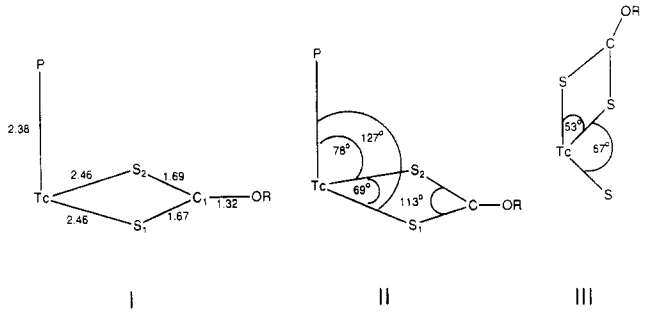

Figure 3. Diagram depicting bond lengths and angles of $[Tc(PPh_3)(S_2COC_4H_9)_3]$ from various perspectives.

from various perspectives are shown in I and II of Figure 3, while III depicts the bond angles looking down the technetium–phosphorus bond, showing the distorted capped octahedral geometry.

An analogous Tc(IV) complex, the tris(dithiocarbamate) dimethylphenylphosphine complex $[Tc(PMe_2Ph)(S_2CNEt_2)_3](PF_6)$, synthesized from $[TcCl_3(PMe_2Ph)_3]$ has recently been reported; however no crystal structure has been reported for a comparison of the bonding parameters.[16]

Summary

In conclusion, we have prepared the first fully characterized technetium xanthate complexes. The functionally versatile, neutral, seven-coordinate complexes show potential as radiopharmaceuticals using the short-lived isotope ^{99m}Tc. The results of the biological studies will be reported elsewhere.

Acknowledgment. We thank Dr. Catherine E. Costello and Chen-Hui Zeng of the NIH Northeast Regional Mass Spectrometry Laboratory for the FAB mass spectra, Dr. Nadine de Vries for her assistance with the ^{99}Tc NMR spectra, and Dr. William M. Davis for his help with the crystallography.

Supplementary Material Available: Listings of final positional parameters for $[Tc(PPh_3)(S_2COBu^n)_3]$ including hydrogen atoms (Table SI), anisotropic thermal parameters (Table SII), intramolecular bond lengths involving non-hydrogen and hydrogen atoms (Tables SIII and SIV), intramolecular bond angles involving non-hydrogen and hydrogen atoms (Tables SV and SVI), and complete X-ray data collection parameters (Table SVIII) (8 pages); listings of final observed and calculated structure factors (Table SVII) (20 pages). Ordering information is given on any current masthead page.

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Reactivity of Coordinated Phosphate Esters: Pentaamminecobalt(III) Complexes

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The reactivity of two phosphate ester complexes designed to test the efficacy of different modes of activation of phosphate esters by metal ions has been investigated. Ethyl 4-nitrophenyl phosphate coordinated to the pentaamminecobalt(III) moiety liberates nitrophenolate in basic solution 10^6-fold faster than the free phosphodiester. The reaction proceeds via attack of coordinated amido ion to yield a four-membered N,O chelate phosphoramidate ethyl ester. The four-membered chelate does not display the enhanced reactivity of the five-membered-ring cyclic ethylene phosphates but decays with cobalt–ligand bond rupture and yields finally free ethyl phosphoramidate. The aminolysis is accompanied by some loss of ethyl 4-nitrophenyl phosphate by the $S_N1(CB)$ mechanism. The binuclear complex (μ-nitrophenyl phosphato)decaamminedicobalt(4+) undergoes aminolysis in basic aqueous media, also by intramolecular attack of coordinated amido ion. The reaction proceeds some 10^2-fold faster than the analogous aminolysis of the mononuclear complex, (4-nitrophenyl phosphato)pentaamminecobalt(1+). The reaction is also accompanied by some $S_N1(CB)$ loss of the intact ligand; in this case, the ligand is the mononuclear complex. This study illuminates some of the modes by which metal ions can enhance the reactivity of phosphate esters. In agreement with other studies, the electrostatic and inductive effects are estimated to contribute $\sim 10^2$-fold to the rate enhancement, while the intramolecularity of the reaction is responsible for the remainder of the observed rate enhancement.

Introduction

Many energy-requiring reactions in living systems obtain their energy from the lysis of phosphoanhydride bonds. The lysis of many phosphate ester, anhydride, and amidate bonds in biological systems is mediated by enzymes that have as a striking feature an almost ubiquitous requirement for metal ions for activity.[1] As part of an ongoing study into the effect of metal ions on the reactivity of phosphate derivatives, the metal complex ions (ethyl 4-nitrophenyl (phosphato)pentaamminecobalt(2+) and (μ-4-nitrophenyl phosphato)decaamminedicobalt(4+) have been synthesized and their reactivity studied.

The (4-nitrophenyl phosphato)pentaamminecobalt(1+), (fluorophosphato)pentaamminecobalt(1+), and (2,4-dinitrophenyl phosphato)pentaamminecobalt(1+) ions studied previously[2-4] react in hydroxide ion solution to yield significant amounts of 4-nitrophenolate, fluoride, and 2,4-dinitrophenolate ions, respectively, by attack of deprotonated ammonia at the phosphorus center. The reaction proceeds via the chelate phosphoramidate, incorporating the phosphorus into a strained four-membered ring. These observations raised several interesting possibilities with regard to metal ion activated phosphoryl transfer.

The demonstration that a cis-coordinated amido ion was an effective nucleophile toward phosphorus centers of coordinated phosphate esters raised the prospect of synthesis of a chelated phosphoramidate ester. Such an ester should be formed, at least as an intermediate, during the aminolysis of a coordinated phosphodiester. The reactivity of this class of compound is of interest because of the presence of the strained four-membered ring incorporating the phosphorus atom. This ring strain might give rise to an increase in the reactivity at the phosphorus atom by lowering the energy of the transition state for the $S_N2(P)$ reaction,[5-7] as observed for the reactions of five-membered-ring organic phosphate esters.[8] The prospect of observing substantial reactivity with phosphate esters involved in four-membered rings

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prompted the synthesis of, and hydrolytic studies on, the (ethyl 4-nitrophenyl phosphato)pentaamminecobalt(2+) ion.

