Kinetics and Mechanism of Aquation and Formation Reactions of Carbonato Complexes. V. Carbon Dioxide Uptake by Hydroxopentaamminecobalt(III) Ion to Form Carbonatopentaamminecobalt(III) Ion[1]

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Abstract: The kinetics of the formation of the complex ion $Co(NH_3)_5CO_3^+$ by reaction of carbonate with the ion $Co(NH_3)_5OH_2^{3+}$ has been studied as a function of acidity in the range $7 < pH < 9$ and of carbonate concentration. The data, combined with the findings of an earlier study of the acid-catalyzed elimination of CO_2 from the pentaamminecarbonato species, suggest a direct CO_2 uptake process according to eq 1. The rate studies were made by stopped-flow technique both by an *equilibration method* suggested by the above reaction scheme and by an *acidification method* in which all the carbonate was initially present as free CO_2. The second-order rate constant k_2 has an average value of $(2.2 \pm 0.4) \times 10^2\ M^{-1}\ sec^{-1}$ at 25° and $I = 0.5\ M$, and its temperature parameters are $\Delta H^\ddagger = 15.3 \pm 0.9$ kcal/mol and $\Delta S^\ddagger = 3.6 \pm 3.0$ cal deg^{-1} mol^{-1} over the range 10–25°. This rapid conversion of molecular CO_2 to bound HCO_3^- by direct addition to hydroxyl coordinated to a metal ion is discussed in relation to the reaction of water or free OH^- ion with CO_2 and the catalyzed hydration of CO_2 effected by carbonic anhydrase.

An earlier study in this series[2] has shown that proton-promoted elimination of carbon dioxide from the monodentate carbonato group of the complex ion $Co(NH_3)_5CO_3^+$ is essentially unidirectional at pH ≤ 5, with a rate of similar magnitude to that of the corresponding reactions of bicarbonate ion or monoalkylcarbonates. At higher pH's, the complex ion decomposition becomes noticeably reversible, especially in the presence of excess carbonate.[3] The purpose of the present work was to make a thorough investigation of the kinetics and mechanism of this reverse process.[4] It has been found that the only significant reactants are the hydroxopentaamminecobalt(III) ion and dissolved carbon dioxide. Thus, unlike most anations of the aquopentaamminecobalt(III) species,[5] metal–oxygen bond cleavage need not be involved and a very rapid direct addition reaction is possible. A new approach is provided concerning some aspects of the aqueous solution chemistry of carbon dioxide in which there has been such a long-standing interest,[6] and some support is afforded for previously proposed concepts of the role of the metal ion in the catalyzed hydration of CO_2 by the enzyme carbonic anhydrase.[7]

Experimental Section

Aquopentaamminecobalt(III) perchlorate was obtained from the complex salt $[Co(NH_3)_5CO_3]NO_3 \cdot H_2O$, prepared according to the method of Lamb and Mysels,[8] by acidification with perchloric acid. The complex which crystallized out first was found to be contaminated with NO_3^-. Repeated crystallization from hot 0.1 M $HClO_4$ was performed until the complex was free of nitrate ion.[9] The purity of the complex was checked by microanalysis and by spectral measurement.[10]

Anal. Calcd for $Co(NH_3)_5H_2O(ClO_4)_3$: N, 15.21; H, 3.72. Found:[11] N, 15.37; H, 3.76.

The buffers and other chemicals were of reagent grade, and deionized and degassed water[12] was used in preparing all solutions. The pH determinations were made with a Beckman Research Model meter, utilizing a water-jacketed sample holder which was thermostated at the reaction temperature. The pH meter was standardized before each experiment with stock buffers.[13] Some of the earlier rate measurements were made on a stopped-flow assembly consisting of a Durrum Model D-130 photometer with a Beckman DU spectrophotometer and a Tectronix Model 5103-N oscilloscope, but most of the data were obtained using the complete Model 110 Durrum stopped-flow assembly. The apparatus was equilibrated at the reaction temperature for at least 4–5 hr prior to use and the solutions were kept in the thermostated drive syringe for 5–10 min before each experiment. In some experiments a photographic record of the oscilloscopic tube was made, while in others the long-lived oscilloscope pattern was traced on transparent paper before transferring to graph paper for analysis. All runs were made at 510 nm, which offers the largest absorbance difference between the reactant and the product.[14]

The rates of the reactions were followed by two different methods, as follows.

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[1] Part IV in this series: T. P. Dasgupta and G. M. Harris, *J. Amer. Chem. Soc.*, **93**, 91 (1971).

