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Toward Synthetic Analogues of Linked Redox and Catalytic Multimetal Sites in Proteins: A Model of the Histidine–Cysteine Bridged Dicopper Array

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Contiguous HisCys residues link a type 1 Cu electron-transfer site to a catalytic Cu-containing site in nitrite reductase and the multicopper oxidases. In efforts to understand the role of the linker in these multimetallic arrays and to design new catalysts, a mixed-valent dicopper complex comprising a bridging thiolate/N-donor ligand that models the CuHisCysCu motif was prepared and characterized by X-ray crystallography. Comparison of spectroscopic and cyclic voltammetry data to those of the mononuclear analogues of each portion of the complex, $LCuSCPh_3$ and LCu(py) (L = β-diketiminate, py = pyridyl), confirmed retention of the dicopper structure in solution.

Electron flow in metalloenzyme catalysis is often facilitated by the juxtaposition of a metal center(s) that undergoes facile redox changes near one(s) that performs substrate functionalization.[1,2] A notable example, among many, can be found in cytochrome *c* oxidase, where Cu_A and heme redox sites are arranged to efficiently funnel electrons to the catalytic heme/Cu_B center where O_2 reduction occurs.[3] While the synthetic modeling approach has provided important insights into the structure and function of such metalloprotein active sites,[4] the usual tactic has been to prepare discrete molecules that replicate the metal-containing site in isolation (a reasonable reductionist strategy). To understand how catalysis proceeds via the cooperative interaction among arrays of redox and catalytic metalloenzyme centers, however, it would be useful to construct and study the properties of molecules or supramolecular entities that incorporate similar collections of redox/catalytic metal sites.[5]

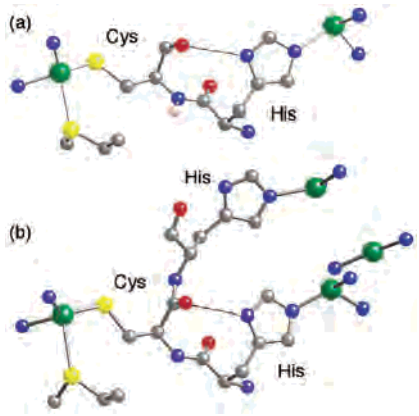

Figure 1. Active site structures emphasizing the HisCys bridge between the type 1 Cu center (left) and the catalytic Cu sites (right) of (a) nitrite reductase (pdb 1AS7) and (b) ascorbate oxidase (pdb 1AOS). The dashed connectors signify hydrogen bonds. Key: green = Cu, blue = N, red = O, yellow = S.

In one such array found in Cu nitrite reductase (NiR, Figure 1a)[6,7] and the multicopper oxidases (Figure 1b),[8,9] a type 1 copper−thiolate site is located ~12.5 Å from a single type 2 Cu center or a Cu apex of a tricopper cluster, respectively. The type 1 site functions to transfer electrons to the other Cu-containing center(s), where nitrite or dioxy-

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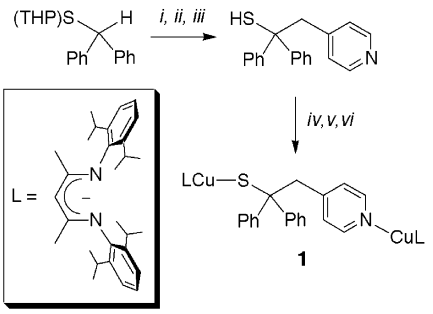
Scheme 1 [a]



[a] Reagents: (i) BuLi, (ii) 0.5 $ClCH_2PyHCl$, (iii) NEt_3, $HOCH_2CH_2SH$, BF_3Et_2O, (iv) MeLi, (v) LCuCl, (vi) LCu(MeCN).

gen reduction occurs.[10] In each protein, the cysteine ligated to the type 1 Cu is adjacent in the amino acid sequence to a histidine that ligates the partner Cu ion. This common bridging HisCys unit is presumed to play an important role in facilitating electron transfer, particularly in view of the extensive unpaired electron spin delocalization onto the cysteine thiolate in oxidized type 1 centers.[11] With the goal of better understanding this role and, ultimately, developing new catalysts, we have targeted for synthesis a model of the "CuHisCysCu" group, and we report herein the successful preparation of such a model complex using a strategy that readily lends itself to future diversification.

