Inorganica Chimica Acta, *181* (1991) 141–143

Inorganica Chimica Acta
LETTER

Colored species formation between mononuclear copper(II) complex and superoxide anion

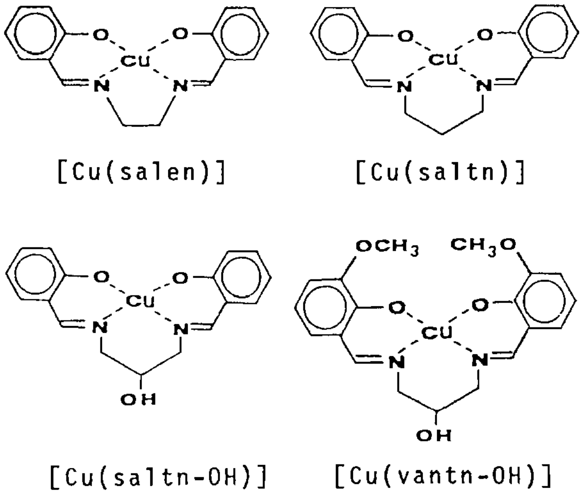
Yuzo Nishida*, Kei Unoura, Izumi Watanabe, Tomohisa Yokomizo
Department of Chemistry, Faculty of Science, Yamagata University, Yamagata 990 (Japan)

and **Yoshikiyo Kato**
Department of Chemistry, Faculty of General Education, Yamagata University, Yamagata 990 (Japan)

(Received November 26, 1990)

Superoxide dismutases, which catalyze the dismutation of the superoxide anion (O_2^-) to molecular oxygen and hydrogen peroxide [1], are found in all free-living organisms except for some oxygen-sensitive obligate anaerobes. O_2^- is the one-electron reduction product of O_2, and is generated as a by-product of oxidative metabolism. Fridovich found that a family of superoxide dismutases having Cu and Zn, or Fe, or Mn at the active site are important for protection against the toxic effect of this free radical [1]. The structure of bovine Cu–Zn superoxide dismutase (SOD) has been determined to 2 Å resolution [2]; the active site Cu(II) and Zn(II) lie 6.3 Å apart and the Cu ligand forms a tetrahedrally distorted square plane. At present, several copper(II) complexes are known to dismutate O_2^- [3–5], however, there is no copper(II) complex which can trap O_2^- at ambient temperature. In this article we wish to report the first example of a colored species formation between a mononuclear copper(II) complex and superoxide anion.

The copper(II) complexes cited in this article are illustrated in Scheme 1.



Scheme 1.

In Fig. 1 the cyclic voltammograms (CV) of [Cu(salen)] (square planar complex) and [Cu(saltn-OH] (distorted tetrahedral complex) [6] are shown. In argon (Ar) atmosphere, [Cu(salen)] and [Cu(saltn-

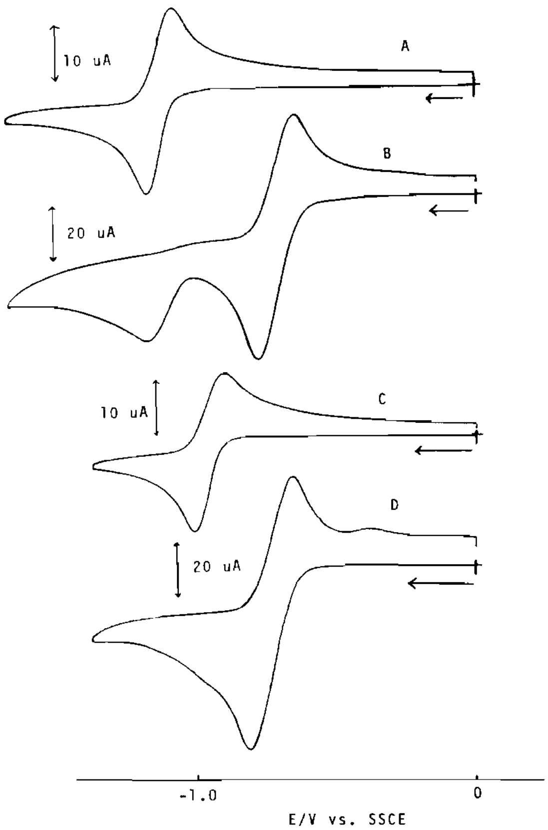


Fig. 1. CV of copper(II) complexes (in dmso, 0.1 M [(n-butyl)$_4$N]BF$_4$ as a supporting electrolyte; 0.002 M copper(II) complex; glassy carbon electrode; scan speed 100 mV/s; 25 °C). A: [Cu(salen)] under Ar, B: [Cu(salen)] under O_2, C: [Cu(saltn-OH)] under Ar, D: [Cu(saltn-OH)] under O_2.