[2] T. P. Dasgupta and G. M. Harris, *ibid.*, **90**, 6360 (1968).

[3] D. J. Francis and R. B. Jordan, *ibid.*, **89**, 5591 (1967).

[4] A preliminary report of this work was presented at the 163rd National Meeting of the American Chemical Society, Boston, Mass., April 1972, see Inor Abstract No. 58.

[5] (a) A. Haim, *Inorg. Chem.*, **9**, 426 (1970); (b) O. E. Klimek, B. Grossman, and A. Haim, *ibid.*, **11**, 2382 (1972).

[6] R. E. Forster, J. T. Edsall, A. B. Otis, and F. J. W. Roughton, Ed., "CO_2: Chemical, Biochemical, and Physiological Aspects," National Aeronautics and Space Administration, Washington, D. C. (Obtainable from U. S. Government Printing Office, Washington, D. C.)

[7] M. E. Riepe and J. H. Wang, *J. Amer. Chem. Soc.*, **89**, 4229 (1967); *J. Biol. Chem.*, **243**, 2779 (1968); J. E. Coleman, *J. Biol. Chem.*, **242**, 5212 (1967); see also ref 6, p 101.

[8] A. B. Lamb and K. J. Mysels, *J. Amer. Chem. Soc.*, **67**, 468 (1945).

[9] It was essential to use nitrate-free salt, since carbonic anhydrase was an additive in many of the experiments, and NO_3^- interferes with the action of carbonic anhydrase: see S. Lindskog, *J. Biol. Chem.*, **238**, 945 (1963).

[10] The molar absorbance index of our preparation of $Co(NH_3)_5H_2O^+$ is 48.0 at 491 nm as compared to 48.6 reported by C. H. Langford and W. R. Muir, *J. Amer. Chem. Soc.*, **89**, 3141 (1967).

[11] Galbraith Laboratories, Inc., Knoxville, Tenn.

[12] The water was purified by passing laboratory distilled water through an IWT research model deionizer and then boiling for 2 hr to remove all dissolved gases.

[13] The primary standards used were Radiometer precision buffer solutions, Type No. S 1510 and S 1500.

[14] Each stopped-flow experiment recorded in this paper is the result of a succession of runs (up to eight) using the same filling of the drive syringes. In the earlier experiments, each of the runs was analyzed separately and the average of the derived rate constants tabulated, but with the Model 110 system the reproducibility becomes exact within a few runs, and these final repetitively identical traces were utilized for the rate constant determination.

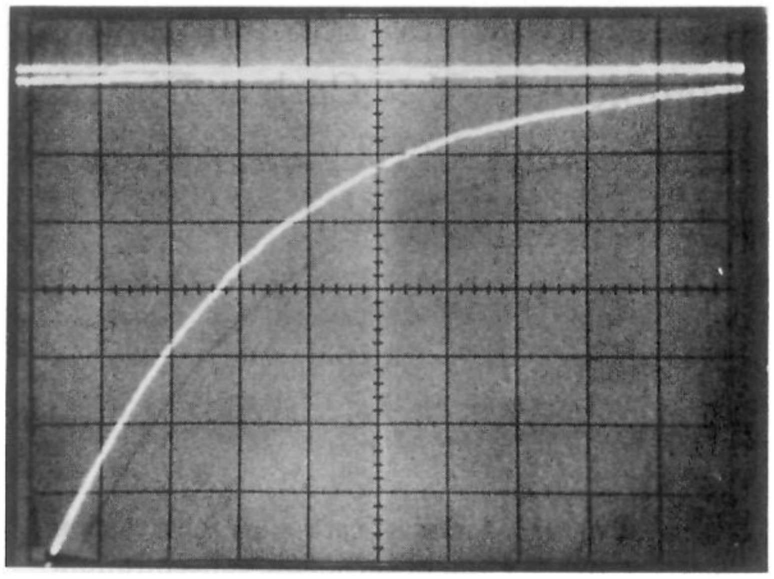

Figure 1. Typical oscilloscope trace of CO_2 uptake at 20° by $Co(NH_3)_5OH^{2+}$ (equilibration method) at pH 7.47, $I = 0.5$ *M* (NaCl), $[HCO_3^-] = 0.05$ *M*, [carbonic anhydrase] = 1 μM. Time scale on abscissa, 1 sec/div. Absorbance on ordinate, arbitrary units.

