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Tryptophan dioxygenase-like catalysis of achiral and chiral manganese(II) porphyrins for dioxygen-inserted indole-ring opening reactions

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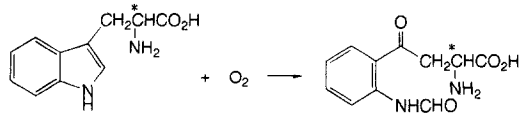
Abstract

The mimetic tryptophan dioxygenase reaction with tetraphenylporphyrinato- or 5α,10α,15α,20α-tetrakis-[*o*-(L-(−)-camphanoylamido)phenyl]porphyrinato-manganese(II), *N*-methylimizadole (as a proximal L-histidine model), and tryptophan analogues (3-methylindole and methyl *N*-acetyl-L (or D)-tryptophanate) in THF or CH_2Cl_2 at 298 K in atmospheric O_2 carried ring-opening products of 2′-formamidoacetophenone and methyl *R*-(−)-2-acetamido-3-(2′-formamidobenzyl)propanoate with 53% and 64% selectivity (23% e.e. of the *R*-(−) isomer), respectively. The key intermediate of ternary *N*-methyl-imizadole-Mn^{III}–O_2^- complex was detected by UV and ESR spectra, and 3-indolyl hydroperoxide was also observed by NMR as a stable precursor of the 2′-formamido-acetophenone formation. The isotopic experiments with 1-deuterated substrate demonstrated that the rate-determining step was the deprotonation process from the indole-ring at the 1-NH position of the substrate by the activated dioxygen (O_2^-). The native tryptophan dioxygenase catalysis was also discussed on the basis of the experimental findings in the present mimetic reactions.

Keywords: Tryptophan dioxygenase model; Metalloporphyrins; Ring-opening dioxygenolysis

1. Introduction

L-Tryptophan 2,3-dioxygenase (TDO) possessing the proximal L-histidine coordinated iron(III) protoporphyrin IX catalyzes the dioxygen-inserted indole-ring cleavage of L-tryptophan (L-Trp) to form L-formylkynurenine (FK) when it has the active iron(II) protoporphyrin IX by the quantitative additions of sodium dithionite or the illumination of light under an aerobic conditions [1–5] [1]



L-Trp FK (1)

[1] It has been reported that the almost inactive ferric(III) TDO must be in the ferrous(II) state to initiate the TDO reaction [3,6]. It has also been reported that the small amount of H_2O_2 can turn the ferric(III) state into active ferrous(II) in TDO [7].

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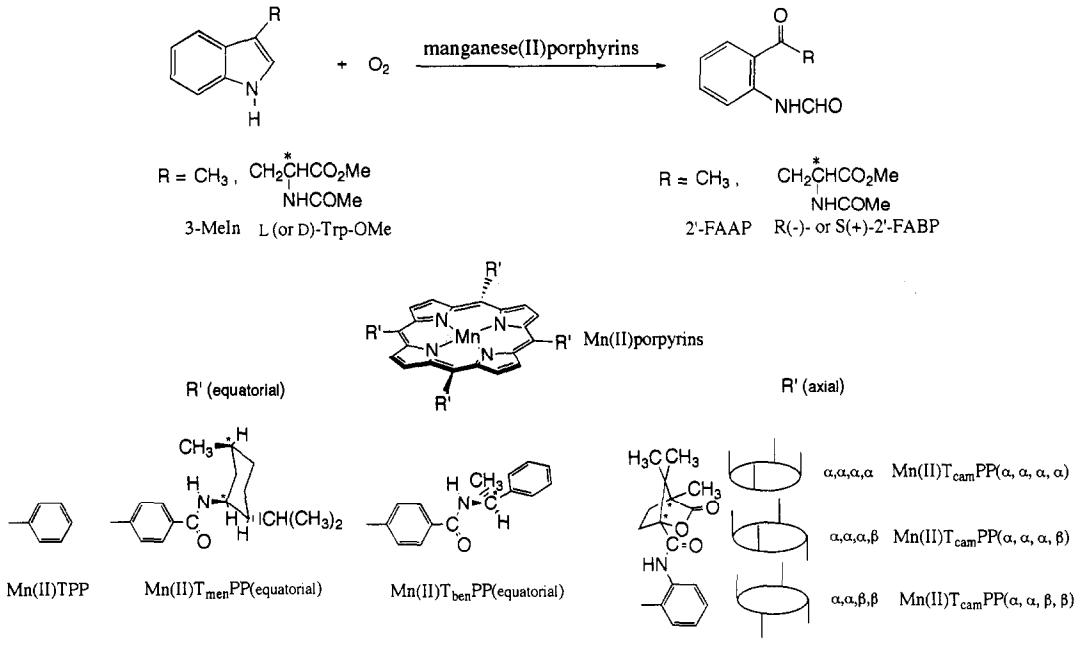
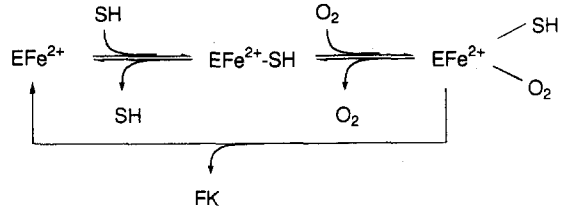


Scheme 1.

