+ copper(II) peptide in the log $K_{12} < -4$ region fit an inner-sphere model better than they fit a diffusion-limited process. Third, the inner-sphere model provides some rationale for the anomalous behavior of the copper(II) tetra-L-alanine and copper(II) tetra-L-valine complexes due to steric interactions.

The steep slope of the free energy dependence for the copper(II) peptide reductions and the lack of a free energy dependence for the copper(II) peptide oxidations in the log $K_{12} < -4$ region are due to a shift of the rate-determining step away from the electron transfer. In the case of the reduction reactions the separation of the products of the electron transfer, the $Ir^{IV}Cl_6$–copper(II) peptide bridged complex, is rate limiting. The rate of the oxidation reactions is limited by substitution of $IrCl_6^{2-}$ for an axial water on the copper(II) peptide.

Acknowledgment. This investigation was supported by Public Health Service Grant No. GM-12152 from the National Institute of General Medical Sciences and by an Eastman Kodak Fellowship (G.D.O.).

Registry No. $Cu^{II}(H_{-2}A_3)^-$, 42179-70-4; $Cu^{II}(H_{-2}L_3)^-$, 29575-61-9; $Cu^{II}(H_{-3}G_3AOCH_3)^-$, 62882-66-0; $Cu^{II}(H_{-3}G_4a)^-$, 76861-95-5; $Cu^{II}(H_{-3}G_3a)^-$, 62801-35-8; $Cu^{II}(H_{-3}PG_2a)^-$, 76861-96-6; $Cu^{II}(H_{-3}G_4)^{2-}$, 57603-18-6; $Cu^{II}(H_{-3}AG_3)^{2-}$, 76899-38-2; $Cu^{II}(H_{-3}A_4)^{2-}$, 62959-95-9; $Cu^{II}(H_{-3}V_4)^{2-}$, 62959-94-8; $Cu^{III}(H_{-2}G_2\beta A)$, 69814-95-5; $Cu^{III}(H_{-2}Aib_3)$, 69990-31-4; $Cu^{III}(H_{-4}C)^-$, 76721-70-5; $IrCl_6^{2-}$, 16918-91-5; $IrCl_6^{3-}$, 14648-50-1.

Contribution from the Department of Chemistry, Purdue University, West Lafayette, Indiana 47907

Reactions of the Trivalent Copper Complex of a Macrocyclic Tetrapeptide, *cyclo*-(β-Alanylglycyl-β-alanylglycyl)

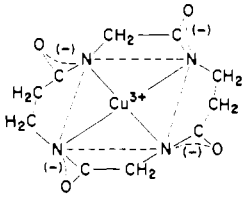
JAMES S. RYBKA and DALE W. MARGERUM*

Received August 1, 1980

The electrode potential for the 14-membered macrocyclic peptide complex $Cu^{III,II}(H_{-4}C)^{1-,2-}$ is +0.48 V. $Cu^{III}(H_{-4}C)^-$ is relatively stable in neutral solution with a half-life of 5.7 weeks at 25.0 °C ($k_d = 2 \times 10^{-7}$ s^{-1}). Its decomposition to copper(II) and oxidized ligand is acid catalyzed and base catalyzed. The latter reaction is first order in [OH^-] and first order in [$Cu^{III}(H_{-4}C)^-$] with $k_{OH} = 1.6 \times 10^{-2}$ M^{-1} s^{-1}. Below pH 1 two intermediate copper(III) macrocyclic species form in rapid succession before a decomposition reaction occurs with a $[H^+]^2$ dependence where $k_{2H} = 1.0 \times 10^{-3}$ M^{-2} s^{-1}. The $Cu^{III}(H_{-4}C)^-$ complex undergoes photochemical decomposition with a quantum yield of 8×10^{-3} at 278 nm. Electron-transfer reactions of $Cu^{II}(H_{-4}C)^{2-}$ with other Cu(III) peptides are rapid. These cross reactions lead to a calculated self-exchange rate constant of 6.0×10^4 M^{-1} s^{-1} for $Cu^{III}(H_{-4}C)^-$ and $Cu^{II}(H_{-4}C)^{2-}$ in 1.0 M $NaClO_4$ at 25.0 °C.

Introduction

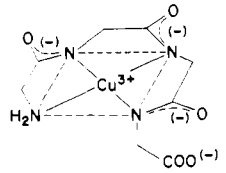
Recent studies have shown that the macrocyclic tetrapeptide C (*cyclo*-(β-alanylglycyl-β-alanylglycyl)) reacts with divalent copper to form a quadruply deprotonated peptide complex.[1] The macrocycle helps to stabilize the $Cu^{II}(H_{-4}C)^{2-}$ complex, slowing down both acid attack on the deprotonated ligand and attack by other ligands on the metal. In the present work the macrocyclic complex is oxidized electrochemically to give the trivalent copper complex $Cu^{III}(H_{-4}C)^-$ with the proposed structure given in I. The electrode potential is only +0.48



I

V for this Cu(III)–Cu(II) couple in accord with a low potential expected[2] due to the presence of four deprotonated peptide donors. The $Cu^{III}(H_{-4}C)^-$ complex is characterized in regard to its UV–visible spectrum, its rate of redox decomposition as a function of pH, its photochemical decomposition, and its rate of electron-transfer reactions.

The trivalent oxidation state of copper has been observed in a number of compounds, many of which are not stable in aqueous solution.[3-6] Stabilization of the higher oxidation state by deprotonated peptide nitrogen donors makes copper(III) more easily accessible and long-lived.[2,7-10] The copper(III)–tetraglycine complex $Cu^{III}(H_{-3}G_4)^-$ (structure II) is



II

stable in neutral solution with a half-life of 5.5 h.[11] The present macrocyclic peptide complex of copper(III) is about 200 times slower in undergoing redox decomposition under the same conditions. The reaction rate is both acid catalyzed and base catalyzed, giving rise to a U-shaped pH profile for the decomposition reaction. However, under all conditions it is slower to decompose than is $Cu^{III}(H_{-3}G_4)^-$.

