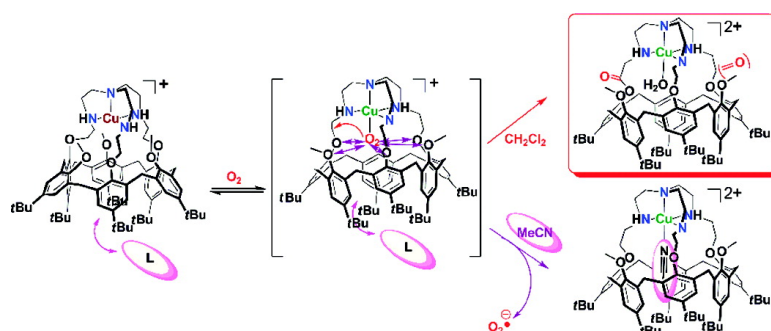


## Dioxygen Activation at a Mononuclear Cu(I) Center Embedded in the Calix[6]arene-Tren Core

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## Dioxygen Activation at a Mononuclear Cu(I) Center Embedded in the Calix[6]arene-Tren Core

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**Abstract:** The reaction of a cuprous center coordinated to a calix[6]arene-based aza-cryptand with dioxygen has been studied. In this system, Cu(I) is bound to a tren unit that caps the calixarene core at the level of the small rim. As a result, although protected from the reaction medium by the macrocycle, the metal center presents a labile site accessible to small guest ligands. Indeed, in the presence of O<sub>2</sub>, it reacts in a very fast and irreversible redox process, leading, ultimately, to Cu(II) species. In the coordinating solvent MeCN, a one electron exchange occurs, yielding the corresponding [CalixtrenCu–MeCN]<sup>2+</sup> complex with concomitant release of superoxide in the reaction medium. In a noncoordinating solvent such as CH<sub>2</sub>Cl<sub>2</sub>, the dioxygen reaction leads to oxygen insertions into the ligand itself. Both reactions are proposed to proceed through the formation of a superoxide-Cu(II) intermediate that is unstable in the Calixtren environment due to second sphere effects. The transiently formed superoxide ligand either undergoes fast substitution for a guest ligand (in MeCN) or intramolecular redox evolutions toward oxygenation of Calixtren. Interestingly, the latter process was shown to occur twice on the same ligand, thus demonstrating a possible catalytic activation of O<sub>2</sub> at a single cuprous center. Altogether, this study illustrates the oxidizing power of a [CuO<sub>2</sub>]<sup>+</sup> adduct and substantiates a mechanism by which copper mono-oxygenases such as DβH and PHM activate O<sub>2</sub> at the Cu<sub>M</sub> center to produce such an intermediate capable of C–H breaking before the electron input provided by the noncoupled Cu<sub>H</sub> center.

### Introduction

Dioxygen activation at a metal center for the functionalization of a C–H bond is a subject of tremendous importance in chemistry,<sup>1–3</sup> as well as in biology.<sup>2–4</sup> The mono-oxygenase activity of many enzymes is based on dioxygen interaction with iron or copper, which allows one to bypass the kinetic barrier that prevents O<sub>2</sub> to “burn” organic matter. Interestingly, subclasses of enzymes that use the same strategies can be identified. For copper enzymes, the activation process is based on the Cu(II)/Cu(I) redox couple, either at a dinuclear or at a

mononuclear center.<sup>5,6</sup> While for coupled dinuclear copper systems,<sup>7</sup> it is now admitted that the activation process occurs through the bielectronic reduction of O<sub>2</sub> by the two Cu(I) centers leading to a Cu<sup>II</sup>O<sub>2</sub>Cu<sup>II</sup> peroxy dinuclear adduct, the reactivity at a mononuclear center is still highly controversial.<sup>6–9</sup> Copper monooxygenases, PHM (peptidylglycine α-hydroxylating monooxygenase)<sup>10</sup> and DβH (dopamine β-hydroxylase),<sup>11</sup> involve a common strategy relying on two noncoupled mononuclear copper centers separated by a solvent cleft (Figure 1, top). Until recently, it was more or less admitted that a two electron reduction of O<sub>2</sub> was necessary to obtain a species reactive enough to break a C–H bond, a Cu(II)–hydroperoxide

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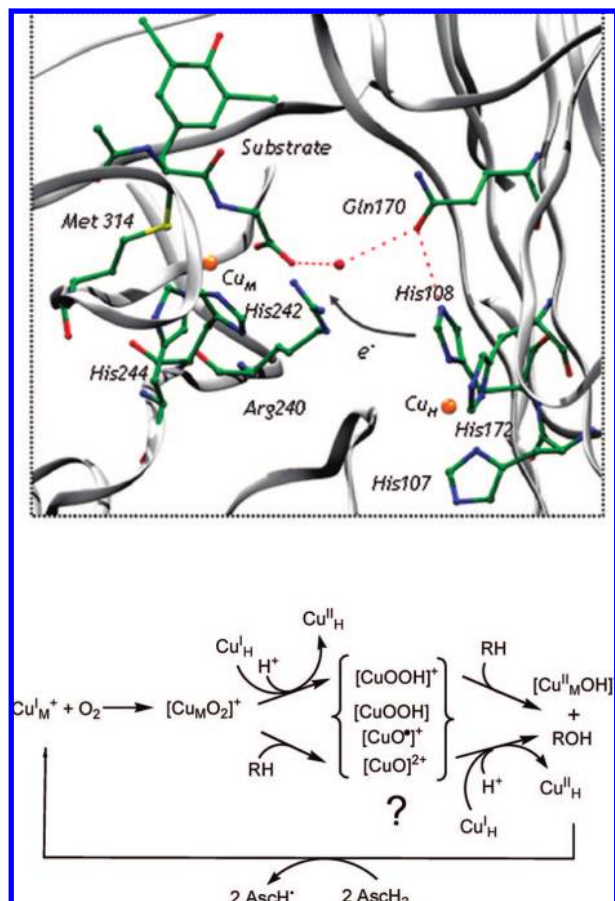
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**Figure 1.** Top: Active site of PHM. Bottom: Alternative pathways for the O insertion into a C–H bond at the active site of the Cu-monooxygenases.

{[Cu<sup>II</sup>(<sup>-</sup>OOH)]<sup>+</sup>}. A large number of investigations have been devoted to this system and have addressed the capability of such an intermediate to achieve the hydroxylation at aliphatic substrates (Figure 1, bottom, upper pathway).<sup>8</sup>

However, more and more biochemical evidence suggests that the oxidation mechanism proceeds through the direct attack of the C–H bond by a copper-superoxo [Cu<sup>II</sup>(O<sub>2</sub><sup>-</sup>)<sup>+</sup>] intermediate (Figure 1, bottom, lower pathway).<sup>10–14</sup> This possibility has recently been explored by quantum chemistry.<sup>15,16</sup> Investigations of the oxidative processes induced by mononuclear

copper–O<sub>2</sub> adducts are difficult to carry out because model compounds usually undergo fast dimerization or exhibit slow reactivity due to sterical crowding around the Cu center.<sup>6</sup> With N<sub>4</sub> ligands, for example, detailed thermodynamic and kinetic studies have shown the formation of a mononuclear [N<sub>4</sub>Cu(O<sub>2</sub>)]<sup>+</sup> adduct followed by its rapid trapping by a second [N<sub>4</sub>Cu(I)]<sup>+</sup> complex leading to the formation of well identified dinuclear Cu(II) peroxy complexes that irreversibly undergo decomposition (Scheme 1).<sup>17–20</sup> As a matter of fact, degradation of copper ligands in the presence of O<sub>2</sub> has often been reported.

