02. The blue complex was isolated after 24 h of reaction at pH 4.0 from a solution that was 0.05 M in each reactant. The addition of 25 mL of 1:1 ethanol:diethyl ether to 50 mL of the solution produced a blue oily substance, which was dissolved in 1 mM HNO_3 (≈1 mL) and slowly dried in a desiccator over silica gel.

Reaction of Pyrophosphate with *cis*-Diaquodiammineplatinum(II). The reaction of pyrophosphate ion with *cis*-$[Pt(NH_3)_2(H_2O)_2]^{2+}$ was carried out under conditions similar to those outlined in the case of the triphosphate reaction. At pH 3.0 the reaction mixture turned blue after 3 h at 40 °C, and a yellow precipitate formed after 6 h. The yellow precipitate formed at an earlier time at lower pH. For example, at pH 1.5, with 1.5 mM platinum complex and 16.0 mM pyrophosphate, the yellow precipitate was observed within 2 h after the reaction was initiated. At pH 1.0 the solution developed a greenish blue color instead of blue as observed at pH 3.0 and 4.0. The blue color was not observed if the reaction was performed at pH 6 or higher. The yellow precipitate from the reaction at pH 3.0 was filtered, washed with ethanol (3 × 2 mL), and dried in vacuo over phosphorus pentoxide. Anal. Found: Pt, 59.0 ± 1.0; PO_4^{3-} 29.8 ± 0.3; ratio on formula basis, 0.96 ± 0.02.

Reaction of (Dihydrogen pyrophosphato)diammineplatinum(II) with *cis*-Diaquodiammineplatinum(II). A solution of (dihydrogen pyrophosphato)diammineplatinum(II) (2.0 mM) was prepared in 0.5 M $NaClO_4$, and the pH was adjusted to 6.0 with 0.5 M NaOH. A solution of *cis*-$[Pt(NH_3)_2(H_2O)_2]^{2+}$ having the same ionic strength and pH as the pyrophosphato complex solution was added such that the concentration of each of the complexes was 1.3 mM. The pH of the mixture dropped to 4.1 within 10 min. A yellow precipitate separated out slowly. No further precipitation was observed after 12 h at 25 °C. The yellow precipitate was filtered, washed, and dried as mentioned earlier. Anal. Found: Pt, 61.5 ± 0.5; PO_4^{3-}, 30.6 ± 0.3; ratio on formula basis, 0.98 ± 0.01.

Kinetic Measurements. The kinetics of aquation of (dihydrogen pyrophosphato)diammineplatinum(II) were followed spectrophotometrically at 252 nm in the pH range 1.0–3.5 in 0.1 M $NaClO_4$. The sample was kept in a thermostated bath (±0.2 °C), and the absorbance was recorded at various time intervals until no further change in absorbance was observed. The rate constants were evaluated from the slopes of the plots of ln $(A - A_\infty)$ vs. time, which were linear for more than 4 half-lives. A few kinetic measurements were carried out at constant pH values with a pH stat. For these measurements, solutions of the pyrophosphato complex (1.0 mM) in 0.1 M $NaClO_4$ were prepared and the desired pH values were established by adding dilute $HClO_4$ or NaOH solutions. As the aquation reaction proceeded, 0.1 M NaOH solution was delivered from a computer-driven syringe in order to maintain a constant pH. Approximate initial rate constants were evaluated from the volume–time kinetic profiles. The kinetics of aquation in the pH range 4–8 were followed by monitoring the appearance or disappearance of phosphorus-31 NMR peaks of the product or reactant. The rate constants were estimated from the slope of the plots of ln $[I_r/(I_p + I_r)]$ vs. time, where I_r and I_p are the integrations of the peaks of the reactant and product, respectively.

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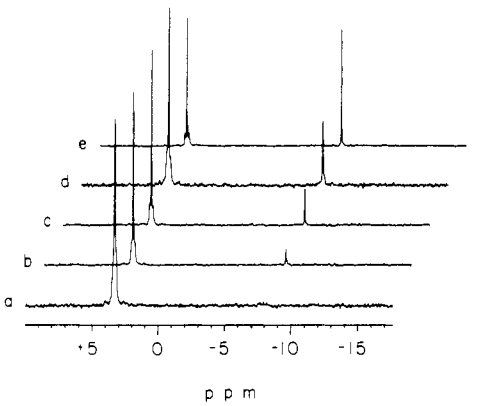

Figure 1. Proton-decoupled 36.3-MHz phosphorus-31 NMR spectra of (dihydrogen pyrophosphato)diammineplatinum(II) at ambient temperature (≈25 °C) and pH 4.5: (a) 0 h; (b) 3 h; (c) 6 h; (d) 24 h; (e) 48 h.

The rate constants can be reproduced to within 10% in the pH range 1–2.5. The agreement between the rate constants obtained spectrophotometrically and those obtained by using the pH stat method was acceptable up to pH 3.5, but at higher, unbuffered pH values the rate constants are subject to larger uncertainty.

The kinetics of the reaction between triphosphate and *cis*-$[Pt(NH_3)_2(H_2O)_2]^{2+}$ were followed spectrophtometrically at either 252 or 260 nm in the pH range 1.0–4.0. The concentration of the triphosphate ion was at least a 10-fold excess over that of the platinum substrate. The platinum concentrations were in the range 2.5–4.5 mM. The ionic strength was maintained at 0.5 M by $NaClO_4$. Since the reaction did not follow pseudo-first-order kinetics, the rate constants were evaluated from the absorbance–time traces by using appropriate rate equations as described in the Results.

The kinetics of the reaction between the pyrophosphate ion and *cis*-$[Pt(NH_3)_2(H_2O)_2]^{2+}$ could not be followed to completion spectrophotometrically due to the precipitation of an insoluble yellow product. Estimates of the rate constants, however, were obtained from the intensities of phosphorus-31 NMR resonances prior to precipitation.

