Oxo- and Hydroxo-Bridged (Porphyrin)iron(III)-Copper(II) Species as Cytochrome *c* Oxidase Models: Acid-Base Interconversions and X-ray Structure of the Fe(III)-(O^{2-})-Cu(II) Complex

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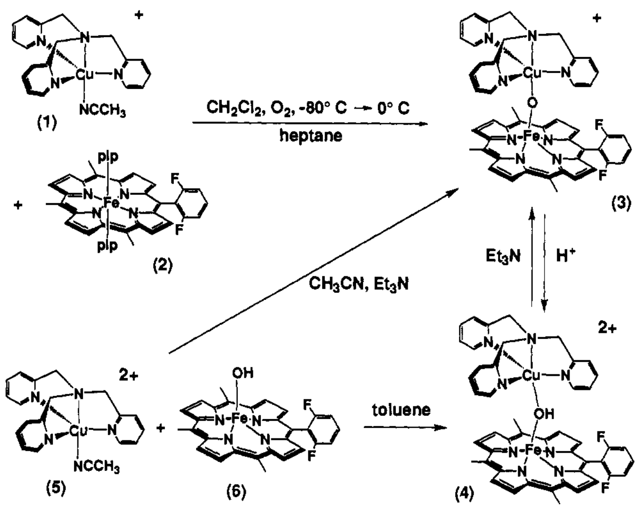
In this communication, we describe the synthesis and interconversion of oxo- and hydroxo-bridged [(F_8-TPP)Fe(III)-X-Cu(II)(TMPA)]$^{n+}$ complexes **3** and **4**, with F_8-TPP = tetrakis(2,6-difluorophenyl)porphyrinate, TMPA = tris(2-pyridylmethyl)-amine, X = O^{2-} (**3**, *n* = 1) and OH^- (**4**, *n* = 2). The X-ray structure of **3** is also described. Such bridged heterodinuclear complexes are of potential interest as structural and functional models for the corresponding cytochrome a_3 heme–copper-(histidyl) moiety, known to be the active site for dioxygen (O_2) binding and reduction in cytochrome *c* oxidases (*C*cOs).[1] The latter are terminal respiratory membrane proteins which effect the exergonic reaction $O_2 + 4e^- + 4H^+ \rightarrow 2H_2O$; the energy is utilized for subsequent ATP synthesis via the pH gradient generated by simultaneous coupled proton pumping across the membrane. Extensive enzyme studies have established the close proximity of the iron and copper atoms. Spectroscopic and kinetic investigations indicate that the activated reduced Fe(II)a_3 and Cu_B(I) react with O_2, each forming M–O_2 adducts,[2] while subsequent electron transfer from the metals at the dinuclear or other sites produces peroxo and/or hydroperoxo, ferryl (i.e., Fe=O), and eventually hydroxo and/or aquo–metal species as reduction products.[1b,3] Additionally, studies on ligand-bound (e.g., azide, CN^-, formate)[4] Fe(III)a_3 and Cu_B(II) oxidized ("resting") forms of the enzyme have been of interest in spectroscopic–magnetic, and thus structural, probing of this unique dinuclear catalytic center. Efforts to generate structurally defined ligand-bridged (porphyrin)iron(III)-X-Cu(II)(ligand) dinuclear species relevant to the biochemical O_2 chemistry or ligand-probe studies have only recently met with success.[5-7]

Scheme I



We previously reported that O_2 reaction of equimolar quantities of [(TMPA)Cu^I(CH_3CN)]$^+$ (**1**) (TMPA = tris(2-pyridylmethyl)-amine) and (F_8-TPP)Fe^{II}-pip_2 (**2**) (F_8-TPP = tetrakis(2,6-difluorophenyl)porphyrin; pip = piperidine) produces magnetically coupled ligand-bridged (porphyrin)iron(III)-X-Cu(II)-(TMPA) complexes, initially thought to be X = O_2^{2-}.[5] We have now unambiguously determined that this product (for F_8-TPP) is an oxo complex [(F_8-TPP)Fe(III)-(O^{2-})-Cu(II)(TMPA)]$^+$ (**3**), a species of considerable interest since it is derived from O_2 and thus represents a functional model for *C*cO.[8] Fe and Cu-edge extended X-ray absorption fine structure (EXAFS) studies on **3** indicate a near-linear Fe–X–Cu arrangement, with short Fe–X (1.72 ± 0.02 Å) and Cu–X (1.82 ± 0.02 Å) distances, X clearly being O. An Fe–Cu distance of $3.54_{ave} \pm 0.07$ Å determined from these experiments also necessitates an essentially linear Fe–X–Cu arrangement.[9] This hint that the reaction product could be an oxo complex (derived from O_2) led us to attempt an acid–base synthesis. In fact, the product obtained by the previous O_2 reaction possesses physical properties (i.e., UV–vis, ^1H NMR, etc.) identical with those of the product isolated by the reaction of the pentacoordinate Cu(II) complex [(TMPA)Cu^{II}(CH_3-CN)]$^{2+}$ (**5**), with labile nitrile ligand, and the hydroxo-Fe(III) complex (F_8-TPP)Fe-OH (**6**),[10] in the presence of triethylamine as base, Scheme I.[11]

