Investigation of the Reactive Oxygen Intermediate in an Arene Hydroxylation Reaction Performed by Xylyl-Bridged Binuclear Copper Complexes

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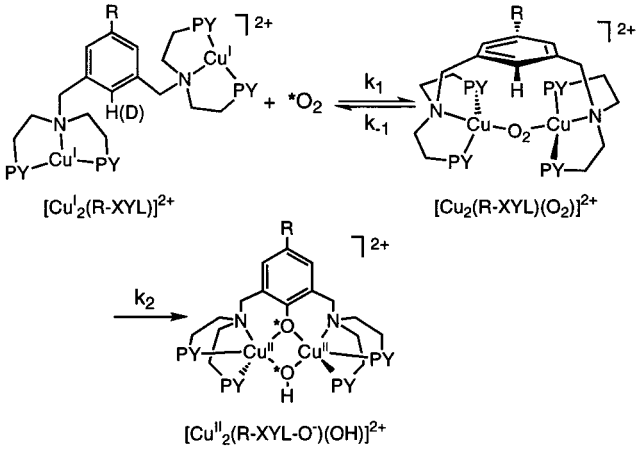
Abstract: The kinetics of the reaction, *via* an oxygen-bound intermediate, of $[Cu^I_2(NO_2\text{-XYL})][ClO_4]_2\cdot CH_3CN$ to $[Cu_2(NO_2\text{-XYL-O-})(OH)]^{2+}$, where the bridging arene is hydroxylated, have been examined with use of resonance Raman spectroscopy. A resonance Raman peak characteristic of peroxide bound in a side-on, μ-η^2:η^2 geometry is observed upon oxygenation of $[Cu^I_2(NO_2\text{-XYL})]$ for both intramolecularly and intermolecularly bridged complexes. The decay of the intramolecularly bridged peroxide stretch at ∼750 cm^{-1} and the growth of the phenolate stretch of the product at 1320 cm^{-1} were monitored over time with use of an excitation wavelength of 406.9 nm. Both the decay of the peroxide stretch and the growth of the phenolate stretch were found to be first order, and the rate constants are consistent, within experimental error, with the peroxide intermediate reacting directly to form the hydroxylated product. The possibility of an unobservable amount of a bis-μ-oxo isomer which is in rapid equilibrium with the side-on peroxide species, and that is responsible for the hydroxylation reaction, is considered. An upper limit for the concentration of the bis-μ-oxo isomer in a solution of $[Cu_2(NO_2\text{-XYL})(O_2)]^{2+}$ was determined. This gives the lower limit for its rate of reaction to form the phenolate product, which is approximately 1000 times faster than the decay of the peroxide intermediate. A comparison of the reactivities of the side-on peroxide and bis-μ-oxo isomers with respect to electrophilic aromatic substitution is made by using frontier molecular orbital theory. This correlation, in conjunction with the estimated, relative rates of reaction for the two isomers to form phenolate product, leads to a molecular mechanism in which the side-on peroxide isomer is likely to be the reactive oxygen intermediate in these systems.

Introduction

Tyrosinase (EC 1.14.18.1) is an oxygenase that occurs widely in bacteria, fungi, plants, and mammals. The enzyme is responsible for the hydroxylation and oxidation of phenols to *o*-quinones.[1] Chemical and spectroscopic studies[2−4] have shown that tyrosinase contains a coupled dicopper active site very similar to that of hemocyanin. Oxygenation of the dicopper(I) core yields a stable oxy form with an absorption spectrum (λ_{max} (ϵ, M^{-1} cm^{-1}) 345 (26 000), 520 (CD), 590 nm (1000)) very similar to that of oxy-hemocyanin (λ_{max} (ϵ, M^{-1} cm^{-1}) 345 (20 000), 485 (CD), 570 nm (1000)), which is known to possess a side-on (or μ-η^2:η^2) peroxo bridging geometry.[5]

A number of copper complexes containing dinucleating ligands where two bis[2-(2-pyridyl)ethyl]amine (PY2) tridentate ligands are linked *via* the amino nitrogen by a xylyl spacer ($[Cu_2(H\text{-XYL})]^{2+}$) have been synthesized.[6] Reaction of dioxygen with $[Cu_2(H\text{-XYL})]^{2+}$ produces a copper(II)-peroxide intermediate that further reacts to yield a hydroxylated phenoxo and hydroxo doubly bridged dicopper(II) complex $[Cu_2(H\text{-XYL-O-})(OH)]^{2+}$ (Scheme 1).[7] The structure of $[Cu_2(H\text{-XYL-O-})(OH)]^{2+}$ has been confirmed by X-ray crystallography, and isotope labeling studies have shown that the source of the

Scheme 1



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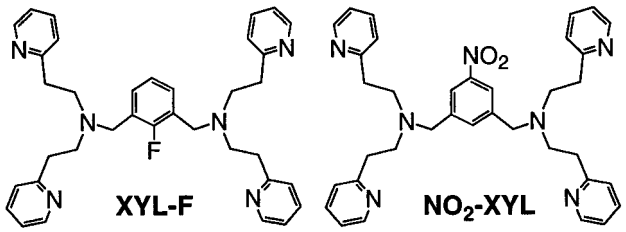
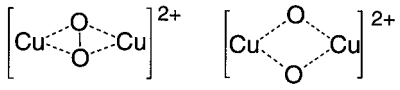
Scheme 2



Scheme 3



oxygen atoms in this compound is dioxygen.[7] This reaction involves the activation of oxygen for hydrocarbon oxygenation and has relevance to the mechanism of reaction of tyrosinase, or other copper monooxygenases.

By substitution of NO_2 *para* to (NO_2-XYL), or F at (XYL-F), the site of arene ring hydroxylation (Scheme 2), it has been possible to slow the hydroxylation process to the point that the substituted $[Cu_2(XYL)(O_2)]^{2+}$ intermediates are stabilized (−80 °C) and can be observed spectroscopically.[6,8] The absorption spectra of $[Cu_2(NO_2\text{-}XYL)(O_2)]^{2+}$ (λ_{max} (ϵ, M^{-1} cm^{-1}) 358 (20 000), 435 (5000), and 530 nm (1200), acetone and dichloromethane) and $[Cu_2(XYL\text{-}F)(O_2)]^{2+}$ (λ_{max} (ϵ, M^{-1} cm^{-1}) 360 (18 700), 435 (4400), 515 nm (1300), acetone and dichloromethane) are similar to those obtained for oxy-hemocyanin and oxy-tyrosinase with the exception of an additional, reasonably intense band at 435 nm.