Table I. Rate of CO_2 Uptake by $Co(NH_3)_5OH^{2+}$ (Equilibration Method); [Carbonate] = 0.05 *M*, [Carbonic Anhydrase] = 1.0 μM, [Complex] = 2.5×10^{-3} *M*, and $I = 0.5$ *M* (NaCl), Phosphate–Citrate Buffer

A. Temperature, 20°				B. Temperature, 25°			
pH	k_{obsd}, sec^{-1}	$10^{-2}k_2$, M^{-1} sec^{-1}	Footnotes	pH	k_{obsd}, sec^{-1}	$10^{-2}k_2$, M^{-1} sec^{-1}	Footnotes
7.11	0.65	1.50		7.12	0.80	1.73	*c, d, e*
7.15	0.63	1.57		7.35	0.66	2.37	*c, d, e*
7.20	0.57	1.53		7.35	0.60	2.07	*c, d, e*
7.30	0.52	1.72		7.36	0.66	2.44	*a, c, e*
7.30	0.48	1.55		7.37	0.58	2.08	*c, d, e*
7.36	0.42	1.50		7.37	0.67	2.56	*a, c*
7.36	0.43	1.55		7.37	0.74	2.94	*a, c*
7.44	0.38	1.61	*a*	7.39	0.52	1.86	
7.47	0.33	1.44		7.40	0.50	1.80	
7.47	0.35	1.56		7.42	0.53	2.09	
7.61	0.27	1.60		7.48	0.45	1.95	
7.61	0.26	1.52		7.51	0.48	2.34	
7.70	0.22	1.56		7.54	0.21	1.01	*f*
7.76	0.20	1.63		7.55	0.41	2.09	*g*
7.76	0.22	1.85		7.72	0.18	1.33	*f*
7.89	0.15	1.62		8.00	0.19	2.74	*c*
7.89	0.16	1.76		8.73	0.035	2.73	*c*
7.90	0.17	1.96		Mean k_2 (25°), (2.3 ± 0.4) × 10^2 M^{-1} sec^{-1}			
7.90	0.15	1.66	*b*				
7.95	0.14	1.76	*b*				
7.96	0.14	1.81	*a*				
7.98	0.15	2.09	*a*				
8.02	0.11	1.56	*c*				
8.05	0.093	1.36	*c*				
8.25	0.071	1.74	*c*				
8.55	0.036	1.78	*c*				
9.15	0.009	2.03	*c*				
Mean k_2 (20°), (1.7 ± 0.2) × 10^2 M^{-1} sec^{-1}							

[a] [Complex] = 1.0×10^{-3} *M*. [b] Borate buffer, 0.05 *M*. [c] Tris buffer, 0.25 *M*. [d] [Complex] = 2.5×10^{-4} *M*. [e] No carbonic anhydrase. [f] Temperature, 15.2°. [g] [Carbonic anhydrase] = 2.0 μM.

Equilibration Method. Weighed amounts of aquopentaammine complex were dissolved either in phosphate–citrate buffer[15] or, in some cases, in 0.5 *M* Tris–HCl buffer,[16] and a 0.1 *M* sodium bicarbonate solution was prepared in a separate flask. The ionic strength of each solution was adjusted to 0.5 *M* with appropriate amounts of $NaNO_3$ or NaCl, and the flasks were thermostated at the required temperature for at least 20 min before introducing into the storage syringes. In runs at the higher pH's, where the equilibrium amounts of CO_2 in the reacting system are not in considerable excess over the complex ion, micromolar amounts of carbonic anhydrase[17] were introduced into the complex solution in order to ensure the maintenance of the CO_2–HCO_3^- equilibrium.[18] In these cases NaCl was utilized to maintain the proper ionic strength.[9] After each series of runs, the pH of the reaction mixture was immediately determined. The pseudo-first-order rate constants, k_{obsd}, were deduced in the conventional fashion from semilogarithmic plots of fraction of reactant remaining *vs.* time as determined from the cleanly exponential oscilloscope traces (see Figure 1 for the record of a typical run).