The detailed mechanism of the TDO reaction (Eq. (1)) has still been unveiled, but the following pathways (Eq. (2)) have hitherto been proposed [3]:



(2)

where EFe^{2+} and SH denote an active TDO carrying the iron(II) protoporphyrin IX {$HemeFe^{II}$} and the L-Trp substrate, respectively. Eq. (2) demonstrates that the TDO-reaction proceeds via the spectrophotometrically assumed ternary substrate–enzyme–dioxygen complex ($SH–EFe^{2+}–O_2$) [2] which is considered to be formed via the enzyme–substrate complex ($EFe^{2+}–SH$) rather than via the enzyme–oxygen one ($EFe^{2+}–O_2$ or $EFe^{3+}–O_2^-$). However, it is not easily accepted that the ternary $SH–EFe^{2+}–O_2$ complex formed from $EFe^{2+}–SH$ and O_2 locates the substrate and O_2 as the fifth and sixth axial ligands, respectively, because the ESR spectroscopic investigation emphasized the intensified coordination of the fifth proximal histidine (His) ligand to the $HemeFe^{II}$ by the addition of SH (L-Trp) to TDO [5]. Eq. (2) also excludes the detailed explanation about the FK formation intramolecularly via the ternary complex, even if the ternary complex is really formed. Recently, Leeds et al. have reported the isotope effects of a solvent (D_2O) on the TDO reaction and the thermodynamic barrier (15.9 kcal mol^{-1}) calculated for the 1-electron oxidation of tryptophan to the free radical by the heme-bound oxygen [8]. Although, Leeds et al. suggested the importance of the indole–proton deprotonation as a partial rate-determining process, they did not give detailed information about the mode of reaction between the substrate–enzyme complex and the heme-bound O_2.

[2] The ternary complex has been assumed by the change of the Soret band at $\lambda_{max} = 432$ nm and Q one $\lambda_{max} = 553$ of the iron(II) protoporphyrin IX to the Soret band at $\lambda_{max} = 418$ nm and Q one $\lambda_{max} = 545$ and 580 nm by the addition of the substrate and O_2.

In this respect, the previously reported mimetic TDO-reactions (the oxidative pyrrole-ring cleavage of indole derivatives) with bis(salicylidene)-ethylenediaminatocobalt(II) (CoSalen) [9], *meso*-tetraphenylporphyrinatocobalt(II) (CoTPP) [10,11], *meso*-tetraphenylporphyrinatoiron(II or III) ($Fe(py)_2TPP$ or Fe-ClTPP) [12,13], manganese(II) phthalocyanine (MnPc) [14], or the in situ prepared catalytic system of CuCl/py [15,16] or $FeCl_2$ (or $FeCl_3$)/py/bpy [17] have not succeeded to interpret the TDO reaction (Eq. (2)) in detail, even though the ternary SH–metal–O_2 complex has been detected in the case of Co^{II}Salen [9], Fe^{III}ClTPP-alkalizing reagent [13], or MnPC [14], together with the ring-opening product.

This article describes the TDO-like catalysis of the following achiral or chiral manganese(II) porphyrins for the ring-opening dioxygenolysis of tryptophan analogues {3-methylindole (3-MeIn) and methyl *N*-acetyl-L (or D)-tryptophanate (L (or D)-Trp-OMe)} to form 2′-formamidoacetophenone (2′-FAAP) or methyl *R*-(−)- or *S*-(+)-2-acetamido-3-(2′-formamidobenzyl)propanoate {*R*-(−)- or *S*-(+)-2′-FABP} (Scheme 1).

2. Experimental

2.1. Materials

3-Methylindole is commercially available, and methyl *N*-acetyl-L (or D)-tryptophanate was prepared according to the literature [15]. *meso*-Tetraphenylporphyrinatometal(II) complexes of M^{II}TPP (M = Mn, Fe, or Co) were obtained as described previously [18,19]. The chiral 5,10,15,20-tetrakis-[*p*-(L(−)-menthoxycarbamoyl)phenyl]porphyrinato manganese(II or III) {$Mn^{II}T_{men}PP$(equatorial), 5,10,15,20-tetrakis-[*p*-(D(+)-α-methylbenzyl)carbamoyl)phenyl]-porphyrinato manganese(II) {$Mn^{II}T_{ben}PP$(equatorial)}, and 5α,10α,15α,20α-, 5α,10α,15α,20β- or 5α,10α,15β,20β-tetrakis-[*o*-(L(−)-camphanoylamido)phenyl]porphyrinato manganese(II) {$Mn^{II}T_{cam}PP(\alpha,\alpha,\alpha,\alpha)$, $Mn^{II}T_{cam}PP(\alpha,\alpha,\alpha,\beta)$, or $Mn^{II}T_{cam}PP(\alpha,\alpha,\beta,\beta)$, respectively} were prepared with the corresponding chiral ligands {T_{men}PP(equatorial)}, {T_{ben}PP(equatorial)}, {$T_{cam}PP(\alpha,\alpha,\alpha,\alpha)$}, {$T_{cam}PP(\alpha,\alpha,\alpha,\beta)$}, or {$T_{cam}PP(\alpha,\alpha,\beta,\beta)$ } instead of with the TPP ligand; the chiral porphyrin ligands were obtained by the amidation of 5,10,15,20-tetrakis-[(*p*-chlorocarbony) or (*o*-amino)]phenyl)-porphyrin with L(−)-methylamine, D(+)-α-methylbenzylamine, or L(−)-camphanic acid.