Recently, novel dicopper bis-μ-oxo cores ($[Cu_2(\mu\text{-}O)_2]^{2+}$) have been structurally and spectroscopically characterized.[9−11] It has been shown that the bis-μ-oxo dicopper cores capped by sterically hindered N,N′,N″-trisubstituted 1,4,7-triazacyclononane (TACN) ligands undergo facile interconversion with a $[Cu_2(\mu\text{-}\eta^2\text{:}\eta^2)\text{-}O_2]^{2+}$ core upon change of solvent.[9] The absorption spectrum obtained for the bis-μ-oxo Cu(II) moiety, $[(TACN^{iPr3})_2Cu_2(\mu\text{-}O)_2]^{2+}$, is very different from that of the side-on peroxide isomer, with bands at 324 (ϵ = 11 000 M^{-1} cm^{-1}) and 448 nm (ϵ = 13 000 M^{-1} cm^{-1}). These findings suggest that the absorption spectra observed for $[Cu_2(NO_2\text{-}XYL)(O_2)]^{2+}$ and $[Cu_2(XYL\text{-}F)(O_2)]^{2+}$ may be due to a mixture of $[Cu_2(\mu\text{-}\eta^2\text{:}\eta^2)\text{-}O_2]^{2+}$ species and $[Cu_2(\mu\text{-}O)_2]^{2+}$ species, the additional band (relative to the side-on bridged structure) at 435 nm being derived from the $[Cu_2(\mu\text{-}O)_2]^{2+}$ core. Either or both of these oxygen species could be responsible for the hydroxylation reaction. The nature of the oxygen intermediates present in $[Cu_2(NO_2\text{-}XYL)(O_2)]^{2+}$ and $[Cu_2(XYL\text{-}F)(O_2)]^{2+}$ and a discussion of their catalytic competence are presented below. This is an important and timely study since, as emphasized in ref 39, it is possible that an unobservable amount of the bis-μ-oxo isomer is responsible for the observed reactivity.

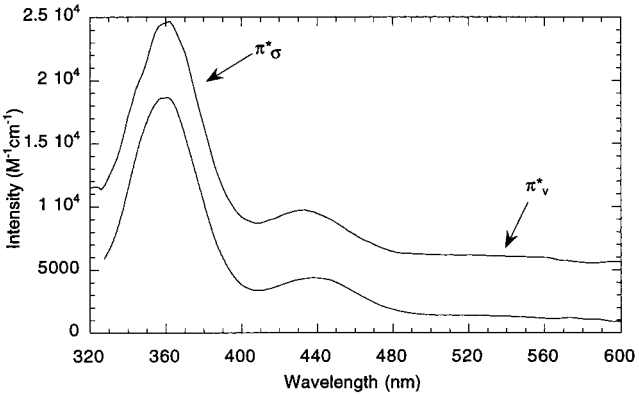


Figure 1. UV−vis absorption spectra of $[Cu_2(NO_2\text{-}XYL)(O_2)][ClO_4]_2$ (top, *y* axis offset by 5000 M^{-1} cm^{-1}, for clarity) and $[Cu_2(XYL\text{-}F)(O_2)][ClO_4]_2$ (bottom) in dichloromethane solution at −80 °C. Peroxide π^*_σ and π^*_v to Cu(II) charge-transfer bands are indicated by arrows. The presence of the π^*_v band was established from higher sensitivity scans and resonance Raman excitation profiles.

Experimental Section

The substituted $[Cu^I_2(XYL)][ClO_4]_2\cdot 1CH_3CN$ precursors for solution samples were prepared as reported previously.[6,8] Resonance Raman spectra were obtained with a Princeton Instruments ST-135 back-illuminated CCD detector on a Spex 1877 CP triple monochromator with 1200, 1800, and 2400 grooves/mm holographic spectrograph gratings. The excitation was provided by Coherent I90C-K Kr+ and Innova Sabre 25/7 Ar+ CW ion lasers. A polarization scrambler was used between the sample and the spectrometer. Spectral resolution was <2 cm^{-1}. Samples for solution Raman spectra were prepared by dissolving substituted $[Cu^I_2(XYL)][ClO_4]_2\cdot 1CH_3CN$ in CH_2Cl_2 or CH_3COCH_3 in an NMR tube and oxygenating at −95 °C. The sample tube was spun with an air-driven NMR spinner and cooled to ∼180 K by a N_2-flow system. Integrated peak intensities used to monitor concentrations were measured relative to the 700 cm^{-1} peak of CH_2Cl_2 or to the 790 cm^{-1} peak of CH_3COCH_3 for solution samples. Isotopic substitution of the O_2^{2-} oxygens was achieved by oxygenation of the complex with $^{18}O_2$ (Isotech, 99% labeled).

Results and Analysis

Identification of the Side-On Peroxide Species. The electronic absorption spectra obtained for $[Cu_2(NO_2\text{-}XYL)(O_2)]^{2+}$ and $[Cu_2(XYL\text{-}F)(O_2)]^{2+}$ in CH_2Cl_2 at ∼200 K are shown in Figure 1. The charge-transfer spectra of $[Cu_2(NO_2\text{-}XYL)(O_2)]^{2+}$ and $[Cu_2(XYL\text{-}F)(O_2)]^{2+}$ are very similar; both exhibit an intense absorption feature at ∼360 nm, a less intense band at 435 nm, and a weak feature at ∼530 nm. These absorption spectra show similarities to those obtained for $[Cu_2(N\mathit{n}PY2)(O_2^{2-})]^{2+}$ (where N*n* are binucleating ligands with PY2 groups linked by *n* $-(CH_2)-$ groups, *n* = 3, 4, 5)[12,13] but are distinct from the absorption spectra of the $\mu\text{-}\eta^2\text{:}\eta^2$ peroxo species in $[Cu(HB(3,5\text{-}R_2pz)_3)]^{2+}\cdot(O_2)$ (where $HB(3,5\text{-}R_2pz)_3$ is hydrotris(3,5-dialkyl-1-pyrazolyl)borate, R = Ph, ^{i}Pr, Bn)[14,15] and those of $[Cu_2(\mu\text{-}O)_2]^{2+}$ cores isolated by Tolman and Stack.[9−11] The absorption bands at 350 and 538 nm observed for $[Cu(HB(3,5\text{-}i\text{-}Pr_2pz)_3)]^{2+}\cdot(O_2)$ have been assigned as per-