Results

Characterization and Kinetics of Aquation of the Pyrophosphato Complex. A yellow solid with an empirical formula $Pt(NH_3)_2H_2P_2O_7 \cdot H_2O$ was isolated from the reaction of *cis*-$Pt(NH_3)_2Cl_2$ with tetrasodium pyrophosphate at pH 8.0. At 25 °C and 0.1 M ionic strength the pH titration curve in the pH range 2.8–11 is characterized by pK_a values of 3.4 ± 0.1 and 4.5 ± 0.1, corresponding to eq 1 and 2.

$$Pt(NH_3)_2H_2P_2O_7 + H_2O \rightleftarrows Pt(NH_3)_2HP_2O_7^- + H_3O^+ \quad (1)$$

$$Pt(NH_3)_2HP_2O_7^- + H_2O \rightleftarrows Pt(NH_3)_2P_2O_7^{2-} + H_3O^+ \quad (2)$$

The UV–visible spectrum of the compound in 0.1 M $NaClO_4$ at its isoionic pH (2.8) shows two bands at 252 and 330 nm with molar absorptivities of 150 and 35 M^{-1} cm^{-1}, respectively. The intensities of both the bands increase with increasing pH. At pH 6.0 the band at 252 nm becomes a shoulder on the rising portion of an intense absorption band located below 230 nm, and above pH 8.5 the shoulder disappears, possibly due to the decomposition of the pyrophosphato chelate at high pH.

The phosphorus-31 NMR spectrum of the compound at pH 8.0 exhibits a singlet and two satellites (from ^{195}Pt, $I = {}^1/_2$, 34% abundant; $^2J_{Pt-P}$ = 15 Hz) centered at 9.0 ppm downfield from free pyrophosphate ion at the same pH. The singlet in the phosphorus-31 NMR spectrum indicates that the two phosphate groups in the complex are magnetically equivalent. The downfield coordination chemical shift of 9.0 ppm may be compared to the 7.2 ppm downfield shift of the chelated pyrophosphate ion in (hydrogen pyrophosphato)tetraamminecobalt(III).[30]

Both the UV–visible and the phosphorus-31 NMR spectra of $Pt(NH_3)_2H_2P_2O_7$ in aqueous solution change with time. The first-order rate constants computed from the absorbance changes at 252 nm correspond to the aquation reaction revealed by the phosphorus-31 NMR spectra shown in Figure 1. The peak and satellites for the pyrophosphato chelate at 3.4 ppm decrease in intensity while a new peak appears at −8.2 ppm. The new peak is in the position for free pyrophosphate ion. The spectral changes in Figure 1 reveal that at pH 4.5 and 25 °C about 40% of the chelate undergoes aquation within 2 days. At pH 8.0, on the other hand, less than 3% of the starting material undergoes aquation within 9 days. The rate constants at various pH values as listed in Table I increase with increasing hydrogen ion concentration.

Table I. Rate Constants for the Aquation of (Dihydrogen pyrophosphato)diammineplatinum(II)[a] in 0.1 M Perchlorate Medium at 25 °C

pH	$10^6 k_{obsd}$, s^{-1}	$10^6 k_{obsd}^{calcd}$,[b] s^{-1}	pH	$10^6 k_{obsd}$, s^{-1}	$10^6 k_{obsd}^{calcd}$,[b] s^{-1}
1.0	8.5	8.6	2.5	5.0 (4[c])	5.4
1.2	8.4	8.4	3.5	4.1 (3[c])	3.7
1.5	8.1	8.1	4.5	3.2	3.4
2.0	7.2	6.9	8.0	<0.1	

[a] The concentration of the platinum complex was in the range 0.4–1.0 mM. [b] The values are calculated according to eq 3. [c] Calculated from the volume–time profiles obtained by using a pH stat.

The aquation process follows the rate law given in eq 3, where k_{obsd} is the observed first-order rate constant. The values of a,

$$k_{obsd} = \frac{a + b[H^+]}{c[H^+] + 1} \quad (3)$$

b, and c obtained from an iterative nonlinear least-squares fit to eq 3 are $a = (3.4 \pm 0.3) \times 10^{-6}$ s^{-1}, $b = (1.6 \pm 0.4) \times 10^{-3}$ M^{-1} s^{-1}, and $c = 177 \pm 40$ M^{-1}.

Platinum(II)-Promoted Hydrolysis of Triphosphate. The total amount of orthophosphate ion obtained at the end of the reaction between *cis*-diaquodiammineplatinum(II) cation and triphosphate anion at pH 3.5 was plotted against the molar ratio, $[P_3O_{10}/Pt]$. The plot exhibited a break point at the molar ratio of 0.55 ± 0.05, indicating that 2 mol of the platinum substrate are required to generate 1 mol of orthophosphate ion.

The reaction of triphosphate ion with *cis*-diaquodiammineplatinum(II) nitrate in the pH range 1.0–5.0 at 0.5 M ionic strength in either nitrate or perchlorate medium forms products that are blue in color. Triphosphate serves as a buffer to limit the pH change (see Experimental Section). In acidic nitrate and perchlorate media the blue products have absorption bands at 615, 475, and 370 nm. An additional absorption band in the UV region at 260 nm was observed in the presence of the perchlorate electrolyte. Since nitrate ion itself has an intense absorption in the UV region, the presence of such a band cannot be discerned in the nitrate medium. The formation of the blue species can be observed within 3 h at 40 °C in the nitrate medium. Under similar conditions, formation of the blue color was not observed before 8 h in perchlorate medium. When the reaction was allowed to proceed in a nitrogen atmosphere in the perchlorate medium, the formation of the blue products was extremely slow; a faint blue color was observed after 12 h at 40 °C.

The absorption spectra recorded during the reaction between triphosphate and *cis*-$[Pt(NH_3)_2(H_2O)_2]^{2+}$ in 0.5 M $NaClO_4$ purged with nitrogen showed that the absorbance in the wavelength region 250–260 nm does not change after 12 h, although the absorbance continues to change in the visible region due to the formation of the blue species. The molar absorptivities of the initial products and the subsequent blue products are therefore the same in the wavelength region 250–260 nm. On the other hand, if the reaction is allowed to proceed in the presence of air, the changes in absorbances in both the visible and the UV region take place simultaneously, and the measured half-lives are the same in both wavelength regions. Under identical experimental conditions, the change in absorbance in the wavelength region 250–260 nm and the time elapsed for the change can be reproduced to within 5%. Since the change in absorbance in the visible region depends upon the amount of air present, the kinetics of the reaction were followed

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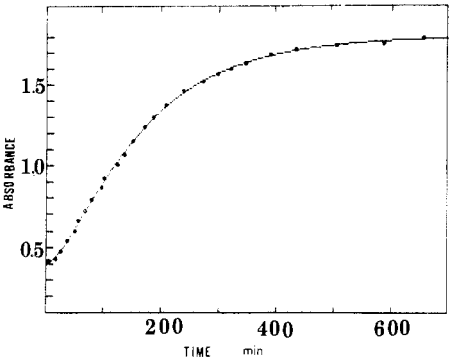


Figure 2. Fit of the absorbance–time data at 252 nm to eq 4 for the reaction between *cis*-diaquodiammineplatinum(II) nitrate (4.5 mM) and triphosphate ion (41.5 mM) at 40 °C and pH 3.9 in 0.5 M $NaClO_4$ in the presence of air. The dotted line was calculated by using fitted values $k_0 = 4.8 \times 10^{-4}$ s^{-1} and $k_1 = 5.1 \times 10^{-5}$ s^{-1} and experimental values $\epsilon_A = 210$ M^{-1} cm^{-1}, $\epsilon_I = 267$ M^{-1} cm^{-1}, and $D_\infty = 1.59$.