2.2. Reaction procedures and product analyses

The dioxygenolyses of 2.5–50 mmol dm^{-3} tryptophan analogues (3-methylindole, *N*-methylindole, and methyl *N*-acetyl-L(or D)-tryptophanate) with 5.0 mmol dm^{-3} achiral metalloporphyrin or 1.0–250 μmol dm^{-3} chiral manganese porphyrins were carried out in THF or CH_2Cl_2 at 298 K for 24 h under O_2 atmosphere. The amounts of unreacted substrates and the products were determined spectrophotometrically (UV in MeOH) after isolation from the reaction mixtures by TLC on silica gel (Merk F254); The R_f and λ_{max} (ϵ) values of 3-methylindole or its ring-opening product 2′-FAAP (2′-formamidoacetophenone) were 0.67 or 0.43 {developing solvent: benzene/ether/heptane (1:1:1)} and 290 (3820) or 318 (4450 cm^{-1} mol^{-1} dm^3), respectively, and those of methyl *N*-acetyl-L (or D)-tryptophanate and its ring-opening product *R*-(−)- or *S*-(+)-2′-FABP (methyl *R*-(−)- or *S*-(+)-2-acetamido-3-(2′-formamido-benzyl)-propanoate) were 0.41 or 0.23 {ethyl acetate/acetone/ether (1:1:1)} and 290 (5230) or 320 nm (3620 cm^{-1} mol^{-1} dm^3), respectively. The enantiomeric excess (e.e.) of the ring-opening product {*R*-(−)- or *S*-(+)-2′-FABP} of methyl-*N*-acetyl-L (or D)-tryptophanate was evaluated from the concentration sum of *R*-(−)- and *S*-(+)-2′-FABP (determined by the UV measurements mentioned above) and their concentration difference measured by the CD spectra in MeOH at λ_{max} = 250

nm ($[\theta] = \pm 850°$ cm^2 dmol^{-1} for pure *S*-(+)- or *R*-(−)-2′-FABP).

2.3. UV, ESR and NMR measurements

UV-visible and ESR spectra for the coordination of O_2, an Trp-analogue, or the both to $Mn^{II}TPP$ were measured in THF at 298 or 77 K, respectively. 100–400 MHz ^{1}H-NMR spectra of the reaction intermediates such as 3-indolyl hydroperoxide were measured in $CDCl_3$ or DMSO-d_6.

3. Results and discussion

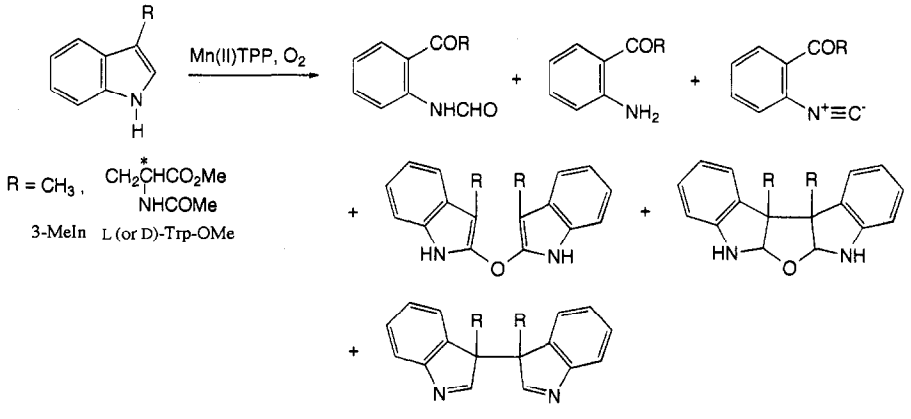
3.1. Catalytic activity of $Mn^{II}TPP$ and reactivities of substrates