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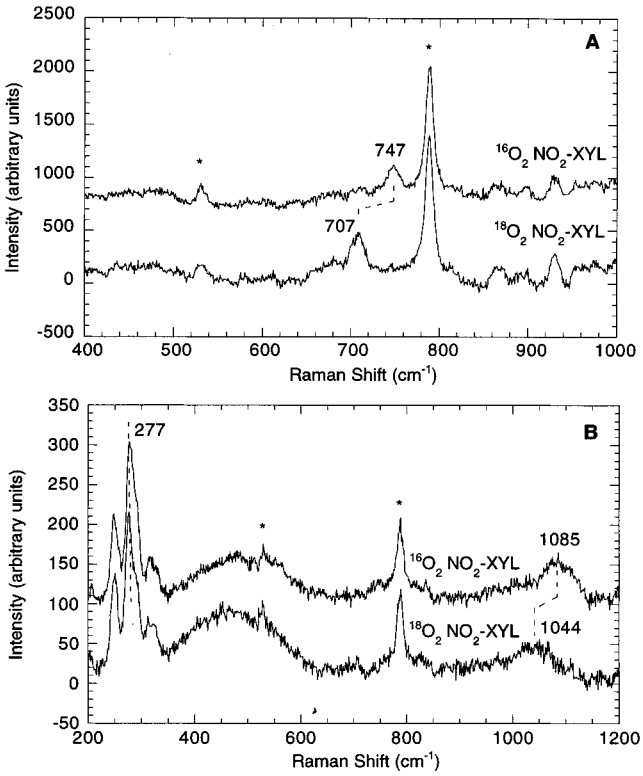

Figure 2. Resonance Raman spectra obtained for ~4 mM solutions of $^{16}O_2$ and $^{18}O_2$ $[Cu_2(NO_2\text{-XYL})(O_2)]^{2+}$ in acetone with an excitation wavelength of (A) 413 and (B) 363.9 nm. Peaks sensitive to oxygen isotope substitution are labeled with their Raman shifts; an asterisk denotes peaks derived from acetone solvent.

oxide π^*_σ and π^*_v to Cu(II) charge-transfer bands, respectively,[15] characteristic of the μ-η^2:η^2 structure; an absorption band at 435 nm is not present in the spectrum of $[Cu(HB(3,5\text{-}i\text{-}Pr_2pz)_3)]^{2+}\cdot(O_2)$.

To probe the origin of the absorption band at 435 nm, resonance Raman spectra of ~4 mM solutions of $[Cu_2(NO_2\text{-XYL})^{16}O_2]^{2+}$ and $[Cu_2(NO_2\text{-XYL})^{18}O_2]^{2+}$ in acetone, at ~180 K, were obtained by exciting into this absorption band, using an excitation wavelength of 413 nm. A single Raman peak is observed at 747 cm^{-1}, which shifts with $^{18}O_2$ substitution to 707 cm^{-1} (Figure 2A). This peak is assigned to the symmetric O−O stretch of a side-on bound peroxide, based upon its frequency and isotope shift (40 cm^{-1}). No oxygen isotope-sensitive peak at ~600 cm^{-1} (in a solution of $[Cu_2(NO_2\text{-XYL})\text{-}O_2]^{2+}$ in acetone or dichloromethane (vide infra)) which is the symmetric stretch of the bis-μ-oxo dicopper core in $[(TACN^{iPr3}\text{-}Cu)_2(\mu\text{-}O)_2](ClO_4)_2$,[10] was observed. Therefore the additional absorption band at 435 nm seen for $[Cu_2(NO_2\text{-XYL})(O_2)]^{2+}$ and $[Cu_2(XYL\text{-F})(O_2)]^{2+}$ is not due to the presence of a bis-μ-oxo species. Raman peaks at 277 and 1085 cm^{-1} are observed (Figure 2B) in the spectrum of an approximately 4 mM solution of $[Cu_2(NO_2\text{-XYL})^{16}O_2]^{2+}$ in acetone, at ~180 K using an excitation wavelength of 363.9 nm (nearly coincident with the highest energy ligand-to-metal charge-transfer (LMCT) band, Figure 1).[16] In analogy to the vibrational data obtained for the side-on peroxide dicopper core in $[Cu(HB(3,5\text{-}Ph_2pz)_3)]_2\cdot(O_2)$,[15] the peak at 1085 cm^{-1} (isotope shift of 41 cm^{-1}) is assigned to the overtone of the asymmetric Cu−O core stretch and the peak at 277 cm^{-1} is assigned to the symmetric Cu−O stretch, which

(16) Enhancement of the O−O stretch from the high-energy LMCT band at ~360 nm is very weak since the donor orbital is strongly Cu−O bonding in character.[15]

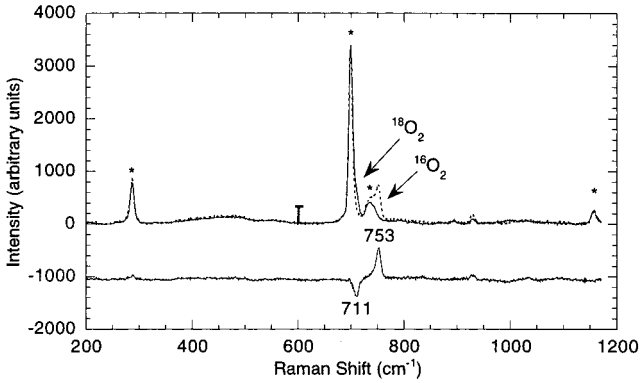

Figure 3. Resonance Raman spectra obtained with an excitation wavelength of 406.9 nm of solutions of $^{16}O_2$ (dashed line, top) and $^{18}O_2$ (solid line, top) $[Cu_2(NO_2\text{-XYL})(O_2)]^{2+}$ in dichloromethane. The bar at ~600 cm^{-1} indicates the intensity of a peak that would be due to the presence of 1% of bis-μ-oxo species in solution. The difference spectrum is shown (bottom) and peaks sensitive to oxygen isotope substitution are labeled with their Raman shifts; an asterisk denotes peaks derived from dichloromethane solvent.

primarily involves copper motion and hence shows no isotope shift upon $^{18}O_2$ substitution. A resonance Raman profile of the 277 cm^{-1}, Cu−O stretch shows it is associated with the 435 nm absorption band, thus this absorption band must be assigned as a $O_2^{2-} \rightarrow$ Cu charge-transfer transition of the side-on bridged structure. The origin of this additional band for the $[Cu_2(\mu\text{-}\eta^2\text{:}\eta^2)\text{-}O_2]^{2+}$ structure is presently being investigated.