Table II. Rate Constants for the Reaction of Triphosphate Ion with *cis*-Diaquodiammineplatinum(II) Nitrate at 40 °C and 0.5 M Ionic Strength ($NaClO_4$)

pH	$[P_3O_{10}]_t$, mM	[Pt], mM	10^4k_0, s^{-1}	10^5k_1, s^{-1}
1.1	25.5	2.1	0.9	2.0
1.4	15.5	1.8	1.0	2.4
1.8	15.5	1.0	1.2	2.8
2.5	15.8	1.5	1.4	3.4
3.4	32.3	3.0	3.2	4.1
3.9	18.2	1.8	2.4	5.3
3.9	31.8	2.3	3.7	4.9
3.9	41.5	4.2	4.5	5.5
3.9	45.0	4.2	5.4	5.2
3.9	58.2	4.2	6.5	5.6
4.4	16.0	1.8	1.8	5.9

spectrophotometrically in the wavelength region 250–260 nm. Due to interference from competing reactions above pH 4.0 as discussed below, the kinetics followed spectrophotometrically were limited to the pH range 1.0–4.0.

Figure 2 shows the absorbance–time profile for the triphosphate reaction using a 10-fold excess of triphosphate over the platinum substrate. The first-order plots constructed from the absorbance time data deviate from linearity within the first half-life. The absorbance–time data, however, can adequately be described by consecutive first-order reactions of the type

$$[Pt(NH_3)_2(H_2O)_2]^{2+} + P_3O_{10}^{n-} \xrightarrow{k_0} (\text{intermediate}) \xrightarrow{k_1} \text{products} \quad (4)$$

The values of the rate constants, k_0 and k_1 were evaluated by using a nonlinear least-squares fit of the integrated rate expression for consecutive first-order reactions[21] as given by eq 5. In this eq

$$D = \epsilon_A[A]_0e^{-k_0t} + \frac{[A]_0\epsilon_1k_0}{k_1 - k_0}(e^{-k_0t} - e^{-k_1t}) + \frac{D_\infty}{k_1 - k_0}(k_0e^{-k_1t} - k_1e^{-k_0t}) + D_\infty \quad (5)$$

$[A]_0$ represents the initial concentration of the starting platinum complex, D and D_∞ are the absorbances at time t and at infinite time, and ϵ_A and ϵ_I are the molar absorptivities of the starting platinum complex and the intermediate. The values of ϵ_I, k_0, and k_1 were determined by fits to eq 5; D_∞, ϵ_A, D, and $[A]_0$ were experimentally determined.

Table II lists the values of k_0 and k_1 at various triphosphate concentrations and pH values.[31] The rate constant k_0 varies linearly with the triphosphate concentration and increases with decreasing hydrogen ion concentration. The second-order rate constant, k_2 (1.10×10^{-2} M^{-1} s^{-1} at pH 3.9), was obtained from a linear least-squares fit using eq 6. The rate constant k_1, which

$$k_0 = k_2[P_3O_{10}^{n-}] \quad (6)$$

is equal to 5.3×10^{-5} s^{-1} at pH 3.9, does not depend on the triphosphate concentration but does increase with increasing pH.

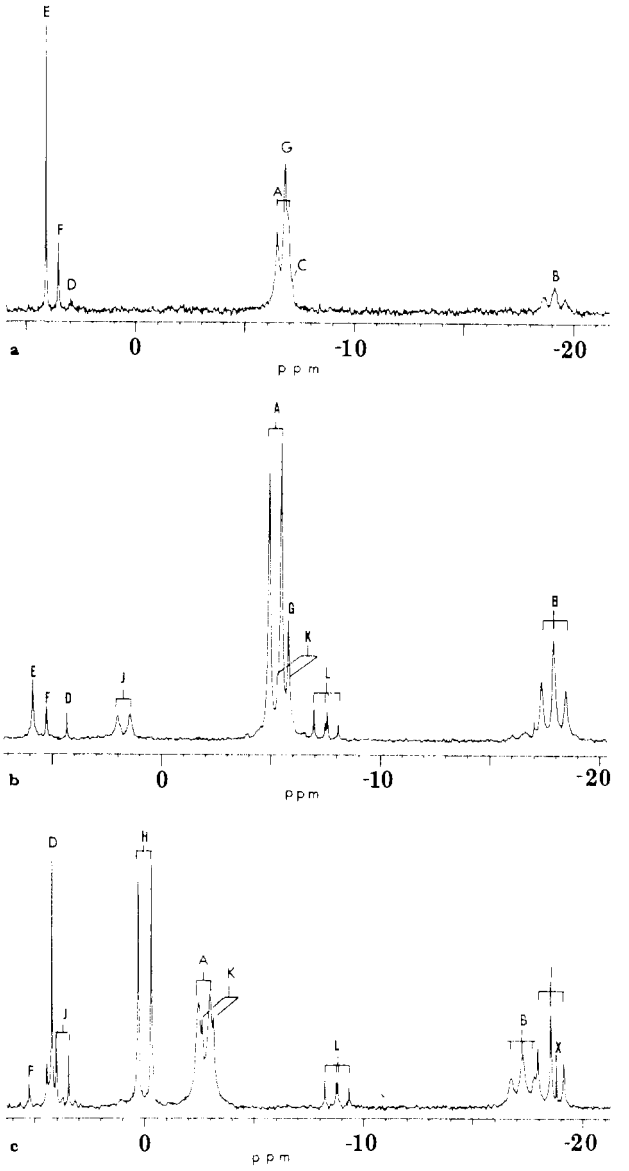
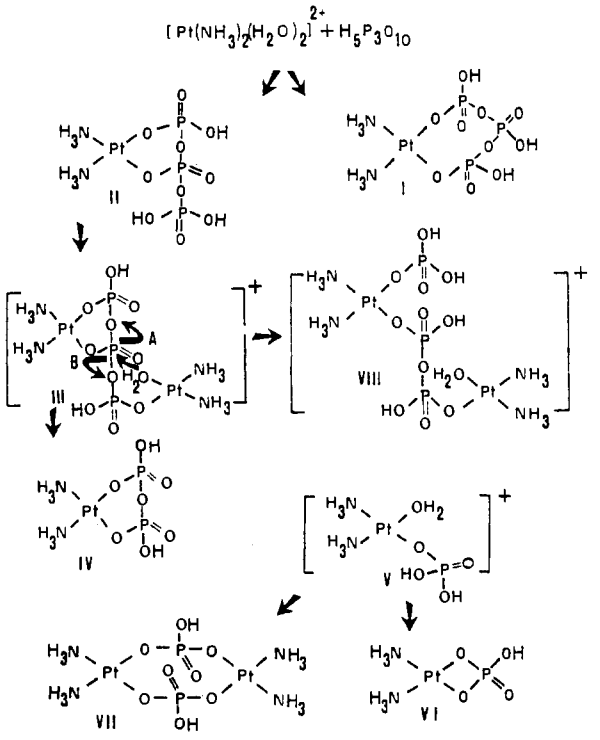


