COMMUNICATION

ChemComm
www.rsc.org/chemcomm

Oxidatively robust monophenolate-copper(II) complexes as potential models of galactose oxidase†

Robertus J. M. Klein Gebbink, Masayuki Watanabe, Russell C. Pratt and T. Daniel P. Stack*
Department of Chemistry, Stanford University, Stanford, CA 94305, USA. E-mail: stack@stanford.edu

Received (in Purdue, IN, USA) 2nd January 2003, Accepted 24th January 2003
First published as an Advance Article on the web 6th February 2003

Cupric complexes of a novel phenanthroline-phenolate ligand have strongly distorted coordination geometries and electrochemical properties conducive to modeling the spectroscopy and reactivity of the enzyme galactose oxidase.

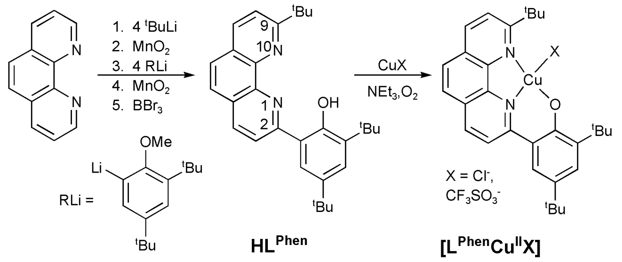
The fungal enzyme galactose oxidase (GOase)[1,2] catalyzes the aerobic oxidation of a broad range of primary alcohols to their corresponding aldehydes, without reacting with secondary alcohols, and produces hydrogen peroxide as the side-product. The enzyme combines both an organic and an inorganic one-electron co-factor—a covalently modified phenoxyl radical of a tyrosine residue and a cupric ion, respectively—in order to perform this type of two-electron oxidation catalysis. Interest in the unique features of GOase not only stems from enzymology but also from (in)organic reaction chemistry, as its chemistry represents a green method of selective oxidation.

Earlier we have reported a first generation of functional and mechanistical models for GOase based on salen-type ligands.[3,4] Several groups have subsequently reported complexes capable of stoichiometric or catalytic aerobic alcohol oxidation by GOase-reminiscent mechanisms,[5–9] and a Cu^{II}-phenoxyl complex has been structurally characterized.[10] Here, we report initial findings in the study of our second generation of complexes designed to be oxidatively and hydrolytically more robust. The phenanthroline-based ligand HL^{Phen} (Scheme 1, center) contains no weak C–H bonds that could provide pathways for oxidative decomposition, and the C=N synthetic linkages, potentially hydrolyzable in the previous generation, are reinforced by incorporation into a heterocycle. To prevent dimerization as previously observed in phenanthroline-phenolate Cu complexes,[11] an additional *tert*-butyl substituent is included at the 9-position. Modeling (DFT, B3LYP/6-31G* geometry optimization) also indicates that a 9-*tert*-butyl substituent crowds the fourth equatorial binding site of the Cu: any resulting distortion to a non-square planar geometry should aid the binding of additional exogeneous ligands (*i.e.* substrates).

The synthesis of HL^{Phen} makes use of the highly modular methodology for phenanthroline derivatization developed by Sauvage *et al.* (Scheme 1).[11,12] Reaction of 1,10-phenanthroline with 4 equiv. of *tert*-butyllithium in toluene, followed by oxidation with MnO_2, yielded 2-*tert*-butyl-1,10-phenanthroline. Subsequent alkylation with 4 equiv. of 2-methoxy-3,5-di-*tert*-butylphenyllithium gave the methoxy-protected ligand MeL^{Phen} after oxidation with MnO_2. Deprotection with BBr_3 in CH_2Cl_2 yielded HL^{Phen} in 15% overall yield. Reaction of HL^{Phen} with equimolar amounts of CuCl and NEt_3 in 1 : 1 dichloromethane/methanol followed by exposure to O_2 resulted in the formation of the green complex [L^{Phen}Cu^{II}Cl] (80% yield). Use of [$Cu^I(CH_3CN)_4$](CF_3SO_3) in the same procedure yielded [L^{Phen}Cu^{II}(CF_3SO_3)], but higher yields of this complex (90%) were achieved by metathesis of [L^{Phen}Cu^{II}Cl] with $AgCF_3SO_3$ in methanol.†

X-ray crystallographic analysis of both Cu complexes (Figs. 1 and S1†)‡ confirmed the prediction of significant distortion of the copper coordination, with remarkable agreement between the calculated and experimental structures (Figure S2).† The 9-*tert*-butyl substituent forces the copper and the fourth ligand out of the phenanthroline plane, resulting in significant tetrahedral distortion of the square planar geometry. The phenolate rings are rotated by 23° and 8° relative to the phenanthroline in the $CF_3SO_3^-$ and Cl^- structures, respectively. In both complexes, the Cu–O(phenolate) bond distance is very short (1.84–1.85 Å), and the two Cu–N distances are disparate (1.92 Å for *cis*-O *versus* 1.98–2.06 Å for *trans*-O). Also notable is the short Cu–O(triflate) distance (2.08 Å).