Possible Presence of a Bis-μ-oxo Complex. It has been proposed that there exists a rapid equilibrium between the two copper core isomers, where O_2 is bound as a side-on peroxide or a bis-μ-oxo species.[9] Thus, in a solution of $[Cu_2(NO_2\text{-XYL})\text{-}O_2]^{2+}$, a small amount of bis-$\mu$-oxo isomer, undetectable with resonance Raman, could be present and responsible for the observed reactivity. The resonance Raman peak at ~600 cm^{-1}, diagnostic of the bis-μ-oxo, dicopper core, shows unusually strong enhancement when exciting into the absorption band at ~400 nm, and therefore is a very sensitive probe for low concentrations of the bis-μ-oxo isomer. Resonance Raman spectra were obtained for solutions of $[Cu_2(NO_2\text{-XYL})]^{2+}$ oxygenated with $^{16}O_2$ and $^{18}O_2$ in dichloromethane to establish an upper limit for the concentration of the bis-μ-oxo isomer that could be present in solution. The difference Raman spectrum in the region of 600 cm^{-1} showed no isotope-sensitive peak (Figure 3). By using the ratio of the intensity of the noise to the CH_2Cl_2 peak at 700 cm^{-1} and comparing this intensity ratio to that of a 1 mM solution of $[(L_{TM})_2Cu_2O_2]^{2+}$ in CH_2Cl_2 (where L_{TM} is tetramethyl-1,2-cylclohexane diamine),[11] the upper limit of the concentration of bis-μ-oxo isomer in a 4 mM solution of $[Cu_2(NO_2\text{-XYL})(O_2)]^{2+}$ was found to be 0.005 mM and the ratio of side-on peroxide isomer to bis-μ-oxo is 1:<0.0013.

Intramolecular Peroxide Formation. During the course of these studies, some interesting concentration effects have been observed. Resonance Raman spectra of a concentrated (~8 mM) solution of $[Cu_2(NO_2\text{-XYL})(O_2)]^{2+}$ in acetone, $^{16}O_2$, and $^{18}O_2$ at 406.9 nm excitation at ~180 K are shown in Figure 4A. Two peaks are observed at 747 and 733 cm^{-1}; the peak at 747 cm^{-1} is very similar to that observed for the low-concentration solution $[Cu_2(NO_2\text{-XYL})(O_2)]^{2+}$. The new peak at lower energy (733 cm^{-1}), which shifts 38 cm^{-1} upon substitution with $^{18}O_2$ and which becomes more intense as the concentration of the Cu(I) solution increases, is assigned to a symmetric O−O stretch of intermolecularly bound μ-η^2:η^2

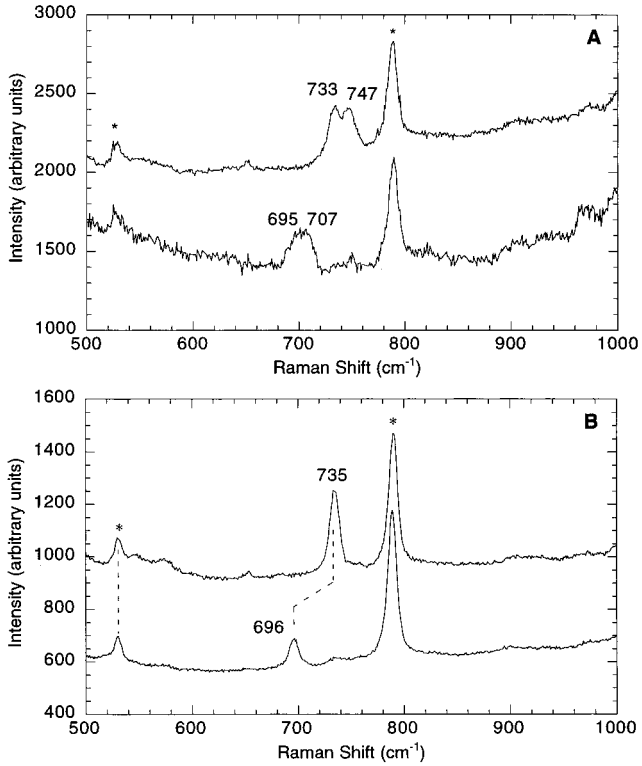

Figure 4. (A) Resonance Raman spectra obtained with an excitation wavelength of 406.9 nm of ~8 mM solutions of $^{16}O_2$ and $^{18}O_2$ [Cu_2-(NO_2-XYL)$(O_2)]^{2+}$ in acetone. Peaks sensitive to oxygen isotope substitution are labeled with their Raman shifts; an asterisk denotes peaks derived from acetone solvent. (B) Resonance Raman spectra obtained with an excitation wavelength of 406.9 nm of solutions of $^{16}O_2$ and $^{18}O_2$ [Cu_2(XYL-F)$(O_2)]^{2+}$ in acetone. Peaks sensitive to oxygen isotope substitution are labeled with their Raman shifts; an asterisk denotes peaks derived from acetone solvent.

peroxide (where peroxide is bridging between the copper atoms of two binuclear molecules). Resonance Raman spectra of [Cu_2-(XYL-F)$(O_2)]^{2+}$ in acetone at ~180 K at 406.9 nm excitation are shown in Figure 4B. The spectrum shows a peak at 735 cm^{-1} that is assigned as the symmetric O−O stretch of an intermolecularly bound peroxide; this is based upon its low frequency which is similar to that observed for the intermolecularly bound peroxide in [Cu_2(NO_2-XYL)$(O_2)]^{2+}$. In contrast to [Cu_2(NO_2-XYL)$(O_2)]^{2+}$, the intermolecular peroxide stretch was observed exclusively in [Cu_2(XYL-F)$(O_2)]^{2+}$ solutions, over the range of ~3−8 mM initial concentrations of the dicopper-(I) complex, indicating that the fluoride substituent at the position of arene ring hydroxylation interferes with the intramolecular binding of O_2. No oxygen isotope sensitive peak indicative of a bis-μ-oxo species was observed at ~600 cm^{-1}.