Acidification Method. The buffered solution of the complex was prepared in the same way as in the equilibrium method and placed in one of the storage syringes. The other reactant was a 0.02 *M* solution of sodium bicarbonate, acidified with perchloric acid to a pH of about 3 in the pH measuring device. Such solutions could not be stored without CO_2 loss, so were freshly made before each series of runs and immediately introduced into the second drive syringe of the stopped-flow machine. After temperature equilibration of the filled drive syringes, runs were made, and the pH of the reaction mixture was determined immediately after each series of runs in the usual manner. No carbonic anhydrase was employed in the *acidification method* runs, since it is essential in these that CO_2 hydrolysis be as slow as possible so that its disappearance by this process be small relative to its uptake by the aquo complex ion, at least during a significant fraction of the latter reaction. Below pH 8, the CO_2 hydrolysis rate is slow enough such that the formation of $Co(NH_3)_5CO_3^+$ goes essentially to completion before the CO_2 concentration has changed appreciably and good first-order plots are obtainable by the conventional procedure. However, above pH 8, the pseudo-first-order rate constants had to be deduced by an initial rate procedure, since the conversion of CO_2 to carbonate by hydrolysis becomes significant during the uptake reaction. For these experiments, the relative absorbance values A_0 at zero time and A_t after a short time interval *t* are determined from the stopped-flow data for the CO_2 uptake reaction. Then relative values of A_∞ are obtained from blank stopped-flow runs using $Co(NH_3)_5CO_3^+$ solutions at the same temperature, complex concentration, and pH as for the corresponding CO_2 uptake runs. For small initial reaction times, one thus obtains the pseudo-first-order rate constants by means of the nonlogarithmic approximation to the conventional first-order rate expression, *viz.*

$$k_{obsd} = (1/t)(A_t - A_0)/(A_\infty - A_0)$$

Results and Discussions

In our study of the decarboxylation of the carbonatopentaamminecobalt(III) ion,[2] it was shown that the reactive species is the protonated form of the carbonato complex, which apparently decomposes unimolecularly to give carbon dioxide and the hydroxopentaammine species. The following mechanism would therefore be expected to describe the overall carboxylation–decarboxylation process

$$\begin{tikzcd}
Co(NH_3)_5OH^{2+} \arrow[dddd, shift left, "K_A"'] & + CO_2 \arrow[r, shift right, "k_2"'] \arrow[dd, shift right, "K_1"'] & Co(NH_3)_5CO_3H^{2+} \arrow[l, shift right, "k_1"'] \arrow[dddd, shift right, "K_C"'] \\
& & \\
& HCO_3^- \arrow[uu, shift right] \arrow[dd, shift right, "K_2"'] & \\
& & \\
Co(NH_3)_5OH_2^{3+} \arrow[uuuu, shift left] & CO_3^{2} \arrow[uu, shift right] & Co(NH_3)_5CO_3^+ \arrow[uuuu, shift right]
\end{tikzcd} \qquad (1)$$

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(15) The 0.5 *M* buffer solutions were made up according to the directions given in the paper by P. J. Ewing, J. M. Markwitz, and I. Rosenthal, *Anal. Chem.*, **28,** 1179 (1956).

(16) Tris = tris(hydroxymethyl)amminomethane. The Tris buffer was made by mixing appropriate quantities of Tris–HCl and Tris–base. The mixing table is available from Sigma Chemical Co., St. Louis, Mo.

(17) Bovine carbonic anhydrase, Grade 1, was used, obtained from Miles Laboratories, Inc., Kankakee, Ill.

(18) Maintenance of this equilibrium is essential in the equilibration method, since $[CO_2]$ must remain constant. This is possible with excess HCO_3^- and catalysis of the dehydration–hydration process ensured by carbonic anhydrase. In absence of the enzyme, depletion of CO_2 results since the uncatalyzed equilibration is rather slow under the conditions of these experiments (ref. 6, p 15).

In this, k_1 and k_2 are the rate constants for CO_2 elimination and uptake, respectively, and the K's are the acid dissociation constants of the various species identified. The rate law corresponding to this equilibration process is

$$d[Co(NH_3)_5CO_3H^{2+}]/dt = k_2[Co(NH_3)_5OH^{2+}][CO_2] - k_1[Co(NH_3)_5CO_3H^{2+}] \quad (2)$$

In a buffered solution of complex aquo ion containing excess carbonate, eq 2 describes a pseudo-first-order equilibration if conditions are such that k_1 and k_2 are rate determining. In these circumstances, the following expression for the observed rate constant applies (h = hydrogen ion concentration, b = total carbonate concentration)

$$k_{obsd} = k_1\left(\frac{h}{h + K_C}\right) + k_2b\left(\frac{K_A}{h + K_A}\right)\left(\frac{h^2}{h^2 + K_1h + K_1K_2}\right) \quad (3)$$

The adequacy of this concept can be tested in two ways by use of the data obtained by the *equilibration method* which are recorded in Table I. Since values are known for k_1 and the various equilibrium constants,[2,19–21] one can compute a new variable $k'_{obsd} = k_{obsd} - k_1h/(h + K_C)$ and a plot of this variable against $K_Ah^2b/(h + K_A)\cdot(h^2 + K_1h + K_1K_2)$ at a given temperature should yield a straight line of intercept zero and slope k_2. Such a plot for the *equilibration method* data at 20° (Table IA) is presented in Figure 2. A least-squares analysis of the experimental data yields an intercept value of 0.012 ± 0.005 and $k_2 = (1.51 \pm 0.03) \times 10^2\ M^{-1}\ sec^{-1}$.