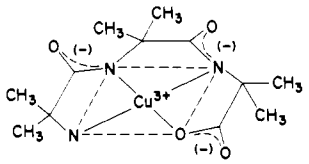
Experimental Section

Reagents. The characterization of the 14-membered macrocyclic peptide (C) and the preparation of the copper(II) complex have been previously described.[1] Electrochemical oxidation to form $Cu^{III}(H_{-4}C)^-$ was accomplished with use of a flow electrolysis apparatus.[10] The

(1) Rybka, J. S.; Margerum, D. W. *Inorg. Chem.* **1980**, *19*, 2784–2790.
(2) Bossu, F. P.; Chellappa, K. L.; Margerum, D. W. *J. Am. Chem. Soc.* **1975**, *97*, 6894–6896.
(3) Scholder, R.; Voelskow, U. *Z. Anorg. Allg. Chem.* **1951**, *266*, 256–268.
(4) Gray, E. T., Jr.; Taylor, R. W.; Margerum, D. W. *Inorg. Chem.* **1977**, *16*, 3047–3055.
(5) Olson, D. C.; Vasilevskis, J. *Inorg. Chem.* **1971**, *10*, 463–470.
(6) Bour, J. J.; Birker, P. J. M. W. L.; Steggerda, J. J. *Inorg. Chem.* **1971**, *10*, 1202–1205.
(7) Margerum, D. W.; Chellappa, K. L.; Bossu, F. P.; Burce, G. L. *J. Am. Chem. Soc.* **1977**, *99*, 2195–2203.
(8) Kirksey, S. T., Jr.; Neubecker, T. A.; Margerum, D. W. *J. Am. Chem. Soc.* **1979**, *101*, 1631–1633.
(9) Margerum, D. W.; Wong, L. F.; Bossu, F. P.; Chellappa, K. L.; Czarnecki, J. J.; Kirksey, S. T., Jr.; Neubecker, T. A. *Adv. Chem. Ser.* **1977**, *No. 162*, 281–303.

0020-1669/81/1320-1453$01.25/0

$Cu^{II}(H_{-4}C)^{2-}$ complex in basic solution, up to 0.1 M NaOH, was oxidized at an applied potential of +0.60 V vs. Ag/AgCl reference electrode. Yields of $Cu^{III}(H_{-4}C)^-$ were nearly 100%, and storage of neutral solutions in the dark can be done for 1 day with less than 2% decomposition. The preparation of the copper(III) complex of the tripeptide of α-aminoisobutyric acid, $Cu^{III}(H_{-2}Aib_3)$ (see structure III), was reported previously.[8] Solutions of $IrCl_6^{2-}$ were prepared from Na_2IrCl_6 as previously described.[12]



III

The solutions for kinetic analysis contained either acetate or borate buffer. Below pH 3 and above pH 11 standard solutions of $HClO_4$ and NaOH were used, respectively. The ionic strength of all solutions was controlled at 1.0 M with $NaClO_4$, which was prepared by the reaction of $HClO_4$ and Na_2CO_3. All measurements were at 25.0 °C.

Apparatus. Cyclic voltammetry was performed with use of a three-electrode system consisting of a carbon paste working electrode, a platinum wire auxiliary electrode, and a standard calomel reference electrode. Voltammograms were generated with a Bioanalytical Systems CV-1 instrument and recorded on a Hewlett-Packard HP7035B X-Y recorder. The $E°$ values were determined as the midpoint between the peak potentials of the cathodic and anodic waves. The observed reduction potential showed a linear increase with $[OH^-]$ concentration above 0.1 M NaOH. This effect was due to the change of media upon the liquid junction potential, and the $E°$ values have been corrected accordingly.

Electron spin resonance spectra were obtained with a Varian E-109, X-band ESR system using a Varian E238 multipurpose cavity. Aqueous samples were studied at ambient temperature with use of a thin quartz cell, S-813, supplied by Scanco of Solvang, Calif. A Beckman R32 spectrometer was used to examine the proton NMR of the oxidized ligand products. The concentration of ligand was about 0.005 M in D_2O. The infrared spectrum was obtained as a Nujol mull on NaCl plates by using a Beckman IR-12 spectrometer. The UV–visible spectra were taken on a Cary 14 spectrophotometer.

The kinetics of $Cu^{III}(H_{-4}C)^-$ decomposition were studied at 365 nm by using a Cary 16 spectrophotometer with a Hewlett-Packard 7101B recorder. Faster reactions were observed by using a Durrum stopped-flow spectrometer interfaced to a Hewlett-Packard 2115A computer.[13] The observed rate constants are the result of at least three replicate experiments with a precision of ±10% from pH 2 to 9 and a precision of 3–5% below pH 2 and above pH 9. The electron-transfer reaction of $Cu^{II}(H_{-4}C)^{2-}$ with $IrCl_6^{2-}$ was studied on a pulsed-flow spectrometer.[14]

The photochemical apparatus is described elsewhere.[15] The method of Parker and Hatchard[16] was used to determine quantum yields at 278 nm. The initial concentration of $Cu^{III}(H_{-4}C)^-$ was 2.6×10^{-5} M, and a 5-cm cell path was used. The Cu(III) complex was assayed spectrometrically at 365 nm prior to and after photolysis and, because of its photochemical sensitivity, was handled in minimal lighting.

An Instrumentation Laboratory Model 245 pH meter with a NaCl-saturated calomel electrode was used for pH measurements, and a correction to give hydrogen ion concentration in 1.0 M ionic strength $NaClO_4$ was determined to be $-\log [H^+] = pH_{read} + 0.28$ by titration of $HClO_4$ with NaOH.

Results and Discussion

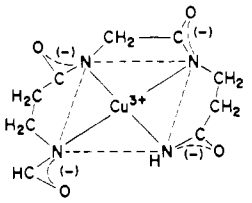
The properties of $Cu^{II}(H_{-4}C)^{2-}$ in solution indicate that there are four deprotonated peptide nitrogens coordinated to the metal in a square-planar geometry (axial water molecules probably give a tetragonally elongated octahedral geometry).[1] Oxidations of copper(II) peptides take place at the metal center with the ligand remaining intact.[2,7] Recent work[17] has shown that the copper(III) complexes lose the axially coordinated solvent molecules, and the proposed geometry for $Cu^{III}(H_{-4}C)^-$ is shown in structure I.