The catalytic activities of the achiral metalloporphyrins {$M^{II}TPP$ (M = Mn, Fe, or Co)} were first examined in the dioxygenolysis of a SH substrate {SH = 3-MeIn (3-methylindole), *N*-MeIn (*N*-methylindole), or Trp-OMe (methyl-*N*-acetyl-LD-tryptophanate)} in THF at 298 K for 24 h under atmospheric O_2. The experimental results shown in Table 1 indicate the following notable points: (a) The present reaction can not proceed without the catalyst. (b) The catalytic activities (reflected by the substrate conversion) follow the order of $Mn^{II} > Fe^{II} > Co^{II}$. Since the coordination of nucleophilic SH (3-MeIn or Trp-OMe) to $M^{II}TPP$ is facilitated by lowering of the energy level of the half-occupied d_{z^2}-orbital on the central metal in the order of $Mn^{II} > Fe^{II} > Co^{II}$ [20]. The order of catalytic activities may reflect the easiness of the SH coordination to $M^{II}TPP$. (c) Trp-OMe is less reactive as compared with MeIn, probably due to the steric hindrance of the bulky Trp-OMe

Table 1
Catalytic activities of metalloporphyrins for the ring-opening dioxygenolysis of methyl *N*-acetyl-LD-tryptophanate (Trp-OMe), 3-methylindole (3-MeIn), or *N*-methylindole (*N*-MeIn) [a]

Catalyst	Substrate	Conv./ %	Yield of ring-opening product [b]/%	Selectivity [c]/ %
none	3-MeIn	0	0	—
$Mn^{II}TPP$	Trp-OMe	77	32	42
	3-MeIn	90	15	17
	N-MeIn	0	0	—
$Fe^{II}TPP$	Trp-OMe	5	trace	—
	3-MeIn	78	9	11
$Co^{II}TPP$	Trp-OMe	0	0	—
	3-MeIn	48	20	42

[a] [Catalyst] = 5.0 mmol dm^{-3} and [substrate] = 50.0 mmol dm^{-3} for the 3 (or *N*)-MeIn (or Trp-OMe), respectively in THF at 298 K for 24 h under atmospheric O_2.
[b] Methyl-2-acetamido-3-(2′-formamidobenzyl)propanoate for Trp-OMe and 2′-formamidoacetophenone for MeIn.
[c] For the ring-opening product.



Scheme 2.

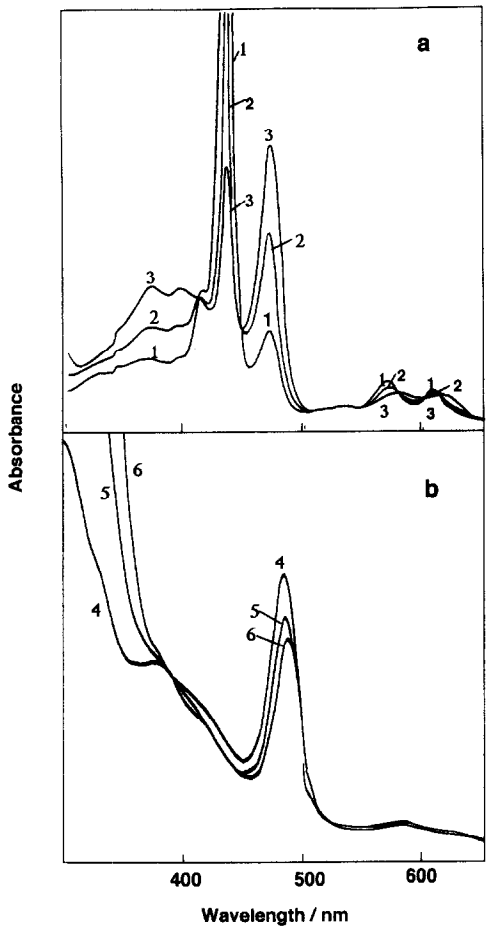


Fig. 1. UV absorption spectra in THF for $Mn^{II}TPP/O_2$ (a) and $Mn^{II}TPP/O_2$ /3-MeIn (b) a: (1) without O_2, (2) after 30 min in air, and (3) after 20 min in O_2. b: $Mn^{II}TPP$ (5.0 μmol dm^{-3}) was reacted with O_2 for 48 h previously. (4) 0.5 min, (5) 6 h, and (6) 24 h after addition of 3-MeIn (50.0 mmol dm^{-3}) to $Mn^{II}TPP/O_2$.

against its coordination to $M^{II}TPP$. (d) The reaction formes other products in addition to the ring-opening ones (2′-FAAP for MeIn and 2′-FABP for Trp-OMe). The reaction products are shown in Scheme 2, but the amounts of ring-opening products such as 2′-aminoacetophenone (or 2-acetamido-3-(2′-aminobenzyl)propanoate) and 2′-isocyanoacetophenone (or 2-acetamido-3-(2′-isocyanobenzyl)propanoate) were negligibly small in the dioxygenolysis of 3-MeIn (or Trp-OMe), respectively.

Another notable point (e) is that *N*-MeIn is not reactive as a Trp-analogue substrate (SH). This indicates the very important fact that *N*-MeIn is not an appropriate SH for the present reaction which requires the N–H bond instead of the N–Me one in the indole-ring of SH for the release of H (as a H^+) from the N–H bond to make a S · radical (as discussed in a later section).

