CHEMISTRY LETTERS, pp. 411-414, 1993.

High Catalytic Activity of trans-$[FeCl_2(cyclam)]^+$ for Peroxidation of Linolenic Acid

Yuzo NISHIDA,* Noriko TANAKA, and Shigeyuki TAKAHASHI
Department of Chemistry, Faculty of Science, Yamagata University,
Yamagata 990

The reaction mixture of trans-$[FeCl_2(cyclam)]^+$ and linolenic acid exhibited high ability for degradation of tetraphenylcyclopentadienone, one of the famous singlet oxygen ($^1\Delta_g$) quenchers, and the peroxidation of linolenic acid was found to proceed in this solution.

Lipoxygenase (linoleate:oxygen oxidoreductase, EC 1.13.11.12) is a non-heme iron dioxygenase which catalyzes the dioxygenation of polyunsaturated fatty acids containing a 1Z,4Z-pentadiene system.[1)] Interest in the mechanism of lipoxygenase has been stimulated these last years by the discovery of the implication of lipoxygenase in the biosynthesis of leukotrienes which have an important role in immediate hypersensitivity and inflammation.[2)] Native lipoxygenase-1 from soybean is colorless (Fe^{2+}) and virtually ESR-silent, and the active form of the enzyme contains iron in the ferric form. The two current hypothesis for the mechanism of lipoxygenase catalysis were reported. Abstraction of hydrogen at C-11 was proposed to be the rate-determining step, which was supported by a primary kinetic deuterium isotope effect.[3)] Hydrogen atom abstraction with reduction of the iron would result in formation of a pentadienyl radical as an intermediate. This mechanism was proposed to account for the fact that active enzyme when treated with substrate linolenic acid in the absence of oxygen lost its characteristic ESR signal at g=6.[4)] Recently an alternative mechanism has been proposed in which electron transfer to the iron does not take place;[5)] this involves an organoiron intermediate in place of the pentadienyl radical and iron(II).

In this report we have found that the reaction mixture of trans-$[FeCl_2(cyclam)]ClO_4$ (cyclam=1,4,8,11-tetraazacyclotetradecane)[6)] and linolenic acid exhibits high ability for degradation of TCPN (TCPN=tetraphenylcyclopentadienone),[7] one of the famous singlet oxygen($^1\Delta_g$) quenchers, and that peroxidation of linolenic acid proceeds in the presence of the trans compound, although the cis compound,[6] cis-[FeCl$_2$(cyclam)]Cl, exhibits no activity for both the reactions.

In Fig. 1, the time course of absorbance at 505 nm(25 ℃) of the reaction mixtures of TCPN, linolenic acid and trans-[FeCl$_2$(cyclam)]$^+$, and TCPN, linolenic acid and cis-[FeCl$_2$(cyclam)]$^+$, are shown. It is known that TCPN shows strong absorbtion band at 505 nm. In the case of of the cis compound, no decrease of the absorbance was detected within several hours. On the other hand, rapid decrease of the absorbance at 505 nm was observed in the mixture with the trans compound. This demonstrates that there is a singlet oxygen($^1\Delta_g$)-like active species in this solution, similar to the case of Ni(II)-β-diketonato/aldehyde system.[8] We have examined the reaction mixtures containing the Fe(III) complex and linolenic acid in terms of the TBA method(TBA=2-thio-barbituric acid);[9] this determines the quantity of malondialdehyde in the solution which may be derived from the peroxidation of unsaturated fatty acid as described in the literature.[10] The results(Fig. 2, 25

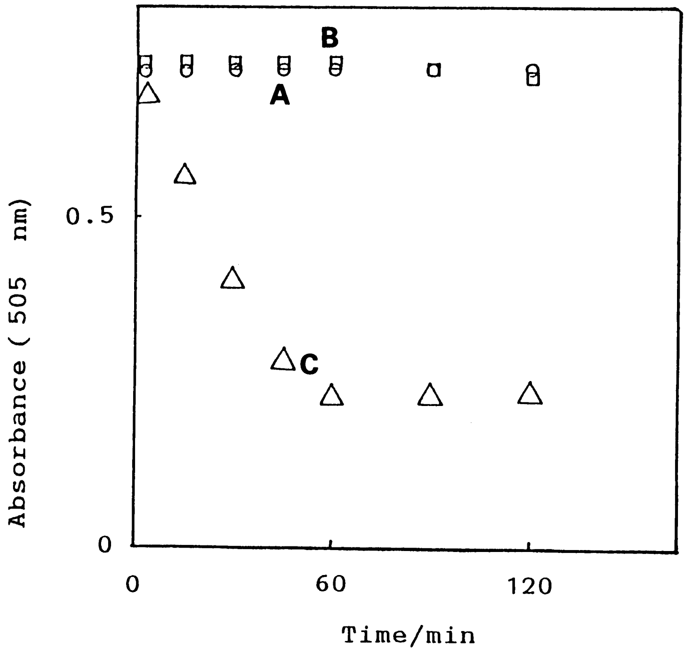

Fig. 1. Time course of the absorbance at 505 nm of the reaction mixture of TCPN(25 cm^3 of 1,2-CH$_2$ClCH$_2$Cl, 1/1500 mol dm^{-3}) and linolenic acid(1.5 cm^3) with iron(III) compound(2 cm^3 of dmso, 1/250 mol dm^{-3}) (B(□):cis-[FeCl$_2$(cyclam)]$^+$ and C(△): trans-[FeCl$_2$(cyclam)]$^+$), and without iron(III) compound, A(○).

℃) are clearly consistent with the result in Fig. 1; that is, only the trans compound can peroxidize the linolenic acid. Thus, the present work is the first report indicating that the peroxidation of linolenic acid by a iron(III) complex is closely related with the formation of a singlet oxygen-like active species.