The reduction potential for this complex was determined as $E° = 0.48 \pm 0.01$ V vs. NHE by cyclic voltammetry. A pseudoreversible

$$Cu^{III}(H_{-4}C)^- + e^- \overset{E°}{\rightleftharpoons} Cu^{II}(H_{-4}C)^{2-} \qquad (1)$$

couple is observed (with a peak to peak separation ΔE_p of 85 ± 10 mV) in the cyclic voltammogram which is similar to other Cu(III,II)–peptide systems.[2] Both $E°$ and ΔE_p are independent of pH from 8.2 to 14.0, indicating that neither of the electroactive species are involved in other equilibria over this pH interval.

The value of 0.48 V for the reduction potential is lower than the $E°$ values of many other copper(III,II) peptide complexes.[2] This can be attributed in part to the presence of four deprotonated-peptide donors, but the 14-membered macrocyclic peptide also contributes to an enhanced thermodynamic stability of the copper(III) oxidation state relative to that of copper(II). This is illustrated by comparison with the open-chained complex of *N*-formyl-β-alanylglycyl-β-alanylamide shown in structure IV which also has a 6,5,6 ring system in



IV

its peptide backbone. This open-chained complex has a reduction potential of 0.62 V, which is 0.14 V higher than the $E°$ for the cyclic peptide complex.[18]

The visible absorption band (λ_{max} 488 nm, ϵ 54 M^{-1} cm^{-1}) for the $Cu^{II}(H_{-4}C)^{2-}$ complex occurs at lower wavelength than for most copper(II)–peptide complexes.[2] The $E°$ and λ_{max} behaviors are consistent with a gain in the crystal field stabilization energy. Copper(III)–donor bond distances are shorter than those of copper(II) with similar ligands.[19,20] The tightness of the macrocyclic ligand cavity favors the smaller radius of copper(III) and accounts for the lower $E°$ value as well as the effect on the d–d transitions for copper(II). These results are also consistent with observations for a series of nickel(II)–macrocyclic amine complexes for which higher energy d–d transitions were observed as the ring size decreased.[21]

(10) Neubecker, T. A.; Kirksey, S. T., Jr.; Chellappa, K. L.; Margerum, D. W. *Inorg. Chem.* **1979**, *18*, 444–448.
(11) Rybka, J. S.; Kurtz, J. L.; Neubecker, T. A.; Margerum, D. W. *Inorg. Chem.* **1980**, *19*, 2791–2796.
(12) Owens, G. D.; Chellappa, K. L.; Margerum, D. W. *Inorg. Chem.* **1979**, *18*, 960–966.
(13) Willis, B. G.; Bittikofer, J. A.; Pardue, H. L.; Margerum, D. W. *Anal. Chem.* **1970**, *42*, 1340–1349.
(14) Owens, G. D.; Taylor, R. W.; Ridley, T. Y.; Margerum, D. W. *Anal. Chem.* **1980**, *52*, 130–138.
(15) Ahn, B.; McMillin, D. R. *Inorg. Chem.* **1978**, *17*, 2253–2258.
(16) Hatchard, C. G.; Parker, C. A. *Proc. R. Soc. London, Ser. A* **1956**, *235*, 518–536.
(17) Youngblood, M. P.; Margerum, D. W. *Inorg. Chem.* **1980**, *19*, 3068–3072.
(18) Raycheba, J. M. T.; Perret, D.; Margerum, D. W., to be submitted for publication.
(19) Margerum, D. W.; Owens, G. D. *Met. Ions Biol. Syst.* **1981**, *12*, Chapter 3.
(20) Diaddario, L. L.; Robinson, W. R.; Margerum, D. W., to be submitted for publication.
(21) Busch, D. H. *Acc. Chem. Res.* **1978**, *11*, 392–400.

Table I. Observed First-Order Rate Constants for the Redox Decomposition of $Cu^{III}(H_{-4}C)^-$ [a]

$-\log [H^+]$ [b]	buffer	k_{obsd}, s^{-1}
2.16	$HClO_4$	8.6×10^{-7} [c]
2.31	$HClO_4$	5.6×10^{-7} [c]
2.46	$HClO_4$	5.8×10^{-7} [c]
2.68	$HClO_4$	5.5×10^{-7} [c]
2.76	$HClO_4$	4.0×10^{-7} [c]
2.86	$HClO_4$	4.7×10^{-7} [c]
3.16	$HClO_4$	4.9×10^{-7} [c]
4.64	0.050 M total acetate	2.0×10^{-7}
8.85	0.5×10^{-3} M total borate	3.0×10^{-7}
8.85	1.0×10^{-3} M total borate	1.9×10^{-7}
8.85	2.5×10^{-3} M total borate	5.1×10^{-7}
8.85	5.0×10^{-3} M total borate	6.3×10^{-7}
8.85	10.0×10^{-4} M total borate	11.3×10^{-7}
11.47	NaOH	6.7×10^{-5}
11.76	NaOH	1.4×10^{-4}
12.06	NaOH	3.2×10^{-4}
12.46	NaOH	8.4×10^{-4}
12.61	NaOH	1.1×10^{-3}
12.76	NaOH	1.6×10^{-3}
12.94	NaOH	3.0×10^{-3}
13.16	NaOH	4.6×10^{-3}
13.46	NaOH	1.0×10^{-2}

[a] $[Cu^{III}(H_{-4}C)^-] = 3 \times 10^{-5}$ M or 6×10^{-5} M, $\mu = 1.0$ M ($NaClO_4$), 25.0 °C, $\lambda = 365$ nm. [b] Using $pK_w = 13.76$ in 1.0 M $NaClO_4$. [c] Initial rate determination.