EPR spectroscopy of both complexes in most solvents gives broadened axial spectra (Fig. 2a; 1 mM in 4 : 1 toluene/acetone; [L^{Phen}Cu^{II}Cl]: $g_\perp = 2.07$, $g_{||} = 2.25$, $A_{||} = 130$ G; [L^{Phen}Cu^{II}(CF_3SO_3)],: $g_1 = 2.01$, $g_2 = 2.075$, $g_3 = 2.24$, $A_3 = 140$ G) consistent with tetrahedrally-distorted square planar geometry about the copper in each case. Again, the ground state configuration calculated for [L^{Phen}Cu^{II}Cl] ($x^2 - y^2$) matches the experimental data. However, the near-identical, well-resolved spectra obtained for methanol solutions of [L^{Phen}Cu^{II}(CF_3SO_3)] and [L^{Phen}Cu^{II}Cl] (Fig. 2b; $g_1 = 2.005$, $g_2 = 2.170$, $g_3 = 2.225$, $A_1 = 140$ G, A_2 = unresolved, $A_3 = 60$ G) are consistent with a rhombically-distorted z^2 ground state, an intriguingly strong perturbation of the neutral complexes that suggests complete solvolysis of the counterions and binding of alcohol ligands.



Scheme 1

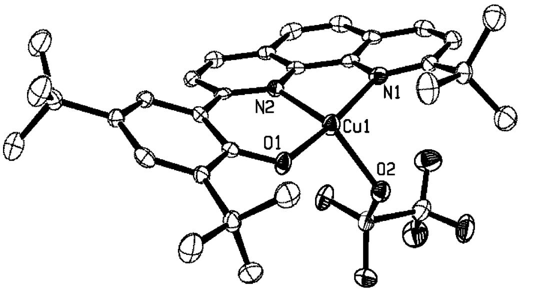


Fig. 1 ORTEP diagram of [L^{Phen}Cu^{II}(CF_3SO_3)] shown with 50% thermal ellipsoids. Hydrogen atoms are omitted for clarity. Selected bond lengths (Å) and angles (°): Cu(1)–N(1) 1.9892(2), Cu(1)–N(2) 1.921(2), Cu(1)–O(1) 1.838(2), Cu(1)–O(2) 2.082(2). N(2)–Cu(1)–(O2) 137.8(1). For [L^{Phen}Cu^{II}Cl]: Cu(1)–N(1) 2.060(5), Cu(1)–N(2) 1.919(5), Cu(1)–O(1) 1.847(4), Cu(1)–Cl(1) 2.277(2), N(2)–Cu(1)–Cl(1) 134.8(2).

---

† Electronic supporting information (ESI) available: Synthetic procedures and instrumental methods, crystallographic data for [L^{Phen}Cu^{II}(CF_3SO_3)] and [L^{Phen}Cu^{II}Cl], and Figures S1, S2 and S3. See http://www.rsc.org/suppdata/cc/b2/b212921c/

DOI: 10.1039/b212921c

UV-Vis experiments show the high affinity of $[\mathrm{L^{Phen}Cu^{II}Cl}]$ for alcoholate ligands: a 1:1 complex is formed with *e.g.* methoxide anion in MeOH (Fig. S3).† The analogous EPR-titration shows the transformation of the rhombic signal of $[\mathrm{L^{Phen}Cu^{II}(MeOH)Cl}]$ into a simple axial signal ($g_\perp = 2.05$; $g_{||} = 2.24$, $A_{||} = 169$ G; Fig. 2c), consistent with the formation of a four-coordinate complex in which both the alcoholate and the phenolate ligand are bound within the equatorial plane. Similar optical results are obtained for the binding of benzyl alcoholate in neat benzyl alcohol. Most interestingly, the methoxide species is not susceptible to oxidation by O_2, whereas the benzyl alcoholate species rapidly decays upon introduction of O_2.