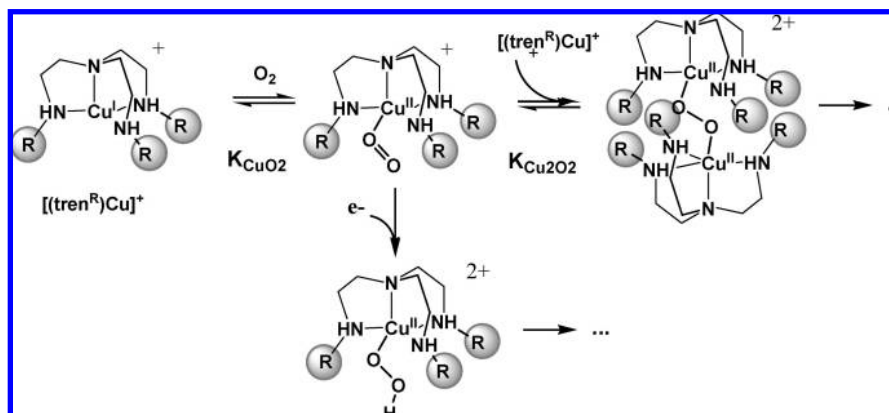
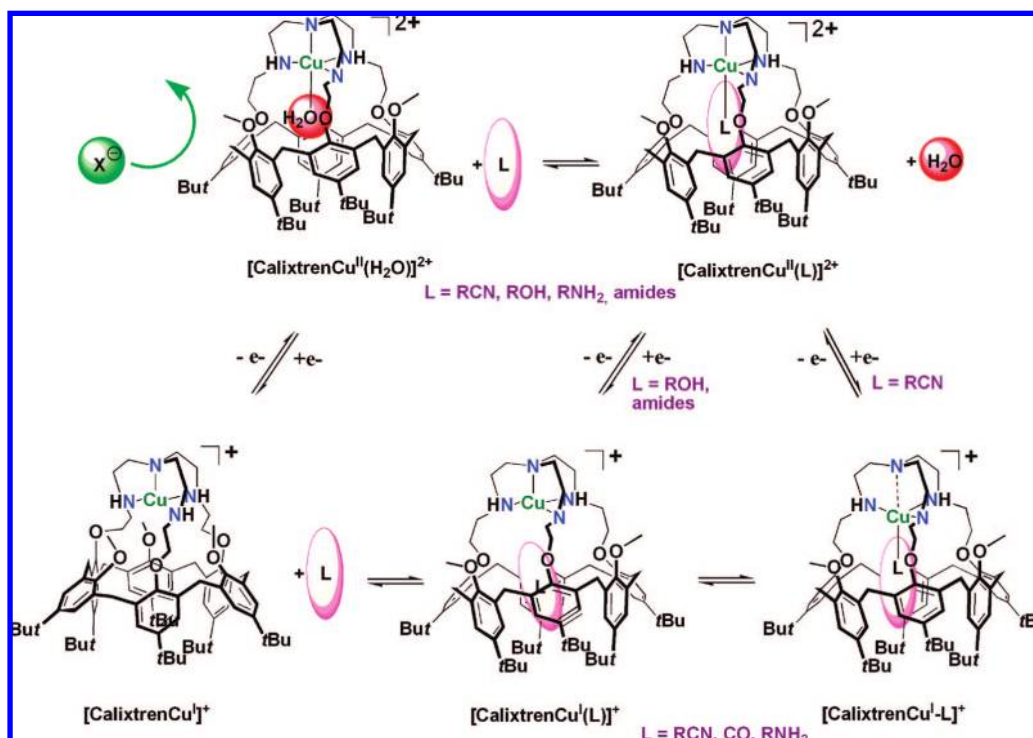
Most recently, isolation at low T of [N<sub>4</sub>Cu(O<sub>2</sub>)]<sup>+</sup> adducts has been successfully achieved thanks to the use of strongly donating N<sub>4</sub> ligands.<sup>9,21,22</sup> It has been shown that these complexes undergo interesting reactive pathways, leading to the oxidation of exogenous substrates<sup>22,23</sup> and/or to the intramolecular oxygen insertion into a C–H bond of the ligand.<sup>24</sup> In some studies, it has been suggested that the [N<sub>4</sub>Cu(O<sub>2</sub>)]<sup>+</sup> adduct, identified as a Cu(II) superoxide complex, is transiently reduced through a one electron process (either by another Cu(I) center or by an H-donor such as a phenol) to yield a Cu<sup>II</sup>OOH intermediate<sup>25–27</sup> proposed to be directly responsible for the observed C–H cleavage.

A fundamental question, however, remains unanswered: is a single isolated cuprous center capable of activating a dioxygen molecule for the oxygenation of an organic substrate? In that context, the challenging modeling of a mononuclear site still appears as a key for a better understanding of its intrinsic reactivity.

We have designed a family of ligands based on calix[6]arene cones capped at the small rim by a nitrogenous coordination core. The so-called funnel complexes present a metal center with a labile coordination site embedded at the small rim of the cavity.<sup>28–30</sup>

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**Scheme 1.** General Pathways for the O<sub>2</sub> Interaction with Previously Described Tren-Based Cu(I) Complexes**Scheme 2.** General Chemical Behavior of Calixtren-Based Cu Complexes in the Absence of O<sub>2</sub>

Importantly, this labile site sits in the heart of the hydrophobic calixarene cone that mimics the active site chamber and the hydrophobic access channel of enzymes. The first member of a third generation of calixarene-based funnel complexes, Calixtren,<sup>31</sup> is a calix[6]arene that is capped by a tren<sup>32</sup> unit, thereby presenting a strong donating N<sub>4</sub> coordination site at the end of the rigidified concave calixarene core (Scheme 2). Its Cu(II) complexes have been first characterized.<sup>33,34</sup> Studies in solution (carried out by electronic and EPR spectroscopies) as well as in the solid state

(XRD) revealed a 5-coordinated metal ion in a distorted trigonal bipyramidal geometry due to its coordination to the N<sub>4</sub> site of the tren cap and to a guest ligand L. More recently, coordination of Cu(I) to Calixtren has been explored and its binding properties toward nonredox ligands studied in detail.<sup>35</sup> On the one hand, the electron-rich metal center was shown to display a relatively strong bonding to CO, due to important  $\pi$ -back-donation from the metal center ( $\nu_{\text{CO}} = 2075 \text{ cm}^{-1}$ ). On the other hand, the cuprous complex displayed only moderate affinity for nitrilo-ligands since, at room temperature (RT) in MeCN, 50% of the complex remained 4-coordinate. Interestingly, guest ligand exchange (L = RCN,

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Scheme 2) in this relatively rigid system proceeds through a two-step dissociative pathway, where Cu–N bond cleavage/formation is differentiated from the nitrilo guest expulsion/inclusion from/into the calixarene cavity. Indeed, variable-temperature NMR studies have highlighted the formation of an intermediate where the guest ligand RCN is trapped in the calixarene cavity without any coordination link to the cuprous center (Scheme 2).

The Cu(II)/Cu(I) electrochemical process was shown to be under the supramolecular control of the binding/unbinding of the guest coordinated to the metal embedded in the calixarene cone acting as a secondary coordination sphere.<sup>33</sup> Depending on the relative affinity of the guest ligand L for Cu(II) and/or Cu(I), three types of redox-driven guest-on ↔ guest-off processes could be managed. In the presence of a good ligand for both oxidation states such as MeCN, the electron exchange process was pseudoreversible due to a fast equilibrium between a 4- and a 5-coordinate Cu(I) species. When the guest was an oxygen-donor (in CH<sub>2</sub>Cl<sub>2</sub> with L = H<sub>2</sub>O, EtOH, or DMF) displaying a high affinity for Cu(II) and no affinity for Cu(I), an irreversible system led to the redox-driven ejection process of the guest at the Cu(I) level. Finally, a redox-driven ligand interchange process of DMF for MeCN at the Cu(I) state allowed the trapping of the thermodynamically less stable Cu(II)–MeCN adduct.

The CalixtrenCu complexes appeared to behave differently from all previously described<sup>20</sup> tren<sup>R</sup>Cu complexes (deprived of a protecting pocket) in a number of ways:

(i) The cuprous complex did not disproportionate in concentrated solutions under an inert atmosphere. This shows that there is no dinuclear contact and no electron transfer between two copper centers, well isolated from one complex to the other.

(ii) It was stable in chlorinated solvents such as dichloromethane or chloroform for days; that is, it did not react to produce a copper(II) chloride complex as other N<sub>4</sub>Cu(I) complexes that reacted within a few minutes.<sup>20,36</sup> Since we have demonstrated that the calixarene cone preserves one site accessible for organic ligands (RCN, RNH<sub>2</sub>), this is not due to a steric protection that prevents contact between the solvent molecule and the Cu(I) center. Rather, it may be ascribed to the isolation of the solvent molecule: the attack of the C–Cl bond by the Cu(I) center would lead to a high energy carboradical that cannot rapidly dimerize since trapped in the calixarene pocket. Such a protective behavior has been previously mentioned in our study relative to the supramolecular modeling of the radical reactive species of Galactose Oxidase.<sup>37</sup>

(iii) Finally, whereas the CalixtrenCu(II) complex behaved as a remarkable receptor for a variety of guests ligands such as nitriles, alcohols, and amides, it appeared to be reluctant to anion binding such as halides or hydroxide.<sup>33</sup> Such an unusual selectivity in favor of neutral guests has been attributed to the presence of the six O atoms belonging to the calixarene core, which point their lone pairs toward the free coordination site of the metal ion, thus making a repulsive crown of high electron density that disfavors anion binding.<sup>33</sup>

All of these observations led us to the conclusion that the calix-ligand confers different reactivity to the Cu(I) center as compared to the classical tren<sup>R</sup> system. Because the cavity of the Calixtren

prohibits dimerization of the copper centers and allows the full control of their fifth coordination site, exploration of the potentially reactive monocopper center included in the calixarene macrocycle appeared very appealing. This drove us to investigate the reaction of the CalixtrenCu(I) complex with O<sub>2</sub>.