The electronic spectrum of $Cu^{III}(H_{-4}C)^-$ consists of a series of charge-transfer bands with absorbance maxima at 365 nm ($\epsilon = 6200 \pm 100$ M^{-1} cm^{-1}) and 250 nm ($\epsilon = 12000$ M^{-1} cm^{-1}) and a shoulder at 275 nm ($\epsilon \sim 10500$ M^{-1} cm^{-1}). The molar absorptivities were determined by titration with ascorbic acid. The peak positions are independent of pH from 0.3 to 12.7, but changes in ϵ are noted below pH 2. Other copper(III) complexes with four nitrogen donors in the plane have λ_{max} and ϵ similar to these values.[2] The tripeptide complexes, which have one carboxylate oxygen in the plane, have slightly red-shifted absorbance maxima with the higher energy transition at 275 nm. The UV spectrum of $Cu^{III}(H_{-4}C)^-$ is different in that it shows a shoulder at 275 nm. This fingerprint has been used to verify the presence of the complex especially after the reactions with other copper(III)–peptide complexes.

Redox Decomposition

In the absence of reducing agents the copper(III)–cyclic peptide complex undergoes a slow reduction of the metal accompanied by ligand oxidation. The loss of $Cu^{III}(H_{-4}C)^-$ obeys eq 2, and the values of k_{obsd} are pH dependent (see Table I).

$$-d[Cu^{III}(H_{-4}C)^-]/dt = k_{obsd}[Cu^{III}(H_{-4}C)^-] \quad (2)$$

Above pH 8. In borate buffer at pH 8.85 the observed first-order rate constant depends upon the buffer concentrations as in eq 3. The borate-dependent rate constant (k_B)

$$k_{obsd} = k'_{obsd} + k_B[\text{borate}]_T \quad (3)$$

equals 9.0×10^{-4} M^{-1} s^{-1}, and the buffer-independent term (k'_{obsd}) equals 2.2×10^{-7} s^{-1}. The pH dependence of k'_{obsd} is shown in Figure 1. The base decomposition can be described by eq 4 and 5 with k'_{obsd} defined by eq 6. The values of k_0 and k_{OH} are 2×10^{-7} s^{-1} and 1.6×10^{-2} M^{-1} s^{-1}, respectively.

$$Cu^{III}(H_{-4}C)^- \xrightarrow{k_0} \text{products} \quad (4)$$

$$Cu^{III}(H_{-4}C)^- + OH^- \xrightarrow{k_{OH}} \text{products} \quad (5)$$

$$k'_{obsd} = k_0 + k_{OH}[OH^-] \quad (6)$$

The decomposition kinetics differ in several respects from those of copper(III)–linear peptide complexes. For $Cu^{III}(H_{-3}G_4)^-$ the redox decomposition reaction is also base catalyzed,[11] but the reactions are more complex because one of the products, $Cu^{II}(H_{-2}G_4)^-$, accelerates the loss of copper(III). The tetraglycine decomposition also has a complex hydroxide dependence due to amine deprotonation to form a $Cu^{III}(H_{-4}G_4)^{2-}$ species. The decomposition reaction of $Cu^{III}(H_{-4}C)^-$ is free of these complications. The predominant form of the copper(II) complex over a wide pH range is $Cu^{II}(H_{-4}C)^{2-}$,[1] which does not catalyze the $Cu^{III}(H_{-4}C)^-$ decomposition, and hence a simple first-order $[OH^-]$ dependence is observed.

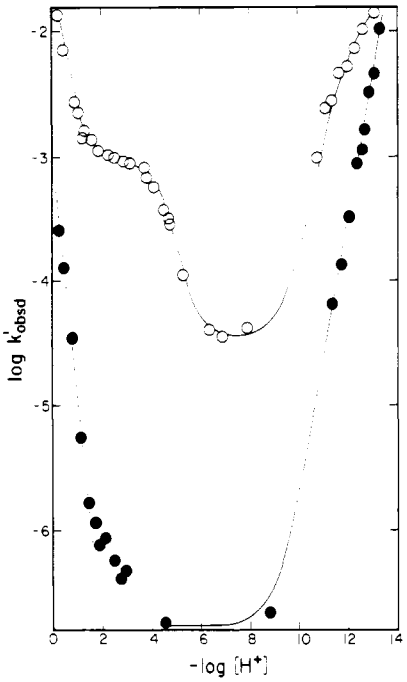


Figure 1. pH profile for the self-redox decomposition of $Cu^{III}(H_{-4}C)^-$ (●) at $\mu = 1.0$ M ($NaClO_4$) and 25.0 °C. The solid line is calculated by using eq 6 with $k_{OH} = 1.6 \times 10^{-2}$ M^{-1} s^{-1} and $k_0 = 2.0 \times 10^{-7}$. The dashed line has a slope of −2. The open circles (○) represent the pH profile for $Cu^{III}(H_{-3}G_4)^-$.

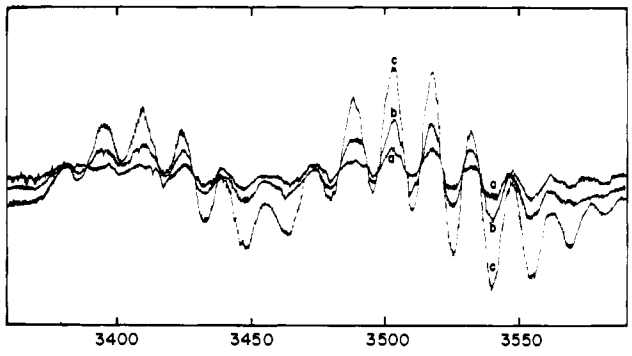
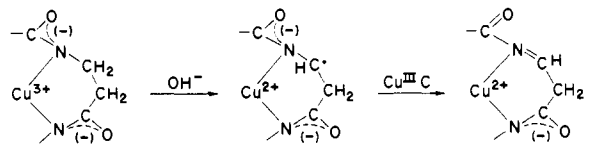


Figure 2. Repetitive ESR scans for $Cu^{III}(H_{-4}C)^-$ self-redox in 0.02 M NaOH, at $\mu = 1.0$ M ($NaClO_4$) and 25.0 °C. $\nu = 9.392$ GHz. Scan time is 8 min with scans initiated at (a) 6 min, (b) 25 min, and (c) 170 min.