Many phosphoryl transfer enzymes require two or more metal ions for activity,[1,9] and the decaamminedicobalt system provides an ideal opportunity to study the effect that two-coordinated metal ions can have on the reactivity of a phosphate ester.

Experimental Section

All chemicals used were analytical grade unless otherwise stated; 4-nitrophenol was recrystallized from ethanol before use.

Electronic spectra were recorded with either a Cary 118C or a Hewlett Packard 8450A spectrophotometer.

^{1}H NMR spectra were recorded with a JEOL FX-200 instrument operating at 200 MHz and referenced with respect to DSS (in D_2O) or TMS (in other solvents). ^{31}P NMR spectra were recorded with JEOL FX-60 and Bruker CXP-200 instruments at 24.21 and 80.98 MHz, respectively, and referenced with respect to external 85% H_3PO_4 (downfield shift is positive).

$HOP(O)(OC_2H_5)(OC_6H_4NO_2)$ was prepared as described.[10]

Synthesis of $[(NH_3)_5CoOP(O)(OC_2H_5)(OC_6H_4NO_2)](ClO_4)_2\cdot H_2O$. To $[(NH_3)_5CoOSO_2CF_3](CF_3SO_3)_2$ (2.1 g) in dry sulfolane was added ethyl 4-nitrophenyl hydrogen phosphate (2.0 g) and several drops of tri-*n*-butylamine. The solution was stirred at 37 °C for 3 h, and the sulfolane and excess phosphate ester were extracted twice with ether. The solid thus obtained was dissolved in water (1600 mL) and absorbed on a Sephadex SP C25 (Na^+ form) column. The column was eluted with a 0.1 M $NaClO_4$ solution, with two minor bands that eluted first being discarded, and then elution with 0.2 M $NaClO_4$ yielded the major band containing the desired product. The solution containing the product was evaporated to ~40 mL and cooled at 4 °C for 16 h. The complex crystallized as fine red needles, which were washed with ethanol and ether and dried in vacuo. Yield: 1.52 g. Anal. Calcd for $C_8H_{26}N_6Cl_2CoO_{15}P$: C, 15.81; H, 4.32; N, 13.84; Cl, 11.68; Co, 9.71; P, 5.10. Found: C, 15.8; H, 4.2; N, 13.7; Cl, 11.6; Co, 9.5; P, 5.1. ^{1}H NMR (D_2O): δ 1.05 (d of tr; J = 1.0 Hz, 7.3 Hz; 3 H), 3.87 (d of quart;[11] $J_{H-H} \sim J_{P-H} \sim 7.3$ Hz; 2 H), 7.15 (d; J = 9.3 Hz; 2 H), 8.09 (d; J = 9.3 Hz; 2 H). ^{31}P{H}NMR (H_2O/D_2O): δ +1.3 (s). ϵ^{max}_{517} = 58.9 M^{-1} cm^{-1}; $\epsilon^{max}_{282} = 9.27 \times 10^3$ cm^{-1}.

Synthesis of $[(NH_3)_5CoOP(O)(OC_6H_4NO_2)OCo(NH_3)_5](ClO_4)_4\cdot 1^1/_2NaClO_4\cdot H_2O$. To $[(NH_3)_5CoOSO_2CF_3](CF_3SO_3)_2$ (4.0 g) in sulfolane (25 mL) was added $(HO)_2P(O)(OC_6H_4NO_2)$ (0.5 g) and 2,4,6-collidine (0.75 mL). The solution was heated to 60 °C for 2 h. The sulfolane was extracted 3 times with ether and the residue dissolved in water (500 mL) and absorbed on a Sephadex SP C-25 (Na^+) column. The column was eluted with aqueous $NaClO_4$ (0.3 M), and bands corresponding to $[(NH_3)_5CoOP(O)_2(OC_6H_4NO_2)]^+$ and $[(NH_3)_5CoOH_2]^{3+}$ were observed. The desired product eluted slowly with 0.5 M $NaClO_4$. This band was collected, evaporated to ~80 mL, and then stored at 4 °C for several days. The red needles of product that formed were collected and washed 5 times with methanol, three times with ether, and then dried in vacuo. Yield: 0.25 g. Anal. Calcd for $C_6H_{36}N_{11}Cl_{5.5}Co_2Na_{1.5}O_{29}P$: C, 6.52; H, 3.29; N, 13.95; Cl, 17.65; Co, 10.65; P, 2.80. Found: C, 6.4; H, 3.4; N, 14.3; Cl, 17.6; Co, 10.7; P, 2.7. ^{1}H NMR (DMSO-d_6) δ 8.18 (d; J = 9 Hz, 2 H), 7.29 (d; J = 9 Hz; 2 H), 3.89, 3.73 (br; 30 H). ^{31}P{H} NMR (H_2O/D_2O): δ +12.6 (s).

Kinetics and Further Experiments. $[(NH_3)_5CoOP(O)(OC_2H_5)(OC_6H_4NO_2)](ClO_4)_2\cdot H_2O$. The kinetics of hydrolysis of the complex was followed spectrophotometrically in aqueous solution by monitoring the release of 4-nitrophenolate at 400 nm. The reaction was followed under pseudo-first-order conditions at a constant ionic strength, 1.0 M ($NaClO_4$). Two methods for initiation of the reaction were used. The first involved use of a rapid hand mixing device, which was required by the rapid rate observed at the higher hydroxide concentrations. A solution of known concentration of the complex (~10^{-4} M) in CO_2-free water was placed in one thermostated compartment; in the other, a $NaOH/NaClO_4$ solution of double the required final concentration. The device mixes equal volumes of these solutions and injects the mixed solution into a thermostated flow-through cell in the light path of the spectrophotometer. The second method involved injection of a solution of the complex of known concentration (10 μL, ~10^{-4} M) into a $NaOH/NaClO_4$ solution (2.00 mL) at the required temperature. The solution was rapidly mixed and placed in the thermostated cell holder of the spectrophotometer. Both methods gave identical results. The data were processed by using a nonlinear least-squares package (LSTSQR). The data sets fitted well to single exponential functions. The yield of 4-nitrophenolate was calculated from the final absorbance of the reaction mixture by using a molar absorbtivity of 18 700 M^{-1} cm^{-1} at 400 nm.