A key challenge to be faced in any effort to synthesize a "CuHisCysCu" unit is preparing a compound that features the low coordination number, unusual geometry, and highly covalent Cu(II)−thiolate interaction that underlie the unique spectroscopic properties of the type 1 site.[12] Such a complex, $LCuSCPh_3$ (L = β-diketiminate shown in Scheme 1), was prepared recently[13] and shown to accurately model the trigonal type 1 biosite structure, including the short and covalent Cu(II)−S(thiolate) bond.[14] We reasoned that the desired CuHisCysCu model might be accessed by replacing one of the phenyl rings in the Ph_3CS^- ligand with a suitable N-donor group that would coordinate a second metal center. Toward this end, we alkylated $(THP)SCPh_2Li$[15] with 4-picolyl chloride (Scheme 1). Rather than generating the free base prior to use in this reaction, we found it expedient to use the commercially available hydrochloride salt and 2 equiv of the lithium reagent; the $(THF)SCPh_2H$ remaining after aqueous workup was recovered in high yield (98%) for subsequent reuse. Removal of the THP group yielded the desired thiol, which was fully characterized, including by an X-ray crystal structure (Figure S1). Sequential treatment

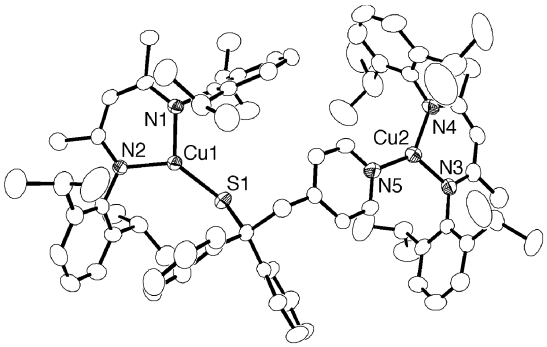


Figure 2. Representation of the X-ray crystal structure of **1**, with all non-hydrogen atoms shown as 50% ellipsoids and heteroatoms labeled. Selected interatomic distances (Å) and angles (deg): Cu1−N1, 1.900(3); Cu1−N2, 1.904(3); Cu1−S1, 2.119(1); Cu2−N3, 1.955(4); Cu2−N4, 1.926(3); Cu2−N5, 1.936(4), Cu1−Cu2, 8.6017(9); N1−Cu1−N2, 97.91(14); N1−Cu1−S1, 127.82(11); N2−Cu1−S1, 129.60(10); N3−Cu2−N4, 99.54(16); N3−Cu2−N5, 127.06(15); N4−Cu2−N5, 133.38(16).

of the deprotonated thiol in THF with LCuCl[13] followed by the Cu(I) complex LCu(MeCN)[16] yielded the mixed-valent dicopper(I,II) complex **1** (69%, dark green crystals).

The X-ray crystal structure of **1** (Figure 2) shows two 3-coordinate Cu sites ligated to the thiolate and pyridyl donors of the bridge.[17] The trigonal geometries of each site are similar to those of the corresponding mononuclear analogues, $LCuSCPh_3$[13] and LCu(py) (Figure S2),[18] and are consistent with their respective Cu(II) and Cu(I) oxidation states. Thus, the average Cu−N distances are 1.94 Å for Cu2 in **1** and 1.95 Å for LCu(py), which are longer than those for Cu1 in **1** (1.90 Å) and for $LCuSCPh_3$ (1.92 Å). The Cu−Cu separation in **1** (8.60 Å) is shorter than in the proteins (∼12.5 Å), consistent with the different number of intervening bonds in the respective bridges (8 vs 11). Finally, the Cu1−S1 distance in **1** (2.119(1) Å) matches the short 2.1243(8) Å bond length in $LCuSCPh_3$, signifying similar Cu−S bonding in the two compounds.