The second procedure is simply to calculate all the k_2 values separately by use of eq 3, and the results of such calculations are included in Table I. It is seen that these computed k_2 values are constant both at 20 and 25° within a reasonable margin of error, and that the average value at 20° is in good agreement with the graphical value, again supporting the concepts summarized by eq 1, 2, and 3.

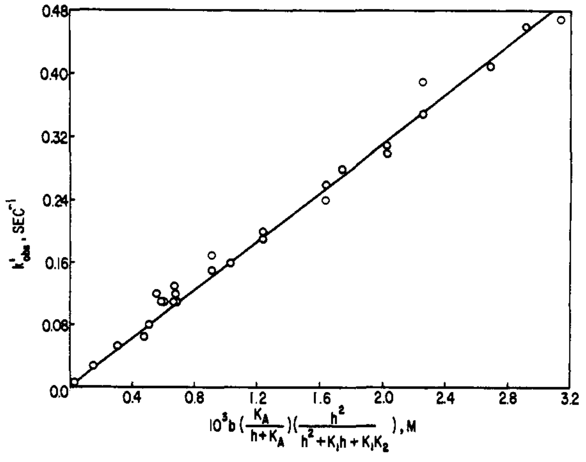

Figure 2. Plot of k'_{obsd} *vs.* $K_Ah^2b/(h + K_A)(h^2 + K_1h + K_1K_2)$ at 20°, $I = 0.5\ M$(NaCl).

In the *acidification method*, the concentration of carbon dioxide is relatively high throughout the period of rate measurement, so that the last term of eq 2 may be ignored, and the observed rate constant is given by the expression

$$k_{obsd} = k_2bK_A/(h + K_A) \quad (4)$$

The data obtained by this method are summarized in Table II, and it is seen that the average values at 20 and 25° are in very good agreement with those obtained by the other technique and recorded in Table I. The Eyring temperature parameters were determined by a computerized weighted least-squares analysis[22] of all the data presented in Tables I and II, which include some results at lower temperatures, in the form of a log (k/T) *vs.* ($1/T$) function. This leads to values of the enthalpy

Table II. Rate of CO_2 Uptake by $Co(NH_3)_5OH^{2+}$ (Acidification Method); $[CO_2] = 0.01\ M$, [Complex] = $2.5 \times 10^{-3}\ M$, and Ionic Strength = 0.5 M (NaCl), Phosphate–Citrate Buffer

A. Temperature, 20°				B. Temperature, 25°			
pH	k_{obsd}, sec^{-1}	$10^{-2}k_2$, M^{-1} sec^{-1}	Footnotes	pH	k_{obsd}, sec^{-1}	$10^{-2}k_2$, M^{-1} sec^{-1}	Footnotes
6.82	0.92	1.47		6.82	1.61	2.58	
6.82	0.87	1.39		6.90	1.18	1.77	*c*
6.82	0.74	1.18	*a, b*	6.92	1.17	1.73	*c*
6.88	0.95	1.45	*a, b*	7.02	1.47	2.03	
6.88	0.83	1.27	*a, b*	7.03	1.68	2.30	
6.90	0.81	1.22	*a, b*	7.03	1.54	2.11	
6.98	0.92	1.31		7.10	0.30	0.40	*d*
7.05	1.07	1.46		7.10	0.32	0.42	*d*
7.10	1.07	1.41		7.20	0.78	0.98	*e*
7.13	1.13	1.47		7.20	0.62	0.78	*e*
7.26	1.17	1.43		7.25	1.35	1.65	
7.35	1.16	1.37		7.30	0.76	0.91	*e*
7.43	1.41	1.62		7.30	0.71	0.85	*e*
7.55	1.37	1.52		7.89	2.06	2.16	
8.39	1.40	1.43	*a*				
8.45	1.30	1.31	*a*				

Mean values of k_2 (M^{-1} sec^{-1}): at 20°, $(1.4 \pm 0.1) \times 10^2$; at 25°, $(2.0 \pm 0.3) \times 10^2$

[a] Tris buffer. [b] [Complex], $1.0 \times 10^{-3}\ M$. [c] [Complex], $1.5 \times 10^{-3}\ M$. [d] Temperature, 10.4°. [e] Temperature, 15.2°.