3.2. Formation of ternary solvent–$Mn^{III}TPP$–O_2^- or substrate–$Mn^{III}TPP$–O_2^- complex

In regard to the ternary complex formation, the $M^{II}TPP/O_2$ system in the absence of SH forms a THF–$Mn^{III}TPP$–O_2^- complex which shifted the Soret (λ_{max} = 433 nm) and Q bands (λ_{max} = 568 and 605 nm) of $Mn^{II}TPP$ to λ_{max} = 469 nm (Soret) and λ_{max} = 574 and 615 nm (Q band) with decreasing the absorbances of the both Soret and Q bands. The addition of SH (MeIn) to the above $M^{II}TPP/O_2$ system (viz. THF–$Mn^{III}TPP$–O_2^-) gave the ternary SH–$Mn^{III}TPP$–O_2^- complex by changing the Soret and Q bands of THF–$Mn^{III}TPP$–O_2^- to λ_{max} = 482 nm (Soret) and λ_{max} = 582 and 640 nm (Q

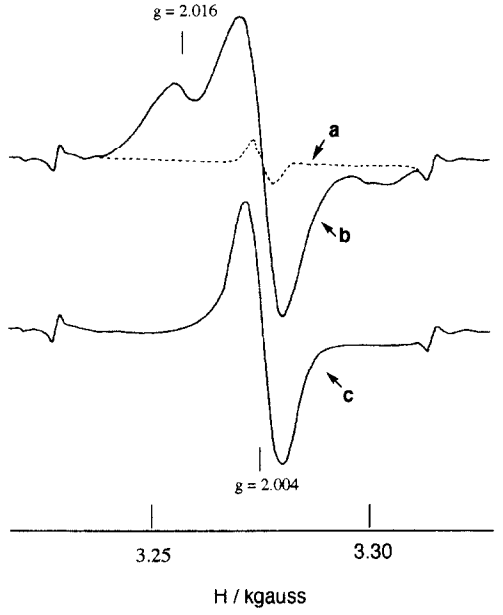
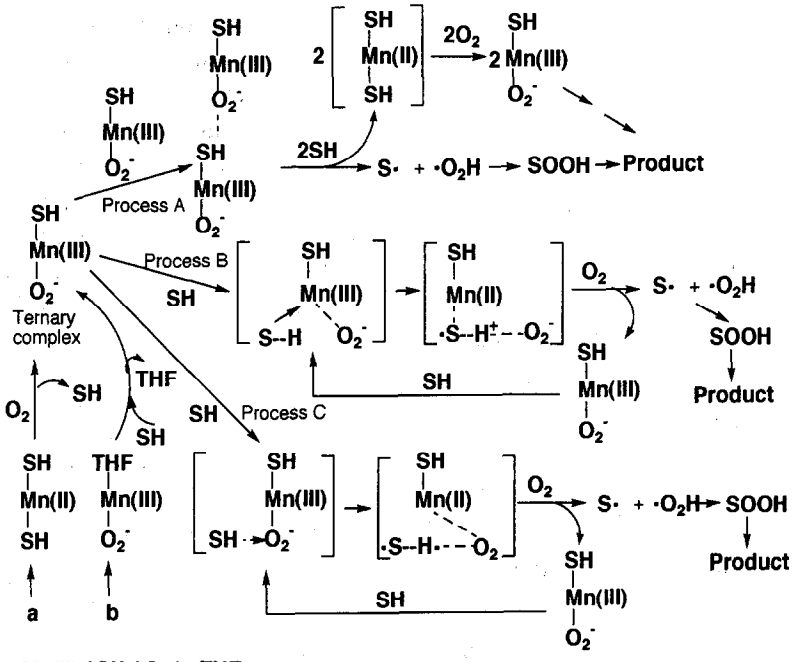


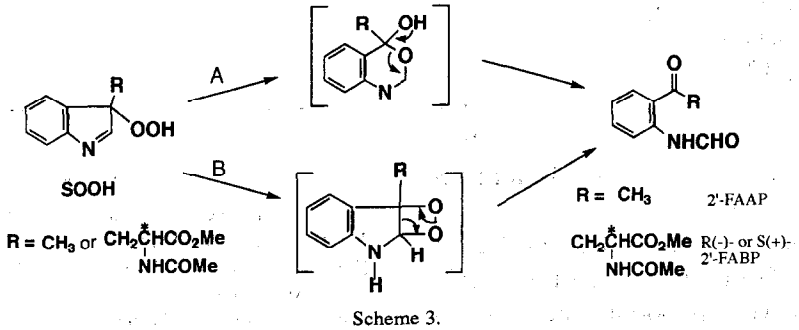
Fig. 2. ESR spectra of (a) $Mn^{II}TPP/O_2$, (b) $Mn^{II}TPP$/3-MeIn/O_2, and (c) $Mn^{II}TPP$/*N*-MeIn/O_2 in THF at 77 K; [$Mn^{II}TPP$] = 10.0 mmol dm^{-3} and [3- or *N*-MeIn] = 1.0 mol dm^{-3}. $Mn^{II}TPP$/3- or *N*-MeIn/O_2 means the mixing of $Mn^{II}TPP$ and 3- or *N*-MeIn for 20 min in deaerated THF before O_2 purge.

band) via $\lambda_{max} = 479$ nm (Soret) and $\lambda_{max} = 580$ and 624 nm (Q band) at the early stage of the mixing time with the lowering of the Soret-band absorbance (Fig. 1).