Kinetics of Hydroxylation. Attack of the 2-position of the arene ring by [Cu_2(R-XYL)$(O_2)]^{2+}$ leads to hydroxylation and formation of the phenoxo and hydroxo doubly bridged dicopper-(II) complex [Cu_2(R-XYL-O-)(OH)$]^{2+}$.[7] The absorption spectrum of [Cu_2(XYL-O-)(OH)$]^{2+}$ has a phenolate to copper(II) charge-transfer band at 440 nm[6] and resonance Raman into this band (using 488.0 nm excitation) shows the symmetric (C−O) vibration of [Cu_2(XYL-O-)(OH)$]^{2+}$ at 1307 cm^{-1}.[17] An intense resonance Raman peak is observed in [Cu_2(NO_2-XYL-O-)(OH)$]^{2+}$at 1320 cm^{-1} with 406.9 nm excitation, which is also assigned to the symmetric C−O stretch. Using resonance Raman spectroscopy it was possible to monitor the time course of the hydroxylation reaction by observing the disappearance of $\nu_{(O-O)}$ at ~750 cm^{-1} of [Cu_2(NO_2-XYL)$(O_2)]^{2+}$ in dichloromethane and acetone, and the appearance of $\nu_{(C-O)}$ at 1320 cm^{-1} of [Cu_2(NO_2-XYL-O-)(OH)$]^{2+}$ using an excitation wavelength of 406.9 nm (Figure 5). Aliquots of a 4 mM stock solution of [Cu_2(NO_2-XYL)$(O_2)]^{2+}$ in dichloromethane and acetone were oxygenated concurrently and maintained at ~195 K. At intervals over a period of several hours resonance Raman spectra were recorded; a fresh aliquot was used for each spectrum to minimize the effects of photodecomposition of the oxygenated complex. The intensities of the O−O stretch and C−O stretch for [Cu_2(NO_2-XYL)$(O_2)]^{2+}$ in dichloromethane and acetone as a function of time are plotted in Figure 6. Over the same time period, spectra of aliquots of a 4mM solution of [Cu_2-(XYL-F)$(O_2)]^{2+}$ in acetone were recorded as a control for the stability of a peroxide bound xylyl system that does not decay *via* hydroxylation of the arene ring. The concentration of [Cu_2-(XYL-F)$(O_2)]^{2+}$ remained constant throughout the experiment, and there was no detectable amount of phenoxo-bridged dicopper complex formed. The decay of the side-on bound peroxide species in [Cu_2(NO_2-XYL)$(O_2)]^{2+}$ in dichloromethane and in acetone at 195 K followed first-order kinetics with rate constants of 6.7×10^{-5} $s^{-1} \pm 0.7 \times 10^{-5}$ s^{-1} and 9.4×10^{-5} $s^{-1} \pm 1.2 \times 10^{-5}$ s^{-1}, respectively, as determined from the intensity of the peroxide stretch at ~750 cm^{-1} with respect to solvent (*vide supra*) and assuming 100% side-on peroxide at time $t = 0$ s. These results are consistent with a rate constant calculated previously for the reaction of [Cu_2(NO_2-XYL)$(O_2)]^{2+}$ to [Cu_2(NO_2-XYL-O-)(OH)$]^{2+}$ at 183 K in dichloromethane of 1.7×10^{-5} s^{-1}.[6] The relative concentration of the phenolate product was determined by comparing the intensity of the $\nu_{(C-O)}$ at 1320 cm^{-1} to the intensity of this peak in a solution of 100% phenolate product; all spectra were normalized by using the intensity of solvent at 700 cm^{-1} (dichloromethane) or 790 cm^{-1} (acetone). The rate constants for the formation of the phenolate product in dichloromethane and acetone of $6.0 \times 10^{-5} \pm 0.7 \times 10^{-5}$ s^{-1} and $7.1 \times 10^{-5} \pm 1.1 \times 10^{-5}$ s^{-1}, respectively, were determined (using a first-order equation for growth) from plotting $\ln(([O-O]_{t=0} - [C-O]_{t=t})/[O-O]_{t=0})$ *vs* time. These data are in accord (within experimental error) with the side-on-peroxide species reacting directly to form the phenolate product. However, the viability of the bis-μ-oxo species as the reactive intermediate needs to be assessed. Therefore, since the upper limit established for the concentration of the bis-μ-oxo isomer is approximately one thousandth of the concentration of the side-on peroxide isomer ([bis-μ-oxo]/[side-on peroxide] = 0.0013), the calculated lower limit of the rates of reaction for the bis-μ-oxo isomer to phenolate product at 195 K in dichloromethane and acetone are 0.05 and 0.06 s^{-1}, respectively, approximately 1000 times faster than the rate that would be associated with the [Cu_2(μ-η^2:η^2)-$O_2]^{2+}$ isomer reacting directly to give product.

Discussion

Resonance Raman spectroscopy has identified two species present in high-concentration solutions of [Cu_2(NO_2-XYL)-$(O_2)]^{2+}$; two peaks assigned as the O−O stretch of peroxide bound in a μ-η^2:η^2 geometry have been observed at 735 and ~750 cm^{-1}. Solutions of initial Cu(I) concentrations in the range of 3−8 mM of [Cu_2(XYL-F)$]^{2+}$ yielded a single Raman peak at 735 cm^{-1} upon oxygenation. An oxygenated, low-concentration solution of [Cu_2(NO_2-XYL)$]^{2+}$ (≤4 mM) showed a single Raman peak at 747 cm^{-1}. Due to the concentration dependence of the peaks at 735 and ~750 cm^{-1} they have been

(17) Pyrz, J. W.; Karlin, K. D.; Sorrell, T. N.; Vogel, G. C.; Que, L. *Inorg. Chem.* **1984**, *23*, 4581−4584.

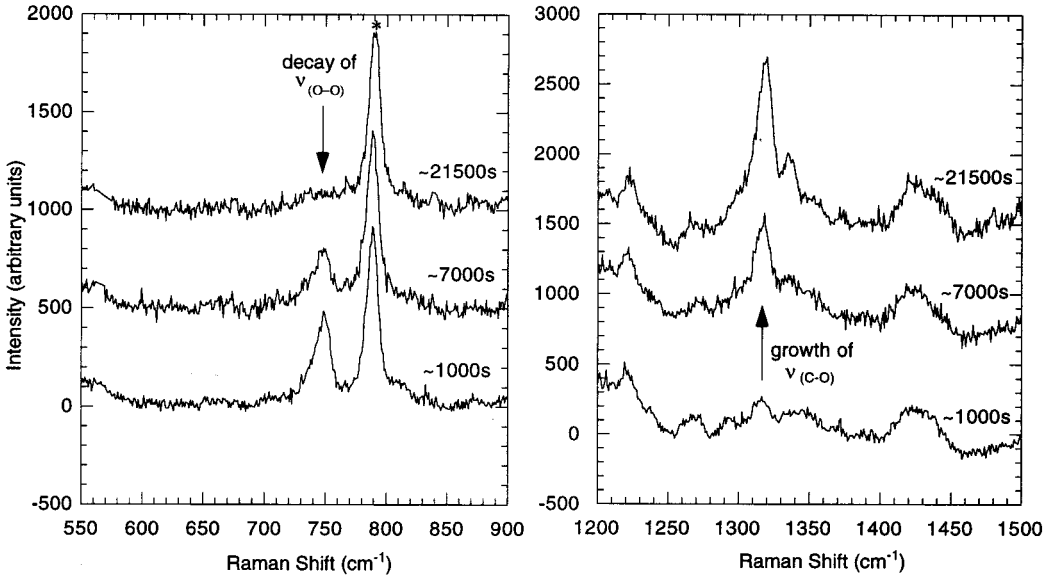

Figure 5. Resonance Raman spectra obtained with an excitation wavelength of 406.9 nm of an ~4 mM solution of $[Cu_2(NO_2\text{-XYL})(O_2)]^{2+}$ in acetone. (Left) The decay of the O−O stretch at 750 cm^{-1}. (Right) The growth of the C−O stretch at 1320 cm^{-1}. An asterisk denotes peaks derived from acetone solvent.