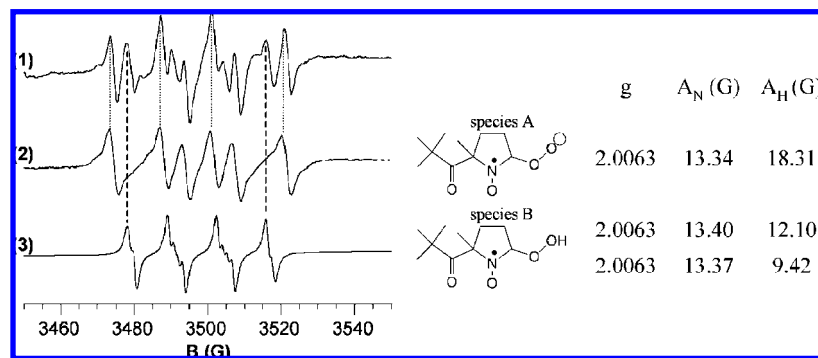
## Reaction with O<sub>2</sub>

Contrary to the Cu(I) complexes based on the first generation of calix[6]-ligands displaying a tris(imidazole)<sup>30</sup> or tris(pyridine)<sup>29</sup> core, the CalixtrenCu(I) complex appeared to be very reactive toward O<sub>2</sub>. As a first analysis, this can be ascribed to the donating strength of the tren cap. However, whereas tren<sup>R</sup>Cu(I) complexes were described as stable in air in the solid state, CalixtrenCu(I) reacts instantaneously in air or with O<sub>2</sub> as shown by the rapid color change to green in the solid state. When dioxygen was introduced into a solution of the [CalixtrenCu]<sup>+</sup> complex at low *T* in various solvents (CH<sub>2</sub>Cl<sub>2</sub> –90 °C, acetone –75 °C, MeCN –40 °C, THF –105 °C), the colorless solution turned blue green instantaneously. This shows that during the very fast reaction of the complex with O<sub>2</sub>, the Cu(I) center has been oxidized to Cu(II). No intermediate could be visually observed whatever the solvent, even at very low *T*. When the same experiments were carried out with the Cu(I)CO complex (obtained upon bubbling CO into the solution before adding O<sub>2</sub>), the reaction was considerably slowed. EPR and UV–vis analyses of the reaction products confirmed the formation of Cu(II) complexes upon the reaction of the Cu(I) compounds with O<sub>2</sub>. Indeed, in each case, an EPR signal was obtained, accounting for ca. 1 equiv of Cu(II) per initial cuprous complex [measured by double integration and compared to an authentic sample of CalixtrenCu(II)]. This was corroborated by the absorption of the oxidized solutions in the visible region. Interestingly, however, the analyses revealed different products as a function of the solvent in which the Cu(I) complex was reacted.

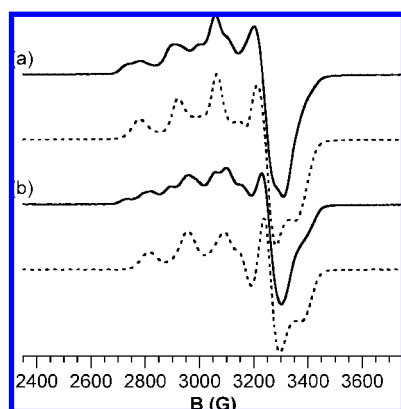
**Analyses of the Products Obtained in MeCN.** When the O<sub>2</sub> reaction was carried out in MeCN at low *T* (ca. 240 K), the EPR and UV–vis spectra of the resulting solution showed the quasi-quantitative formation of the well-known<sup>33</sup> dicationic [CalixtrenCu<sup>II</sup>–NCMe]<sup>2+</sup> complex as the sole species. ESI mass and NMR analyses of the ligand isolated after decomplexation (with KCN) indicated intact Calixtren. The reaction of the cuprous complex with dioxygen was followed at RT by EPR spectroscopy in the presence of the radical trap 5-*tert*-butyl-oxycarbonyl 5-methyl-1-pyrroline *N*-oxide (BMPO). Upon progressive diffusion of O<sub>2</sub> (via a syringe) into the EPR cell containing the Cu(I) complex, signals at around *g* = 2 appeared and grew (Figure 2). These signals correspond to the (BMPO–OOH)<sup>•</sup> and (BMPO–O<sub>2</sub>)<sup>•–</sup> adducts that are produced under such solvent conditions. Indeed, the signal corresponding to (BMPO–OOH)<sup>•</sup> (B) has been identified by comparison to the one reported in the literature<sup>39</sup> when H<sub>2</sub>O is the solvent, whereas the signal corresponding to (BMPO–O<sub>2</sub>)<sup>•–</sup> (A) has been identified by comparison to the signal recorded with a solution of BMPO in dry MeCN to which KO<sub>2</sub> and crown ether (18-C6) had been added. The same experiment carried out in the absence of the Cu(I) complex gave no EPR signal.

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- (38) Izzet, G.; Zeng, X.; Akdas, H.; Marrot, J.; Re naud, O. *Chem Commun.* **2007**, 810–812.

- (39) BMPO was used because it is a good radical trap for superoxide but does not react with carboradicals. The signal attribution to adducts A and B has been further substantiated by comparison to the signals recorded from a solution of BMPO in dry MeCN containing various bases or acids (such as trifluoroacetic acid, triethylamine, lutidine, . . .), to which KO<sub>2</sub> and crown ether (18-C6) have been added. See: Zhao, H.; Joseph, J.; Zhang, H.; Karoui, H.; Kalyanaraman, B. *Free Radical Biol. Med.* **2001**, *31*, 599–606.



**Figure 2.** EPR spectra (X band, 9.4 GHz, RT) of (1) a MeCN solution of  $[\text{CalixtrenCu}]^+$  ( $\sim 10^{-3}$  M) reacted with dioxygen in the presence of the radical trap BMPO ( $5 \times 10^{-2}$  M), (2) a solution of BMPO ( $5 \times 10^{-2}$  M),  $\text{KO}_2$  ( $2 \times 10^{-3}$  M), and 18-C-6 ( $2 \times 10^{-3}$  M) in MeCN (species A,  $(\text{BMPO}-\text{O}_2)^{\cdot-}$ ), and (3) radical  $(\text{BMPO}-\text{OOH})^{\cdot}$  (species B). EPR data for species A have been obtained by simulation of spectrum 2 with Xsophe. Species B is a mixture of two diastereoisomers (45/55). Spectrum 3 is the simulation spectrum of species B with EPR data given in ref 39.



**Figure 3.** Comparative EPR spectra (X band, 9.4 GHz, 100 K) of the reaction products of complex  $[\text{CalixtrenCu}]^+$  reacted with  $\text{O}_2$  in  $\text{CH}_2\text{Cl}_2$  (–) with the well-known  $[\text{CalixtrenCu-L}]^{2+}$  (---). (a) Crude reaction mixture ( $L = \text{H}_2\text{O}$ ); (b) after addition of MeCN to the EPR tube ( $L = \text{MeCN}$ ).

This experience shows that the low  $T$  reaction of  $[\text{CalixtrenCu}]^+$  with dioxygen in MeCN proceeds through the mono-electronic reduction of  $\text{O}_2$  by the Cu(I) center and leads to the clean formation of the dicationic  $[\text{CalixtrenCu}^{\text{II}}-\text{NCMe}]^{2+}$  complex with concomitant release of superoxide (Scheme 2, top pathway). When reacted at RT, the analytical data of the crude solution were qualitatively similar, except for the observation of a minor product detected by ESI mass spectrometry at  $M + 16$ , which will be identified below.