The hydrolysis of $[(NH_3)_5CoOP(O)(OC_2H_5)(OC_6H_4NO_2)]^{2+}$ was also followed by ^{31}P NMR spectroscopy. $[(NH_3)_5CoOP(O)(OC_2H_5)(OC_6H_4NO_2)](ClO_4)_2\cdot H_2O$ (60 mg) and NaCl (82 mg) were dissolved in H_2O (1.35 mL) containing Na_3PO_4 (0.02 M) and D_2O (0.4 mL). The solution was cooled in an ice bath and NaOH (0.25 mL, 2 M) added, and then it was placed in the probe of the spectrometer at 5 °C and consecutive spectra accumulated. (Acquisition parameters: acquisition frequency 80.98 MHz, spectral width 9 kHz, acquire 8K data points, zero fill to 16K points, pulse repetition time 0.5 s, 120 scans per spectrum.)

The second step of the reaction was also followed by ^{31}P NMR spectroscopy at 25 °C. $[(NH_3)_5CoOP(O)(OC_2H_5)(OC_6H_4NO_2)](ClO_4)\cdot H_2O$ (30 mg) was dissolved in H_2O (1.35 mL) and D_2O (0.4 mL) containing Na_3PO_4 (~0.05 M), and NaCl was added to make the final ionic strength 1.0 M. The solution was cooled in an ice bath and then a solution of NaOH (0.25 mL, 1, 2, 4, 6, or 8 M) added. The reaction was allowed to proceed for 5–10 min, and then the solution was warmed to 25 °C and placed in the probe of the CXP-200 spectrometer thermostated at 25 ± 1 °C. ^{31}P NMR spectra were accumulated every 5 min for ~1 h. (Acquisition parameters: acquisition frequency 80.98 MHz, spectral width 9 kHz, accumulate 8192 data points, transform to 16K data points, pulse angle 90°, pulse repetition time 0.5 s, number of scans per spectrum 600). The spectra were integrated, and the integral of the signals was normalized with respect to the integral of the standard (PO_4^{3-}). The rate of decay of the signal of interest was determined by plotting the log of the normalized integral of the signal versus time.

$[(NH_3)_5CoOP(O)(OC_2H_5)(OC_6H_4NO_2)](ClO_4)_2\cdot H_2O$ (50 mg) was dissolved in $H_2{}^{18}O$ (0.9 mL, 9.8% ^{18}O) and cooled to ~0 °C in an ice bath, NaOH (0.1 mL, 5 M) was added, and the reaction was allowed to proceed in the ice bath for 10 min. The solution was then warmed to ~27 °C and allowed to react for a further 30 min. The solution was passed through a Dowex 50W-X2 (Na^+) column (0.4 cm × 3 cm). The column was then washed with D_2O (1 mL) and the effluent and washings combined in a 10-mm NMR tube and a ^{31}P NMR spectrum recorded. (Acquisition parameters: acquisition frequency 80.98 MHz, spectral width 2 kHz, accumulate 16K data points, pulse angle 90°, pulse repetition time 4.1 s, 300 scans, resolution enhancement by trapezoidal multiplication).

The rate of hydrolysis of the uncoordinated ligand was determined at 25 °C, μ = 1.0 M ($NaClO_4$) by the initial rate method. The reaction was monitored at 400 nm. The initial concentrations of both $^-(O)_2P(OC_2H_5)(OC_6H_4NO_2)$ and NaOH were varied from run to run. Only data from the first ~1% of the reaction were used in the calculation of the initial rate.

$[(NH_3)_5CoOP(O)(OC_2H_5)(OC_6H_4NO_2)](ClO_4)_2\cdot H_2O$ (40 mg) was dissolved in H_2O (1.0 mL) at ~0 °C in an ice bath, and ice cold NaOH (1.0 mL, 1.0 M) was added and the reaction allowed to proceed at ~0 °C for 10 min. Concentrated HCl (0.5 mL) was then added; after 2 min the solution was diluted to 200 mL and the cationic products were absorbed on a Sephadex SP-C-25 (Na^+ form) column (0.8 × 8.0 cm). Elution with 0.1 M NaCl yielded apparently only two bands; a fast moving purplish band (~80% of the product) and a slow moving red product (~20%). The major purplish band was collected and reduced in volume, and ^{31}P NMR spectra were recorded of the product(s) at various pH's. In another experiment, the major band was collected, and after the pH was adjusted to ~10 by addition of NH_3, the solution was reduced by the addition of $Co(ClO_4)_2\cdot 6H_2O$ (~1 mg) and KCN (0.2 g) and a ^{31}P NMR spectrum of the reduced products recorded. The reduction procedure was checked for its effect on phosphoramidate esters by reduction of the complex initially formed by hydrolysis at 0 °C before addition of HCl, where the identity of the phosphorus-containing ligand was known.

$[(NH_3)_5CoOP(O)(OC_6H_4NO_2)OCo(NH_3)_5](ClO_4)_4\cdot 1^1/_2NaClO_4\cdot H_2O$. A solution of known concentration of the complex $[(NH_3)_5CoOP(O)(OC_6H_4NO_2)OCo(NH_3)_5]^{4+}$ (10 mL, ~2×10^{-2} M) was added to a $NaOH/NaClO_4$ solution (2.00 mL) in a spectrophotometric cell at the required temperature, and the new solution was rapidly mixed and placed in the spectrophotometer. The change in absorbance at 400 nm was monitored with time. The data sets were processed by the LSTSQR program; all data sets fitted single exponential functions. The quoted rates and errors are the mean and standard deviation of at least three determinations. The yield of 4-nitrophenolate was determined from the infinity absorbance value by using a molar absorbtivity of 18 700 M^{-1} cm^{-1}.

