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Co-Oxidations of 1,3-Diphenylisobenzofuran by the Haber-Weiss Reaction. Is Singlet Oxygen Concerned in This Oxidation?

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Summary; The reaction of $O_2^{-\cdot}$ with H_2O_2 in the presence of 1,3-diphenylisobenzofuran was studied. o-Dibenzoylbenzene was obtained in 82 % yield, which decreased to 52 % when dimethoxyethane was presence. Additions of β-carotene or 1,4-diazabicyclo-[2,2,2]-octane also inhibited the production of o-dibenzoylbenezene. These results show that singlet oxygen may be a considerable species generated by the Haber-Weiss reaction.

Much current research is concerned with the generation of $^1O_2$ in some biological oxidations. Especially, peroxidation of lipids has been of considerable interest in the relation of $O_2^{-\cdot}$ and $^1O_2$. Pederson and Aust (1) reported that $O_2^{-\cdot}$ gave rise in turn to $^1O_2$ which was immediate cause of lipid peroxidation. Recently, Kellogg and Fridovich (2) proporsed that $^1O_2$ might be generated by the Haber-Weiss reaction ($\underline{1}$), which has been proporsed as a source of OH radical (3),

$$O_2^{-\cdot} + H_2O_2 \longrightarrow {}^1O_2 + \cdot OH + OH^- \qquad (\underline{1})$$

In relation to biological investigations, there have been many searches for $^1O_2$ using KO_2 as $O_2^{-\cdot}$ sources in DMSO-H_2O system (4), but no reports regarding the confirmation of $^1O_2$ generation

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Abbreviations; DPBF, 1,3-diphenylisobenzofuran. DBB, o-dibenzoylbezene. DABCO, 1,4-diazabicyclo-[2,2,2]-octane. DME, dimthoxyethane. DMSO, dimethylsulfoxide.

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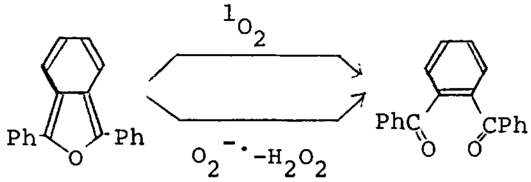
from $O_2^{-\cdot}$ in DMSO-H_2O_2 system. In order to certify this possibility, co-oxidation of DPBF, which reacts smoothly with $^1O_2$ (5), and its inhibition by β-carotene and DABCO were sutdied. The



concentrations of $O_2^{-\cdot}$ and H_2O_2 used in this study were higher than those in respiring cells.

Materials and Methods

DPBF (6) was prepared by the methods in the literature, and was confirmed not to have any carbonyl compounds by ir spectrum. Potassium superoxide was obtained from Ventron Alfa Products, 18-crown-6-ether from Nisso Research Chemicals, β-carotene and DABCO from Wako Pure Chemical Industries. These were used without further purification. Commercial available H_2O_2 from Wako was used after the determination of its concentration by iodometric titration. DMSO from WAKO was used after vacuum distillation from CaH_2 under nitrogen. DME from Tokyo Kasei was used after dried over sodium.

To a 20 ml of DPBF solution (3 mM) in 1:1 mixed solvent of DMSO and DME, 50 mg of powdered KO_2 was suspended. Five ml of 30 % H_2O_2 was added to a vigorous stirred solution in an open reactor in dark. KO_2 was completely disappeared within 5 min. The mixture was extracted with CH_2Cl_2. The extract was washed with NaCl saturated solution and then dried over $MgSO_4$. The yield of DBB was determined by using high pressure liquid chromatography.

All procedures were carefully completed as soon as possible since DPBF was a sensitive compound to various oxidants.

Results

Co-oxidations of DPBF

Oxidation of DPBF was carried out in $O_2^{-\cdot}$-H_2O_2 system to give a DBB in 52 % yield. Meanwhile, oxidations of 1,2-dimethylcyclohexene and 9,10-diphenylanthracene under the same reaction conditions did not give the corresponding oxidized products in more than significant amounts.

Control experiments indicated that DPBF was less reactive to the respective species itself such as molecular oxygen, KO_2, and H_2O_2 under the reaction conditions used in this study (run 4, 5, and 6 in table).

Table Co-oxidations of DPBF

Run	Reaction System[a)]	DBB %[b)]
1	KO_2 + H_2O_2	52
2	KO_2 + H_2O	25
3	KOH + H_2O_2	17
4	KO_2 + crown ether	trace
5	H_2O_2	trace
6	O_2 + KOH + crown ether	trace

a) The amounts of the reagents used were the following; run 1, KO_2 50 mg and H_2O_2(30 %) 5 ml; run 2, KO_2 50 mg and H_2O 5 ml; run 3, KOH 17 mg and H_2O_2(30 %) 5 ml; run 4, KO_2 50 mg and 18-crown-6-ether 90 mg; run 5, H_2O_2(30 %) 5 ml; run 6, KOH 17 mg, 18-crown-6-ether 90 mg, and O_2 was saturated in the solvent.
b) The yield was calculated based on DPBF used.