**Analyses of the Products Obtained in  $\text{CH}_2\text{Cl}_2$ .** When performed in a  $\text{CH}_2\text{Cl}_2$  solution, either at low  $T$  or RT, the reaction of the  $[\text{CalixtrenCu}]^+$  complex with  $\text{O}_2$  led to different analytical data. EPR spectra of the oxidized solution showed the presence of several Cu(II) species (Figure 3). One corresponds to the well-known  $[\text{CalixtrenCu}-\text{OH}_2]^{2+}$  complex,<sup>33</sup> whereas other signals attest to the formation of one or more new Cu(II) species. When MeCN was added into the same EPR tube (after the  $\text{O}_2$  reaction), the signature of  $[\text{CalixtrenCu}-\text{NCMe}]^{2+}$  resulting from the substitution of the aqua guest ligand for MeCN was associated even more clearly with other signals relevant to unknown new Cu(II) complexes. This suggests that, whereas the Calixtren-based Cu(I) complex was quantitatively oxidized into  $[\text{CalixtrenCu}-\text{NCMe}]^{2+}$  in the coordinating solvent MeCN, in the noncoordinating solvent  $\text{CH}_2\text{Cl}_2$ , a different reactive pathway produced new Cu(II) species. Knowing that Calixtren-based Cu(II) complexes are reluctant to bind anions such as hydroxide or chloride, we thus speculated that the new

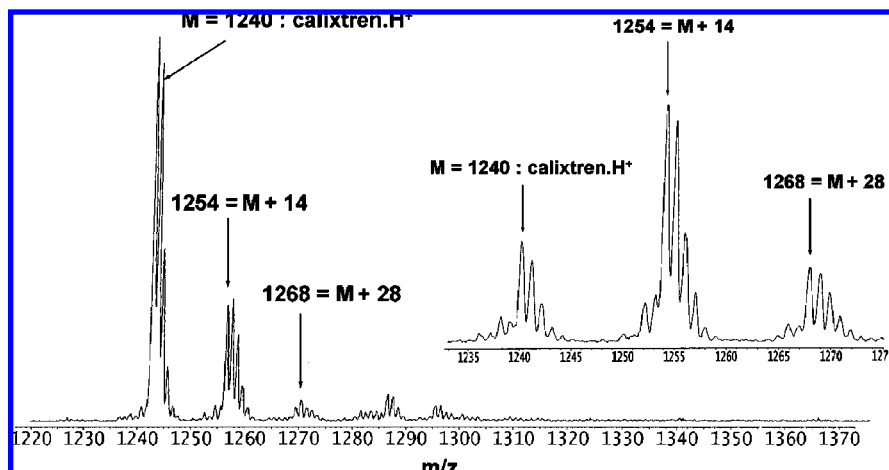
spectroscopic signatures are due to a chemical modification of the Calixtren ligand itself.

After demetalation of the products obtained by reaction at low  $T$  (addition of KCN), the solution was analyzed by  $^1\text{H}$  NMR spectroscopy. The corresponding spectrum clearly showed a structurally modified calix-ligand, which, however, could not readily be identified.<sup>40</sup> ESI mass analyses of the low  $T$  reacted solution treated with KCN showed the presence of new groups of peaks, all presenting a molecular weight higher than that of the ligand [ $M = \text{Calixtren} + \text{H}^+$  ( $m/z = 1240$ )]. The most intense peak at  $M + 14$  corresponds to the incorporation of one oxygen atom and removal of two H atoms. The product corresponding to  $M - 2$  is not observed, indicating the absence of an imino-derivative of Calixtren. The spectrum also showed the presence of a minor product at  $M + 28$ . The formulation of these compounds as  $[\text{Calixtren} + (\text{O}-2\text{H})_n]^+$  attests to a formal  $n \times 4$  electron Calixtren oxidation induced by the reaction of  $\text{O}_2$  with its Cu(I) complex. When the oxygenation reaction of the Cu(I) complex was carried out at RT, the peaks corresponding to the same  $m/z$  were observed, with, however, different ratios. Indeed, peaks at  $M + 14$  and  $M + 28$  were more intense than that of the starting material ( $M$ ). This shows that the oxidative process leading to the oxidation of Calixtren is facilitated at a higher temperature (Figure 4, inset).

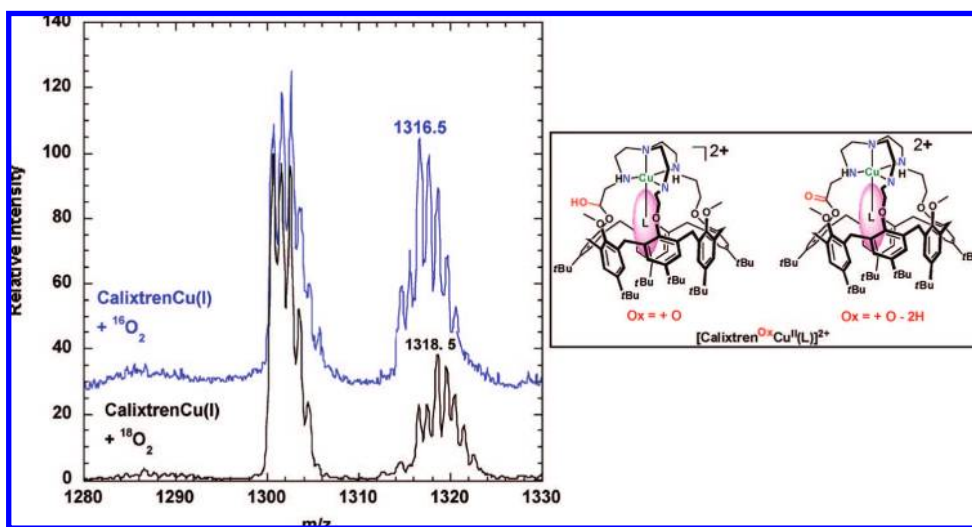
When carried out in a  $\text{CH}_2\text{Cl}_2/\text{acetone}$  (1:1 v/v) mixture, the oxygenation of  $[\text{CalixtrenCu}]^+$  at 200 K indicated a slightly different ESI mass profile. Indeed, whereas the ( $M + 14$ ) product was still detectable, a peak corresponding to just one O atom insertion ( $M + 16$ ) became the most intense.

When directly analyzed after short reaction times in  $\text{CH}_2\text{Cl}_2$ , the ESI mass analyses of the crude metallated products obtained before decomplexation revealed a new complex peak at  $m/z$  1316 in addition to the one corresponding to the cupric complex  $[\text{Calixtren}(-\text{H})\text{Cu}]^+$  at  $m/z$  1300.5.<sup>41</sup> This new feature corresponds to the overlapping/combination of two peaks attributed to  $[\text{Calixtren}(-\text{H})\text{Cu} + \text{O}]^+$  ( $m/z = 1316.5$ ) and  $[\text{Calixtren}(-\text{H})\text{Cu} + \text{O} - 2\text{H}]^+$  ( $m/z = 1314.5$ ), in agreement with theoretical isotopic patterns. The same solution analyzed in MeCN showed four sets of peaks: two of them correspond to  $[\text{CalixtrenCu}]^{2+}$  and  $[\text{CalixtrenCu}-\text{NCMe}]^{2+}$  [at  $m/z = 650.9$  and  $m/z = 671.4$  ( $z = 2$ ), respectively], the other two attributed to the corresponding mono-oxygenated products [ $m/z = 658.9$

(40) The same procedure carried out with the reaction products resulting from the reaction in MeCN showed unmodified Calixtren ligand (a nice sharp  $^1\text{H}$  spectrum attesting to a  $C_{3v}$  symmetrical calixarene core). See Figure S1.



**Figure 4.** ESI mass spectra of the reaction mixture  $\{[\text{CalixtrenCu}]^+ + \text{O}_2$  in dichloromethane,  $T = 200 \text{ K}$ ) after treatment with KCN. Inset: ESI profile of the same reaction carried out at RT.



**Figure 5.** Left: Comparative mass spectra of the crude reaction mixture after reaction of the CalixtrenCu(I) complex with  $^{16}\text{O}_2$  (top) and  $^{18}\text{O}_2$  (bottom) in dichloromethane.<sup>41</sup> Right: Proposed structures for the oxygenated products corresponding to  $(M + \text{O})$  and  $(M + \text{O} - 2\text{H})$ .<sup>42</sup>

and 679.3]. This shows that not only Cu(II) remains coordinated to the oxidized ligand but it also retains its ability to bind a guest ligand. Together with the above-reported EPR analyses, these observations suggest that the coordination core is only little affected by the oxygenation of Calixtren under these experimental conditions.