Figure 3. Proton-decoupled 36.3-MHz phosphorus-31 NMR spectra at the end of the reaction (48 h) between *cis*-diaquodiammineplatinum(II) (10.0 mM) and triphosphate ion at room temperature: (a) pH 1.0, $[P_3O_{10}]_t = 20.0$ mM; (b) pH 5.0, $[P_3O_{10}]_t = 40.0$ mM; (c) pH 8.0, $[P_3O_{10}]_t = 40.0$ mM.

The progress of the reaction was also monitored by phosphorus-31 NMR spectroscopy in the pH range 1.0–8.0. One set of products is observed in the pH range 1–4 while a different set is observed in the pH range 7–8. The NMR spectrum of the products at pH 5 contains the peaks corresponding to the products observed in both the lower and upper pH ranges. Figure 3a shows the spectrum recorded at the end of the reaction at pH 1.0 in 0.5 M $NaClO_4$. The doublet, peak A, at −6.88 ppm ($J = 19.0$ Hz) and the triplet, peak B, at −19.36 ppm ($J = 19.0$ Hz) are due to free triphosphate ion, and the peak C at −7.23 ppm is due to an impurity of pyrophosphate ion that was present in the starting material. The singlet, peak D, at 2.83 ppm arises from the pyrophosphato chelate discussed earlier. The signals E, F, and G at 3.96, 3.41, and −6.95 ppm, respectively, appear coincident with the formation of the blue product. At pH 4.0, the spectrum of

(31) The reactions were followed in solutions buffered by the excess triphosphate concentration so that pH values usually decreased by 0.2 or less during the reaction. Since the dependence of the rate upon pH is not steep (see Table II), the uncertainty associated with this small pH change is small. Taking into account this uncertainty along with other experimental uncertainties, the reproducibility of the rate constants was better than 8%.

Scheme I



the products has the same three signals, E, F, and G, except that all the peaks appear 0.4–0.6 ppm downfield from their position at pH 1.0. At the beginning of the reaction at pH 4.0, two doublets at 4.60 ppm (J = 20.8 Hz) and 0.02 ppm (J = 21.0 Hz) and a doublet of doublets centered at −8.51 ppm (J = 21.0, 20.8 Hz) were observed. These two doublets and the doublet of doublets disappeared with the formation of peaks E, F, and G. On the basis of the chemical shifts, coupling constants, and stoichiometric data, these initial peaks were attributed to the formation of a triphosphato-bridged diplatinum species[22] (shown as III in Scheme I). The phosphorus-31 NMR spectrum of a blue product[32] isolated at the end of the reaction at pH 3.0 exhibits peaks in the same locations as signals E and G along with small peaks due to free triphosphate ion (Figure 4, supplementary material). The remaining peak, F, in Figure 3a is due to an orthophosphato complex of platinum(II).[21]

The phosphorus-31 NMR spectrum at the end of the reaction at pH 5.0 is shown in Figure 3b. Peaks D, E, F, and G appeared at 4.17, 5.71, 5.10, and −5.94 ppm, showing a downfield shift of 0.34, 1.75, 0.69, and 1.01 ppm, respectively, relative to their positions at pH 1.0. In addition to these peaks, a doublet, J, at 1.58 ppm (J = 20.7 Hz), a peak, K, at −5.45 ppm, and a doublet of doublets, L, centered at −7.65 ppm (J = 20.7, 21.0 Hz) were observed. Peaks J–L are due to the formation of the β,γ-chelate (II in Scheme I) as observed earlier.[20] Peak K is a doublet of the uncomplexed α-phosphate group, which is partly hidden under peak G.

At pH 8.0, the products of the reaction are light yellow rather than blue; the phosphorus-31 NMR spectrum of the reaction mixture at the end of the reaction at this pH is shown in Figure 3c. Peaks J and K for the β,γ-chelate are shifted downfield from their positions at pH 5.0 while L has an upfield shift: J is found at 3.77 ppm (J = 21.2 Hz); K, at −2.89 ppm (J = 21.4 Hz); L, at −8.76 ppm (J = 21.2, 21.4 Hz). Doublet H at 0.02 ppm (J = 21.5 Hz) and triplet I at −18.54 ppm (J = 21.5 Hz) are due to the α,γ-chelate. Peak D with the two satellites at 4.20 ppm

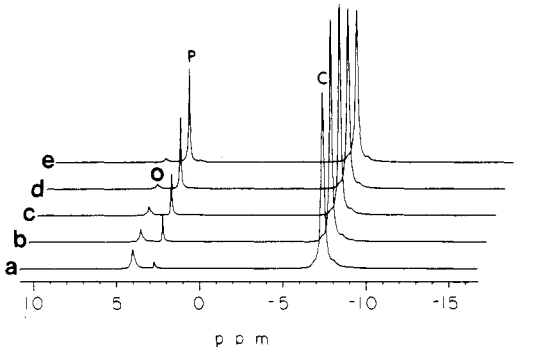


Figure 5. Proton-decoupled 36.3-MHz phosphorus-31 NMR spectra of the reaction between pyrophosphate ion (40.0 mM) and *cis*-diaquodiammineplatinum(II) (10.0 mM) at pH 4.0 and 55 °C: (a) 0.5 h; (b) 1.5 h; (c) 3.0 h; (d) 6.0 h; (e) 10.5 h.

arises from the pyrophosphato chelate and the peak, F, at 5.20 ppm is due to the orthophosphato complex. No peaks are observed at pH 8 that correspond to peaks E and G that were observed at lower pH. In addition to these peaks, a yellow product with a platinum:phosphate stoichiometric ratio of 1.03 was isolated from the reaction.