The course of the reaction can be followed with ESR spectroscopy to monitor the formation of the copper(II) complexes produced. Figure 2 shows repetitive ESR scans of $Cu^{III}(H_{-4}C)^-$ decomposition in 0.02 M NaOH. An important feature of these spectra is the nitrogen hyperfine coupling. The number of lines is consistent with four nitrogen atoms about the copper(II) center, and the sharpness of the lines is similar to those of $Cu^{II}(H_{-4}C)^{2-}$ while other copper(II) peptides do not display this high a degree of resolution.[1]

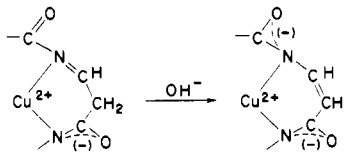
After the decomposition reaction in 5×10^{-3} M NaOH is complete, the visible spectrum of the products is similar to a

Scheme I



I V

Scheme II



VI

sample prepared by ascorbic acid reduction. Both solutions have an absorbance maximum at 488 nm, with no other peaks at higher wavelength, and the absorbances of these two solutions agree within 5%. (The visible absorption band for $Cu^{II}(H_{-4}C)^{2-}$ is at a significantly lower wavelength than other copper(II)–peptide complexes; e.g., the quadruply deprotonated complex of *N*-formyltriglycylamide has a peak at 507 nm and triply deprotonated complexes of tetraglycine and pentaglycine have peaks at 515 nm and 510 nm, respectively.[2,22])

Reduction of the copper(III) center is accompanied by ligand oxidation. Scheme I shows a proposed reduction mechanism similar to that observed for copper(III)–linear peptide complexes.[23] In this scheme a methylene hydrogen abstraction leads to formation of a copper(II) ligand radical that reacts with another copper(III) complex to yield one intact ligand and one dehydropeptide of the starting ligand (as in structure V). In base the dehydropeptide hydrolyzes to form a peptide amide and carbonyl products. If this scheme were applied to the cyclic peptide complex, we would expect as products 50% intact cyclic ligand and 50% hydrolyzed to form either *N*-glyoxal-β-alanylglycyl-β-alaninamide or $HCOCH_2$-$CONHCH_2CONHCH_2CH_2CONHCH_2CONH_2$ (if oxidation were at the β-alanyl residue). The resulting mixture should have ESR and visible spectral properties indicating the presence of a copper(II)–linear peptide, but each of these experiments shows features that are very similar to those of $Cu^{II}(H_{-4}C)^{2-}$. These spectral characteristics suggest that ligand oxidation was not accompanied by bond cleavage and opening of the 14-membered macrocyclic ring.

The other possibility for ligand oxidation is presented in Scheme II. This path is similar to Scheme I in that a dehydropeptide is formed on one of the β-alanyl residues, but now further reaction via another proton loss from a carbon atom will lead to the formation of a carbon–carbon double bond as in structure VI. The β-alanyl residue that has been oxidized would now be coplanar with the two peptide linkages on either side of it. Molecular models show that a macrocyclic olefin should accommodate copper(II) in a planar complex similar to $Cu^{II}(H_{-4}C)^{2-}$. Glycylglycylhistidine undergoes an oxidative decarboxylation by copper(II) hydroxide in the presence of O_2 to form a copper(II) complex in which an olefin is formed in the six-membered chelate ring.[24]

When the base reaction mixture is lyophilized and washed with anhydrous ethanol to form a neutral powder, its infrared spectrum can be compared to that of $Na_2Cu(H_{-4}C)$. The results show some evidence for a band at 1700 cm^{-1} where an olefinic stretch would be expected; however the band is weak and is only a shoulder on the carbonyl stretching band. If the base reaction products are acidified to dissociate the metal, passed down a Chelex resin in the Na^+ form, and then lyophilized, the oxidized ligand products can be obtained in the absence of Cu(II). An NMR spectrum of this sample with the concentration of total ligand about 0.005 M in D_2O shows two unresolved doublets at 5.40 and 5.73 ppm vs. Me_4Si. Other features are consistent with the NMR of the free ligand in trifluoroacetic acid.[1] These two peaks are assigned to the olefinic protons, whose chemical shifts are typically 5–7 ppm. The ESR, visible, IR, and NMR spectra of the products are consistent with those proposed in Scheme II.

Table II. Summary of the $Cu^{III}(H_{-4}C)^-$ Reactions in Acid[a]

−log [H⁺]	% ΔA init. absorbance drop	2nd reacn		final reacn	
		% ΔA	k_{obsd}, s^{-1}	% ΔA	k_{obsd},[b] s^{-1}
0.29	35	15	3.9 ± 0.2	50	2.7×10^{-4}
0.46	28	17	4.5 ± 1.2	55	1.3×10^{-4}
0.76	11	6	6 ± 3	60	3.5×10^{-5}
1.16	8	15	5.0 ± 0.9	65	5.6×10^{-6}
1.46	0	11	5.4 ± 1.0	70	1.7×10^{-6}
1.76	0	16	2.6 ± 1.6	80	1.2×10^{-6}
1.98				98	7.2×10^{-7}

[a] $[Cu^{III}(H_{-4}C)^-] = 7.3 \times 10^{-6}$ M or 3.0×10^{-5} M, $\mu = 1.0$ M ($NaClO_4$), 25.0 °C, λ = 365 nm. [b] Average precision of rate constants was ±3%.