The reaction of $[(NH_3)_5CoOP(O)(OC_6H_4NO_2)OCo(NH_3)_5]^{4+}$ in hydroxide solution was also followed by ^{31}P NMR spectroscopy. $[(NH_3)_5CoOP(O)(OC_6H_4NO_2)OCo(NH_3)_5](ClO_4)_4\cdot 1^1/_2NaClO_4\cdot H_2O$

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Table I. Hydrolysis of $[(NH_3)_5CoOP(O)(OC_2H_5)(OC_6H_4NO_2)]^{2+}$ (μ = 1.0 M ($NaClO_4$))

temp, °C	[NaOH], M	10^3k_{obs}, s^{-1}	yield of 4-nitrophenolate, %	10^3k_{NP}, s^{-1}	10^3k_{CB}, s^{-1}
5	0.05	2.12	76.2	1.62	0.50
15	0.05	7.83	66.2	5.18	2.65
25	0.05	28.5	54.3	15.5	13.0
35	0.05	91.3	42.6	38.9	52.4
25	0.01	5.13	53	2.72	2.41
25	0.025	12.93	53	6.85	6.08
25	0.10	53.1	51	27.1	26.0
25	0.25	135	53	72	63
25	0.50	276	49	135	141

(30–50 mg) was dissolved in H_2O (1.35 mL) and D_2O (0.4 mL) containing Na_3PO_4 (0.015 M) and NaCl to make the final ionic strength 1.0 M. A spectrum was recorded, then NaOH (0.25 mL) of the required concentration was added, and consecutive spectra were recorded at the required temperature, 25 or 15 °C. The integrals of the signals were normalized with respect to the standard (PO_4^{3-}) and then plotted on a log scale versus time to yield rate constants for the reaction. (Acquisition parameters: acquisition frequency 80.98 MHz, sweep width 9 kHz, 8K points per spectrum, zero fill to 16K points, pulse repetition time 0.5 s, 600 transients per spectrum.)

Results

$[(NH_3)_5CoOP(O)(OC_2H_5)(OC_6H_4NO_2)](ClO_4)_2\cdot H_2O$. The complex was synthesized by heating $[(NH_3)_5CoOSO_2CF_3](CF_3SO_3)_2$ and $HOP(O)(OC_2H_5)(OC_6H_4NO_2)$ in an inert solvent and isolated after ion-exchange chromatography. The complex displays a ^{31}P NMR chemical shift of +1.3 ppm, which is ~6 ppm downfield from the ionized free ester, a magnitude expected for monodentate coordination of phosphate derivatives to Co(III).

The hydrolysis of the complex was followed spectrophotometrically at 400 nm over the hydroxide concentration range, 0.01–0.5 M (μ = 1.0 M $NaClO_4$). The release of nitrophenolate followed the rate law $v = k_1[(NH_3)_5CoOP(O)(OC_2H_5)(OC_6H_4NO_2^{2+}][OH^-]$ with a second-order rate constant k_1 of (5.48 ± 0.03) × 10^{-1} L mol^{-1} s^{-1} at 25 °C. The temperature dependence of the reaction was investigated over the range 5–35 °C. The yield of 4-nitrophenolate increased with decreasing temperature. The yield of 4-nitrophenolate at each temperature was determined and the observed rate constant partitioned into rate constants for two competing reactions, one producing 4-nitrophenolate, the other not (Table I). The activation parameters for the two processes were determined by linear regression of plots of ln k versus $1/T$: for the 4-nitrophenolate-producing reaction, $\Delta H^{\ddagger}$ = 75 ± 2 kJ mol^{-1} and $\Delta S^{\ddagger}$ = −27 ± 6 J K^{-1} mol^{-1}; for the conjugate base pathway, $\Delta H^{\ddagger}$ = 108 ± 2 kJ mol^{-1} and $\Delta S^{\ddagger}$ = 82 ± 5 J K^{-1} mol^{-1}.

^{31}P NMR studies on the reaction showed that at ~5 °C the starting material gave two products, one with a chemical shift of 25.6 ppm, chelate ethyl phosphoramidate complex (vide infra) (80 ± 3%), and the free diester, $^-OP(O)(OC_2H_5)(OC_6H_4NO_2)$ (−4.5 ppm).

The origin of the −4.5 ppm ^{31}P NMR signal was verified by the addition of authentic $HOP(O)(OC_2H_5)(OC_6H_4NO_2)$ to the sample and observing the increase in the signal intensity. Not only were the chemical shifts of the product and authentic $^-OP(O)(OC_2H_5)(OC_6H_4NO_2)$ ion identical but the *P*–O–C–*H* coupling constants were also identical.

The rate of decomposition of the chelate in hydroxide solution was followed by ^{31}P NMR spectroscopy at 25 °C. The initial reaction to yield the chelate was conducted at low temperature (~0 °C) to maximize its yield. The rate of loss of the chelate was determined at four hydroxide ion concentrations from 0.25 to 1.0 M and at constant ionic strength of 1.0 M. The rate of its decomposition, ~10^{-3} s^{-1}, was independent of hydroxide concentration in the range studied and was accompanied by the production of some insoluble cobalt oxide.

The question of whether the ring opening reaction proceeds with Co–O or P–O fission is a vital one. To answer this question, a tracer experiment was conducted. The reaction of $[(NH_3)_5CoOP(O)(OC_2H_5)(OC_6H_4NO_2)]^{2+}$ in 0.5 M NaOH was conducted in 8.8% ^{18}O-labeled water. The ultimate phosphorus-containing products, ethyl phosphoramidate and $^-OP(O)(OC_2H_5)(OC_6H_4NO_2)$, were removed together from the reaction mixture by ion-exchange chromatography, and high-resolution $^{31}P\{H\}$ NMR spectra of the two products were recorded at 80.98 MHz (Figure 1). This tracer experiment relies on the fact that ^{18}O-substituted phosphate derivatives display ^{31}P NMR chemical shifts typically between 0.02 and 0.04 ppm upfield relative to the unsubstituted phosphate. The spectrum of $^-OP(O)(OC_2H_5)(OC_6H_4NO_2)$ consisted of a singlet of 0.6 Hz width at half peak height. As expected for a $S_N1(CB)$ reaction, there was no incorporation of ^{18}O label into the product. The spectrum of ethyl phosphoramidate also consisted of a single peak with a half-height width of 2.4 Hz. This makes the observation of the satellite peak more difficult, since it is quite possible that the upfield satellite peak was obscured by the broadness of the major signal. However, close inspection of the peak reveals that it is symmetrical and it appears unlikely that a satellite of 8.8% intensity could be obscured totally under the wing of this peak. The reason for the broadness of the ethyl phosphoramidate resonance is unknown although it may be due to that fact that the quadrupolar nitrogen atom (^{14}N, I = 1) is directly bonded to the P atom. The question of whether the products exchange oxygen or not was not determined directly; however, it is known generally that phosphate esters and amidates exchange oxygen very slowly under the conditions of this experiment.[12]