Oxidations of DPBF in $O_2^{-\cdot}$-H_2O and OH^--H_2O_2 systems were carried out, because the dismutation of $O_2^{-\cdot}$ (2) and the base catalyzed disproportionation of H_2O_2 (3), which were reported as

$$2\ O_2^{-\cdot} \longrightarrow {}^1O_2 + O_2^{2-} \qquad (\underline{2})$$

$$2\ H_2O_2 \xrightarrow{OH^-} {}^1O_2 + 2\ H_2O \qquad (\underline{3})$$

the sources of ${}^1O_2$ (4), were possible to occur competitively with the Haber-Weiss reaction in the $O_2^{-\cdot}$-H_2O_2 system (run 2 and 3 in table). Efficiency of the systems for the oxidations of DPBF was following order; $O_2^{-\cdot}$-H_2O_2 > $O_2^{-\cdot}$-H_2O > OH^--H_2O_2.

<u>Inhibitions of co-oxidation of DPBF by β-carotene and DABCO</u>

Formation of DBB examined in the presence of β-carotene and DABCO, which have been extensively used as quenchers of ${}^1O_2$ (7). When either of them was present in the $O_2^{-\cdot}$-H_2O_2 system, the yield of DBB clearly decreased with the amounts of the quenchers as shown in fig. On the contrary, no inhibitions were observed in the $O_2^{-\cdot}$-H_2O and OH^--H_2O_2 systems.

<u>Solvent effects for the co-oxidation of DPBF in $O_2^{-\cdot}$-H_2O_2 system</u>

If the Haber-Weiss reaction is occurred in the $O_2^{-\cdot}$-H_2O_2 system, OH radical may contribute to the oxidation of DPBF (8).

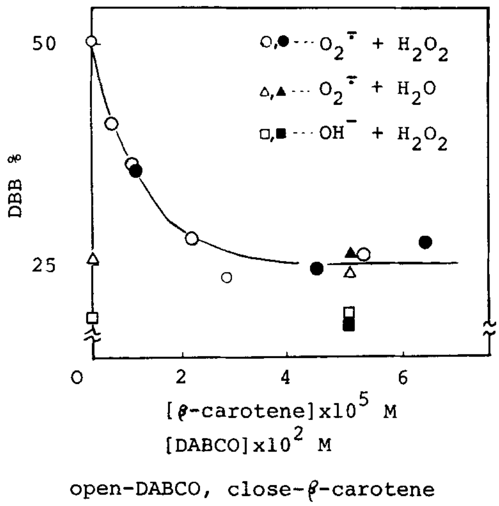


Fig. 1 Effects of β-carotene and DABCO on the formation of DBB.

DMSO and DME routinely used as the solvents are known as the scavengers of OH radical (9). The yield of DBB in DMSO solvent was 82 %, meanwhile that in DME solvent was 50 % and was equivalent to it in a 1:1 mixed solvent system. Apparently, DME functioned more effective than DMSO as an inhibitor of the oxidation of DPBF under the reaction conditions.

Discussion

Generally the possibility that $^1O_2$ may be involved in some oxidation processes has been indicated by the following studies; those based on measuring chemiluminescence due to decay of $^1O_2$, and chemical trapping and quenching of $^1O_2$. The conclusions described following are attributed to the studies of trapping by DPBF and quenching by β-carotene and DABCO.

Rate constant of the Haber-Weiss reaction (10) recently determined is very low, for example 2.25 $M^{-1}sec^{-1}$ (11), in the dilute concentrations of both $O_2^{-\cdot}$ and H_2O_2. Despite of the presence of such results, it was obvious that both $O_2^{-\cdot}$ and H_2O_2

were essential to the effective oxidation of DPBF, and that $O_2^{-\bullet}$ operated together with H_2O_2 to generate some active species. This discrepancy may be explained by the following two factors; 1) the concentrations of $O_2^{-\bullet}$ and H_2O_2 were exceedingly high, 2) the greater part of the reaction solvent consisted of polar organic compounds.

Partial inhibition of oxidation of DPBF by efficient quenchers such as β-carotene and DABCO was found only in the $O_2^{-\bullet}$-H_2O_2 system. In addition, the amounts of DBB produced were qualitatively in accord with the respective rate constants for trapping by DPBF (k_r=2.5x10^9 $M^{-1}sec^{-1}$ (5) and for quenching by β-carotene (k_q=3x10^{10} $M^{-1}sec^{-1}$ (7) and by DABCO (k_q=1.6x10^7 M^{-1} sec^{-1} (7). Moreover, the ratio of quenching rate constants of β-carotene to DABCO obtained, approximately 10^3, was exactly equal to that in photo-sensitized oxidations, 2x10^3. The results suggested that $^1O_2$ or some species mimicking $^1O_2$ may prompt a part of the oxidation of DPBF.

No inhibition by quenchers was found in $O_2^{-\bullet}$-H_2O and OH^--H_2O_2 systems, although the reactions of <u>2</u> and <u>3</u> were known as the sources of $^1O_2$ (4). Perhaps the amounts of $^1O_2$ generated were so small as to be involved within the experimental error. In $O_2^{-\bullet}$-H_2O system, the quenching of $^1O_2$ by $O_2^{-\bullet}$ may be occurring (12). If such a quenching was occurred in $O_2^{-\bullet}$-H_2O_2 the system, yield of DBB might be changed by the following factors; 1) the amounts of KO_2, 2) the concentration of H_2O_2, 3) the concentration of DPBF.

In conclusion, the amounts of DBB inhibited by β-carotene or DABCO are at least induced by the reaction of DPBF with $^1O_2$ or some species mimicking $^1O_2$ which appears to be produced by the Haber-Weiss reaction. This reaction could sufficiently compete with the dismutation of $O_2^{-\cdot}$ in DMSO-H_2O_2 system, and may become important in more hydrophobic media.

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