Below pH 4. The redox decomposition of $Cu^{III}(H_{-4}C)^-$ is acid catalyzed, but the reactions are not as simple kinetically as the base reaction. Above pH 2 the initial absorbances are in agreement with the calculated values based on the ε value. The initial rate method was used to obtain the first-order rate constant, k'_{obsd}, because the reactions were quite slow. As Figure 1 shows these rate constants are larger than k_0 (eq 4) and increase with decreasing pH. Other copper(III)–peptide complexes in this pH region show protonation of a peptide oxygen without metal–nitrogen bond cleavage.[11] This "outside" protonation has also been observed for nickel(II) and copper(II) peptide complexes.[25,26] These reactions are shown in eq 7 and 8, for which we can only estimate $pK_H \sim 2.5$ and $k_1 \leq 10^{-6}$ s^{-1}.

$$Cu^{III}(H_{-4}C)^- + H^+ \underset{}{\overset{K_H}{\rightleftharpoons}} Cu^{III}(H_{-4}C)H \quad (7)$$

$$Cu^{III}(H_{-4}C)H \xrightarrow{k_1} \text{products} \quad (8)$$

Below pH 2 the copper(III)–cyclic peptide complex undergoes two reactions without the loss of copper(III) before the self-redox occurs. These reactions are summarized in Table II. The first reaction is a very fast absorbance drop at 365 nm that shows a pH dependence in its extent and accounts for as much as 35% of the absorbance change in 0.5 M $HClO_4$. The second reaction is first order in [Cu(III)] with a rate constant of 4 s^{-1}. The rate constant and the extent of the second reaction (~15% of the absorbance change) are independent of acidity but can be seen only below pH 2. As a test of whether these two reactions involve the loss of copper(III) they were allowed to go to completion in 0.5 M $HClO_4$ and then (after 15 s) the solution was quenched with 2 M sodium acetate to raise the pH. All of the initial copper(III) absorbance was recovered, indicating that these first two reactions are reversible and do not lead to copper(III) reduction. The first reaction may be the formation of a $Cu^{III}(H_{-4}C)H_2^+$ species. We know that one outside protonation causes only

(22) Billo, E. J. *Inorg. Nucl. Chem. Lett.* **1974**, *10*, 613–617.
(23) Kurtz, J. L.; Burce, G. L.; Margerum, D. W. *Inorg. Chem.* **1978**, *17*, 2454–2461.
(24) DeMeester, P.; Hodgson, D. J. *Inorg. Chem.* **1978**, *17*, 440–444.
(25) Paniago, E. B.; Margerum, D. W. *J. Am. Chem. Soc.* **1972**, *94*, 6704–6710.
(26) Wong, L. F.; Cooper, J. C.; Margerum, D. W. *J. Am. Chem. Soc.* **1976**, *98*, 7268–7274.

small spectral changes, but it may be that the addition of two protons causes the large $\Delta\epsilon$ observed. The second reaction, which is acid independent, must be the conversion from outside protonation to inside protonation; i.e., peptide nitrogen protonation with breaking or distortion of the bond to the metal (eq 10).

$$Cu^{III}(H_{-4}C)H + H^+ \underset{}{\overset{K_{2H}}{\rightleftharpoons}} Cu^{III}(H_{-4}C)H_2^+ \quad (9)$$

$$\left.\begin{matrix} Cu^{III}(H_{-4}C)H \\ Cu^{III}(H_{-4}C)H_2^+ \end{matrix}\right\} \underset{}{\overset{k_2}{\rightleftharpoons}} \left\{\begin{matrix} Cu^{III}(H_{-3}C) \\ Cu^{III}(H_{-3}C)H^+ \end{matrix}\right. \quad (10)$$

After the preceding reactions are complete, the final (much slower) reaction results in the loss of copper(III). This reaction is first order in [Cu(III)]; the rate constants for the reaction are given in Table II and show a second-order $[H^+]$ dependence below pH 1.5 with a third-order rate constant, $k_{2H} = 1.0 \times 10^{-3}$ M^{-2} s^{-1} (see eq 11). The second-order hydrogen

$$k'_{obsd} = k_{2H}[H^+]^2 \quad (11)$$

ion dependence may result from additional unwrapping of the ligand from the metal prior to redox, or the redox step itself may involve H^+.

After the acid decomposition reaction is complete and the solutions are made basic (to pH 12.5), copper(II)–peptide complexes are observed which have one absorbance maximum at 488 nm with no appreciable increases in absorbance at higher wavelength. As was true for the base decomposition reaction, this result indicates the absence of linear peptide fragments due to cleavage of the macrocyclic peptide backbone. Proton NMR of the peptide product (after removal of Cu^{2+}) showed the same two unresolved peaks as the base decomposition product. These results indicate that the acid redox decomposition reaction products are the same as those in base. This result is not totally unexpected because the products of redox decomposition for $Cu^{III}(H_{-3}G_4)^-$ are the same in both acid and base with the distribution changing at higher pH due to the autocatalytic nature of that reaction.[11]

A remarkable feature of the pH profile for the redox decomposition reaction of $Cu^{III}(H_{-4}C)^-$ is the minimal rate constant at neutral pH, with a value of k_0 of 2.0×10^{-7} s^{-1}, corresponding to a half-life of nearly 6 weeks. This represents a factor of almost 200 in kinetic stabilization relative to $Cu^{III}(H_{-3}G_4)^-$ as shown in Figure 1. For $Cu^{III}(H_{-3}G_4)^-$ the acid redox decomposition reaction also is preceded by partial dissociation of the complex to form a species of higher reduction potential.[11] The macrocyclic peptide ligand helps to stabilize the copper(II) complex $Cu^{II}(H_{-4}C)^{2-}$ by slowing down both the acid attack upon the deprotonated ligand and the nucleophilic attack upon the metal.[1] For the trivalent complex $Cu^{III}(H_{-4}C)^-$, the increased kinetic stabilization could be attributed to either the lower $E°$ value for the complex or to the importance of ligand substitution kinetics in the self-decomposition process.

Photochemical Redox. The redox decomposition of copper(III) peptides is also photochemically catalyzed. For $Cu^{III}(H_{-4}C)^-$ the neutral decomposition is 50 times faster under a 100-W tungsten lamp than in the dark. The average value of the quantum yield at 278 nm is $\Phi = (8 \pm 1) \times 10^{-3}$ molecules of Cu(III) reduced per quantum. This value is less than that for some other copper(III) peptides, e.g., $\Phi_{278} = 0.3$ for $Cu^{III}(H_{-2}Aib_3)$[27] and $\Phi_{365} = 0.06$ for $Cu^{III}(H_{-3}G_4)^-$,[11] but still represents a significant photochemical pathway.