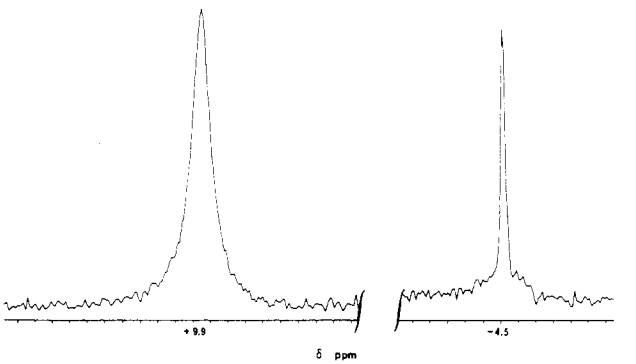

Figure 1. ^{31}P NMR spectrum of the anionic products of the hydrolysis of $[(NH_3)_5CoOP(O)(OC_2H_5)(OC_6H_4NO_2)]^{2+}$ in 8.8% $H_2{}^{18}O$. Conditions are as per the Experimental Section. Scale: 2 Hz/division.

The hydrolysis of the free $^-OP(O)(OC_2H_5)(OC_6H_4NO_2)$ ion was followed at 400 nm by the initial rate method. The reaction in the presence of hydroxide ion obeyed the rate law

$$v = k_1[^-OP(O)(OC_2H_5)(OC_6H_4NO_2)][OH^-]$$

with k_1 = (3.3 ± 0.3) × 10^{-7} L mol^{-1} s^{-1} at 25 °C and μ = 1.0 M. The rate constant includes both attack at phosphorus and at aromatic carbon, and the extent of attack at phosphorus has not been determined in this case, although, in hydroxide ion solution, hydrolysis of the methyl 2,4-dinitrophenyl phosphate ion occurs with 55% P–O cleavage.[13]

$[(NH_3)_5CoOP(O)(OC_6H_4NO_2)OCo(NH_3)_5](ClO_4)_4$ Reactivity. The complex $[(NH_3)_5CoOP(O)(OC_6H_4NO_2)(OCo(NH_3)_5]^{4+}$ was synthesized by heating $(HO)_2P(O)(OC_6H_4NO_2)$ and an excess of $[(NH_3)_5CoOSO_2CF_3](CF_3SO_3)_2$ in an inert solvent and purifying the resulting mixture by cation-exchange chromatography. The desired product was readily separated by virtue of its high charge and crystallized from the perchlorate solution.

The hydrolysis of this complex was followed spectrophotometrically at 400 nm over a range of hydroxide concentrations. The rate of release of 4-nitrophenolate from $[(NH_3)_5CoOP(O)(OC_6H_4NO_2)OCo(NH_3)_5]^{4+}$ was first order in hydroxide up to 0.5 M (Table II), yielding a second-order rate constant of (6.23

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Table II. Hydrolysis of $[(NH_3)_5CoOP(O)(OC_6H_4NO_2)OCo(NH_3)_5]^{4+}$ (μ = 1.0 M ($NaClO_4$))

[NaOH], M	temp, °C	10^3k_{obs}, s^{-1}	yield of 4-nitrophenolate, %
0.1	5	0.346 ± 0.001	70.3
0.1	15	1.43 ± 0.01	66.4
0.1	25	5.83 ± 0.1	59.0
0.1	35	20.7 ± 0.6	49.5
0.01	25	0.53 ± 0.01	
0.05	25	2.87 ± 0.01	
0.2	25	11.9 ± 0.1	
0.5	25	31.5 ± 0.4	

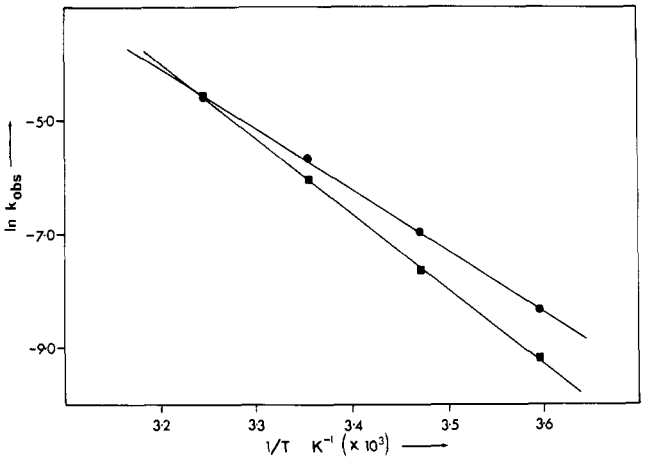


Figure 2. Eyring plot, ln k_{obs} versus $1/T$, for both reactions of $[(NH_3)_5CoOP(O)(OC_6H_4NO_2)OCo(NH_3)_5]^{4+}$ in [NaOH] = 0.1 M: ●, 4-nitrophenolate-producing reaction; ■, $S_N1(CB)$ reaction.

± .07) × 10^{-2} L mol^{-1} s^{-1} at 25 °C and μ = 1.0 M ($NaClO_4$).

The temperature dependence of the reaction was determined, the yield of 4-nitrophenolate decreasing with increasing temperature. The observed rate constant for each hydroxide concentration was partitioned into rate constants for two competing reactions by determining the amount of 4-nitrophenolate produced. A plot of ln k_{obs} versus $1/T$ was linear for both reactions (Figure 2). The activation parameters for the two processes were as follows: 4-nitrophenolate production, $\Delta H^{\ddagger}$ 87 ± 2 kJ mol^{-1} and $\Delta S^{\ddagger}$ = −1 ± 4 J K^{-1} mol^{-1}; the other reaction, $\Delta H^{\ddagger}$ = 108 ± 2 kJ mol^{-1} and $\Delta S^{\ddagger}$ = 67 ± 6 J K^{-1} mol^{-1}.