Platinum(II)-Promoted Hydrolysis of Pyrophosphate. Like the triphosphato reaction, the reaction of pyrophosphate with *cis*-$[Pt(NH_3)_2(H_2O)_2]^{2+}$ in the pH range 1–5 produces a blue product. The blue species has absorption bands at 610, 475, and 370 nm. However, unlike in the triphosphato reaction, a yellow precipitate appears while the reaction is in progress. The rate of formation of this yellow precipitate depends upon the pH. With platinum and pyrophosphate concentrations of 2.0 and 20.2 mM, the formation of the precipitate was observed within 2 h at pH 1.5, while such a precipitate formed after 6 h at pH 3.5. Moreover, the amount of precipitate obtained at pH 3.5 appeared to be smaller than the amount of precipitate obtained at pH 1.5.

Figure 5 shows the phosphorus-31 NMR spectra recorded at various time intervals during the progress of the reaction. Small amounts of yellow precipitate were filtered out before the spectra were recorded at 6.0 and 10.5 h. In addition to peak C at −7.39 ppm for free pyrophosphate ion, two additional peaks, O at 4.03 and P at 2.75 ppm, were observed as soon as the first spectrum could be recorded. Peak O decreased in intensity with time while the intensity of the peak P increased. Peak O is apparently due to the formation of an intermediate that decays to the final product (peak P). The observation of the intermediate and the final products is consistent with the kinetics for consecutive reactions of the type observed for triphosphato reaction. The complete analysis of the kinetics for the pyrophosphato reaction, however, was not possible because of the precipitation of one of the products. The rate constant for the decay of the intermediate at pH 4.0 and 55 °C was estimated to be 10^{-4} s^{-1} from the intensities of the phosphorus-31 resonances of products prior to the precipitation. Since the data were taken from the initial 3 h of the reaction, the estimated value is a lower limit because of the consecutive kinetic behavior of the reaction. This estimated value of the rate constant is at least 4 orders of magnitude greater than the corresponding rate constant for the hydrolysis of free pyrophosphate ion.[33]

The reaction of (dihydrogen pyrophosphato)diammineplatinum(II) with *cis*-$[Pt(NH_3)_2(H_2O)_2]^{2+}$ at pH 5.0 results in the formation of a yellow precipitate. The phosphorus-31 NMR spectrum of the reaction mixture within 10 min of mixing the reactants (prior to precipitation) exhibits two peaks, one at 5.31 ppm with two satellites 18 Hz apart and the other at 18.23 ppm. The spectra recorded after filtration of the precipitate did not exhibit the peak at 18.23 ppm. The peak at 5.3 ppm is 1.1 ppm downfield from the position of the peak for the pyrophosphato chelate, and the peak at 18.23 ppm is 13 ppm downfield from the position of the orthophosphato complex (peak F in Figure 3).

(32) Bose, R. N.; Viola, R. E.; Cornelius, R. D., manuscript in preparation, dealing with the characterization of this blue product along with other phosphate blues and their epr and electrochemical properties. Since the formation of the blues takes place in acidic nitrate and acidic perchlorate media in the presence or, much more slowly, in the absence of air, it is possible that nitrate (or perchlorate) oxidizes platinum(II).

(33) Van Wazer, J. R.; Griffith, E. J.; McCullough, J. F. *J. Am. Chem. Soc.* **1955**, *77*, 287–291.

Discussion

Kinetics of Aquation of (Dihydrogen pyrophosphato)diammineplatinum(II). All the physical measurements reported in the Results for the solid pyrophosphato complex are consistent with the formulation of the compound as a pyrophosphato complex in which pyrophosphate is a bidentate ligand coordinated through both of the phosphate groups. The two-bond coupling constant, $^2J_{P-Pt}$, obtained from the phosphorus-31 NMR spectrum of the compound is 15 Hz. This coupling constant is much smaller than the coupling constants found in the phosphate-bridged dinuclear metal–metal-bonded platinum(III) complexes,[18] for which two-bond coupling constants in the range 40–90 Hz have been reported. The difference in coupling constants between the phosphate-bridged dinuclear complex and the pyrophosphato chelate is understandable in view of the differences in the ligand, the binding mode, and the oxidation state of the platinum.

A mechanism consistent with our kinetics and NMR data for the aquation of the pyrophosphato chelate is presented as steps 7 through 11.

$$Pt(NH_3)_2H_2P_2O_7 + H_3O^+ \overset{K_b}{\rightleftharpoons} [Pt(NH_3)_2H_3P_2O_7]^+ + H_2O \quad (7)$$

$$Pt(NH_3)_2H_2P_2O_7 + H_2O \underset{k_r}{\overset{k_f}{\rightleftharpoons}} Pt(NH_3)_2(H_2O)H_2P_2O_7 \quad (8)$$

$$[Pt(NH_3)_2H_3P_2O_7]^+ + H_2O \underset{k'_r}{\overset{k'_f}{\rightleftharpoons}} [Pt(NH_3)_2(H_2O)H_3P_2O_7]^+ \quad (9)$$

$$Pt(NH_3)_2(H_2O)H_2P_2O_7 \xrightarrow{k_{aq}} \text{products} \quad (10)$$

$$[Pt(NH_3)_2(H_2O)H_3P_2O_7]^+ \xrightarrow{k'_{aq}} \text{products} \quad (11)$$

The rate expression given by eq 12 can be derived for this mechanism.

$$k_0 = \frac{k_{aq}k_f}{(k_r + k_{aq})(K_b[H^+] + 1)} + \frac{k'_{aq}k'_fK_b[H^+]}{(k'_r + k'_{aq})(K_b[H^+] + 1)} \quad (12)$$

Under conditions for which $k_r >> k_{aq}$ and $k'_r >> k'_{aq}$, this equation simplifies to the form of eq 3. The values of k_f and K_b are then equal to the constants a (3.4×10^{-6} s^{-1}) and c (177 M^{-1}), respectively, and the value of k'_f can be calculated to be 9.0×10^{-6} by using the value of b ($b = k'_fK_b$). According to this analysis, the rate of aquation is limited by the initial aquation process.

An alternative mechanism may be considered in which the order of the aquation and protonation steps differs. Such a mechanism also results in a rate law having the functional form of eq 3, and the rate of aquation of the complex would still be limited by the initial aquation step.

