Chem. Pharm. Bull. **39**(7) 1894—1896 (1991) Vol. 39, No. 7

COOXIDATION OF ENONES DURING REACTION OF SUPEROXIDE WITH ACIDIC AROMATIC AMINES

Hiromasa TAKIZAWA, Tetsuo NAGANO and Masaaki HIROBE*
Faculty of Pharmaceutical Sciences, University of Tokyo, Hongo, Bunkyo-ku, Tokyo 113, Japan

Enones can be epoxidized by $O_2^{-\bullet}$ in the presence of various compounds having an acidic >NH group. Cyclic voltammetry and ESR study show that $O_2^{-\bullet}$ was disproportionated into H_2O_2 and O_2 by these compounds. Furthermore, H_2O_2 was deprotonated by unreacted $O_2^{-\bullet}$ and HOO^- was formed, which is widely known to be the effective oxidant in the epoxidation of enones. This confirms the strong basicity of $O_2^{-\bullet}$ in aprotic media, and we propose an alternative scheme for the Haber–Weiss reaction in the absence of metal ions.

KEYWORDS superoxide; Haber-Weiss reaction; hydroxyl radical; hydrogen peroxide; hydroperoxy anion; hydroperoxy radical; enone; oxidation

It has become clear that superoxide ($O_2^{-\bullet}$), a one-electron reduction product of dioxygen (O_2), plays important roles in various biological processes.[1] In spite of its involvement as a key intermediate, the chemical reactivity of $O_2^{-\bullet}$ itself is reported to be rather innocuous.[2] We have reported that $O_2^{-\bullet}$ can oxidize olefins and substrates having labile hydrogens in the presence of acyl halides,[3] polyhalides,[4] or CO_2.[5] In these reactions, $O_2^{-\bullet}$ reacts with halides to form peroxy intermediates which are more highly reactive than $O_2^{-\bullet}$ alone.[6] Furthermore, the phosphate moiety of nucleotides enhances the reactivity of $O_2^{-\bullet}$ in the nucleic base release reaction.[7]

A more intensive study of the $O_2^{-\bullet}$–adjunct agents system led us to the finding that $O_2^{-\bullet}$ can oxidize enones to the corresponding epoxides in the presence of nucleic bases and other compounds having an acidic >NH group (Table I).

These compounds were found to disproportionate $O_2^{-\bullet}$ into H_2O_2 and O_2 in DMSO. In this system, $O_2^{-\bullet}$ was shown to act as a base to form hydroperoxy anion (HOO^-) and hydroperoxy radical ($HOO\bullet$) through deprotonation of H_2O_2 (Chart 1). The reaction of $O_2^{-\bullet}$ with H_2O_2 is well known to produce the hydroxyl radical ($HO\bullet$) in the presence of Fe ions. This is called the Fe-catalyzed Haber-Weiss reaction (Chart 2).[8] In fact, adding Fe^{2+} to our system (KO_2-adjunct agent) produced no epoxide and the characteristic ESR signal of the PBN-CH_3 adduct was observed, ensuring $HO\bullet$ formation.[9] However, in the absence of a metal ion, the epoxidation of enones proceeded.

We used chalcone and its derivative (4'-methoxychalcone) as substrates. The reactivity of these

Table I. Epoxidation of Chalcone by KO_2–Adjunct Agent System

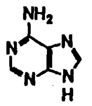
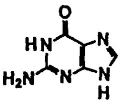
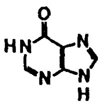
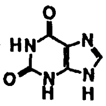
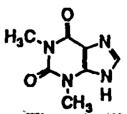
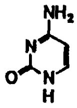
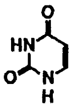
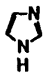
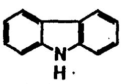
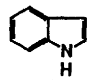
Adjunct agent	 Adenine	 Guanine	 Hypoxanthine	 Xanthine	 Theophylline
Yield% (Recovery%)	85 (2)	31 (59)	34 (44)	28 (59)	77 (0)
Adjunct agent	 Cytosine	 Uracil	 Imidazole	 Carbazole	 Indole
Yield% (Recovery%)	53 (38)	80 (0)	61 (38)	54 (11)	21 (54)

Chart 1

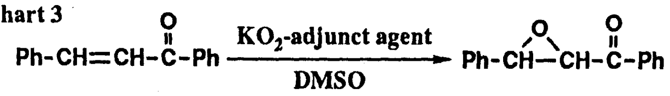
$$H_2O_2 + O_2^{-} \longrightarrow HOO\bullet + HOO^{-}$$

substrates was quite similar. Enones are decomposed by $O_2^{-\bullet}$ via base-catalyzed autoxidation, as reported by Frimer et al.[10] However, in the presence of the compounds shown in Table I, epoxidation of chalcone proceeded in fairly high yield (Chart 3).