$^{18}\text{O}_2$  labeling confirmed the exclusive incorporation of an O atom into the ligand from dioxygen, not from  $\text{H}_2\text{O}$ . The resulting ESI-MS spectrum of  $[\text{CalixtrenCu}]^+$  in the presence of  $^{18}\text{O}_2$  displayed a complex fragment as above but shifted by 2 emu as compared to the one obtained with  $^{16}\text{O}_2$ . Again, on the basis of theoretical isotopic pattern comparisons, the peak was attributed to the mixture of  $[\text{Calixtren}(-\text{H})\text{Cu} + ^{18}\text{O}]^+$  ( $m/z = 1318.5$ ) and  $[\text{Calixtren}(-\text{H})\text{Cu} + ^{18}\text{O} - 2\text{H}]^+$  ( $m/z = 1316.5$ ). Furthermore, a careful comparison of the MS-MS spectra of the  $[\text{Calixtren}(-\text{H})\text{Cu} + ^{16}\text{O}]^+$  and  $[\text{Calixtren}(-\text{H})\text{Cu} + ^{18}\text{O}]^+$  fragments showed a common peak at 1298.6 (see Figure S2), which corresponds to the loss of a water molecule in each case. This establishes that the oxygen atom in the monooxygenated compound is not linked to either an aromatic or a bridging methylene unit of the calixarene skeleton because, in such positions, a water loss would be impossible. Such a loss of water was not observed for the fragmentation of the peaks corre-

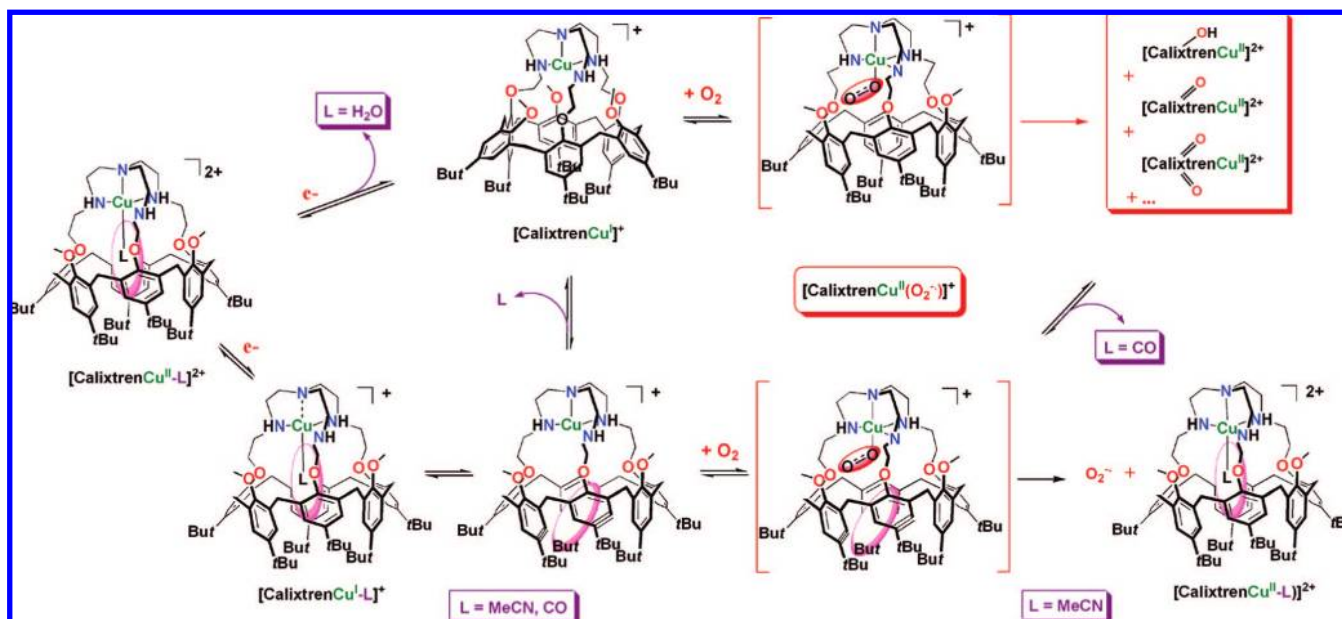
sponding to  $[\text{Calixtren}(-\text{H})\text{Cu} + ^{16}\text{O} - 2\text{H}]^+$  and  $[\text{Calixtren}(-\text{H})\text{Cu} + ^{18}\text{O} - 2\text{H}]^+$ , which is clearly consistent with the formation of a keto moiety. The presence of the three fragments showed that the oxidation reaction was only partial.

Finally, FTIR analyses of the products isolated after KCN decomplexation (which corresponds to the major formation of the  $\{M + (\text{O} - 2\text{H})_n\}$  products), showed a relatively strong absorption at  $1735 \text{ cm}^{-1}$  together with a weak one at  $1262 \text{ cm}^{-1}$  that did not exist for the starting Calixtren ligand (Figure S3). These new absorptions are indicative of the formation of an ester function. Therefore, the oxygenation site is proposed to be the methylene groups belonging to the aza cap, situated in  $\beta$ -position relative to the coordinating N atom, and in  $\alpha$ -position of the calixarene phenoxyl group (see Figure 5, right). Such a position is consistent with the fact that the  $N_4$  environment provided by the oxidized tren cap still allows Cu-coordination (see above).<sup>42</sup> All of these observations converge to the conclusion that the reaction of CalixtrenCu(I) with  $\text{O}_2$  in  $\text{CH}_2\text{Cl}_2$  leads to the insertion of an O atom into a C-H bond and is followed by the dehydrogenation of the resulting hydroxylated moiety (an hemiacetal) into an ester function.

At this point, it is worth noting that the addition of  $\text{KO}_2$  (with 18-C6) to the Cu(I) complex did not oxidize either the copper center



**Scheme 3.** Reaction Pathways of Complex  $[\text{CalixtrenCu}^{\text{I}}(\text{L})]^+$  with Dioxygen with the Proposed Transient Formation of a Reactive Cu(II) Superoxo Adduct<sup>a</sup>



<sup>a</sup> Top pathway: In pure CH<sub>2</sub>Cl<sub>2</sub> or PhCN. Bottom pathway: In pure MeCN or in CH<sub>2</sub>Cl<sub>2</sub> or PhCN solutions containing MeCN or CO.

or the ligand. When the Cu(II) complex was reacted with excess KO<sub>2</sub> or 1–5 equiv of H<sub>2</sub>O<sub>2</sub>, the ligand remained untouched. Only after reaction with a large excess of H<sub>2</sub>O<sub>2</sub> (ca. 100 equiv) was some degradation of the ligand observed, with lower MW compounds assignable to dehydrogenated products, but no peaks at +14 or +16. This indicates that Cu(I) is not appreciably reactive toward superoxide and that its reaction with O<sub>2</sub> leads to different reactive species than does the reaction of Cu(II) with H<sub>2</sub>O<sub>2</sub>, thus excluding an outer sphere process for the oxygenation of the calixtren ligand. Also importantly, the addition of a coordinating guest to the solution such as MeCN protected the ligand since, according to ESI analyses, the ratio of oxidized ligand versus intact ligand decreased. This was not the case with CO, which slowed the Cu/O<sub>2</sub> reaction but did not protect the ligand toward oxidative degradation. Finally, when carried out at RT in pure CH<sub>2</sub>Cl<sub>2</sub> in the presence of the radical spin trap BMPO, almost no EPR signal at *g* = 2 could be detected, thus showing that release of superoxide is, at the best, a minor pathway for the reaction of [CalixtrenCu]<sup>+</sup> with O<sub>2</sub> in a noncoordinating solvent. This also suggests that the active species is not a free radical diffusing into the solution, but rather a metal-centered active species.

All of these results drive us to propose that the reaction of CalixtrenCu(I) with O<sub>2</sub> proceeds through the transient formation of a [CuO<sub>2</sub>]<sup>+</sup> adduct, the evolution of which depends on the reaction medium: release of superoxide into the reaction medium favored by the presence of a guest ligand well suited for Cu(II) binding (MeCN, not CO) versus intramolecular redox evolution of the [CuO<sub>2</sub>]<sup>+</sup> adduct toward *O* insertion into the tren cap (Scheme 3).