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(19) Comprehensive additional studies in this laboratory (K. Hyde, D. A. Palmer, unpublished) of the aquation of $Co(NH_3)_5CO_3^+$ at pH 3 and $I = 0.5\ M$ ($NaNO_3$) over the temperature range 7–37° have yielded average values for k_1 of 0.42, 0.65, and 1.10 sec^{-1} at 15, 20, and 25°, respectively. These figures have been adopted for the *equilibration method* computations.

(20) The magnitudes of K_1 and K_2 are well known at zero ionic strength and various temperatures (see F. S. Nakayama, *J. Inorg. Nucl. Chem.*, **33**, 1287 (1971)), but data concerning these constants in the range $0.1\ M < I < 0.5\ M$ are very sparse indeed. We have used the best fit equation of J. M. Edmond and J. M. T. M. Gieskes, *Geochim. Cosmochim. Acta*, **34**, 1261 (1970), to compute values for pK_1 at $I = 0.5\ M$ of 6.10, 6.06, and 6.03 at 15, 20, and 25°, respectively. These values are consistent with some other scattered data at nonzero ionic strengths: see G. Schwarzenbach, *Helv. Chim. Acta*, **40**, 907 (1957); also, R. Näsänen, P. Meriläinen, and K. Leppänen, *Acta Chem. Scand.*, **15**, 913 (1961). A value for pK_2 of 9.8 at 25° and $I = 0.5\ M$ is quoted by J. N. Butler and R. Huston, *J. Phys. Chem.*, **74**, 2976 (1970), which we have adopted for this and the lower temperatures. Actually, K_2 contributes negligibly to the calculation of k_2 below pH ~9, so the magnitude assigned to it is not very significant.

(21) We obtain a value for pK_C of 6.7 at 25° and $I = 0.5\ M$, both by titration (ref 2) and from our most recent kinetic data on the acid decomposition of $Co(NH_3)_5CO_3^+$ (see ref 19). Acid–base titration of a sample of our aquopentaamminecobalt(III) complex yields a value for pK_A of 6.6 at 25° and $I = 0.5\ M$, which is consistent with the recently published figure of 6.4 obtained at 25° and $I = 1.0\ M$: F. Yajima, A. Yamasaki, and S. Fujiwara, *Inorg. Chem.*, 10, 2350 (1971). In view of the small temperature variation of K_1 and K_2 (ref 20), we have adopted the fixed values pK_C = 6.7 and pK_A = 6.6 over the temperature range of our experiments.

(22) Computer program provided by Dr. R. J. Buchacek of this laboratory, as obtained from laboratory of Professor G. Gordon, University of Iowa, Iowa City, Iowa.

and entropy of activation of 15.3 ± 0.9 kcal/mol and 3.6 ± 3.0 cal deg^{-1} mol^{-1}, respectively, and a calculated figure for k_2 at 25° of $(2.2 \pm 0.4) \times 10^2$ M^{-1} sec^{-1}.

The results presented and discussed above leave little room for doubt that the CO_2 uptake consists of a simple addition of molecular CO_2 by attachment of carbon to the oxygen atom of the hydroxopentaammine complex ion in a manner closely analogous to what appears to be the mechanism of cyanate addition to the aquo complex ion.[23] The possibility that direct replacement of the OH group on the metal by carbonate takes place is excluded by the principle of microscopic reversibility since the decarboxylation of $Co(NH_3)_5CO_3^+$ in acid solution occurs without Co–O bond fission.[24] However, this does not exclude the possibility of a contribution to the carboxylation process by bimolecular reaction of HCO_3^- with the hydroxopentaammine ion involving an activated complex in which bond making occurs between carbon and the oxygen atom attached to the metal ion in concert with bond fission between carbon and one of the oxygen atoms of the incoming HCO_3^- group. This would be very similar to the formation of nitritopentaamminecobalt(III) ion by addition of N_2O_3 to the hydroxo complex by concerted N–O bond formation and N–N bond cleavage.[25] This possibility was tested by a series of runs at 20°, employing the equilibration method, in which the total carbonate concentration was varied widely, yielding the results given in Table III. A moderate upward trend is noted in the values of k_2 in the systems self-buffered by carbonate, but this deviation is seen to be somewhat less pronounced in the presence of other buffers. It seems probable that the small accelerating effect of a large excess of carbonate is specific to this medium, though a minor contribution to the carboxylation process by direct HCO_3^- addition in the manner just discussed is not excluded. However, the over-riding consideration is that k_2 as derived from eq 3 is a good constant over the range 7.1 < pH < 9.1 at constant low total carbonate concentration (see Table I) although within this range the $[CO_2]$ as fixed by the last term in brackets in eq 3 varies by a factor of nearly 100. It is worth noting that a limited set of unpublished results,[26] obtained recently for this same reaction at 20° and pH ~8 with carbonate self-buffering but at a lower ionic strength, yields k_2 values in fair agreement with our low-carbonate value.