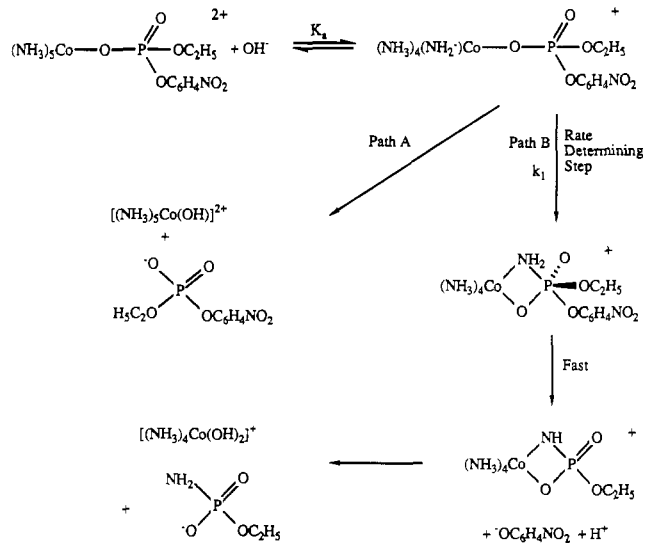
The reaction was also followed by ^{31}P NMR spectroscopy. The product of the non-4-nitrophenolate-releasing pathway had a chemical shift identical with that of the $[(NH_3)_5CoOP(O)_2(OC_6H_4NO_2)]^+$ ion.[2] This is consistent with the previous results, where the $S_N1(CB)$ loss of the phosphate ligand is competitive with 4-nitrophenolate release[2] except that, in this case, the ligand lost is actually another metal ion complex. The product, $[(NH_3)_5CoOP(O)_2(OC_6H_4NO_2)]^+$, does not react further on the time scale of these experiments, as was expected.[2]

The species initially observed in the 4-nitrophenolate-producing pathway ($\delta(^{31}P)$ 17.9) reacts more slowly to yield a compound with a chemical shift of 18.6 ppm. The 18.6 ppm signal is coincident with the signal of added $[(NH_3)_5CoO_3PNH_2]^+$ synthesized by intermolecular attack of NH_3 on $[(NH_3)_5CoOP(O)_2(OC_6H_4(NO_2)_2)]^+$, as described previously.[4] The identification of the 18.6 ppm signal as an O-bonded phosphoramidate complex suggests that the 17.9 ppm precursor is a binuclear N,O-bridged phosphoramidate species. This is in accord with the observation that coordination of phosphoramidates to Co(III) via the NH_2 group leads to an upfield shift of the order of 1 ppm.[2,4]

Discussion

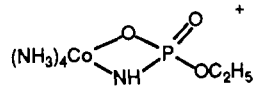
$[(NH_3)_5CoOP(O)(OC_2H_5)(OC_6H_4NO_2)](ClO_4)_2\cdot H_2O$. A mechanism for the decay of the diester complex that fits the rate law, the bifurcated path, the ^{31}P NMR spectroscopy, and the tracer experiment is shown in Scheme I. The non-nitrophenolate-yielding path gave $^-OP(O)(OC_2H_5)(OC_6H_4NO_2)$ as the only phosphorus-containing product observed by ^{31}P NMR; the cobalt-containing product was presumably $[(NH_3)_5CoOH]^{2+}$, and

Scheme I



such a path is readily accounted for by the common $S_N1(CB)$ mechanism of cobalt(III) amines[13] and subsequent decomposition of $(NH_3)_5CoOH^{2+}$ in strong base.

The initial product of the 4-nitrophenolate-producing reaction was a species that had a chemical shift of +25.6 ppm. The identity of this product was assigned, by analogy with results from the $[(NH_3)_5CoOP(O)_2(OC_6H_4NO_2)]^+$ system, to the chelate



The complex is presumed to be a monocation (deprotonated on the bridging N), since it would be expected to be more acidic than chelate phosphoramidate, which has a $pK_a \sim 13.1$, and its ^{31}P NMR chemical shift is also independent of hydroxide concentration in the range 0.25–1.0 M unlike that of the chelate phosphoramidate.[4] The assignment of the initial product as the chelate is supported by its hydrolytic behavior. In hydroxide solution, the complex decomposed slowly to yield ethyl phosphoramidate (δ +9.5) and cobalt oxide. Despite numerous attempts, the complex was never isolated as a pure solid, and its identity was inferred from the experiments discussed below. The presence of the P–N bond in the decomposition product suggests that this bond also existed in the precursor complex. When the complex, initially produced in the base hydrolysis at ~0 °C (δ 25.6), was hydrolyzed in acid (2.5 M HCl) and chromatographed on a cation-exchange column (Sephadex SP-C25), ^{31}P NMR spectra of the cationic products (pH 2–3) showed two triplets (J ~ 6 Hz) at +8.5 and +7.0 ppm. When these products were reduced with $[Co(CN)_6]^{4-}$ (produced in situ), two products were observed in the ^{31}P NMR spectrum; the signals, of approximately equal intensity, at +9.5 and +3.9 ppm, correspond to ethyl phosphoramidate and ethyl phosphate, respectively. The reduction procedure does not affect the integrity of phosphoramidates; reduction of the complex initially formed by base hydrolysis at 0 °C (δ 25.6) yields only ethyl phosphoramidate. These experiments indicate that the phosphorus-containing products eluted from the cation-exchange column were a N-bonded ethyl phosphoramidate complex and an ethyl phosphate complex. The presence of ethyl phosphate in the reduced solution implies that the acid hydrolysis of the precursor complex (δ 25.6) yielded an ethyl phosphate complex; this in turn implies that the precursor complex possessed the Co–O–P linkage. The presence of ethyl phosphoramidate in the reduced solution shows that its immediate precursor complex was N-bonded, since free phosphoramidates are hydrolyzed extremely rapidly in acidic conditions.[15] Coordination of phosphoramidates through the nitrogen protects the P–N bond from

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acid hydrolysis by blocking the protonation site. These results and arguments indicate that both the Co–O–P and the Co–N–P links exist in the intermediate and therefore support the chelate structure.