Mechanism of Platinum(II)-Promoted Hydrolysis of Triphosphate. The spectra at the end of the reaction in the pH range 1–8 all contain signals due to the formation of ortho- and pyrophosphato complexes of platinum. In the pH region 1–4 these ortho- and pyrophosphato complexes are blue in color due to the subsequent oxidation of the products as described below. At pH 8.0 such an oxidation is apparently not favorable, and the only products observed are ortho- and pyrophosphato complexes of platinum(II). The rates at which these hydrolyzed products are formed are at least 100 times faster (vide infra) than the hydrolysis of the free triphosphate ion under comparable experimental conditions[33] in the absence of metal complexes.

The formation of the various hydrolyzed products and chelates from the reaction of triphosphate ion with *cis*-$[Pt(NH_3)_2(H_2O)_2]^{2+}$ can be understood in terms of Scheme I. The β,γ-chelate, II, which is formed initially, reacts with another *cis*-$[Pt(NH_3)_2(H_2O)_2]^{2+}$ to form the binuclear intermediate III. The cleavage of a phosphorus–oxygen bond of III in either position a or position b leads to the formation of products as indicated by path A or B. The pyrophosphato chelate, IV, formed by path A was readily observed in the phosphorus-31 NMR spectrum when the reaction proceeded at pH 8.0. The other product of reaction A is an aquo orthophosphato complex, V. Although this species can be observed in the NMR spectrum (peak F in Figure 3), the intensity of its peak is less than one-half of the intensity of the peak for the pyrophosphato chelate, IV. Further reaction of product V yields a yellow precipitate with a platinum:phosphate stoichiometric ratio of 1.03 ± 0.02, which is consistent with the formulation of either a bis(orthophosphato)-bridged diplatinum species, VII, or an orthophosphato chelate, VI.

The phosphorus-31 NMR spectrum of the blue product isolated from the reaction in the pH region 1–5 exhibits two signals, E and G, the positions of which are pH-dependent. The integrated intensity ratio of these two peaks, G:E, was found to be 2.0 ± 0.1. The average oxidation state of platinum in this blue product is +2.3 ± 0.2. The formation of tetranuclear platinum blues with α-pyridone and other amide ligands with partial Pt–Pt axial bonds between the two bridged dimeric species is now well established,[34] and the average oxidation state in these blues has been reported to be +2.25. The results of the reaction through path B would be a species, VIII, in which pyrophosphate bridges between the two platinum atoms and orthophosphate is a monodentate ligand coordinated to a single platinum atom. The formation of an axial Pt–Pt bond between the two molecules of VIII with the oxidation of one of the four platinum(II) ions to platinum(III) is consistent with the redox titration, EPR spectra,[32] and NMR data. The singlet E is attributed to the coordinated orthophosphate and the peak G to the bridging pyrophosphate ligand. The chemical shifts of these two peaks cannot be directly compared with those of platinum(II) phosphates due to the difference in the oxidation state of the platinum species.

A mechanism consistent with the kinetics and NMR data in the pH range 1–4 where dimerization of *cis*-$[Pt(NH_3)_2(H_2O)_2]^{2+}$ is very slow[25] can be represented by eq 13–17. The species

$$H_4P_3O_{10}^- + H_2O \overset{K_1}{\rightleftharpoons} H_3P_3O_{10}^{2-} + H_3O^+ \quad (13)$$

$$H_3P_3O_{10}^{2-} + H_2O \overset{K_2}{\rightleftharpoons} H_2P_3O_{10}^{3-} + H_3O^+ \quad (14)$$

$$[Pt(NH_3)_2(H_2O)_2]^{2+} + H_2P_3O_{10}^{3-} \underset{k_{-3}}{\overset{k_3}{\rightleftharpoons}} [Pt(NH_3)_2H_2P_3O_{10}]^- \quad (15)$$

$$[Pt(NH_3)_2(H_2O)_2]^{2+} + [Pt(NH_3)_2H_2P_3O_{10}]^- \xrightarrow{k_4} [Pt_2(NH_3)_4(H_2O)H_2P_3O_{10}]^+ \quad (16)$$

$$[Pt_2(NH_3)_4(H_2O)H_2P_3O_{10}]^+ \xrightarrow{k_5} \text{products} \quad (17)$$

$[Pt(NH_3)_2H_2P_3O_{10}]^-$ and $[Pt_2(NH_3)_4(H_2O)H_2P_3O_{10}]^+$ represent a (triphosphato)diammineplatinum(II) chelate and a triphosphato-bridged diplatinum complex. Since the intermediate has been characterized as a triphosphato-bridged diplatinum species,[22] the first step of the reaction observed kinetically can be represented by eq 13–16. Assuming a steady-state approximation for the species $[Pt(NH_3)_2H_2P_3O_{10}]^-$, the expression for the rate of formation of the intermediate based on eq 13–16 is given by eq 18 where $[P_3O_{10}]_t$ represents the total triphosphate concentration. Under conditions for which $k_4[Pt(NH_3)_2(H_2O)_2^{2+}] >> k_{-3}$, the rate of formation of the intermediate would be second order at a constant pH and first order with respect to each of the reactants. Taking an upper estimate of 10^{-5} s^{-1} for the rate

$$\text{formation of intermediate} = \frac{k_4k_3[Pt(NH_3)_2(H_2O)_2^{2+}]^2[P_3O_{10}]_t}{k_{-3} + k_4[Pt(NH_3)_2(H_2O)_2^{2+}]} \times \left(\frac{K_1K_2}{K_1K_2 + K_1[H^+] + [H^+]^2}\right) \quad (18)$$

(34) Barton, K. J.; Caravana, C.; Lippard, S. J. *J. Am. Chem. Soc.* **1979**, *101*, 7269–7277. Ginsberg, A. P.; O'Halloran, T. V.; Fanwick, P. E.; Hollis, L. S.; Lippard, S. J. *Ibid.* **1984**, *106*, 5430–5439. Hollis, L. S.; Lippard, S. J. *Inorg. Chem.* **1983**, *22*, 2600–2604, 2605–2614.

constant for aquation of the triphosphato chelate (similar to that for the pyrophosphato chelate) and assuming k_4 is similar in magnitude to k_3 (0.1 M^{-1} s^{-1}; discussed below), it can be shown that at the lowest platinum concentration $k_4[Pt(NH_3)_2(H_2O)_2^{2+}]$ is at least 25 times greater than k_{-3}. The observed second-order rate constant k_2 is therefore simply the rate constant for the formation of $[Pt(NH_3)_2H_2P_3O_{10}]^-$. Although a single-step formation of the chelate is shown in eq 13, formation of a monodentate triphosphato complex followed by rapid ring closure is also possible.