Electron-Transfer Reactions

In order to be able to determine the self-exchange rate for $Cu^{III,II}(H_{-4}C)^{1-,2-}$, we reacted $Cu^{II}(H_{-4}C)^{2-}$ with the copper(III) complex of the tripeptide of α-aminoisobutyric acid, $Cu^{III}(H_{-2}Aib_3)$, which has an $E°$ value of 0.66 V.[8] The self-exchange rate between $Cu^{III}(H_{-2}Aib_3)$ and $Cu^{II}(H_{-2}Aib_3)^-$ has been determined by NMR line-broadening experiments.[28] The reaction of eq 12 has a positive $E°$ value of 0.18 V. The

$$Cu^{III}(H_{-2}Aib_3) + Cu^{II}(H_{-4}C)^{2-} \xrightarrow{k_{12}} Cu^{II}(H_{-2}Aib)_3^- + Cu^{III}(H_{-4}C)^- \quad (12)$$

electronic spectra of the two Cu(III) complexes are sufficiently different to permit the reaction to be monitored by the formation of $Cu^{III}(H_{-4}C)^-$ at 250 nm where the ϵ values for $Cu^{III}(H_{-4}C)^-$ and $Cu^{III}(H_{-2}Aib_3)$ are 12100 and 7140 M^{-1} cm^{-1}, respectively.

The reaction was conducted under second-order unequal concentration conditions. For a reaction under these conditions, the integrated rate expression is given by eq 13, where

$$\frac{1}{C_B^0 - C_A^0} \ln \left[\frac{C_A^0(C_B^0 - x)}{C_B^0(C_A^0 - x)} \right] = kt \quad (13)$$

C_A^0 and C_B^0 represent the initial concentrations of species A and B and x is the amount of product at any time, t. This expression can be converted to the observable parameters of absorbance at any time (A_t), initial absorbance (A_0), and the final absorbance (A_∞), giving eq 14, where q is defined as

$$\ln \left[\frac{qA_\infty - A_t + A_0(1 - q)}{q(A_\infty - A_t)} \right] = C_A^0(q - 1)kt \quad (14)$$

C_B^0/C_A^0. For these experiments, $[Cu^{II}(H_{-4}C)^{2-}]_0 = 3.1 \times 10^{-5}$ M and $[Cu^{III}(H_{-2}Aib_3)]_0 = 2.1 \times 10^{-5}$ M, so $q = 1.48$. The observed second-order rate constant, $k_{12} = (1.55 \pm 0.06) \times 10^6$ M^{-1} s^{-1}, is the result of seven replicate experiments.

The kinetic self-exchange rate constant for $Cu^{III,II}(H_{-4}C)^{1-,2-}$ (k_{11}) can be calculated from the Marcus theory for outer-sphere electron transfer by using eq 15 and 16.[29] The value

$$k_{12} = (k_{11}k_{22}K_{12}f)^{1/2} \quad (15)$$

$$\log f = (\log K_{12})^2/[4 \log (k_{11}k_{22}/Z^2)] \quad (16)$$

of k_{22} for $Cu^{III,II}(H_{-2}Aib_3)^{0,-}$ is 5.5×10^4 M^{-1} s^{-1},[26] and the log K_{12} value is 3.05. The parameter Z is a collisional frequency term (10^{11} M^{-1} s^{-1}). Iteration of eq 15 and 16 produced a self-consistent set of values for k_{11} and f as 6.0×10^4 M^{-1} s^{-1} and 0.65, respectively. This value for k_{11} is similar to that found for $Cu^{III,II}(H_{-2}Aib_3)^{0,-}$ in the NMR line-broadening experiments.[28] It is interesting that the calculated electron-transfer self-exchange rate constants are the same for the macrocyclic and open-chain peptide complexes.

Another electron-transfer reaction of $Cu^{II}(H_{-4}C)^{2-}$ was performed with $IrCl_6^{2-}$ (eq 17) for which the equilibrium lies

$$Cu^{II}(H_{-4}C)^{2-} + IrCl_6^{2-} \underset{k_{21}}{\overset{k_{12}}{\rightleftharpoons}} Cu^{III}(H_{-4}C)^- + IrCl_6^{3-} \quad (17)$$

far to the right. The reaction was conducted under second-order equal-concentration conditions while we monitored the loss of absorbance due to $IrCl_6^{2-}$, which has an absorbance maximum at 490 nm. Data were obtained with a pulsed-flow spectrometer for measuring very rapid reactions in solution.[14] The experimental data are presented in Table III. The flow velocity was varied randomly to minimize any effects due to instrumental drift. The theoretical data treatment for a reaction under second-order equal-concentration conditions has been established.[30] However, this treatment cannot be reliably applied to reactions whose half-lives are much less than 1 ms,

(27) Hamburg, A. W.; Margerum, D. W., unpublished results.

(28) Koval, C. A.; Margerum, D. W. *Inorg. Chem.*, in press.
(29) Marcus, R. A. *Annu. Rev. Phys. Chem.* **1964**, *15*, 155–196.
(30) Gerisher, H.; Heim, W. *Z. Phys. Chem. (Wiesbaden)* **1965**, *46*, 345–352.

Table III. Data for the $Cu^{II}(H_{-4}C)^{2-}$ + $IrCl_6^{2-}$ Reaction Using the Pulsed-Flow Instrument[a]

velocity, m/s	abs[b]	$10^4 t_{1/2}$, s	velocity, m/s	abs[b]	$10^4 t_{1/2}$, s
8.37	0.0503	1.8	7.83	0.0502	1.8
4.68	0.0455	2.0	9.11	0.0499	1.7
6.26	0.0435	1.6	6.99	0.0468	1.6

[a] $A_0(IrCl_6^{2-}) = 0.2155$, $A_0(Cu^{II}(H_{-4}C)^{2-}) = 0.0043$, $A_\infty = 0.0228$, [Cu(II)] = [Ir(IV)] = 1.47×10^{-5} M, $\lambda = 490$ nm, 25.0 °C, $\mu = 0.1$ M ($NaClO_4$), pH 8.00. [b] Absorbance corrected for media effects.