This similarity in Cu−S interactions between **1** and $LCuSCPh_3$ is supported by the congruence of their UV−vis and EPR spectra (Figure S3).[19] In particular, the low energy $RS^- \rightarrow$ Cu(II) CT absorption (∼750 nm) and the low $g_{||}$ and $A_{||}^{Cu}$ and high A^N values that were used to define the unique electronic structure of $LCuSCPh_3$[14] are replicated in **1**. The spectral data also point to the retention of the structure of **1**

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(17) X-ray data for **1**: monoclinic, space group $P2_1/c$, a = 9.9400(7) Å, b = 21.4515(15) Å, c = 34.032(2) Å, β = 93.661(2)°, V = 7241.8(9) Å^3, Z = 4, ρ_{calcd} = 1.149 g/cm^3. Non-hydrogen atoms were refined with anisotropic thermal parameters, and hydrogen atoms were in idealized positions with riding thermal parameters. Full-matrix least-squares refinement on F^2 converged with R1 = 0.0607, wR2 = 0.1286, GOF = 0.990 for 12782 independent reflections with $I > 2\sigma(I)$ and 772 parameters.
(18) Prepared from reaction of LCu(MeCN) with pyridine.
(19) (a) Data for **1**: UV−vis (pentane) [λ_{max}, nm (ϵ, M^{-1} cm^{-1})] 489 (2260), 747 (5700). EPR (toluene, 20 K, 9.59 GHz) $g_{||}$ = 2.17, $A_{||}^{Cu} = 113 \times 10^{-4}$ cm^{-1}, $g_\perp$ = 2.03, $A^N = 13 \times 10^{-4}$ cm^{-1}. (b) Data for $LCuSCPh_3$ (ref 13): UV−vis (heptane) [λ_{max}, nm (ϵ, M^{-1} cm^{-1})] 427 (1100), 561 (1300), 749 (5800). EPR (toluene, 20 K, 9.61 GHz) $g_{||}$ = 2.17, $A_{||}^{Cu} = 111 \times 10^{-4}$ cm^{-1}, $g_\perp$ = 2.04, $A^N = 13 \times 10^{-4}$ cm^{-1}.

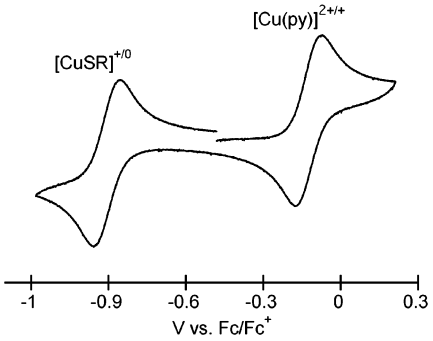

Figure 3. Cyclic voltammogram of **1** in THF. See Table 1 for conditions.

Table 1. Electrochemical Data from Cyclic Voltammetry[a]

complex	$E_{1/2}$ (V vs Fc/Fc^+)	ΔE_p (mV)[b]
1	−0.911	98
	−0.112	104
$LCuSCPh_3$[c]	−0.986	101
LCu(py)	−0.108	108

[a] Measured in THF with 0.2 M Bu_4NPF_6, Pt electrode, $Ag/AgNO_3$ reference, ambient temperature. [b] Scan rate 100 mV s^{-1}. [c] Values similar to those reported previously (ref 13).

in solution, which was further corroborated by cyclic voltammetry (Figure 3 and Table 1). Two well-separated quasireversible waves ($i_a \approx i_b$) of equal size are consistent with independent Cu(I)/Cu(II) redox processes for each site in **1**. These waves are assigned by comparison to data for $LCuSCPh_3$ and LCu(py) (Table 1), with the $E_{1/2}$ differences between the two different kinds of sites ($\Delta \sim 0.8$ V) being readily attributed to ligand charge effects (anionic thiolate vs neutral pyridyl) like those described elsewhere for a range of β-diketiminate−Cu compounds.[16,20]

In conclusion, we have prepared a mixed-valent copper complex comprising a bridging thiolate/N-donor ligand that models the CuHisCysCu motif found in NiR and the multicopper oxidases. With proof in hand of the feasibility of the synthetic strategy involving sequential attachment of LCu fragments to the thiolate and then the N-donor of the bridging ligand, other multimetal combinations can be targeted (including, for instance, porphyrin models of heme sites), and studies of electron transfer and catalysis with these combinations may be initiated.

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Supporting Information Available: Experimental details, Figures S1−S3 showing X-ray structures of $HSC(Ph)_2CH_2py$ and LCu(py) and spectra (PDF), and X-ray data (CIF). This material is available free of charge via the Internet at http://pubs.acs.org.

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