### Electrochemical Study under O<sub>2</sub>

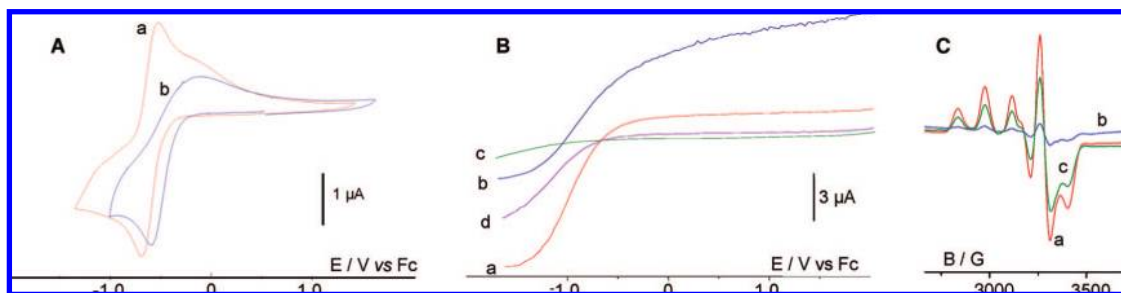
The interaction between [CalixtrenCu<sup>I</sup>(L)]<sup>+</sup> and O<sub>2</sub> has been scrutinized by electrochemistry in different solvents, mainly MeCN and CH<sub>2</sub>Cl<sub>2</sub>. Cyclic voltammetry (CV) studies, conducted with a solution of the Cu(II) complex, allowed one to follow the transient formation of the Cu(I) complex at the electrode and its fate in the absence or presence of O<sub>2</sub>. Alternatively, the evolution of a solution of the Cu(I) complex, prepared in the bulk by quantitative

electrolysis, was monitored after addition of O<sub>2</sub> by CV or rotating disk electrode voltammetry (RDEV). All of these experiments were conducted with extremely dry solvents (H<sub>2</sub>O < 1–3 ppm in the dry box) at a relatively low concentration of complex (ca. 1 mM) and O<sub>2</sub> (when introduced in solution).

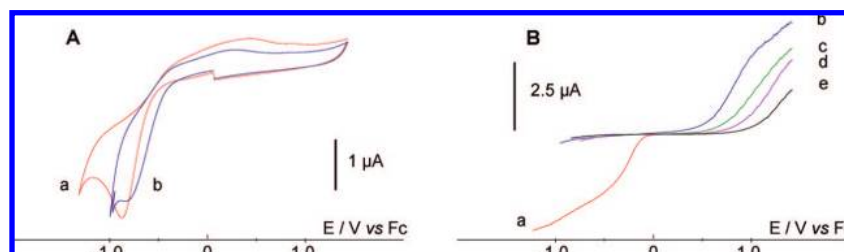
In pure MeCN under N<sub>2</sub>, the [CalixtrenCu<sup>II</sup>–NCMe]<sup>2+</sup>/[CalixtrenCu<sup>I</sup>(MeCN)]<sup>+</sup> system is reversible at any scan rate. In the presence of O<sub>2</sub>, the behavior is scan rate (*v*) dependent. At *v* > 0.2 V s<sup>-1</sup>, the CVs remained unchanged. At lower *v*, the CVs became irreversible under O<sub>2</sub> with a broadening and disappearance of the reoxidation peak as shown in Figure 6. Successive scans showed a passivation of the Pt electrode. The reaction of the Cu(I) complex in the bulk was followed by RDEV. After an exhaustive electrolysis of [CalixtrenCu<sup>II</sup>–NCMe]<sup>2+</sup> at a reduction potential *E* = –0.85 V (*n* ≅ 1 F/mol), the Cu(I) complex was quantitatively obtained as shown by the appearance of the corresponding reduction wave by RDEV voltammetry. The position of the wave, exactly the same as that of the Cu(II) derivative, indicated that [CalixtrenCu<sup>I</sup>(MeCN)]<sup>+</sup> was the only species detected in solution. After interaction with O<sub>2</sub> (at RT), the complex in its initial form [CalixtrenCu<sup>II</sup>–NCMe]<sup>2+</sup> was restored up to a ca. 70% ratio<sup>43</sup> as indicated by the RDEV wave (curve d, Figure 6A). The

- (41) When analyzed in dichloromethane, the CalixtrenCu(II) complexes always give a peak at 1300.5 corresponding to the monocation [Calixtren(H)Cu]<sup>+</sup>. The <sup>16</sup>O<sub>2</sub>-oxygenated product (+16) is then characterized by a peak at 1616.5, formally [Calixtren(H)Cu]<sup>+</sup> + O.
- (42) Oxygenation in α-position of the nitrogen atom would lead to an amido function (for the M + 14 product), which would strongly change the coordination environment of the Cu center. Most consistent is the formation of an ester moiety in β-position, which indeed should be quite sensitive to acid-base treatments.
- (43) The cuprous complex with an empty cavity, [CalixtrenCu(∅)]<sup>+</sup>, whose existence in a 1:1 ratio in pure MeCN at RT has been established by <sup>1</sup>H NMR spectroscopy (see ref 35) is not detected under these experimental conditions, due to fast binding/unbinding of MeCN relative to the CV scan rate (see ref 33). However, the fact that only 70% of the Cu(II) complex is recovered in this experiment carried out at RT, as compared to the ca. 100% yield in the above-described experiment run at low *T*, is assignable to the enthalpically favored presence of MeCN in the calixarene pocket.





**Figure 6.** Solutions of [CalixtrenCuII-L]<sup>2+</sup> complex (ca. 1 mM) in MeCN (L = MeCN), NBu<sub>4</sub>-PF<sub>6</sub>, 0.2 M. (A) Cyclic voltammetry at a Pt electrode,  $\nu = 0.02 \text{ V s}^{-1}$ , (a) under N<sub>2</sub>, (b) under 1 atm O<sub>2</sub>. (B) Rotating disk electrode voltammetry at a Pt electrode,  $\nu = 0.01 \text{ V s}^{-1}$ , (a) before and (b) after electrolysis under N<sub>2</sub>, (c) after O<sub>2</sub> bubbling, and (d) after polishing of the electrode. (C) EPR spectra of MeCN solutions of [CalixtrenCuII-NCMe]<sup>2+</sup>: (a) before electrolysis, (b) after reduction electrolysis, and (c) after O<sub>2</sub> bubbling.



**Figure 7.** Solution of the [CalixtrenCuII-OH<sub>2</sub>]<sup>2+</sup> complex (ca. 1 mM) in CH<sub>2</sub>Cl<sub>2</sub>, NBu<sub>4</sub>-PF<sub>6</sub>, 0.2 M, at a Pt electrode. (A) Cyclic voltammetry at  $\nu = 0.02 \text{ V s}^{-1}$ , (a) under N<sub>2</sub>, (b) under 1 atm O<sub>2</sub>. (B) Rotating disk electrode voltammetry,  $\nu = 0.01 \text{ V s}^{-1}$ ; (a) before and (b) after electrolysis under N<sub>2</sub>, (c, d, e) after interaction with O<sub>2</sub>: 2, 5, 30–60 min, respectively.

passivation of the electrode during the interaction of the Cu(I) complex with O<sub>2</sub> was also detected here. If the Pt electrode remained immersed in the solution during this process, no current was detected (curve c, Figure 6A) unless the electrode was repolished or removed from the solution during the reaction (curve d, Figure 6A). Under these conditions, the reaction with O<sub>2</sub> was almost instantaneous. It was estimated roughly to ca. 5 s from the CV. The evolution of the complex was also followed by EPR (Figure 6B). The intense signal characteristic of [CalixtrenCuII-NCMe]<sup>2+</sup> almost completely disappeared after electrolysis of the solution, attesting to the reduction of the copper center. After bubbling O<sub>2</sub> into the solution, the initial signal was recovered with an intensity corresponding to >70% of the starting Cu(II) solution, corroborating the reoxidation to the initial form. These observations are in complete agreement with those made above as they also clearly show that the interaction of the cuprous complex with O<sub>2</sub> leads principally to the reoxidation to the initial [CalixtrenCuII-NCMe]<sup>2+</sup> with production of a reduced species of O<sub>2</sub>, which is responsible for the passivation of the Pt electrode.<sup>44</sup> The loss of ca. 30% of the complex after reoxidation by O<sub>2</sub> may be ascribed to the formation of the damaged complex [Calixtren<sup>Ox</sup>CuII]<sup>2+</sup>, not detected at the electrode.