and so an empirical scheme has been developed which is based on a linear dependence of the absorbance quotient, $(A - A_\infty)/(A_0 - A_\infty)$, on the first half-life of a known system.[14,31] On this basis the values of $t_{1/2}$ in Table III were determined. The average half-life for this reaction is $(1.7 \pm 0.3) \times 10^{-4}$ s. The observed second-order rate constant is $k_{12} = 4.0 \times 10^8$ M^{-1} s^{-1}, since the initial concentration of the reactants is 1.47×10^{-5} M. This value is almost a factor of 4 larger than the value calculated from eq 15 which gives k_{12}^{calcd} equal to 1.1×10^8 M^{-1} s^{-1} on the basis of $k_{11} = 6.0 \times 10^4$ M^{-1} s^{-1}, k_{22}-$(Ir^{IV,III}Cl_6^{2-,3-}) = 2.3 \times 10^5$ M^{-1} s^{-1},[32] log $K_{12} = 6.95$, and $f = 0.096$. On the other hand the observed k_{12} value is a factor of 10 less than the diffusion-limiting rate constant for 2-,2- reactants. As discussed elsewhere[33] for a series of Cu(II) peptides reacting with $IrCl_6^{2-}$, the value of 4×10^8 M^{-1} s^{-1} corresponds to a limiting rate constant expected for a model in which axial substitution of water in the $Cu^{II}(H_{-4}C)^{2-}$ complex is the rate-determining step. Although the behavior of $IrCl_6^{2-}$ with $Cu^{II}(H_{-4}C)^{2-}$ is only a factor of 4 different from the Marcus predictions, the behavior is consistent with that of other copper(II) peptides with $IrCl_6^{2-}$ where the reaction does not correspond to an outer-sphere electron-transfer process.[33]

Conclusions

The macrocyclic peptide ligand helps to stabilize the copper(III) complex both thermodynamically and kinetically. The reduction potential of 0.48 V for $Cu^{III,II}(H_{-4}C)^{1-,2-}$ is less than that predicted for four deprotonated peptide donors alone and the tightness of the metal cavity appears to favor the higher oxidation state.

The redox decomposition reactions are much slower than those of corresponding open-chain peptide complexes of copper(III). The kinetic results for the acid decomposition suggest that ligand substitution in equatorial positions is involved prior to the redox steps and that the macrocyclic ligand makes these processes more difficult.

The electron-exchange reactions for $Cu^{III,II}(H_{-4}C)^{1-,2-}$ are rapid, and the value of 6.0×10^4 M^{-1} s^{-1} calculated for the self-exchange agrees with other Cu(III,II) self-exchange rate constants.

Acknowledgment. This investigation was supported by Public Health Service Grant No. GM12152 from the National Institute of General Medical Sciences.

Registry No. $Cu^{III}(H_{-4}C)^-$, 76721-70-5; $Cu^{II}(H_{-4}C)^{2-}$, 74185-28-7; $IrCl_6^{2-}$, 16918-91-5.

(31) Gerisher, H.; Holzwarth, J.; Seifert, D.; Strohmaier, L. *Ber. Bunsenges. Phys. Chem.* **1972**, *76*, 11-16.
(32) Horwitz, P.; Kustin, K.; *Trans. Faraday Soc.* **1966**, *62*, 427-432.
(33) Owens, G. D.; Margerum, D. W., *Inorg. Chem.*, companion paper in this issue.

Contribution from the Radiation Laboratory, University of Notre Dame, Notre Dame, Indiana 46556

Anion Radical Oxidation of Nickel(II) Macrocyclic Complexes. Pulse Radiolysis of (2,12-Dimethyl-3,7,11,17-tetraazabicyclo[11.3.1]heptadeca-1(17),13,15-triene)nickel(II) in Sodium Bromide Solution

P. MORLIERE and L. K. PATTERSON*

Received May 6, 1980

The oxidation of $Ni^{II}(CR+4H)$ [CR+4H = 2,12-dimethyl-3,7,11,17-tetraazabicyclo[11.3.1]heptadeca-1(17),13,15-triene, α isomer] by Br_2^- and subsequent chemistry of Ni(III) intermediates have been studied by pulse radiolysis. The initial electron transfer is diffusion controlled and results in an initial product which exhibits characteristics of a Ni(III)-Br^- adduct. The stability of this adduct, especially in acidic media, is dependent on bulk bromide concentration, demonstrating an equilibrium process of the form $Ni^{III}(CR+4H)(Br^-)(H_2O) + H_2O \rightleftarrows Ni^{III}(CR+4H)(H_2O)_2 + Br^-$, with $K_{eq} = 360$ M^{-1}. The pK of dissociation for the aquated species is 4.0 ± 0.8. At pH >5 displacement of the bound bromide is first order in $[OH^-]$ with a rate constant of 6×10^9 M^{-1} s^{-1}. Disappearance of the $Ni^{III}(CR+4H)(OH^-)$ intermediate is also pH dependent, suggesting amine dissociation in the ligand and formation of an unstable ligand radical leading to a final product with increased ligand unsaturation.

Introduction

The interest in transition-metal complexes with uncommon and frequently unstable oxidation states and their associated chemistry has grown considerably during recent years. Such complexes are mentioned as intermediates in both catalytic reactions and biological processes.[1] A special group of these are the nickel(II) complexes with macrocyclic ligands which form species of high stability.[2] Electrochemical oxidations of the Ni(II) metal center in these complexes have been reported for various macrocyclic ligands in nonaqueous solvents where the lifetimes of Ni(III) complexes can be quite long.[2-5] This large stability permits detailed characterization of Ni(III) compounds by EPR and optical spectroscopy. With a few exceptions, Ni(III) complexes are generally short-lived in aqueous solutions, and only a limited number of investigations have been reported.[6-8]

(1) D. Meyerstein, *Acc. Chem. Res.*, **11**, 43 (1978).
(2) F. V. Lovecchio, E. S. Gore, and D. H. Busch, *J. Am. Chem. Soc.*, **96**, 3109 (1974).
(3) D. C. Olson and J. Vasilevskis, *Inorg. Chem.*, **8**, 1611 (1969).
(4) E. K. Barefield, F. Lovecchio, N. E. Tokel, E. Ochiai, and D. H. Busch, *Inorg. Chem.*, **11**, 283 (1972).
(5) A. Wolberg and J. Manassen, *Inorg. Chem.*, **9**, 2365 (1970).

0020-1669/81/1320-1458$01.25/0