Chart 2

$$H_2O_2 + O_2^{-\bullet} \xrightarrow[\text{Haber-Weiss reaction}]{Fe^{n+}} HO\bullet + HO^- + O_2$$

Chart 3



$$\text{Ph-CH=CH-}\overset{O}{\overset{\|}{C}}\text{-Ph} \xrightarrow[\text{DMSO}]{KO_2\text{-adjunct agent}} \text{Ph-CH}\overset{O}{-}\text{CH-}\overset{O}{\overset{\|}{C}}\text{-Ph}$$

Additive effects of adenine and adenosine on cyclic voltammogram of O_2 show that $O_2^{-\bullet}$ reacts with adenine, but not with adenosine (Fig. 1). $O_2^{-\bullet}$ was detected by ESR even after several hours without conversion into H_2O_2. But in the KO_2–adenine system, $O_2^{-\bullet}$ was not detected soon after the start of the reaction, and a large amount of H_2O_2 was formed (Fig. 2). This indicates that adenine can disproportionate $O_2^{-\bullet}$ into H_2O_2 and O_2 and that the acidic proton (of N-9 position) played an important role as a proton source.

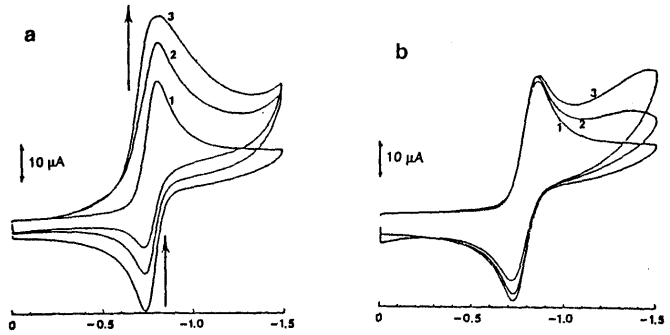


Fig. 1 Effect of Nucleic Bases on Cyclic Voltammogram of O_2.

Cyclic voltammograms of O_2 (curve 1), O_2 plus 0.5 mM adenine (a) or adenosine (b) (curve 2), and O_2 plus 1.0 mM adenine or adenosine (curve 3) in DMSO, containing 0.1 M tetraethylammonium perchlorate as a supporting electrolyte. Measurements were done using the platinum electrode at a scan rate of 0.05V/s.

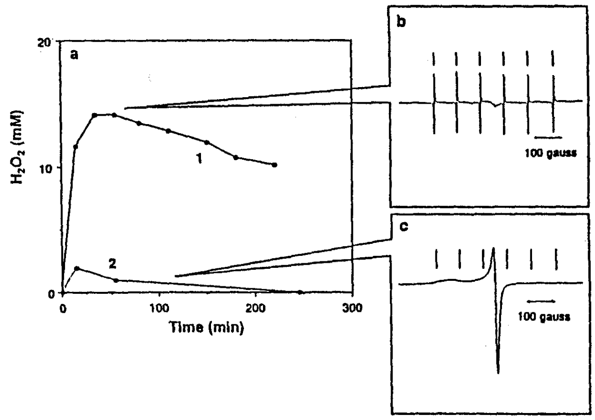


Fig. 2 Amounts of $O_2^{-\bullet}$ and H_2O_2 during the Reaction of KO_2 and Adenine or Adenosine

(a): line 1, amounts of H_2O_2 during the reaction of KO_2 and adenine; line 2, amounts of H_2O_2 during the reaction of KO_2 and adenosine. (b): ESR spectra of $O_2^{-\bullet}$ during the reaction of KO_2 and adenine. (c); ESR spectra of $O_2^{-\bullet}$ during the reaction of KO_2 and adenosine. H_2O_2 was measured by the method of Allen et al.[16] ESR spectra were recorded at 77K under the conditions as follows: microwave power 10 mW, modulation frequency 100 kHz with an amplitude of 6.3G, gain 4x100 (b), 4x1 (c), and response time 0.3s. The six arrows indicate the signal of Mn^{2+}.