In CH<sub>2</sub>Cl<sub>2</sub>, the Cu(II) center is coordinated to a water molecule, while the Cu(I) displays an empty cavity. Accordingly, CV and RDEV associate two peaks or two waves in an irreversible process, a reduction process at  $E_{pc}$  or  $E_{1/2} \approx -0.9 \text{ V}$  for [CalixtrenCuII-OH<sub>2</sub>]<sup>2+</sup> and a reoxidation peak much more positive at  $E_{pa}$  or  $E_{1/2} \approx 0.03 \text{ V}$  for the empty cavity complex, [CalixtrenCu<sup>I</sup>(∅)]<sup>+</sup> (Figure 7A). The two species interconvert

electrochemically, indicating a fully chemically reversible process. The effect of the presence of O<sub>2</sub> in solution on the CV was examined. Under these experimental conditions (extremely dry solution, relatively low concentration in complex, ca. 1 mM, solution not saturated in O<sub>2</sub>), no evidence for the transient formation of a [Cu-O<sub>2</sub>]<sup>+</sup> adduct could be obtained as no noticeable difference of behavior was detected at a scan rate as low as 0.02 V s<sup>-1</sup> (Figure 7A). However, when the Cu(I) solution obtained by electrolytic reduction was subjected to O<sub>2</sub>, the RDEV oxidation wave observed for the empty cavity cuprous complex progressively vanished. This process was completed after ca. 30 min. (Figure 7B, curve b to c) and was associated with a total disappearance of the electrochemical characteristics of the CalixtrenCu complexes. Only a broad oxidation wave at a higher potential was observable with no reduction feature either by CV or by RDEV, which suggests that, under these experimental conditions, the CalixtrenCu complexes have been destroyed. In this case, no problem of passivation of the electrode was observed, suggesting that no free radical was released into the solution.<sup>44</sup>

The reactivity toward O<sub>2</sub> was also examined in dry benzonitrile, which does not coordinate the cuprous center in the Calixtren system.<sup>45</sup> Accordingly, the electrochemical behavior in this solvent was the same as that in CH<sub>2</sub>Cl<sub>2</sub>, with, under O<sub>2</sub>, a slow disappearance of the CalixtrenCu waves and appearance of a broad process at higher potential. Interestingly, when a few equivalents of MeCN was added to the electrolyzed Cu(I) solution before O<sub>2</sub> bubbling, reappearance of [CalixtrenCuII-NCMe]<sup>2+</sup> in a small ratio was observed, once the oxygenation completed. These results thus substantiate the fact that interaction between O<sub>2</sub> and the empty cavity complex

(44) Passivation of the Pt electrode is also observed when H<sub>2</sub>O<sub>2</sub> is added to the MeCN solution of the complex. Thus, it might be related to the disproportionation of O<sub>2</sub><sup>-</sup> that is released at the electrode during the reaction with O<sub>2</sub>. See: (a) Andrieux, C. P.; Hapiot, P.; Savéant, J.-M. *J. Am. Chem. Soc.* **1987**, *109*, 3768–3775, and refs cited therein.

(45) It has been shown for the closely related tris(pyridine) system that the relative affinity of the Cu(I) center embedded in a hexa(‘Bu)-calix[6]arene cavity for PhCN versus MeCN is 1/500 due to its weaker binding ability and its larger size (see ref 29b).

[CalixtrenCu<sup>I</sup>(∅)]<sup>+</sup> leads to species that are different from the initial [CalixtrenCu<sup>II</sup>-L]<sup>2+</sup> complex due to ligand oxidation. They also show that interception of the putative [CuO<sub>2</sub>]<sup>+</sup> intermediate by MeCN is faster than the intramolecular redox process leading to ligand oxygenation, thus illustrating the protecting effect of the nitrilo guest with regard to the preservation of the Calixtren ligand integrity during the Cu(I)-O<sub>2</sub> interaction process. Finally, when a trace of water was added to the electrolyzed PhCN Cu(I) solution, the reaction with O<sub>2</sub> proceeded almost instantaneously. This indicates that water is not innocent in the reaction, although its role has not been clarified yet.

## Discussion

The cuprous complex [CalixtrenCu]<sup>+</sup> displays a very high reactivity toward dioxygen, as predicted by its high affinity for CO. From a thermodynamic point of view, this must be related to the strong electron-donating tren ligand, which, as a first coordination sphere, makes the cuprous center a strongly reducing agent. This is best illustrated by its redox potential, which is fairly negative ( $E^0 = -0.68$  V vs Fc<sup>+</sup>/Fc in MeCN;  $E_{pc} = -0.94$  V/ $E_{pa} = 0.03$  V in CH<sub>2</sub>Cl<sub>2</sub>).<sup>33</sup> Kinetically, however, such a high reactivity must be related to the presence of a labile site, which, although included in the calixarene cavity, remains accessible for exogenous ligand binding, such as to O<sub>2</sub>.

The fact that the Cu(I) complex readily reacts with O<sub>2</sub> in the solid state strongly supports a reactive behavior that is based on the O<sub>2</sub> interaction at a single Cu(I) center. In solution as well, the approach of a second copper center at a distance close enough to allow the formation of a Cu<sub>2</sub>O<sub>2</sub> dinuclear adduct is precluded by the calixarene core. The complex displayed a high reactivity toward O<sub>2</sub> in the absence {[CalixtrenCu<sup>I</sup>(∅)]<sup>+</sup>} as well as in the presence of a MeCN guest ligand inside the cavity {[Cu<sup>I</sup>(calixtren)(MeCN)]<sup>+</sup>}, the reaction being rapidly and irreversibly driven, even at very low *T*, to the final oxidized compound(s), identified as calixarene-based Cu(II) complexes. However, the reaction products were different, which attests to different evolution pathways. This is most consistent with an inner sphere process because outer sphere electron exchanges should lead to the same reaction products whatever the nature of the guest. As well, the redox potential of the O<sub>2</sub>/O<sub>2</sub><sup>-</sup> reversible process in MeCN,  $E^0 = -1.13$  V, as compared to that (vide supra) of the [CalixtrenCu<sup>II</sup>-NCMe]<sup>2+</sup>/[Calixtren-Cu<sup>I</sup>(NCMe)]<sup>+</sup> process, which is higher in CH<sub>2</sub>Cl<sub>2</sub> as indicated by the  $E_{pa}$  irreversible peak, is not compatible with an outer sphere electron transfer. Hence, the key step of the mechanism is proposed to be the formation of a common [CalixtrenCu(O<sub>2</sub>)]<sup>+</sup> adduct, presumed to be a superoxide-Cu(II) species,<sup>21,22</sup> the evolution of which depends on the reaction medium (Scheme 3).

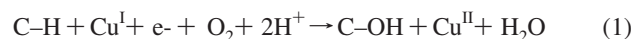
This adduct, however, could not be accumulated or transiently detected. This is surprising in view of the thermodynamic data previously reported for O<sub>2</sub> binding to a tren derivative, which predicted a relatively good thermodynamic stability for the closely related [tren<sup>Me2</sup>Cu-O<sub>2</sub>]<sup>+</sup> complex at RT.<sup>46</sup> Such a contrasting behavior may be explained by the repelling effect for anions of the calixarene core, which is not only π-basic but also provides a crown of six oxygen atoms surrounding the inner binding site.<sup>33,38</sup> Such an electron-rich second coordination

sphere must destabilize/activate the Cu(II)-bound superoxide moiety directly resulting from the mono-electronic reduction of O<sub>2</sub> by the trenCu(I) center. Indeed, EPR and electrochemical studies in MeCN have unambiguously established the formation of free superoxide as a first product of evolution of the [CalixtrenCu<sup>II</sup>(O<sub>2</sub><sup>-</sup>)]<sup>+</sup> adduct, concurrently with [Calixtren-Cu<sup>II</sup>-NCMe]<sup>2+</sup>. In such a solvent well suited for Cu(II) coordination, electrostatic repulsion between the O<sub>2</sub><sup>-</sup> anionic ligand and the *O*-rich calixarene small rim and formation of the stable Cu(II)-MeCN adduct may be proposed to be the driving forces for the fast ejection of the superoxide anion from the calixarene core, with no oxidative damage of the ligand.

In contrast, in CH<sub>2</sub>Cl<sub>2</sub> (or PhCN) in the absence of any molecule susceptible to displace O<sub>2</sub><sup>-</sup> other than H<sub>2</sub>O at a low concentration, the putative [CalixtrenCu<sup>II</sup>(O<sub>2</sub><sup>-</sup>)]<sup>+</sup> intermediate undergoes the intramolecular redox process leading to the insertion of an oxygen atom into the ligand. Indeed, isotopic labeling experiments have confirmed that the inserted *O* atom has been provided by O<sub>2</sub>, not residual water. Although we have not been able to definitely identify the precise site of attack,<sup>42</sup> the ESI mass analyses and spectroscopic data are most consistent with the oxygenation of the OCH<sub>2</sub> moieties linking the tren cap to the calixarene core. Three products have been identified, one corresponding to the insertion of one O atom (*M* + 16), another denoting a subsequent dehydrogenation (*M* + 14), and a third likely stemming from a second oxygenation-dehydrogenation sequence (*M* + 28). Very interestingly, the detection of products corresponding to the formal 4e<sup>-</sup> oxidation of the ligand (*M* + 14) and 8e<sup>-</sup> oxidation of the ligand (*M* + 28) indicates that the oxidative process can run catalytically.