The pK_a values of triphosphoric acid, $H_5P_3O_{10}$, are <0.5, 1.0, 2.0, 5.7, and 8.6.[35] Since no saturation effect was observed in the rate up to pH 4.0, at least one deprotonated site involved must have a pK_a greater than 3. Taking the pK_a value as $pK_2 = 5.7$ and assuming $pK_1 = 1.0$, an estimate of k_3 from eq 18 can be obtained as 0.7 M^{-1} s^{-1}.

In the pH range 5–8, a competition between the phosphato complex formation and hydroxo-bridged dimer formation is possible. Since the reaction in this pH range was monitored by phosphorus-31 NMR, the amount of hydroxo-bridged dimer and higher oligomer is unknown. However, from the intensities of the phosphorus-31 resonances, it was found that 80% of the initial platinum complex has been converted to the various phosphato products. Therefore, the hydroxo-bridged complexes should have consumed less than 20% of the initial platinum(II) complex. It remains a possibility, however, that these hydroxo-bridged oligomers readily react with the triphosphate ligand to form the phosphato complexes.

The first-order rate constant for the decay of the intermediate is attributed to the hydrolysis of the coordinated triphosphate. The rate constant increases with increasing pH. A mechanism consistent with the interpretation that the coordinated hydroxide ion acts as a nucleophile in order to hydrolyze the phosphate ligand is shown by eq 19 and 20. Assuming that the acid dissociation

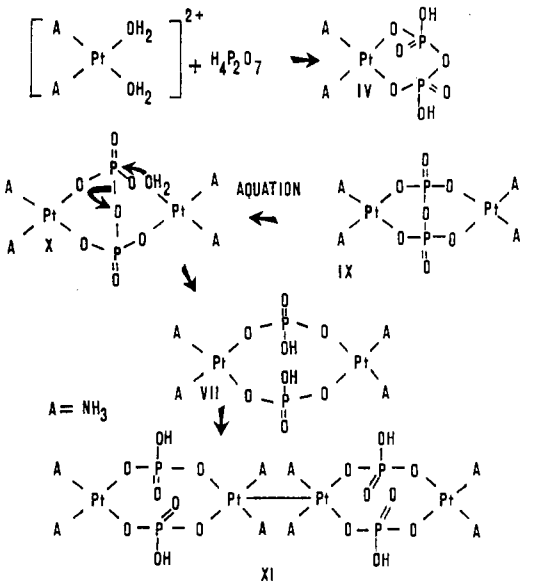
$$[Pt_2(NH_3)_4(H_2O)H_2P_3O_{10}]^+ + H_2O \overset{K}{\rightleftharpoons} [Pt_2(NH_3)_4(OH)H_2P_3O_{10}] + H_3O^+ \quad (19)$$

$$[Pt_2(NH_3)_4(OH)H_2P_3O_{10}] \xrightarrow{k_h} \text{hydrolyzed products} \quad (20)$$

constant for the aquated intermediate, $[Pt(NH_3)_4(H_2O)(H_2P_3O_{10})]^+$, is close to the first acid ionization constant of *cis*-$[Pt(NH_3)_2(H_2O)_2]^{2+}$ (pK = 5.7),[36] an estimate of 5×10^{-4} for k_h can be obtained.

Mechanism of Platinum(II)-Promoted Hydrolysis of Pyrophosphate Ion. The position of peak O is 0.63 ppm downfield from the resonance of the pyrophosphato chelate reported earlier, and peak P is 1.2 ppm downfield from the position of the free orthophosphate peak. The downfield location of peak O from that of the pyrophosphato chelate is consistent with the formation of a pyrophosphato-bridged diplatinum species (either IX or X in Scheme II).[37] The crystal structure of such a pyrophosphato-bridged double chelate has been reported by Stanko.[15] Aquation of this double chelate followed by hydrolysis by the coordinated

Scheme II



aquo or hydroxo group would lead to the formation of a bis(orthophosphato)-bridged diplatinum complex, VII. The estimated rate constant for the decay of the intermediate, 10^{-4} s^{-1}, would be consistent with the rate constant for such an aquation reaction. The product VII, a bis(orthophosphato)diplatinum(II) complex, would yield a blue species after oxidation and the formation of a partial Pt–Pt bond. The structure of such a blue can be formulated as a dimer (or oligomer) of XI. Under the conditions where formation of the blue species is slow, precipitation of the product, VII, occurred due to its low solubility. The analytical data of the yellow precipitate are consistent with the product VII. The formation of the orthophosphato platinum blue starting from pyrophosphate ion can also be supported by the fact that the orthophosphato blue prepared from orthophosphate ion and *cis*-$Pt(NH_3)_2Cl_2$ has the same absorption and EPR spectra as the blue that resulted from the pyrophosphato reaction.[21,32]

The formation of the pyrophosphato-bridged double chelate and the hydrolyzed phosphato-bridged diplatinum complex can also be supported from the direct reaction of (dihydrogenpyrophosphato)diammineplatinum(II) with *cis*-(diaquodiammineplatinum(II). The peak at 1.1 ppm downfield from the location for the pyrophosphato chelate is due to the formation of the pyrophosphato-bridged diplatinum species, IX. The peak at 18.23 ppm is attributed to the orthophosphato-bridged species, VII, which precipitates as a yellow solid. The analysis of this yellow precipitate is consistent with such a formulation. Moreover, in the case of (orthophosphato)cobalt(III) complexes, the phosphorus-31 resonance of the bridged orthophosphate ion appears at about 10 ppm downfield from the resonance of the monodentate orthophosphato complex.[10] In the present case, the position of the peak at 18.23 ppm is 13 ppm downfield from the peak position of the monodentate complex and is consistent with the assignment as the bis(orthophosphato)-bridged diplatinum product, VII.

Alternatively, it is possible that peak O is due to a pyrophosphato-bridged "semidouble" chelate, X, which undergoes intramolecular phosphate hydrolysis to yield the products. The phosphate groups in this dinuclear structure are not magnetically equivalent, yet their chemical shifts may be close enough to give rise to a single broad peak as seen in Figure 5.

Conclusion

On the surface, the results of this study on the acceleration of phosphate hydrolysis by platinum(II) appear to be different than the results of previous studies using cobalt(III). The accelerated hydrolysis in the presence of cobalt(III) has been observed only when the ratio of the concentration of cobalt(III) to that of phosphate is greater than 1. In the present study, accelerated hydrolysis has been found to occur even in the presence of a pseudo-first-order concentration of phosphate.