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(8) Large changes in the IR spectrum of **3** occur when using $^{18}O_2$ in the synthesis.[5]

(9) (a) Unusually large intensities corresponding to the Fe–Cu and Cu–Fe interactions were observed in the Fourier transforms at ~*R* = 3.6 Å. These could be simulated only by assuming a strong multiple scattering interaction (focusing effect) mediated by a near-linear Fe–O–Cu connectivity. The average ∠Fe–O–Cu observed from least-squares refinement at each absorption edge was 173 ±7°. The detailed X-ray absorption spectroscopic study will be presented elsewhere; manuscript in preparation. (b) The distinctive intense peak characterizing the linear Fe–O–Cu moiety was not something observed in enzyme EXAFS studies;[1e] observation of it would be independent of the enzyme metal-ion content or models used in the data analysis.

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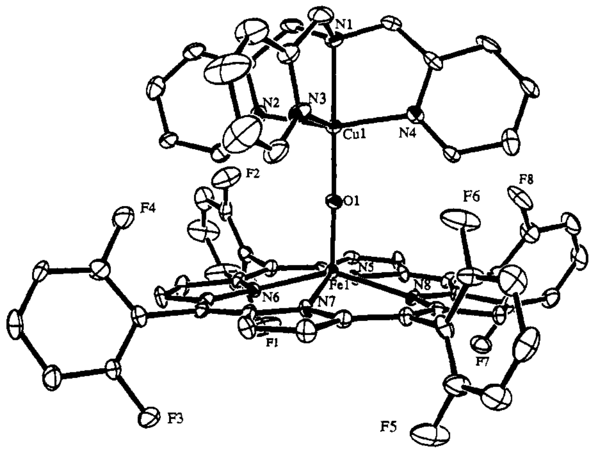

Figure 1. ORTEP (30% ellipsoids) view of cationic portion of $3\text{-}(ClO_4)\cdot 3C_7H_8\cdot 2CH_3CN$. Selected bond distances (Å): Cu1–O1, 1.856(5); Cu1–N1, 2.100(7); Cu1–N2, 2.009(7); Cu1–N3, 2.172(7); Cu1–N4, 1.984(8); Fe1–O1, 1.740(5); Fe–N, 2.099(7)–2.117(6); Fe1···Cu1, 3.596(2). Selected angles (deg): Fe1–O1–Cu1, 178.2(4); O1–Cu1–N1, 176.7(3); N2–Cu1–N4, 158.4(3); N5–Fe1–N7, 150.8(2); N6–Fe1–N8, 148.2(2).

X-ray quality crystals of **3** were obtained from acetonitrile/toluene under argon. The extraordinary structure[12] reveals the expected near-linear Fe(III)-oxo-Cu(II) coordination, Figure 1. The iron coordination is typical for high-spin (porphyrin)Fe(III) and close to that observed for *μ*-oxo iron dinuclear complexes $\{(TPP)Fe\}_2O$[13] and $\{(F_8\text{-}TPP)Fe\}_2O$.[14] The oxo ligand takes up the fifth position always available in Cu(II) complexes with TMPA,[15] but the coordination is distorted from the more usual trigonal-bipyramidal geometry; instead, a square-based pyramidal arrangement (N3 axial) is observed, and the three pyridine rings appear to fall in between difluorophenyl groups of the porphyrin, thus leaving one "slot" free.

Complete magnetic Mössbauer and multifield saturation magnetization studies of **3** (solutions and solid state) have been carried out,[16] concluding that the compound has an overall $S = 2$ ground state with the high-spin Fe(III) ($S = {}^5/_2$) antiferromagnetically coupled through the oxo ligand to the $S = {}^1/_2$ Cu(II) ion. The detailed analyses indicate that $J = -87\ \text{cm}^{-1}$ from $H = -2J\mathbf{S}_1\cdot\mathbf{S}_2$, a value which is lower than the $-J \geq 200\ \text{cm}^{-1}$ (e.g., $-2J \geq 400\ \text{cm}^{-1}$)[17] usually ascribed to "resting" oxidized forms of the cytochrome *c* oxidase enzyme.[17d]

The method of synthesis of **3** suggested to us that a hydroxo analogue might be synthetically accessible. Indeed, when **5** suspended in toluene is reacted with **6** under argon without added base, the hydroxo complex $[(F_8\text{-}TPP)Fe(III)\text{-}(OH^-)\text{-}Cu(II)(TMPA)]^{2+}$ (**4**) was isolated (85%) (Scheme I).[18] Its UV–vis properties are much like those of **6** (Soret; $\lambda_{max} = 408$ nm in CH_2Cl_2), but ^1H NMR data[18] and zero-field Mössbauer parameters ($\delta = 0.39 \pm 0.02$ mm/s and $\Delta E_Q = 0.68 \pm 0.03$ mm/s typical for high-spin ($\{porphyrin)Fe(III)\}_2\text{-}O$))[13b] reveal substantial differences from the oxo complex **3** as well as a recently described *μ*-hydroxo complex $\{[Fe(OEP)]_2(OH)\}ClO_4$ (OEP = octaethylporphyrinate),[19] indicating an influence of the Cu(II) ion binding and the bridging hydroxo ligand. Preliminary magnetic Mössbauer data also indicate magnetic coupling to the Fe with an integer-spin ground state. However, protonation of oxo complex **3** to give the hydroxo complex **4** appears to cause weakening of the Fe–Cu magnetic coupling; the room-temperature magnetic moment for **4** is $5.5 \pm 0.1\ \mu_B$ (solution, Evans method in CH_2Cl_2), compared to $5.1 \pm 0.1\ \mu_B$ (both solid and solution) for **3**.[16a] Temperature-dependent solid- and/or solution-state measurements will be needed to verify this supposition.