Table III. Rate of CO_2 Uptake by $Co(NH_3)_5OH^{2+}$ (Equilibration Method); Effect of Variation of [Carbonate] at 20°; [Complex] = 2.5×10^{-3} M; $I = 0.5$ M (NaCl); [Carbonic Anhydrase] = 1.0 μM

pH	[Carbonate], M	k_{obsd}, sec^{-1}	$10^{-2}k_2$, M^{-1} sec^{-1}	Footnotes
8.72	0.05	0.028	2.19	*a*
8.92	0.05	0.020	2.66	*b*
8.05	0.10	0.24	2.21	*a*
8.04	0.10	0.22	1.95	*c*
8.04	0.20	0.43	2.04	*c*
7.96	0.25	0.89	2.92	*a*
8.05	0.50	1.55	3.17	*a*
8.17	0.03	0.049	1.26	*d*
8.23	0.045	0.062	1.51	*d*
8.23	0.060	0.083	1.68	*d*
8.15	0.075	0.13	1.87	*d*

[a] Self-buffered by carbonate. [b] Borate buffer, 0.025 M. [c] Tris buffer, 0.25 M. [d] Reference 26.

We are now able to discuss quantitatively the nature of the final equilibrium described by eq 1. At 25°, one derives from our data

$$K_E = \frac{[Co(NH_3)_5OH^{2+}][CO_2]}{[Co(NH_3)_5CO_3H^{2+}]} = \frac{k_1}{k_2} = 5 \times 10^{-3} \quad (5)$$

The absorbance measurements quoted in Table III of ref 3 enable the calculation of a value for K_E at $I = 1.0$ M ($KClO_4$). The figure so obtained[27] as an average from the 11 measurements made is 2×10^{-3}, in quite reasonable agreement with that above, considering the uncertainties in the values[20,21] of K_I and K_C, and the ionic medium differences in the two approaches.

Finally, it is of some interest to compare the second-order rate constants for various naturally occurring CO_2 hydration processes with our k_2 value. The appropriate figures are given in Table IV. One possibly significant factor in comparing the reactivities to carbon dioxide of water, hydroxopentaamminecobalt(III) ion, and hydroxyl ion might be their relative basicities, which are in a ratio[28] of approximately $1:10^7:10^{16}$. The rate constants do fall in this order, but obviously not in any simple proportion, since while OH^- converts CO_2 to carbonate 10^7 times more rapidly than does water, it is only 40 times more effective than the cobalt complex. Obviously, other factors are of significance, per-

Table IV. Rate Constant Data for Various Reactions of Carbon Dioxide at 25° in Aqueous Solution

Reaction	k, M^{-1} sec^{-1}	$\Delta H^{\ddagger}$, kcal/mol	$\Delta S^{\ddagger}$, cal deg^{-1} mol^{-1}	Footnotes
(1) $CO_2 + H_2O \rightarrow H_2CO_3$	7×10^{-4}	18.4	−11.2	*a, b*
(2) $Co(NH_3)_5OH^{2+} + CO_2 \rightarrow Co(NH_3)_5CO_3H^{2+}$	2.2×10^2	15.3	3.6	*c*
(3) $CO_2 + OH^- \rightarrow HCO_3^-$	8.5×10^3	12.7	2.0	*d*
(4) Conversion of CO_2 to HCO_3^- in presence of bovine carbonic anhydrase	$\sim 4 \times 10^7$	~6	~0	*e*

[a] First-order constant is 0.04 sec^{-1} (ref 6, p 20). [b] Converted to second-order constant, assuming $[H_2O] = 55.5$ M. [c] This work. [d] Reference 6, p 21. [e] $k = k_{enz}$, as determined from data given in ref 31, Table I. $\Delta H^{\ddagger}$ and $\Delta S^{\ddagger}$ estimated from data quoted by R. P. Davis in "The Enzymes," Academic Press, New York, N. Y., 1961, p 556.