On the other hand, the ESR spectrum of the $Mn^{II}TPP/O_2$ system in THF had a weak signal of O_2^- of the THF–$Mn^{III}TPP$–O_2^- complex at $g = 2.004$. This O_2^- signal at $g = 2.004$ was greatly intensified with the ternary complex of SH–$Mn^{III}TPP$–O_2^-, when O_2 was poured into the preliminary mixed system of $Mn^{II}TPP$ and SH (MeIn) in deaerated THF (Fig. 2). The ESR of $Mn^{II}TPP$/SH or SH/O_2 system per se did not show a peak in ESR, but the unreactive *N*-MeIn also gave the O_2^- signal at $g = 2.004$ through the formation of the ternary *N*-MeIn–$Mn^{III}TPP$–O_2^- complex. It is noteworthy here that the mixed system of $Mn^{II}TPP$/SH (3-MeIn)/O_2 had an ESR signal (at $g = 2.016$) indicating the carbon (C·) or peroxy COO· radical of 3-MeIn. The ESR signal at $g = 2.016$, however, does not lead to an idea that the ternary SH–$Mn^{III}TPP$–O_2^- complex turns to ·S (or ·O_2S)–Mn^{II}–O_2^- via the deprotonation of



a. Mn(II) / SH / O_2 in THF
b. Mn(II) / O_2 / SH in THF
Mn(III) = Mn(III)TPP
SH = 3-MeIn orTrp



Scheme 3.

SH from SH–Mn^{III}TPP–O_2^- (or via the subsequent reaction of ·S–Mn^{II}–O_2^- with O_2), because the rate-determining deprotonation from the coordinated SH does not occur easily without third bodies such as O_2^- (as discussed bellow).

Thus, the activation of O_2 by the Mn^{II}TPP catalyst through the ternary complex formation in the reaction system was confirmed by UV and ESR spectra. It is necessary, therefore, to clarify how activated O_2 (O_2^-) behaves for the present ring-opening of SH (3-MeIn or Trp-OMe).

3.3. *Mn^{II}TPP-catalyzed reaction process via ternary SH–Mn^{III}TPP–O_2^- complex*

There are three possible modes of the reaction between the ternary SH–Mn^{III}TPP–O_2^- complex and free (or coordinated) SH for the formation of the ring-opening product (Scheme 3). The formation process of the ternary complex per se is also dependent on the starting system. The Mn^{II}TPP/SH/O_2 system can start to make the ternary SH–Mn^{III}TPP–O_2^- complex after O_2 purge into the previously mixed Mn^{II}TPP/SH, while the Mn^{II}TPP/O_2/SH system commences to obtain the same ternary complex after the addition of SH into the Mn^{II}TPP/O_2 system. In both systems, the ternary SH–Mn^{III}TPP–O_2^- complex was detected clearly by the ESR measurements.

Process A in Scheme 3 indicates an intermolecular reaction between two ternary SH–Mn^{III}TPP–O_2^- complexes, while process B (or C) shows the reaction between the ternary SH–Mn^{III}TPP–O_2^- complex and SH in the inner shell of the complex. The former case (A) indicates a second-order kinetic dependency on the Mn^{II}TPP concentration, while the latter case (B or C) would take a first-order one. In this regard, the present dioxygenolysis of SH (3-MeIn or Trp-OMe) with Mn^{II}TPP is of the latter case as shown in Fig. 3 (in which is also shown the treatment of the present mimetic TDO-reaction by the pseudo-first-order rate law).

The processes B and C can also produce the S · (3-methylindolyl) radical which was detected by ESR at $g = 2.016$ as the S · species or the oxygenated SOO · one. The activated O_2 (O_2^-) in the former pathway (B) abstracts H from SH as a proton (H^+) with the electron back-donation from SH (not from O_2^-) to Mn^{III} in the complex, while the activated O_2^- extracts the H radical from SH and returns the electron to Mn^{III} in the latter process (C). Both of the