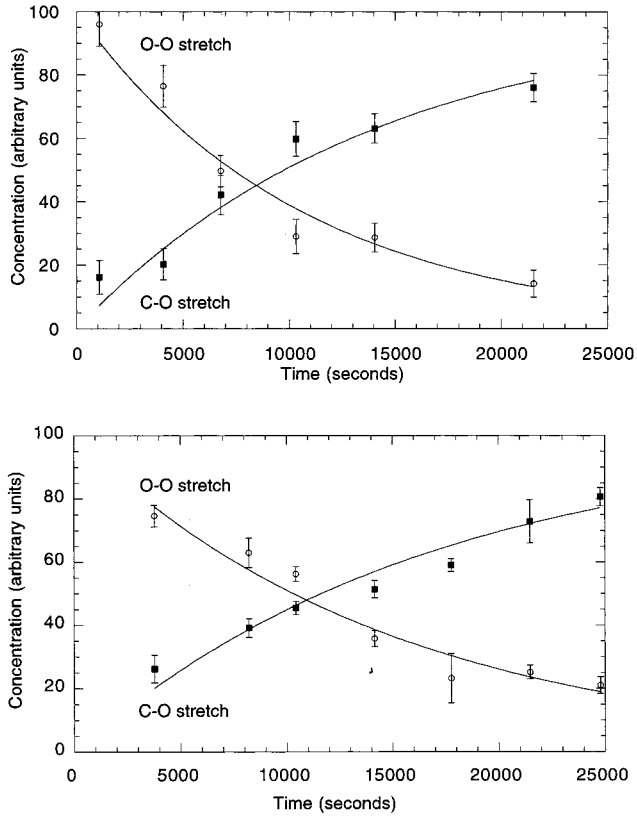

Figure 6. Concentration of side-on peroxide and phenolate product as determined from the integrated intensities of $\nu_{(O-O)}$ and $\nu_{(C-O)}$ as a function of time for $[Cu_2(NO_2\text{-XYL})(O_2)]^{2+}$ in acetone (top) and dichloromethane (bottom).

assigned to intermolecular and intramolecular peroxide-bridged copper complexes, respectively. The possibility of intermolecular oxygenation occurring with these *m*-xylyl containing dinucleating ligands has been proposed previously.[6] Analysis of the dinucleating ligand (obtained by extraction) after room-temperature oxygenation of solutions of $[Cu^I_2(R\text{-XYL-H})]^{2+}$ (where R = NO_2, ^{t}Bu, F, and CN) in dichloromethane showed that 10−25% (depending on the concentration of Cu(I) used in the reaction solution) of the product did not contain a hydroxylated benzene ring. It was postulated that at higher concentrations of the dicopper(I) complex an intermolecular O_2-binding process may become favorable and the resulting cluster species is incapable of arene hydroxylation due to the unfavorable geometry of the bridging oxygen for xylyl attack.[6] Therefore the appearance of a resonance Raman peak associated with an intermolecular peroxide generated upon oxygenation of a high-concentration solution of the binuclear copper(I) complex $[Cu_2(NO_2\text{-XYL})]^{2+}$ is consistent with previous results. In a similar study, only a small amount (<3%) of hydroxylated product was recovered from an oxygenated solution of $[Cu_2(XYL\text{-F})O_2]^{2+}$ in CH_2Cl_2; the major product from isolation was intact $[Cu_2(XYL\text{-F})]^{2+}$.[8] This result was ascribed to the presence of an electronically deactivated ring and a strong C−F bond; however, the data obtained from resonance Raman indicate that the major product from oxygenation contains peroxide bound intermolecularly, and therefore inactive toward arene ring hydroxylation.

Resonance Raman data obtained for $[Cu_2(NO_2\text{-XYL})(O_2)]^{2+}$ and $[Cu_2(XYL\text{-F})(O_2)]^{2+}$ show peaks associated with dioxygen bound as peroxide in a μ-η^2:η^2 geometry. No vibrations associated with the presence of a $[Cu_2(\mu\text{-}O)_2]^{2+}$ core have been observed for oxygenated solutions for a range of initial Cu(I) concentrations. Oxygenation of a related ligand system, where 4,7-diisopropyl-1,4,7-triazacyclononane ligands are tethered by a *m*-xylyl linker ($[(m\text{-XYL}^i\text{Pr4})Cu_2(CH_3CN)]^{2+}$), yielded both an *intermolecularly* bridged bis-μ-oxo complex ("dimer-of-dimer" and/or oligmeric complexes) and an intramolecularly bridged side-on peroxide isomer.[18,19] Oxygenation of related diisopropyl-substituted TACN ligands, in which one of the nitrogen donors is bonded to CH_2R, gave binuclear, bis-μ-oxo cores,[18] with the exception of $[(m\text{-XYL}^i\text{Pr4})Cu_2(CH_3CN)]^{2+}$-(*vide supra*) indicating that the relatively stiff *m*-xylyl linker holds the copper atoms sufficiently far apart to inhibit the formation of a binuclear, intramolecular bis-μ-oxo species, characterized by a Cu−Cu distance of approximately 2.8 Å. It is interesting to note that for solutions of $[Cu_2(NO_2\text{-XYL})(O_2)]^{2+}$ and $[Cu_2(XYL\text{-F})(O_2)]^{2+}$ in acetone and dichloromethane, there

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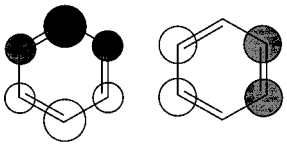
(19) Mahapatra, S.; Kaderli, S.; Llobet, A.; Neuhold, Y.-M.; Palanché, T.; Halfen, J. A.; Young, V. G.; Kaden, T. A.; Que, L.; Zuberbühler, A. D.; Tolman, W. B. *Inorg. Chem.* **1997**, *36*, 6343−6356.

Scheme 4



is no evidence for the presence of a bis-μ-oxo species even when dioxygen is bound intermolecularly and hence when the constraints on the Cu−Cu distance are relaxed. Therefore it appears that neither the electronic structure of the N-donor ligands nor the geometric constraints of the *m*-xylyl linker favor formation of a bis-μ-oxo isomer for the complexes $[Cu_2(NO_2\text{-}XYL)(O_2)]^{2+}$ and $[Cu_2(XYL\text{-}F)(O_2)]^{2+}$.