Cyclic voltammograms of $[\mathrm{L^{Phen}Cu^{II}Cl}]$ and $[\mathrm{L^{Phen}Cu^{II}(CF_3SO_3)}]$ (1 mM, 0.1 M Bu_4NClO_4, CH_2Cl_2, 100 mV s^{-1}) show a single reversible process at +0.56 V *vs.* Fc^+/Fc ascribed to one-electron oxidation of the phenolic moiety. UV-Vis titration with the strong one-electron oxidant tris(4-bromophenyl)aminium hexachloroantimonate (TPA) (CH_2Cl_2/CH_3CN, −80 °C) shows that one electron per $[\mathrm{L^{Phen}Cu}]^+$ unit can be removed. The strong absorption bands of the oxidized complexes at 415 and 475 nm ($\varepsilon \sim 8000$) are indicative of the formation of phenoxyl radical species (Fig. 3).[13–15] In addition, the complexes formed are essentially EPR-silent in analogy to the antiferromagnetically coupled Cu^{II}-phenoxyl radical in the oxidized form of GOase.[14,16]

In summary, a new generation of copper-phenolate complexes with a design focused on chemical robustness have been synthesized and characterized. The formation of one-electron-oxidized forms of $[\mathrm{L^{Phen}Cu^{II}Cl}]$ and $[\mathrm{L^{Phen}Cu^{II}(CF_3SO_3)}]$, mimicking the oxidized form of galactose oxidase, and the affinity of the neutral forms for alcohol- and alcoholate ligands show promise for further studies of alcohol oxidation.

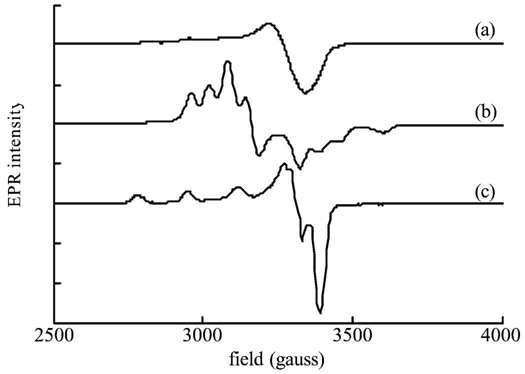


Fig. 2 X-band EPR spectra (1 mM solutions, 77 K, 9.51 GHz) of $[\mathrm{L^{Phen}Cu^{II}Cl}]$ in 4:1 toluene/acetone (a) and in methanol (b), and $[\mathrm{L^{Phen}Cu^{II}(OMe)}]$ in methanol (c).

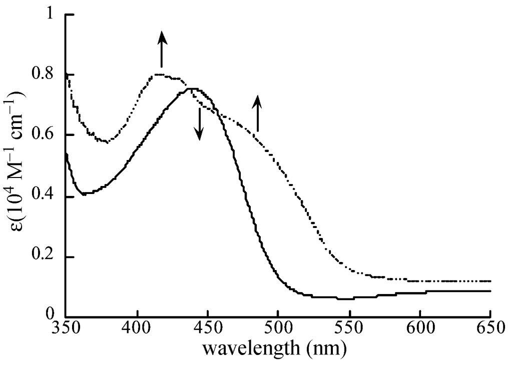


Fig. 3 UV-Vis spectra (CH_2Cl_2/CH_3CN, −80 °C, 0.11 mM) of $[\mathrm{L^{Phen}Cu^{II}Cl}]$ (solid) and $[\mathrm{L^{Phen}Cu^{II}Cl}]^+$ (dashed); arrows indicate spectral changes observed upon oxidation with TPA.

We acknowledge the Netherlands Organization for Scientific Research (NWO) for a postdoctoral fellowship (R.J.M.K.G.) and the Japan Atomic Energy Research Institute (JAERI) for a fellowship to M.W. R.C.P. is the grateful recipient of an Urbanek Family Stanford Graduate Fellowship. Additional funding was provided by the NIH (T.D.P.S., GM50730).

Notes and references

‡ *Crystal data* for $[\mathrm{L^{Phen}Cu^{II}(CF_3SO_3)}]$: $C_{31}H_{35}CuF_3N_2O_4S$, olive green plate, $M = 652.21$, orthorhombic, space group *Pbca*(#61), $a = 14.549(0)$, $b = 14.583(0)$, $c = 28.183(1)$, $V = 5979.4(2)$ Å^3, $Z = 8$, $D_{\mathrm{calc}} = 1.45$ g cm^{-3}, $T = 150$ K, $F(000) = 2712.00$, μ(Mo-Kα) = 8.6 cm^{-1}, 24474 reflections collected, 4290 unique ($R_{\mathrm{int}} = 0.049$). The final agreement factors are $R_1 = 0.030$ for 2805 data with $F_0 > 4\sigma(F_0)$ and $wR_2 = 0.064$ for all data. CCDC 201829.