We have investigated the reaction mechanism on the peroxidation of linolenic acid in terms of ESR spectra. In the spectrum of the trans compound, the signal due to the low-spin species is observed(g=3.2 — 2.3), and the spectrum also indicates that the cis compound is of high-spin type. The addition of linolenic acid to the solution of the trans compound under an aerobic condition has lead to appearance of a new ESR signal at g≒ 4.3, suggesting the formation of some high-spin species in the solution. This implies that the linolenic acid may interact with the iron(III) ion of the trans compound. No radical species was detected in the ESR spectrum of the reaction mixture of linolenic acid and the trans compound at the initial stage.

We also have measured the cyclic voltammograms of both the trans and the cis compounds.[11] It is noteworthy that the electrochemical properties of both compounds(reduction potential, Fe(III)→Fe(II), its reversibility, etc.) are very similar to each other under argon,

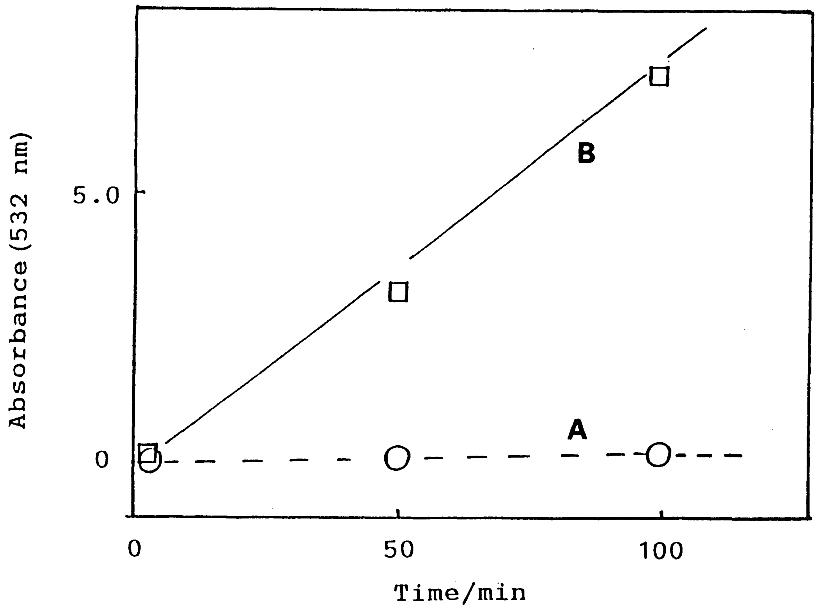

Fig. 2. Time course of the absorbance at 532 nm of reaction mixture(iron complex(1 cm^3 in dmso, 1/250 mol dm^{-3}), 25 cm^3 of 1,2-$ClCH_2CH_2$-Cl, and 0.5 cm^3 of linolenic acid) treated by TBA(see text). A(○): cis-$[FeCl_2(cyclam)]^+$; B(□): trans-$[FeCl_2(cyclam)]^+$.

however, that CV behaviour of the trans compound is greatly different from that of the cis compound under dioxygen atmosphere; that is, the reaction between the iron(II) species formed at -0.07 V(vs.SSCE) and dioxygen was observed only for the trans compound.

Based on these facts, we would like to propose a reaction mechanism for peroxidation of linolenic acid by the trans compound; at first linolenic acid approaches to a iron(III) ion, and dioxygen molecule may interact with the iron(III) ion loosely, and this loosely bound oxygen may be activated to contain some degree of singlet oxygen character, as suggested in Ni(II)-β-diketonato/aldehyde system.8) This activated oxygen may react with the substrate, leading to the peroxidation of linolenic acid. The formation of an oxygenated species seems to be specific for the trans compound, as indicated in the present electrochemical data, and also as suggested in the reaction with H_2O_2.12)

References

1) G. A. Veldink and J. F. G. Vliegenhart, Adv. Inorg. Biochem., 6, 139(1984).
2) B. Samuelsson, Science, 220, 568(1983).
3) M. R. Egmond, G. A. Veldink, J. F. G. Vliegenhart, and J. Boldingh, Biochem. Biophys. Res. Commun., 54, 1178(1973).
4) J. J. M. C. de Groot, G. J. Garssen, J. F. G. Vliegenhart, J. Boldingh, and B. F. van Gelder, Biochim. Biophys. Acta, 337, 71 (1975); 326, 279(1975).
5) E. J. Corey and R. Nagata, J. Am. Chem. Soc., 109, 8107(1987).
6) P. Chan and C. K. Coon, J. Chem. Soc., Dalton Trans., 1976, 858.
7) H. W.-S. Chan, J. Am. Chem. Soc., 93, 2357(1971).
8) Y. Nishida, T. Fujimoto, and N. Tanaka, Chem. Lett., 1992, 1291.
9) Y. Nishida and K. Yamada, J. Chem. Soc., Dalton Trans., 1990, 3639.
10) B. Halliwell and J. M. C. Gutteridge, "Free Radicals in Biology and Medicine," Oxford University Press, London(1985), Chap. 4.
11) The electrochemical measurements were done as follows: solvent, dimethylsulphoxide; supporting electrolyte, 0.1 mol dm^{-3} $(n\text{-}C_4H_9)_4$-NBF_4; metal compound, 0.1 mol dm^{-3}; temperature, 25 ℃; glassy carbon electrode; reference electrode, saturated sodium chloride calomel electrode(SSCE).
12) W. Nam, R. Ho, and J. S. Valentine, J. Am. Chem. Soc., 113, 7052 (1991)

(Received November 27, 1992)