It has been proposed[4,6] that the hydroxylation of the arene ring proceeds *via* direct electrophilic attack of the ring by the oxygen-bound intermediate as evidenced by the lack of a deuterium kinetic isotope effect at the position of arene ring hydroxylation (thereby precluding H-atom abstraction as the rate-determining step),[20] the increase in the rate of product formation with the addition of electron-donating substituents to the arene ring[6] and identification of products from an NIH shift reaction when methyl is substituted at the 2-position of the arene ring.[21] Reactivity studies have shown that both the side-on peroxide complexes $[Cu_2(N\mathit{n}PY_2)(O_2)]^{2+}$ and the bis-μ-oxo complex $[(TACN^{Bn3}Cu)_2(\mu\text{-}O)_2]^{2+}$ exhibit electrophilic behavior: $[Cu_2(N\mathit{n}PY_2)(O_2)]^{2+}$ reacts with PPh_3 to yield O=PPh_3 and oxidizes 2,4-di-*tert*-butylphenol to 3,3′,5,5′-tetra-*tert*-butyl-2,2′-dihydroxybiphenyl under an inert atmosphere;[22] $[(TACN^{Bn3}Cu)_2(\mu\text{-}O)_2]^{2+}$ reacts with ferrocene in the presence of $HBF_4{\cdot}EtO_2$ to give $[Cp_2Fe]^+$ and generates 2 equiv of monomeric Cu(II)−semiquinone complexes upon reaction with catechols.[23] The analysis of the kinetic data obtained for the reaction of $[Cu_2(NO_2\text{-}XYL)(O_2)]^{2+}$ to $[Cu_2(NO_2\text{-}XYL\text{-}O\text{-})(OH)]^{2+}$ suggests that either the side-on peroxide isomer reacts directly to form the phenolate product at a rate of $\sim 1 \times 10^{-5}$ s^{-1} or, to be the dominant species responsible for the formation of the product, a small amount of bis-μ-oxo isomer present in equilibrium reacts >1000 times faster than the observed decay of the peroxide species.

If the bis-μ-oxo isomer is the reactive oxygen intermediate, its greater reactivity must be ascribed to a more electrophilic character than the side-on peroxide isomer. A consideration of the reactivities of the bis-μ-oxo and side-on peroxide isomers can be made with use of frontier molecular orbital theory. In general the important frontier orbitals for electrophilic attack are the LUMO (lowest unoccupied molecular orbital) of the electrophile and the HOMO (highest occupied molecular orbital) of the nucleophile, *i.e.* the LUMO of the Cu_2O_2 core and the HOMO of arene ring. A small HOMO−LUMO splitting will yield a large interaction energy for the reaction. Also the symmetry of the interacting orbitals and the coefficients of these orbitals in the relevant frontier molecular orbitals have to be considered. If the arene ring of $[Cu_2(H\text{-}XYL)(O_2)]^{2+}$ is approximated to benzene, the HOMO is degenerate and comprises the MO's shown in Scheme 4.

The energy of the HOMO, estimated from the ionization potential, is −9.2 eV and the LUMO is to much higher energy (*ca.* −1.5 eV). Results of electronic structure calculations for the side-on peroxide core[35] and the bis-μ-oxo core[32,38] can be used to establish the frontier molecular orbital description and thus gain insight into the relative reactivities of the two cores. To facilitate comparison of the molecular orbital descriptions of the two isomers, results were used from SCF-Xα-SW density functional theory[24−29] for both Cu_2O_2 cores with the same ligand set: $[(NH_3)_4Cu_2(\mu\text{-}\eta^2\text{:}\eta^2)O_2]^{2+}$ [35] and $[(NH_3)_4Cu_2(\mu\text{-}O)_2]^{2+}$.[30,31] The LUMO of the $\mu\text{-}\eta^2\text{:}\eta^2$ peroxide isomer ($LUMO_{side-on\ peroxide}$) is calculated to have an energy of −5.4 eV and comprises an antibonding interaction between Cu $d_{x^2-y^2}$ (70%) and O_2^{2-} π^*_σ (14.5%). The LUMO of the bis-μ-oxo core ($LUMO_{bis-\mu-oxo}$) is described by 62% Cu $d_{x^2-y^2}$ and 20% O_2^{2-} σ^* in an antibonding configuration at −6.0 eV.[30] However, the LUMO+1 of the bis-μ-oxo core is situated only ∼0.2 eV above the LUMO and is very similar to the LUMO of the side-on peroxide isomer in both orbitals and coefficients (68% Cu $d_{x^2-y^2}$ and 14% π^*_σ).[31] Thus, the possible Cu_2O_2 core molecular orbitals which can participate in the reaction contain the O_2^{2-} π^*_σ and σ^* orbitals. From the X-ray crystal structure of the product (Figure 7A), it can be seen that the O−O−C2−C5 vector is close to linear but the arene ring is tilted approximately 35° to the plane of the Cu_2O_2 plane. A reaction pathway involving the σ^* (oxygen component of $LUMO_{bis-\mu-oxo}$) is possible for both the bis-μ-oxo and the side-on peroxide isomers; although the σ^* containing molecular orbital for the side-on peroxide core is calculated to be at a much higher energy (−1.0 eV) than that of the bis-μ-oxo core (−6.0 eV), it has a greater oxygen σ^* orbital coefficient of ∼60%. Approach of the σ^* along the C5−C2−O2−O1 axis would lead to a symmetry-forbidden overlap (no net bonding) with the π orbitals of $HOMO_{benzene}$ (Figure 7B, bottom). However, the oxygen σ^* orbital could interact with $HOMO_{benzene}$ if it approached the π orbitals of the arene ring either above or below the plane. For a nonzero overlap between σ^* and $HOMO_{benzene}$ the arene ring would have to tilt away from the Cu_2O_2 core (Scheme 5, bottom). Due to the square-based pyramidal geometry of the 5-coordinate Cu(II)'s, the angled orientation of the arene ring (and hence the amino nitrogens oriented on the same side of the Cu_2O_2 core) would require the two tridentate nitrogen donor units to be in an "eclipsed" configuration, and the *cis* axial ligands on the opposite side of the Cu_2O_2 plane to the arene ring, to minimize steric repulsion between the arene ring and the pyridine rings. In fact, an X-ray crystal structure of a related

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(31) Cramer *et al.* have performed calculations using multiconfigurational *ab initio* theory on the side-on peroxide and bis-μ-oxo isomers; the LUMO of the bis-μ-oxo and side-on peroxide isomers are described by an antibonding interaction between the Cu $d_{x^2-y^2}$ and oxygen π^*_σ, with the LUMO(side-on peroxide) ∼1.1 eV below that of the LUMO(bis-μ-oxo).[32] The descriptions of the energies and composition of the frontier molecular orbitals for both the side-on peroxide and bis-μ-oxo cores appear to be relatively independent of the type of calculation used (*ab initio* and Density Functional Theory).