The previous studies of this type all involved intramolecular aminolysis of coordinated monoesters. In those studies, the chelate phophoramidate in base decayed to the N-bound monodentate phosphoramidate, which then decayed slowly to the free phosphoramidate.[2,3] In this case, the chelate phosphoramidate ester apparently decays to free ethyl phosphoramidate with no intermediate observable by the ^{31}P NMR spectroscopy. That result is not surprising, since ethyl phosphoramidate ion would be expected to be a better leaving group than phosphoramidate ion from the Co(III) center. The rate of decay of the chelate intermediate was followed by ^{31}P NMR spectroscopy at a variety of hydroxide ion concentrations from 0.25 up to 1.0 M, and it was independent of base concentration over the range studied. Such a path can be accommodated, since the chelate complex will be deprotonated at the bridging nitrogen in the experimental conditions used here, i.e. pH >12. Such a deprotonated nitrogen is akin to that required for the $S_N1(CB)$ reaction of cobalt(III) amine complexes.[14] It is possible therefore that this chelate is the reactive deprotonated coordinated amine complex required for the $S_N1(CB)$ process, and it leads first to Co–O bond rupture followed by rapid Co–N rupture. The rate of release of nitrophenolate from $^-OP(O)(OC_2H_5)(OC_6H_4NO_2)$ in hydroxide ion solution is enhanced more than 10^6-fold upon coordination to the $(NH_3)_5Co$ moiety. The reaction to liberate nitrophenolate occurs via intramolecular attack of coordinated amido ion. If the reaction proceeds as shown in Scheme I, then the derived rate law for appearance of nitrophenolate would be

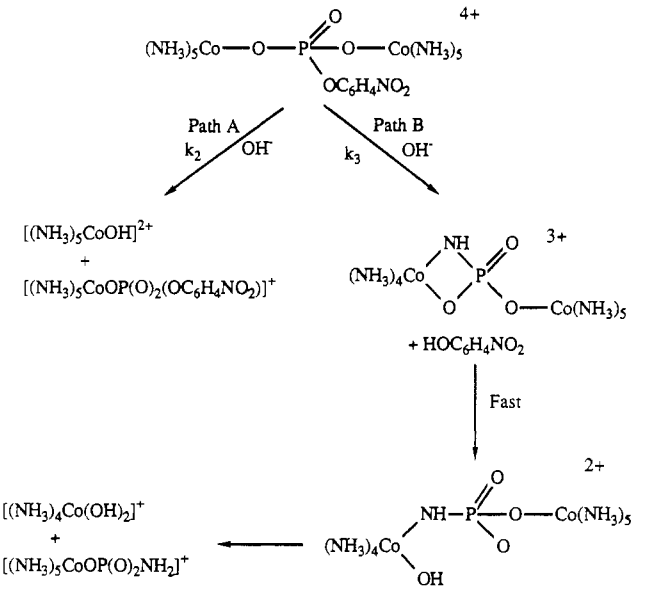
$$v = k_1K_a[(NH_3)_5CoOP(O)(OC_2H_5)(OC_6H_4NO_2)^{2+}][OH^-]$$

From this rate law and with the assumption that the pK_a of Co(III)-bound ammines is ~16 for dipositive complexes,[16,17] the rate of 4-nitrophenolate release from the phosphorus center is ~20 s^{-1}. This is $\sim 10^8$-fold faster than the rate constant for 4-nitrophenolate release from the free ligand in a 1 M hydroxide ion solution. Given the results of previous studies and those here, it is likely that addition of the amido ion and decay of the five-coordinate aminophosphorane ester are near-concerted processes and the aminophosphorane ester approaches an activated complex. At best it will be a very short lived intermediate. The addition of the amido ion therefore is likely to be rate determining.

The attack of cis-coordinated amido ion on the phosphorus center yields a chelated phosphoramidate ester. This molecule should be rather strained; for example the chelate phosphate ion has an in-ring O–P–O angle of 98.7° at the tetracoordinate phosphorus.[6,18] For comparison the corresponding angle[19] in the reactive methyl ethylene phosphate is 99°. The replacement of an oxygen with a nitrogen in the strained ring is not expected to relieve the ring strain to any great degree. However, the ^{18}O tracer experiment shows that reaction of the complex almost certainly proceeds with Co–Ligand bond rupture; i.e., there was little or no reaction at the strained phosphorus center. By way of comparison, the strain at the N–Co–O angle is likely to be substantial (~76°) compared with the strain-free angle (90°).[6,18]

These results may call into question the assumption,[5,6,7] which until now has gone unchallenged, that the chelate phosphate ester will be extremely reactive. It should be pointed out, however, that the observations made herein, i.e. that the chelate ethylphosphate is apparently unreactive, must be tempered with the understanding

Scheme II. Proposed Reaction Sequence for the $[(NH_3)_5CoOP(O)(OC_6H_4NO_2)OCo(NH_3)_5]^{4+}$ Ion in Hydroxide Solution



that the relatively rapid cobalt–ligand bond rupture may be obscuring some slightly slower reaction involving the strained phosphorus center, and this problem is addressed elsewhere.[10]

$[(NH_3)_5CoOP(O)(OC_6H_4NO_2)OCo(NH_3)_5](ClO_4)_4$ Reactivity. The hydrolysis of the complex was first order in both complex and hydroxide ion, and as expected, the reaction proceeded via two paths (Scheme II). The $S_N1(CB)$ path yielded $[(NH_3)_5CoOH]^{2+}$ and $[(NH_3)_5CoOP(O)_2(OC_6H_4NO_2)]^+$. The other path liberated nitrophenolate ion, and by analogy with the previous reactions the initially expected phosphorus-containing product would be the N,O chelate phosphoramidate with an attached pentaamminecobalt moiety. This species might be expected to ring open to the N,O-bridging phosphoramidate. The chemical shift of such a ring-opened species was predicted to be ~16–18 ppm.[20] Thus, the signal at 17.9 was tentatively assigned as the N,O-bridging phosphoramidate. In a slower subsequent reaction the N,O-bridging phosphoramidate (δ 17.9) was lysed to yield a species with a ^{31}P NMR chemical shift of ~19 ppm. Previous experience suggested that a chemical shift change of this magnitude and direction was due to the loss of the Co(III) ion coordinated via the nitrogen of the phosphoramidate; i.e., the new species was an O-coordinated phosphoramidate complex. This was confirmed by the addition of a sample of genuine[4] $[(NH_3)_5CoOP(O)_2NH_2]^+$ to the solution and observing the increase in the 19 ppm signal. Therefore, the reaction is proposed to proceed as in Scheme II.