---

*Author to whom correspondence should be addressed.

0020-1693/91/$3.50

OH)] exhibit a quasi-reversible wave at −1.14 and −0.97 V (versus SSCE, saturated sodium chloride calomel electrode) in dimethyl sulphoxide (dmso), respectively (cf. traces A and C in Fig. 1). These should correspond to the Cu(II)/Cu(I) redox couple [7]. In the presence of O_2 (saturated in dmso; the concentration of O_2 is 2.1 mM [8] and the O_2 molecule undergoes quasi-reversible reduction at −0.71 V in dmso), some changes were observed in the CV. In the case of [Cu(salen)], the reoxidation peak (Cu(I)→Cu(II)) disappeared as seen in Fig. 1, implying that the copper(I) species formed may react with O_2 or O_2^-, although the reduction peak (Cu(II)→Cu(I)) was always observed (50–1000 mV/s scan rate) in the presence of the O_2 molecule. On the other hand, the reduction peak of [Cu(saltn-OH)] disappeared in the presence of O_2, as shown in trace D of Fig. 1. This suggests that the complex [Cu(saltn-OH)] is reacting with the O_2^- ion, which is formed electrochemically at −0.71 V.

In order to confirm the above suggestion, we have investigated the reaction between the copper(II) complex and superoxide anion produced by the electrochemical reduction of O_2 molecule in dmso, the concentration of O_2^- being determined from cyclic voltammetry based on the results reported by Sawyer and Roberts, and Stein *et al.* [8]. When a dmso solution of O_2^- (2.8 mM) was added to a dmso solution of [Cu(salen)] (2 mM), no change was observed for the color and d–d bands in the absorption spectra. On the other hand, the green solution (cf. trace A in Fig. 2) of [Cu(saltn-OH)] (1∼2 mM) turned to brown immediately on the addition of O_2^- solution (2.6∼5.2 mM), but this brown solution is unstable at room temperature, decomposing within a few minutes. We followed this color change using spectroscopic and stopped-flow techniques at 25 °C. The results indicated that the addition of O_2^- caused the instantaneous increase of absorbance around 650 nm and in the range 500–550 nm as shown in Fig. 2, and these absorption bands disappeared rapidly (cf. Fig. 3), yielding a yellowish-green solution whose d–d bands are essentially similar to those of the original green solution (cf. Fig. 2). These facts demonstrate that O_2^- is reacting with [Cu(saltn-OH)], which is consistent with the result obtained by CV. The fact that all the distorted tetrahedral complexes, [Cu(saltn)], [Cu(saltn-OH)] and [Cu(vantn-OH)], exhibit high catalytic activity for the decomposition of O_2^- (SOD-like function)*, whereas such a function is not observed for the square planar complex, [Cu(salen)], is consistent with the above conclusion. The formation of a brown species was not detected in the reaction mixture of [Cu(saltn)] and O_2^-, and [Cu(vantn-OH)] and O_2^-, the structural and electrochemical properties of these two compounds being very similar to those of [Cu(saltn-OH)]. For the cases of the [Cu(saltn)] and [Cu(vantn-OH)] complexes, a decrease of d–d band intensities (*c.* 650 nm) was observed on the addition of O_2^- anion in the initial stage, and a similar decrease of the d–d band was also observed when the complex [Cu(saltn-OH)] was reduced by the electrochemical method. These suggest that the brown species observed for [Cu(saltn-OH)] and O_2^- should not be due to the formation of copper(I) species, but to an adduct formation between them.