The clear relationship between **3** and **4** is revealed by their reversible acid–base interconversion, Scheme I. Thus, dissolution of isolated **4** in dichloromethane solvent and addition of 1.0 equiv of triethylamine produces the authentic compound **3** quantitatively (by UV–vis and ^1H NMR spectroscopies), also isolable as an analytically pure material in 50% yield. Furthermore, addition of triflic acid (1.0 equiv) to a solution of **3** in dichloromethane causes an instantaneous complete change to hydroxo species **4**, as judged by the changes in ^1H NMR and UV–vis spectra.

In conclusion, a novel essentially linear (porphyrin)iron-oxo-Cu(II) complex (**3**) has been synthesized, either from O_2 reaction with corresponding reduced iron and copper species or by an acid–base reaction of oxidized metal complexes. A hydroxo analogue is related by protonation of the oxo group, showing that appropriate choice of porphyrin and copper ligand allows for the stabilization of such bridged species. These complexes are potential structural and/or functional models for cytochrome *c* oxidase reaction intermediates or fully reacted and protonated oxidized states. While an oxo bridge has previously been suggested as a possibility for the oxidized coupled "resting"-state enzyme,[20] the linear Fe–O–Cu structure does not seem consistent with the enzyme EXAFS results.[1e,9b] Our further efforts will focus on complete characterization of these and other derivatives, as well as on the mechanism of the O_2-derived reaction. A question of considerable interest is whether there are $M\text{-}(O_2^{2-})\text{-}M'$ (M, M′ = Fe or Cu) intermediates involved.

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Supplementary Material Available: Positional and isotropic thermal parameters for $[(F_8\text{-}TPP)Fe(III)\text{-}(O^{2-})\text{-}Cu(II)\text{-}(TMPA)](ClO_4)\cdot 3C_7H_8\cdot 2CH_3CN$ (**3-ClO₄**) (4 pages). Ordering information is given on any current masthead page.

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(11) $[(F_8\text{-}TPP)Fe\text{-}(O^{2-})\text{-}Cu(TMPA)](ClO_4)\cdot{}^1/_4C_7H_{16}$ ($3\text{-}(ClO_4\cdot{}^1/_4C_7H_{16})$): ^1H NMR (CD_3CN) δ 65 (pyrrole), 9.5, 9.1 (*m*-phenyl), 7.8 (*p*-phenyl), 1.26, 0.86 (C_7H_{16}). Anal. Calcd for $C_{63.75}H_{42}ClCuF_8FeN_8O_5$: C, 58.58; H, 3.24; N, 8.57. Found: C, 58.23; H, 3.22; N, 8.28.

(12) Crystal data: $[(F_8\text{-}TPP)Fe\text{-}O\text{-}Cu(TMPA)](ClO_4)\cdot 3C_7H_8\cdot 2CH_3CN$, $3\text{-}(ClO_4)\cdot 3C_7H_8\cdot 2CH_3CN$: MW = 1640.4, orthorhombic, $a = 20.017(7)$, $b = 24.530(8)$, and $c = 15.451(4)$ Å, $V = 7587(4)$ Å^3, space group $P2_12_12_1$, $Z = 4$, $F(000) = 3376$, $D_c = 1.44$ g cm^{-3}, μ(Mo Kα) = 5.85 cm^{-1}. The data were collected ($3.5 \leq 2\theta \leq 50°$) at −90 °C on a Rigaku AFC6S diffractometer using Mo Kα ($\lambda = 0.71069$ Å) radiation. 7344 reflections were collected, and 4915 ($I \geq 3\ \sigma(I)$) were used in the analysis $R = 0.050$, $R_w = 0.054$.

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(18) $[(F_8\text{-}TPP)Fe\text{-}(OH^-)\text{-}Cu(TMPA)](ClO_4)_2\cdot C_7H_8$ ($4\text{-}(ClO_4)_2\cdot C_7H_8$): ^1H NMR (CD_2Cl_2) δ 69 (pyrrole), 11.5, 10.8 (*m*-phenyl), 7.7 (*p*-phenyl), 7.2, 2.1 (C_7H_8). Anal. Calcd for $C_{69}H_{47}Cl_2CuF_8FeN_8O_9$: C, 56.21; H, 3.21; N, 7.60. Found: C, 56.10; H, 3.20; N, 7.55.

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