The rate constant for the intramolecular aminolysis reaction of this dinuclear bridging phosphate ester species is 3.4×10^{-2} L mol^{-1} s^{-1} at 25 °C and μ = 1.0 M. Using the arguments set out above and the estimated pK_a for the ammine ligands in the 4+ ion of ~15,[17] one can estimate the rate constant for the attack of amido ion on the phosphorus center of $[(NH_3)_5CoOP(O)(OC_6H_4NO_2)OCo(NH_3)_5]^{4+}$ to be ~0.3 s^{-1}.

The rate constant for the corresponding reaction of the mononuclear complex was 3.6×10^{-4} L mol^{-1} s^{-1} under the same conditions.[2] This 100-fold increase in rate is due solely to the effect of the additional metal ion. The origin of this effect is probably due to several factors; the additional metal ion coordinated to the phosphate ester must increase the electrophilicity of the phosphorus atom.[21] There is also a statistical effect which is simply that there

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(20) ^{31}P NMR chemical shifts of phosphate derivatives seem to obey simple additivity rules. For example, coordination via oxygen shifts the chemical shift of a phosphate derivative 6–8 ppm downfield and coordination of a phosphoramidate via the N shifts the chemical shift ~1 ppm upfield. O-protonation shifts the chemical shift ~1–2 ppm upfield where N-protonation shifts the chemical shift ~15–18 ppm upfield.

is twice the number of possible nucleophiles in the dinuclear complex; in the absence of other effects this would only double the rate of reaction of the mononuclear complex. In addition to the two effects already mentioned, the difference in charge between the two complexes must effect the pK_a of the ammine ligands, which are required to be deprotonated to act as nucleophiles, but this is spread over the two cobalt centers so the effect is not large. The lower pK_a of the ammines in the dinuclear complex means that at a given hydroxide concentration there is a higher concentration of the active species. This will be offset to some degree by the expectation that as usual for the $S_N2(P)$ reactions of phosphate esters, the rate constant for the reaction will be dependant on the basicity of the nucleophile.[13,22] Thus, the lower basicity of the ammines in the complex $[(NH_3)_5CoOP(O)(OC_6H_4NO_2)OCo(NH_3)_5]^{4+}$ (pK_a estimated to be ~ 15)[17] will mean a lowering of the rate constant for attack of the amido ion.

Attack of the amido ion on the phosphorus center of this dinuclear complex occurs some 80-fold slower than the corresponding reaction for $[(NH_3)_5CoOP(O)(OC_2H_5)(OC_6H_4NO_2)]^{2+}$. This implies that further esterification of a phosphate ester has a greater effect on the electrophilic character of the phosphorus atom than coordination of even a relatively small trivalent metal ion (effective ionic radius for Co(III) is 0.55 Å).[23] Even so, the effect of the additional metal ion leads to a useful increase in the rate of ester hydrolysis, and the effect is similar to that observed in a previous example[24] where two metal ions were coordinated to the same phosphate ester residue, where the intramolecular nucleophile was OH^-.

The chemistry detailed here is typified by the attack of a coordinated amido ion at the phosphorus center, almost certainly as the rate-determining step. The rate enhancement for the intramolecular reaction is of the order of 10^8–10^{10}-fold compared with that of the free ester under the same conditions. Much of this effect comes from the intramolecularity of the nucleophilic attack, but a factor of $\sim 10^2$ can be ascribed to the effect of each metal ion on the P center in promoting the process. These factors are in spite of the strain engendered by the formation of a four-membered chelate ring. They do point to even larger factors for intramolecular paths involving coordinated nucleophiles, which are ideally oriented such as might be achieved in the enzyme cleft. They also point to the reason for the metal ions in the vicinity of substrate in such enzymic systems. The sum of such effects is well on the way to accounting for much of the rate increase observed in the enzymic reactions.

Acknowledgment. We wish to express our thanks to the NMR service of the Australian National University and to the ANU Microanalytical Laboratory.

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Intramolecular Attack of Amido Ion on Phosphate Esters Coordinated to the Pentaammineiridium(III) Moiety

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The phosphodiester metal ion complexes (ethyl 4-nitrophenyl phosphato)pentaammineiridium(III) and bis(4-nitrophenyl phosphato)pentaammineiridium(III) have been synthesized and their reactivities under basic conditions studied. Both complexes react predominantly via intramolecular attack of a deprotonated coordinated ammonia to liberate 4-nitrophenolate ion. The four-membered phosphoramidato chelate ring thus formed rapidly ring opens, probably via P–O and P–N rupture, to yield N-bonded phosphoramidate monoester and O-bonded phosphate ester complexes. The rate constant for intramolecular attack of amido ion is enhanced considerably relative to the reactivity of the uncoordinated substrate under the same conditions. However, the intramolecular reactions are slower in the Ir(III) complexes compared with analogous Co(III) complexes by $\sim 10^3$-fold. The strain induced by chelation of phosphate monoesters by metal ions in enzymes has been proposed as a possible explanation for the large increases observed in the rate of enzymic hydrolysis over the nonenzymic reactions. This study however raises doubt about the supposed reactivity of such strained phosphate ester chelates primarily because the rate of ring opening of the chelate appears to be much greater than that of exocyclic ester hydrolysis.

Introduction

The work of Westheimer and others in the 1950s and 1960s showed that the five-membered-ring cyclic phosphate esters were hydrolyzed up to 10^8-fold faster than the corresponding acyclic esters.[1] The rapid rate of hydrolysis of the cyclic esters was attributed to the strain in the ring, which destabilized the phosphate ester and stabilized the activated complex on the way to the phosphorane intermediate relative to the acyclic ester reactivity. Since that time, several groups of workers have proposed that the enzymic hydrolysis of phosphate monoesters (which require metal ion cofactors[2]) might occur via the intermediacy of the chelate phosphate ester;[3-5] i.e., the metal ion chelates the phosphate ester, forming a highly strained four-membered ring that reacts rapidly for reasons similar to those advanced for the rate of reaction of the organic cyclic phosphate esters.

Several groups of workers have tried to test this hypothesis by attempting the synthesis of chelate phosphate esters and studying the reactivity of such a species.[3,5,6] Several Co(III) complexes of phosphate ion are known,[7] and in instances where chelation is possible the chelate is quite stable in the pH region 4–9; beyond this region the monodentate and fully dissociated complexes are the most stable.[7] These observations encouraged workers to attempt the synthesis of Co(III) chelate phosphate esters. However, none of these attempts have been successful to date.

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