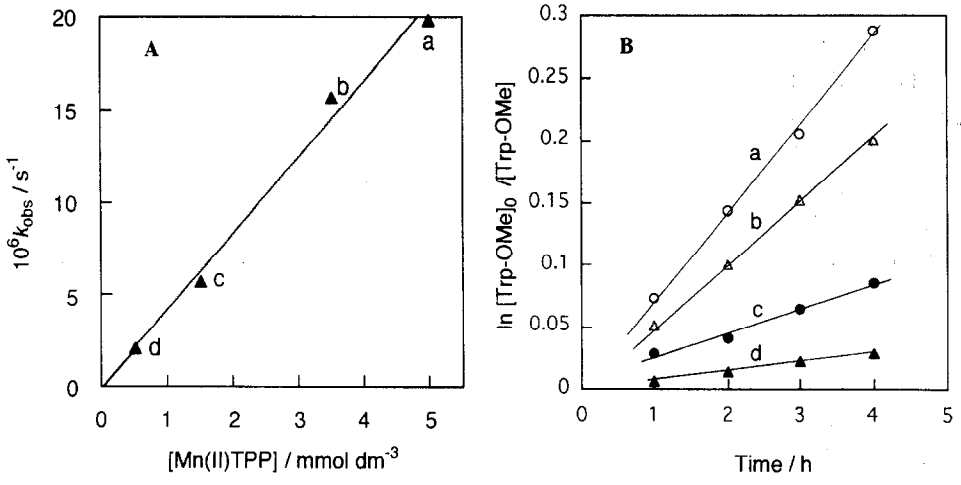


Fig. 3. Rate dependence on the catalyst concentration for the Mn^{II}TPP-catalyzed dioxygenolysis of 3-MeIn (50.0 mmol dm^{-3}) in THF at 298 K (A) and the pseudo-first-order rate expression of the present reactions (B). (a) 5.0 mmol dm^{-3}, (b) 3.5 mmol dm^{-3}, (c) 1.5 mmol dm^{-3}, and (d) 0.5 mmol dm^{-3} of [Mn^{II}TPP].

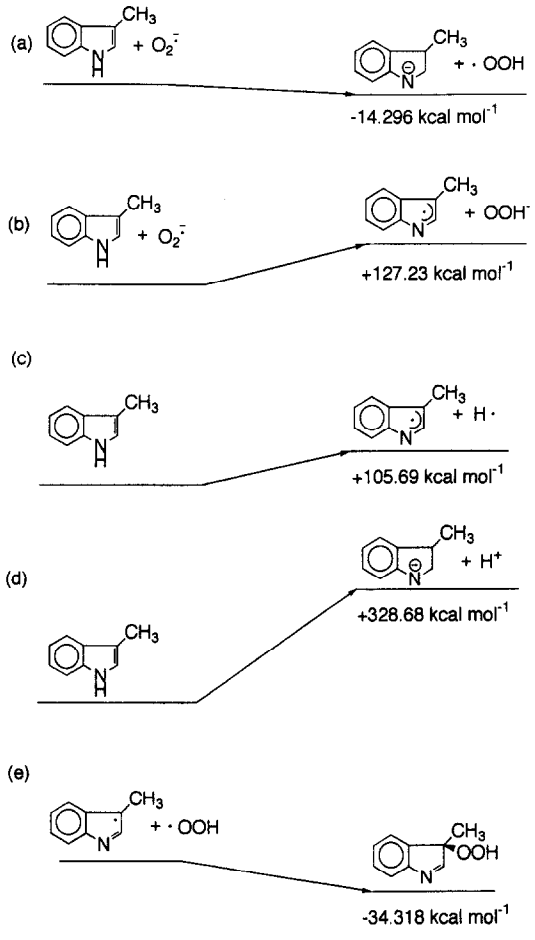


Fig. 4. MOPAC PM3 calculations of the reaction between 3-MeIn and O_2^- or between 3-indolyl and HO_2 radicals.

processes B and C can reproduce the ternary complex of SH–Mn^{III}TPP–O_2^-. The molecular orbital calculations of the H^+ or H · abstraction from SH (3-MeIn) by O_2^- performed with MOPAC PM3 (ver. 6.03) [21] supported process B (deprotonation by O_2^-) as the energetically plausible one (see Fig. 4).

3.4. Isotope effect of 1-deuterated substrate on the reaction

The use of SD (1-deuterated Trp-OMe) instead of SH (undeuterated Trp-OMe) in the dioxygenolysis reaction with Mn^{II}TPP in THF decreased the reaction rate appreciably without considerable change of the selectivity of the ring-opening product (2′-FABP) in the range of 35.4–41.6% (Table 2). Since the pseudo-first-order rate ratio, $k_H/k_D = 1.8$, indicates a first-order isotope effect, the deprotonation at the N-1 position of the substrate SH is a rate-determining step in the present reaction which proceeds via process B.

3.5. Formation of SOOH (hydroperoxide) as a precursor of the ring-opening product

In regard to the formation of the hydroperoxide (SOOH) in process B, the NMR measurements of the Mn^{II}TPP-catalyzed oxygenolysis of SH (3-MeIn) along the reaction time detected SOOH (3-indolyl hydroperoxide) as a stable intermediate (Fig. 5). In the NMR peaks of the reaction mixtures along the reaction time (A–D), the methyl (3-Me) signal at 2.35 ppm decreased gradually with increasing the two methyl signals of SOOH at 1.60 ppm and the ring-opening product (2′-FAAP) at 2.74 ppm; On the other hand, the peroxy–proton signal of SOOH was also observed at 7.80 ppm along A–D.The methyl or proton signal of SOOH (1.60 or 7.80 ppm, respectively) was appreciably diminished by the addition of CF_3CO_2H (hydroperoxide decomposer) to the reaction

Table 2
Dioxygenolysis of 1-deuterated and unduetrated Trp-OMe with Mn^{II}TPP [a]

Substrate	Conv./%	Yield of 2′-FABP/%	Selectivity of 2′-FABP/%	$10^6 k_{obs}/s^{-1}$	k_H/k_D
1-deuterated Trp-OMe	30	11	35	4.2	1.8
undeuterated Trp-OMe	18	7	41	2.3	

[a] [Mn^{II}TPP] = 1.0 mmol dm^{-3} and [substrate] = 20.0 mmol dm^{-3} in THF at 298 K in O_2 for 24 h.