**Reactive Pathways.** As discussed in the Introduction, the oxidizing species that have been proposed to be directly responsible for C-H bond breaking process are either superoxides, peroxides, or even oxides.<sup>6,8,9</sup> In our system, as the Cu centers are isolated from each other, formation of peroxides or oxides would require abstraction of a hydrogen atom or electron(s) from the reaction medium, most probably the solvent, before the attack of the ligand itself. This seems unlikely from both a thermodynamic and a kinetic point of view: the OCH<sub>2</sub> moiety of the ligand is by far the more oxidizable and the more accessible group if compared to a solvent such as CH<sub>2</sub>Cl<sub>2</sub>. Furthermore, as above stated, replacing the couple Cu(I)/O<sub>2</sub> by Cu(II)/H<sub>2</sub>O<sub>2</sub> led to either no reactivity at low concentration or dehydrogenation products at high concentration. Therefore, we propose the reactive species directly responsible for the C-H bond breaking to be the superoxide intermediate [Cu<sup>II</sup>O<sub>2</sub>]<sup>+</sup>.

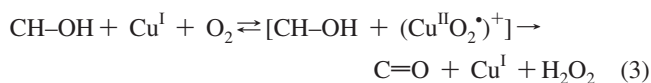
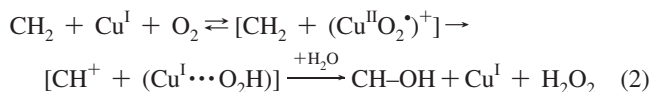
For Cu mono-oxygenases, the hydroxylation process is associated with the formation of water as a coproduct and thus requires an additional electron according to eq 1:



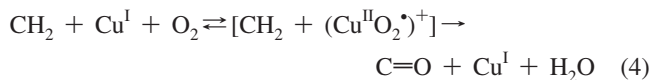
A second electron input allows the regeneration the active Cu(I) species. So, the enzyme sacrifices one-half of the oxidizing power of O<sub>2</sub> by injecting two electrons in the system. With our complex, the mechanism is obviously different. We do not have the controlled e<sup>-</sup> inputs as in the enzyme, and we do not know if the oxidation process is associated with the release of H<sub>2</sub>O or H<sub>2</sub>O<sub>2</sub> or to the co-oxidation of one component of the reaction medium.

The formal equations for the formation of the oxygenated products may correspond to the sequential 2e<sup>-</sup> oxidations of the substrate according to eqs 2 and 3 (possibly via radical intermediates):

(46) The thermodynamic parameters for Cu(I)/O<sub>2</sub> binding (see Scheme 1) with the hexa-*N*-methylated tren ligand are the following (see Scheme 1 and ref 20d): *T*, 183 K;  $K_{\text{CuO}_2}$ ,  $1.35 \times 10^9$  M<sup>-1</sup>;  $K_{\text{Cu}_2\text{O}_2}$ ,  $1.4 \times 10^8$  M<sup>-1</sup>. *T*, 298 K;  $K_{\text{CuO}_2}$ ,  $1.55 \times 10^1$  M<sup>-1</sup>;  $K_{\text{Cu}_2\text{O}_2}$ ,  $2.5 \times 10^3$  M<sup>-1</sup>.

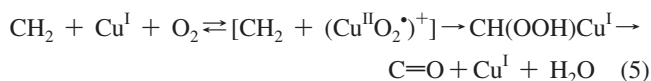


and/or to the direct 4e<sup>-</sup> oxidation pathway according to eq 4:



Either pathway would explain the [M + 28] product as Cu remains cuprous after the first cycle. However, as we know that the isolated product is Cu(II), we must admit that somehow there is a one electron redox process that stops the reaction at the Cu(II) state. Yet, we do not know either what is the sacrificial species (solvent, H<sub>2</sub>O<sub>2</sub>?).

Another possibility is the formation of an alkylperoxide intermediate that could either directly yield the keto product or be intercepted by the solvent, according to eqs 5 and 6:



The latter proposal is very appealing as it would explain the solvent effect observed by mass analyses (M + 14 in CH<sub>2</sub>Cl<sub>2</sub>, M + 16 in acetone as major products).

## Conclusion

We have herein described a system based on a calix[6]-cryptand that isolates a Cu center but simultaneously maintains a free coordination site accessible to small exogenous ligands such as a nitrile, H<sub>2</sub>O, CO, and O<sub>2</sub>. Hence, this system constitutes a unique tool for exploring the intrinsic reactivity of a monocopper center toward O<sub>2</sub>. Importantly, the Calixtren core not only provides an electron-rich environment for the metal ion, but also a second coordination sphere (the oxygen crown of the small rim and a π-basic hydrophobic cavity) that acts as a funnel for small neutral guest but as a repellent for anions.

The resulting strongly reducing Cu(I) center is very reactive toward O<sub>2</sub>. The reaction, identified as an inner-sphere process, is proposed to transiently lead to a [CuO<sub>2</sub>]<sup>+</sup> adduct. According to all previously reported studies on N<sub>4</sub>Cu/O<sub>2</sub>, its nature can be assigned as a Cu(II)-superoxide entity resulting from a one electron exchange between Cu(I) and O<sub>2</sub>. Such an adduct, in our system, is not stable and could not be observed, even at low *T*. This may well be due to the anionic character of the transiently formed superoxide entity and its repulsive interaction with the second coordination sphere. Indeed, in the presence of a guest ligand well suited for stabilizing the Cu(II) state, O<sub>2</sub><sup>-</sup> is readily ejected from the complex into the reaction medium. However, most interestingly, in the absence of such a guest ligand, the transiently coordinated superoxide leads to an intramolecular attack of the Calixtren ligand leading to the insertion of oxygen atoms into a C–H bond of the tren cap. This result is of the most importance as it unambiguously

demonstrates that an isolated cuprous center reacts with O<sub>2</sub> to produce a species that is capable per se of oxygenating an organic moiety, without the need of an external electron input. Such a result highly substantiates the hypothesis according to which, in copper monooxygenases, the [CuO<sub>2</sub>]<sup>+</sup> adduct attacks first the C–H bond of the substrate, before the electron input from the second Cu center (see Figure 1, bottom, lower pathway). Also, very interestingly and quite unexpectedly, the insertion of two O atoms on the same ligand associated with the loss of four hydrogen atoms indicates that the process can run catalytically, either through a multiple two-electron process per O<sub>2</sub> [then associated with the release of H<sub>2</sub>O<sub>2</sub> and regeneration of Cu(I)] or through a multiple four-electron process per O<sub>2</sub> [then associated with the release of H<sub>2</sub>O and regeneration of Cu(I)]. More work is needed to clarify which pathway is followed, and we are planning to explore the reactivity of this system toward external substrates.

## Experimental Section

The CalixtrenCu(I) and Cu(II) complexes were prepared as previously described.<sup>33,35</sup> The CalixtrenCu(I) was too sensitive to dioxygen to be stored for more than a day, even in the solid state and in a dry box, so each time it was synthesized and isolated just before carrying out the experiments with O<sub>2</sub>.

EPR spectra were recorded on a spectrometer operating at X-band frequency (9.44 GHz) equipped with an shq001 cavity fitted with a liquid nitrogen cryostat for low temperature experiments or with a multicapillary sample cell for RT studies. For liquid nitrogen temperature experiments, the following instrument settings were used: field modulation amplitude frequency of 100 kHz, field modulation amplitude of 0.6 mT, time constant of 0.04 s, sampling time 20 ms, 2048 sampling points, field sweep 0.15 T, microwave power of 10 mW at 80 K. At RT, 100 spectra were successively recorded over 2115 s using the following experiment setting: field modulation amplitude frequency of 100 kHz, field modulation amplitude of 0.2 mT, time constant of 0.04 s, sampling time 20 ms, 512 sampling points, field sweep 10 mT, microwave power of 10 mW.

ESI-MS spectra have been obtained using a LCQ trap ion spectrometer.

The electrochemical studies of the copper complexes were performed in a glovebox (Jacomex) (O<sub>2</sub> < 1 ppm, H<sub>2</sub>O < 1 ppm) with a home-designed three-electrodes cell (WE, Pt; RE, Ag; CE, Cu). The potential of the cell was controlled by an AUTOLAB PGSTAT 100 (Ecochemie) potentiostat monitored by a computer. All solvents used (CH<sub>2</sub>Cl<sub>2</sub>, MeCN, PhCN) were distilled and kept under N<sub>2</sub>. The supporting salt, NBu<sub>4</sub>PF<sub>6</sub>, was purified and dried under vacuum for 48 h at 100 °C, then kept under N<sub>2</sub> in the glovebox. All experiments were performed with solutions of ca. 10<sup>-3</sup> M in electroactive species. All potential values are reported in the text versus the ferrocene/ferrocenium couple.

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**Supporting Information Available:** Complementary FTIR, ESI mass, and <sup>1</sup>H NMR spectra of the oxygenated products. This material is available free of charge via the Internet at <http://pubs.acs.org>.

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