Crystal data for $[\mathrm{L^{Phen}Cu^{II}Cl}]\cdot CH_2Cl_2$: $C_{30}H_{35}ClCuN_2O\cdot CH_2Cl_2$, green needle, $M = 621.50$, triclinic, space group P-1(#2), $a = 8.8538(9)$, $b = 13.1058(14)$, $c = 13.7598(16)$, $\alpha = 88.321(4)$, $\beta = 74.910(3)$, $\gamma = 77.254(4)$, $V = 1502.9(5)$ Å^3, $Z = 2$, $D_{\mathrm{calc}} = 1.37$ g cm^{-3}, $T = 150$ K, $F(000) = 646$, μ(Mo-Kα) = 10.2 cm^{-1}, 6970 reflections collected, 4245 unique ($R_{\mathrm{int}} = 0.053$). The final agreement factors are $R_1 = 0.063$ for 2077 data with $F > 4\sigma(F)$ and $wR_2 = 0.154$ for all data. CCDC 201828. See http://www.rsc.org/suppdata/cc/b2/b212921c/ for crystallographic files in CIF or other electronic format.

1 J. P. Klinman, *Chem. Rev.*, 1996, **96**, 2541–2561.
2 J. W. Whittaker and M. M. Whittaker, *Pure Appl. Chem.*, 1998, **70**, 903–910.
3 Y. Wang and T. D. P. Stack, *J. Am. Chem. Soc.*, 1996, **118**, 13097–13098.
4 Y. Wang, J. L. DuBois, B. Hedman, K. O. Hodgson and T. D. P. Stack, *Science*, 1998, **279**, 537–540.
5 P. Chaudhuri, M. Hess, U. Florke and K. Wieghardt, *Angew. Chem., Int. Ed.*, 1998, **37**, 2217–2220.
6 P. Chaudhuri, M. Hess, T. Weyhermuller and K. Wieghardt, *Angew. Chem., Int. Ed.*, 1999, **38**, 1095–1098.
7 P. Chaudhuri, M. Hess, J. Muller, K. Hildenbrand, E. Bill, T. Weyhermuller and K. Wieghardt, *J. Am. Chem. Soc.*, 1999, **121**, 9599–9610.
8 S. Itoh, M. Taki, S. Takayama, S. Nagatomo, T. Kitagawa, N. Sakurada, R. Arakawa and S. Fukuzumi, *Angew. Chem., Int. Ed.*, 1999, **38**, 2774–2776.
9 F. Thomas, G. Gellon, I. Gautier-Luneau, E. Saint-Aman and J.-L. Pierre, *Angew. Chem., Int. Ed.*, 2002, **41**, 3047–3050.
10 L. Benisvy, A. J. Blake, D. Collison, E. S. Davies, C. D. Garner, E. J. L. McInnes, J. McMaster, G. Whittaker and C. Wilson, *Chem. Commun.*, 2001, 18.
11 B. M. Holligan, J. C. Jeffery and M. D. Ward, *J. Chem. Soc., Dalton Trans.*, 1992, 3337–3344.
12 C. O. Dietrich-Buchecker, P. A. Marnot and J. P. Sauvage, *Tetrahedron Lett.*, 1982, **23**, 5291–5294.
13 J. A. Halfen, B. A. Jazdzewski, S. Mahapatra, L. M. Berreau, E. C. Wilkinson, L. Que and W. B. Tolman, *J. Am. Chem. Soc.*, 1997, **119**, 8217–8227.
14 J. Muller, T. Weyhermuller, E. Bill, P. Hildebrandt, L. OuldMoussa, T. Glaser and K. Wieghardt, *Angew. Chem., Int. Ed.*, 1998, **37**, 616–619.
15 A. Sokolowski, H. Leutbecher, T. Weyhermuller, R. Schnepf, E. Both, E. Bill, P. Hildebrandt and K. Wieghardt, *J. Biol. Inorg. Chem.*, 1997, **2**, 444–453.
16 M. M. Whittaker and J. W. Whittaker, *Biophys. J.*, 1993, **64**, 762–772.