As shown in the previous paper [10], in distorted tetrahedral copper(II) complexes one unpaired electron lies mainly in the $d_{x^2-y^2}$ orbital, and some lobes of this d orbital are not screened by the ligand

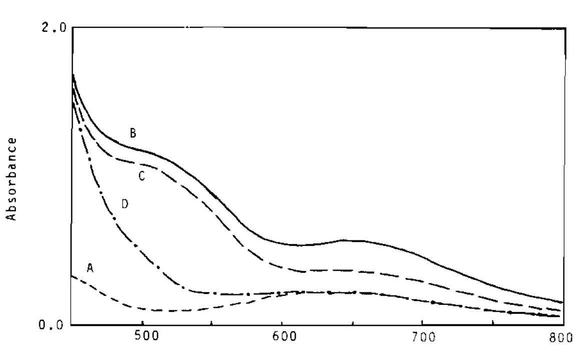


Fig. 2. Absorption spectra of reaction mixture (in dmso; equal quantities of O_2^- (2.6 mM) and [Cu(saltn-OH)] (2 mM) were mixed). A: [Cu(saltn-OH)] (1 mM), B: 9 s after mixing, C: 25 s after mixing, D: 153 s after mixing.

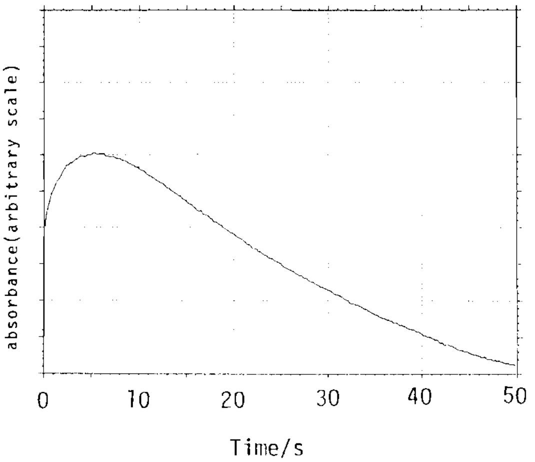
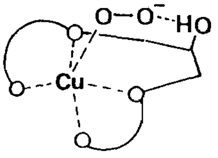


Fig. 3. Time course of absorbance at 650 nm in mixed solution of [Cu(saltn-OH)] (2 mM) and superoxide anion (2.6 mM) in dmso at 25 °C.

---

*The content of superoxide anion in the reaction mixture (in dmso) was determined by the NBT method [9].

atoms. The present result clearly indicates that the presence of such a d orbital is necessary for the interaction of these copper(II) compounds with superoxide anion. Based on this discussion, it seems reasonable to assume that the structure of the brown species is as illustrated below; O_2^- coordinates to the copper atom through the interaction with the unscreened d orbital, and the OH group in the ligand system may stabilize the $Cu–O_2^-$ adduct, perhaps through the formation of a hydrogen bond (see below) or a partial hydrogen atom transfer.



Acknowledgements

We are thankful for support from the Inamori Foundation and a Grant-in-aid for Scientific Research No. 02403012 from the Ministry of Education, Science and Culture.

References

1 I. Fridovich, *Adv. Inorg. Biochem., 1* (1979) 67.
2 J. A. Tainer, E. Getzoff, K. M. Beem, J. S. Richardson and D. C. Richardson, *J. Mol. Biol., 160* (1982) 181.
3 L. R. Alvare, K. Goda and T. Kimura, *Biochim. Biophys. Res. Commun., 69* (1976) 687.
4 R. F. Pasternack, A. Banth, J. M. Pasternack and C. S. J. Johnson, *J. Inorg. Biochem., 15* (1981) 261.
5 E. Kimura, T. Koike and M. Kodama, *Inorg. Chem., 25* (1986) 2242.
6 N. Kitajima, K. Whang, Y. Moro-oka, A. Uchida and Y. Sasada, *J. Chem. Soc., Chem. Commun.*, (1986) 1504.
7 G. S. Patterson and R. H. Holm, *Bioinorg. Chem., 4* (1975) 257.
8 D. T. Sawyer and J. L. Roberts, Jr., *J. Electroanal. Chem., 12* (1966) 90; J. Stein, J. P. Fackler, Jr., G. J. McClune, J. A. Fee and L. T. Chan, *Inorg. Chem., 18* (1979) 3511.
9 C. Beauchamps and I. Fridovich, *Anal. Biochem., 44* (1971) 276.
10 Y. Nishida, M. Takeuchi, N. Oishi and S. Kida, *Inorg. Chim. Acta, 96* (1985) 81; Y. Nishida, *Z. Naturforsch., Teil C, 42* (1987) 1358.