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[27] Data are given for the initial and final absorbances,[3] from which one may deduce the ratio $[Co(NH_3)_5OH^{2+}]/[Co(NH_3)_5CO_3^+]$ at various pH's at equilibrium in a solution of fixed total carbonate concentration. Use of the constants pK_I and pK_C quoted earlier (ref 20 and 21) enables calculation of the K_E of eq 5, since

$$K_E = \frac{[Co(NH_3)_5OH^{2+}][HCO_3^-]}{[Co(NH_3)_5CO_3^+]}\frac{K_C}{K_I}$$

[28] This ratio is determined by consideration of the equilibrium constants for the reactions: $H_3O^+ + H_2O \rightleftarrows H_2O + H_3O^+$, $Co(NH_3)_5OH_2^{3+} + H_2O \rightleftarrows Co(NH_3)_5OH^{2+} + H_3O^+$, and $H_3O^+ + OH^- \rightleftarrows H_2O + H_2O$.

haps of a steric nature as suggested by the moderately large negative $\Delta S^{\ddagger}$ value for the water reaction as compared to essentially zero $\Delta S^{\ddagger}$ values for the other two species. A recent study of CO_2-catalyzed oxygen exchange of the $Cr(NH_3)_5OH^{2+}$ ion[29] has enabled an estimate of the rate constant for the reaction of this species with CO_2 at 25° and $I = 0.1\ M$. The suggested lower limit of 11 $M^{-1}\ sec^{-1}$ fits satisfactorily into the qualitative pattern with respect to relative basicity noted above, since the pK_a of the chromium(III) aquo complex ion is indeed somewhat smaller[30] than that of its cobalt(III) analog.

Turning now to the carbonic anhydrase catalyzed process, hydroxyl coordinated to zinc has long been believed to be the significant reactant,[7] a concept supported by a recent review of the metal ion function in this enzyme,[31] although a contrary opinion based on nmr data has also appeared.[32] The evidence is strong[31] that the pK of the presumed aquo–zinc(II) complex of the enzyme is probably about 7, very close to that of the cobalt(III) species of our study. It seems quite logical to expect that it is the metalloenzyme in its hydroxo form which does in fact react directly with CO_2 to yield the bicarbonate product as does the pentaamminecobalt(III) analog. However, there is no problem of the bicarbonate product being "trapped" on the metal ion of the metalloenzyme, due to the much greater lability of Zn(II) complexes as compared to Co(III),[33,34] so that rapid "turnover" of CO_2 to free HCO_3^- is possible. Furthermore, the data of Table IV indicate that the major factor facilitating rate acceleration by the metalloenzyme is the greatly decreased enthalpy of activation as compared to either $Co(NH_3)_5OH^{2+}$ or the free OH^- ion. Thus one need not invoke special stereochemical factors to explain the enzyme activity other than an ability to get the reacting species together effectively, which is of course the basic assumption of the Michaelis–Menten mechanism.[31] Nevertheless, a conclusion along these lines cannot be very definitive until more comprehensive data are available concerning the temperature parameters of the enzyme-catalyzed reaction.

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Aminolysis of Cobalt(III) Activated Esters. Stabilization of an Amine–Alcohol Intermediate

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Abstract: The reaction between isopropylglycinate bis(ethylenediamine)cobalt(III) perchlorate and glycine ethyl ester in dimethyl sulfoxide to form ethylglycylglycinate bis(ethylenediamine)cobalt(III) perchlorate and isopropyl alcohol occurs in two stages. The first reaction follows a rate law $k_{obsd} = k_1$[amine] with $k_1 = 14 \pm 1\ M^{-1}\ sec^{-1}$ at 25°, $\mu = 0.015$, and is attributed to addition of glycine ethyl ester to the carbonyl center. The second reaction follows the rate law $k_{obsd} = Kk[\text{amine}]/(1 + K[\text{amine}])$ with $K = 16\ M$, $k = 1.4 \times 10^{-2}\ sec^{-1}$ at 25°, and $\mu = 0.015$. It is accounted for in terms of the general acid catalyzed removal of isopropyl alcohol from the conjugate base of the amine–alcohol intermediate. Infrared and quenching experiments have also been used to identify the intermediate and to investigate some of its properties.

The mechanism of aminolysis of organic esters still appears to be a viable issue. Whether the reaction is concerted or proceeds *via* an addition–elimination process remains uncertain in most instances.[1,2] Certainly amine–alcohol intermediates are required or implied in the addition of amines to some carbonyl centers[3–6] and their properties as a function of pH have been inferred by generating them from different sources and observing the breakdown products.[3,5] Even so in no single case of ester aminolysis has the amine–alcohol intermediate been directly observed and the rates and rate laws for its formation and decay established.

The possibility of observing such an intermediate arises by chelating to a metal an amino acid ester *via* the amino group and carbonyl oxygen atom, structure

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