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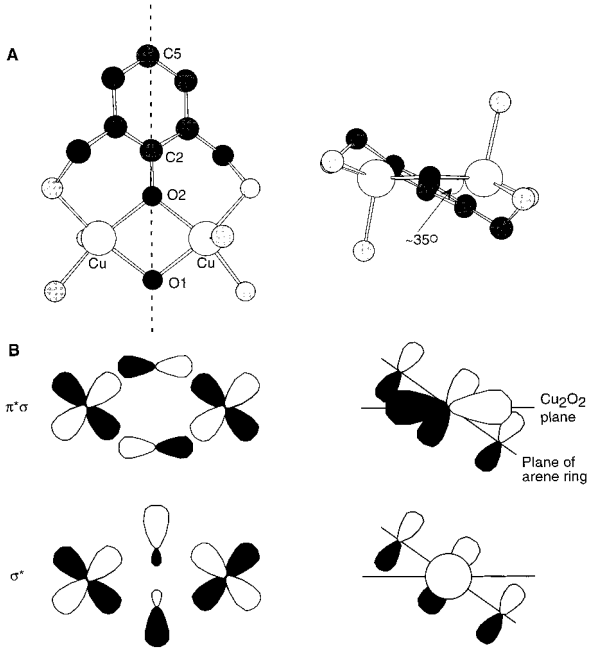
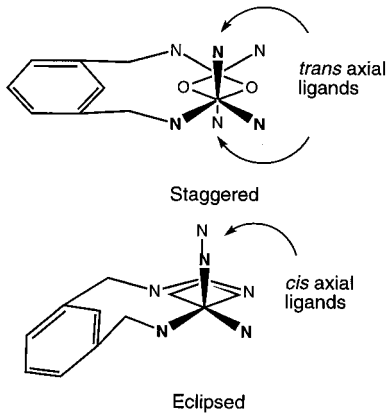


Figure 7. (A) Views of the X-ray crystal structure of $[Cu_2(H\text{-}XYL)(O\text{-})(OH)]^{2+}$ (left) perpendicular to the Cu_2O_2 plane and (right) parallel to the Cu_2O_2 plane. Ligand backbone removed for clarity. (B) (Left) Illustrations of the molecular orbitals containing π^*_σ (top) and σ^* (bottom) viewed from perpendicular to the Cu_2O_2 plane. (Right) Interaction of the π^*_σ (top) and σ^* (bottom) with the HOMO of benzene viewed along the O1−O2 axis.

Scheme 5



system (where one carbon atom is removed from the xylyl linker) $[Cu_2(UN\text{−}O\text{-})(OH)]^{2+}$ shows that the equivalent C5−C2−O2−O1 vector is no longer linear; the arene ring is at an angle of 143° to the Cu_2O_2 core, and the axial pyridines are oriented *cis* and on the opposite side to the arene ring.[33] The crystal structure of the product $Cu_2(H\text{-}XYL\text{-}O\text{-})(OH)]^{2+}$ [7] shows the axial pyridines to be in a *trans* configuration, indicating the arene ring remained approximately parallel to the Cu_2O_2 core during attack, thereby decreasing the likelihood of an angled approach by the O_2^{2-} σ^* containing molecular orbital.

Due to the dihedral angle of ~35° of the arene ring with respect to the Cu_2O_2 plane, the O_2^{2-} π^*_σ orbital, present as approximately 14% of both the $LUMO_{side-on\ peroxide}$ and $LUMO{+}1_{bis-\mu-oxo}$ would have a symmetry allowed interaction with $HOMO_{benzene}$ (Figure 7B, top). Since the energies of $LUMO_{side-on\ peroxide}$ and $LUMO{+}1_{bis-\mu-oxo}$ are separated by only ~0.4 eV, and the coefficients of these MO's are very similar, the reactivity of both isomers toward electrophilic attack of the arene ring via π^*_σ is expected to be comparable.[34] The same conclusion can be reached by using the results of the calculations of Cramer *et al.*[32] due to the similarity of the LUMO for the bis-μ-oxo and side-on peroxide cores. Further, from copper K-edge studies,[40] there is significant additional charge donation from copper to the oxo in the bis-μ-oxo isomer relative to the side-on peroxide structure, which is consistent with SCF-Xα calculations,[30] and would disfavor electrophilic attack by the oxo-bridge. Certainly there appears to be little evidence to suggest that the bis-μ-oxo isomer is more reactive in arene ring hydroxylation and hence the "fast" reaction rate of the bis-μ-oxo isomer in comparison to the side-on peroxide species (which is required by the large concentration difference in solution observed with resonance Raman) is not supported by the MO description of the relative reactivities. However, hydrogen atom abstraction reactions which may proceed *via* the σ^* molecular orbital of the Cu_2O_2 core appear to be favored by the bis-μ-oxo isomer due to the deeper energy of the σ^* containing MO (−6.0 eV) compared with that of the side-on peroxide core (−1.0 eV), as observed experimentally by Tolman *et al.*[23,19]

Tyrosinase exhibits striking similarities to the coupled binuclear copper site in hemocyanin.[3] The main difference between the two sites appears to be the higher accessibility of the tyrosinase active site to exogenous ligands.[35] Tyrosinase activates O_2 for reaction with phenols oxygenating them to *o*-quinones.[1] The side-on bound peroxide is activated for cleavage due to the presence of some peroxide σ^* character in the HOMO, which weakens the O−O bond.[36] It has been demonstrated that the monophenol substrate binds directly to the copper at the active site and undergoes an associative rearrangement toward the equatorial plane for hydroxylation.[2] Since the phenol substrate is a good electron donor ligand, coordination of substrate to the copper center would shift electron density into the LUMO, which is antibonding with respect to both the Cu−O and O−O bonds. The question of whether bond scission occurs prior to or concomitant with substrate activation is as yet unanswered.[37]

However, the observation of inter- and intramolecular side-on peroxide isomers in solution with use of resonance Raman, the results obtained from the kinetic study of the reaction of $[Cu_2(NO_2\text{-}XYL)(O_2)]^{2+}$ to $[Cu_2(NO_2\text{-}XYL\text{-}O\text{-})(OH)]^{2+}$ and the application of frontier molecular orbital theory to the relative reactivity of the side-on peroxide and bis-μ-oxo isomers indicate that hydroxylation of the arene ring in $[Cu_2(XYL)(O_2)]^{2+}$ complexes is likely to occur via a π^*_σ mechanism involving direct attack by the $\mu\text{-}\eta^2\text{:}\eta^2$ peroxo dicopper core.

Acknowledgment. This work was supported by grants from the National Institutes of Health (DK31450 (E.I.S.) and GM28962 (K.D.K.)).

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