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(37) One reviewer has pointed out that the small chemical shift difference (0.63 ppm) between peak O and the signal due to (dihydrogen pyrophosphato)platinum(II) seems to be too small to correspond to a bridged diplatinum(II, II) such as species IX. The oxidation state of the intermediate, however, is not known. The final product has an average oxidation state for platinum of 2.25. If the intermediate has an oxidation state other than 2.0, we have no reference point from which to measure the chemical shift change. The possibility that peak O is due to the pyrophosphate chelate IV and that it appeared 0.63 ppm downfield due to the small variation in pH during the reaction can also be ruled out. The phosphorus-31 NMR spectra of the pyrophosphato chelate show an upfield shift of 0.82 ppm when the pH is changed from 8.0 to 3.5. The hydrolysis of pyrophosphate in the presence of platinum complexes under the condition of excess pyrophosphate is accompanied by a decrease in pH of 0.2 or less at room temperature. Even if the pH change were larger at 55 °C, the position of the peak for the pyrophosphato chelate is expected to be shifted upfield rather than downfield.

On the mechanistic level, however, the differences lie in the rate at which processes occur rather than in the nature of mechanistic steps. The mechanism of the accelerated hydrolysis of polyphosphates in the presence of either platinum(II) or cobalt(III) can be described by three steps: (1) coordination of a metal ion to the polyphosphate to reduce the nucleophilicity of the phosphate ligand; (2) coordination of a second metal ion having cis coordination positions available; (3) hydrolytic attack by the cis hydroxide ion on the polyphosphate chain. The strength of binding in the first step determines the stoichiometry necessary for the accelerated hydrolysis to be observed. Apparently the hard cobalt(III) forms a tight complex with the polyphosphate so that an excess concentration of metal ion must be present before step 2 can occur. The softer platinum(II) forms a weaker complex, leaving metal ions available for step 2 even in the presence of excess phosphate. The weaker binding also accounts for the possibility of Scheme II, in which aquation must occur before hydrolysis. The slower aquation in the case of cobalt(III) for a complex such as IX prevents bidentate pyrophosphate from being rapidly hydrolyzed by the presence of a second metal ion.

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Registry No. *cis*-$[Pt(NH_3)_2(H_2O)_2]^{2+}$, 20115-64-4; $Pt(NH_3)_2H_2P_2O_7$, 98736-88-0; $H_4P_2O_7$, 2466-09-3; $H_5P_3O_{10}$, 10380-08-2.

Supplementary Material Available: Figure 4, ambient-temperature (≈25 °C) 36.3-MHz phosphorus-31 NMR spectrum of blue product isolated from the reaction of triphosphate ion with *cis*-diaquodiammineplatinum(II) at pH 3.0 (1 page). Ordering information is given on any current masthead page.

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Copper(II) Coordination Chemistry in Bovine Plasma Amine Oxidase: Azide and Thiocyanate Binding

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Azide and thiocyanate binding to the Cu(II) sites in bovine plasma amine oxidase have been investigated by absorption, CD, and EPR spectroscopy. Anion complexes of the resting enzyme display characteristic absorption bands: N_3^-, λ = 385 nm ($\Delta\epsilon$ = 3200 M^{-1} cm^{-1}); SCN^-, λ = 365 nm ($\Delta\epsilon$ = 2400 M^{-1} cm^{-1}). The energies and intensities of these bands are consistent with N_3^-/SCN^- → Cu(II) ligand-to-metal charge-transfer assignments, where the anions are coordinated within the equatorial plane in a tetragonal Cu(II) complex. EPR data support a tetragonal structure for the Cu(II)–anion complex and further indicate that the Cu(II) electronic ground state is not greatly perturbed by anion binding. Azide binding produces CD bands at 700 (d–d), 400 (LMCT), and 480 nm; this last result suggests that the absorption and CD bands displayed by the resting enzyme in the 400–500-nm range are due in part to Cu(II) electronic transitions. Azide and thiocyanate also bind to the substrate-reduced enzyme, producing LMCT transitions that are quite similar to those observed for anion complexes of the resting enzyme. Although simple titration curves are not obtained for anion binding in phosphate buffers, it is clear that the substrate-reduced amine oxidase has a lower anion affinity than the resting form. Taken together, the data establish that anion binding is a Cu(II) ligand-substitution reaction that does not lead to major structural changes in the Cu(II) site. Beef plasma amine oxidase is inhibited by N_3^- and SCN^- but not by F^-, Cl^-, or I^-. When O_2 is present in saturating concentrations, the anion inhibition pattern changes from mixed to uncompetitive, as the amine concentration increases. Thus anion binding to both the resting enzyme and at least one other enzyme form (probably a reduced species) decreases substrate oxidation rates.

Elucidating the active-site structure and mechanism of copper-containing amine oxidases is of considerable interest. These enzymes catalyze the two-electron oxidative deamination of primary amines to aldehydes.[1-4] Dioxygen serves as the electron acceptor, being reduced to hydrogen peroxide. The overall reaction is

$$RCH_2NH_2 + O_2 + H_2O \rightarrow RCHO + H_2O_2 + NH_3$$

A wide variety of mono-, di-, and polyamines can serve as substrates, depending on the enzyme source, e.g. fungi, plants, and mammals.[1-4] Copper-containing amine oxidases contain two Cu(II) ions per enzyme molecule, which is composed of two noncovalently bound subunits with a total molecular weight of ~180000. In addition, these enzymes contain another cofactor that reacts with carbonyl reagents and is reduced by substrates.[5] Copper is generally thought to participate in the reoxidation of the substrate-reduced enzyme.[1-4] No Cu(I) intermediates were detected by rapid-freeze EPR experiments during the reoxidation of benzylamine-reduced pig plasma amine oxidase.[6] This result, together with other data, led Knowles and co-workers to propose that Cu(II)-coordinated hydroxide acts as a nucleophile to assist in hydride transfer to O_2.[7] On the other hand, some results suggest that a Cu(II)/Cu(I) redox cycle is possible.[4,8] There are reasonable circumstances whereby a catalytically competent Cu(I) enzyme form would not have been detected under the conditions of the EPR experiment. Further complicating this picture are NMR relaxation results obtained with pig kidney diamine oxidase, which indicated that neither the substrate amino groups nor NH_3 binds near Cu(II).[9] Thus the exact mechanistic role of copper in amine oxidases is still somewhat obscure, and it has even been suggested that copper is not directly involved in catalysis.[9] Some structural information on the copper site is available. On the basis of EPR g values and the energies of the d–d bands, amine oxidase

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