We have also examined the correlation between the epoxidation yields and the pKa values in DMSO of these compounds.[11] In contrast to uracil, adenine, imidazole, carbazole and indole (pKa=14.1, 14.2, 18.6, 19.9, 20.95, respectively), pyrrole, 2-pyrrolidone and 2-piperidone (23.0, 24.2, 26.4, respectively) did not react with $O_2^{-\bullet}$, thus no H_2O_2 was formed, and no epoxidation of chalcone proceeded. Adenine, after deprotonation, was most probably converted into stable potassium salt, as Frimer et al. have reported,[12] since the consumption of adenine and the appearance of its oxidized products do not appear in HPLC. This shows that $O_2^{-\bullet}$ can abstract a proton from weakly acidic compounds whose pKa values are as high as *ca* 22.

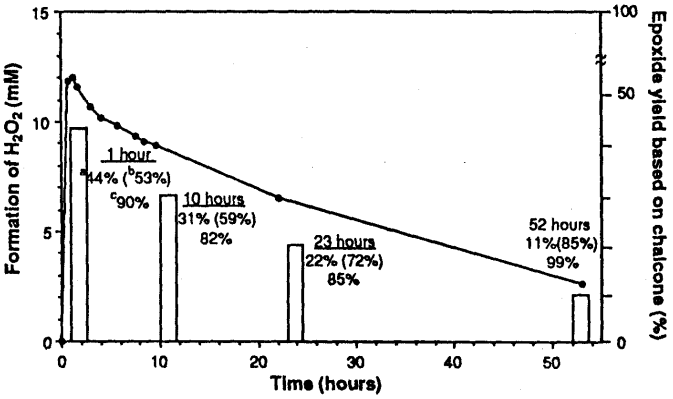


Fig. 3 Time Course of H_2O_2 Formation and H_2O_2 Concentration-Dependent Epoxidation of Chalcone.

Curve with solid circles shows H_2O_2 concentration in DMSO 90 ml containing adenine 4.2 mmol and KO_2 4.2 mmol (solution A). Open column shows the yield of chalcone epoxidation, when chalcone 0.35 mmol, 18-crown-6-ether 0.35 mmol and a small portion of KO_2 were dissolved in 15 ml of the solution A. [a]Yield of epoxide based on chalcone. [b]Chalcone recovery. [c]Yield of epoxide based on H_2O_2.

The H_2O_2 formed was then deprotonated by unreacted $O_2^{-\bullet}$ which acted as a strong base, and HOO^- was produced (Chart 1). HOO^- reacts with enones to form the corresponding epoxides.[13)]

Figure 3 shows the time course of the formation of H_2O_2 in the KO_2-adenine system. The DMSO solution containing H_2O_2 produced from KO_2 and adenine with no $O_2^{-\bullet}$ remaining did not oxidize chalcone to the epoxide. However, the epoxidation did proceed upon addition of a small portion of KO_2 into the solution at 1 h, 10 h, 23 h, and 52 h after the first mixing of KO_2 and adenine. The yields based on H_2O_2 concentration were as high as 80~90% at each stage. The epoxide yield based on chalcone completely depended on the H_2O_2 concentration (Fig. 3). These data clearly show that H_2O_2 formed from the disproportionation of $O_2^{-\bullet}$ was deprotonated by $O_2^{-\bullet}$ to produce HOO^-, which was the ultimate reactant in the epoxidation of chalcone.

The Haber-Weiss reaction in the presence of Fe ion has been unanimously regarded as the most important one in the research concerning $O_2^{-\bullet}$. A number of reports have concluded that $HO\bullet$ produced through this reaction must be the ultimate toxin of reactive oxygen species.[14)] The rate of the reaction is thought to be too slow in the absence of metal ions,[15)] but no extensive investigation has been made in this respect. Our investigation carried out in the absence of metal ions in organic solvent shows the formation of HOO^- and $HOO\bullet$, during which $O_2^{-\bullet}$ acted as a base. This is the alternative scheme for the Haber-Weiss reaction in the absence of metal ion, and proves meaningful in the field of $O_2^{-\bullet}$ study.

ACKNOWLEDGEMENT The authors are grateful for a research grant from the Ministry of Education , Science and Culture of Japan.

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(Received May 13, 1991)