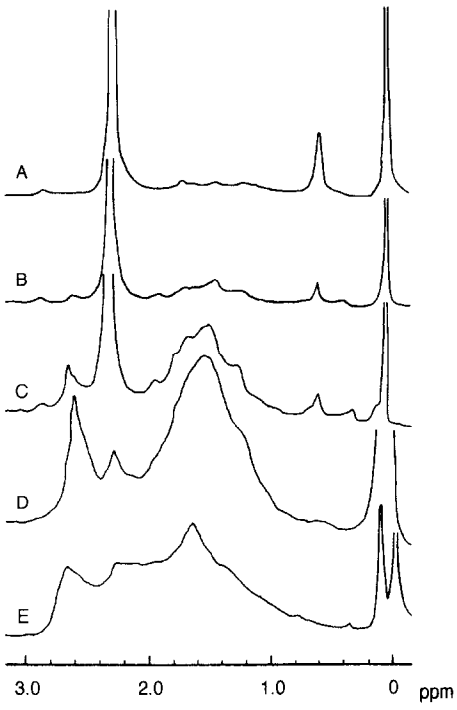


Fig. 5. ^{1}H-NMR methyl-signal change by the formation of 3-methylindolyl hydroperoxide in the dioxygenolysis of 3-MeIn (0.52 mol dm^{-3}) with Mn^{II}TPP (14.0 mmol dm^{-3}) in $CDCl_3$ at 298 K after (A) 20 min, (B) 40 min, (C) 6 h, and (D) 24 h. (E) indicates CF_3CO_2H addition to (D) (after 3 h).

mixtures (in D) including SOOH. Thus, the hydroperoxide SOOH was conformed in the present reaction which proceeds through the reaction between the ternary SH–Mn^{III}TPP–O_2^- complex and SH via the rate-determining deprotonation from SH by the activated dioxygen (O_2^-). In Scheme 3, the formation of the ring-opening product 2′-FAAP (or 2′-FABP) from SOOH proceeds via the Criegee-type step A rather than via the dioxetane-type path B, because the latter path is thermodynamically unfavorable [22,23].

3.6. Dependence of ring-opening product formation on reaction systems

Although SH (3-MeIn or Trp-OMe) was found to actually behaves as a basic ligand of the ternary SH–Mn^{III}TPP–O_2^- complex in the present reaction, it is of interest here to examine that another unreactive ligands such as *N*-methylimidazole (denoted by *N*-MeIm or His′) is useful or useless instead of SH as a model of the proximal His in TDO. In the dioxygenolysis of SH (3-MeIn) by Mn^{II}TPP with or without His′ (*N*-MeIm) in CH_2Cl_2 under the same reaction conditions denoted in Table 1, the yield and selectivity of the ring-opening product (2′-FAAP) were considerably changed by both the concentration ratio ([Mn^{II}TPP]/[His′] = 0–60) and the reaction system as shown in Table 3.

In the reaction system of Cat/O_2/SH or Cat/SH/O_2, the addition of SH before O_2 purge (viz. Cat/SH/O_2) was found slightly more effective than the Cat/O_2/SH system in terms of the 2′-FAAP production. This supports that the Cat/SH/O_2 system produced 2′-FAAP through the reaction between the ternary SH–Mn^{II}TPP–O_2^- complex and SH, while the Cat/O_2/SH system (the addition of SH after O_2 purge) can give a chance for the direct reaction between O_2^- in the THF–Mn^{III}TPP–O_2^- complex (detected by ESR) and SH to form

Table 3
The dioxygenolysis of 3-MeIn (SH) with Mn^{II}TPP (Cat) in the presence of *N*-MeIm (His′) [a]

[His′]/[Cat]	System [b]	Conv./%	Yield of 2′-FAAP/%	Selectivity of 2′-FAAP/%
0	Cat/O_2/SH	99	4	4
0	Cat/SH/O_2	94	9	10
20	Cat/His′/O_2/SH	94	50	53
20	Cat/His′/SH/O_2	89	8	9
60	Cat/His′/O_2/SH	95	7	7

[a] [Cat (Mn^{II}TPP)] = 5.0 mmol dm^{-3} and [SH (3-MeIn)] = 50.0 mmol dm^{-3} in CH_2Cl_2 at 298 K in O_2 for 24 h.
[b] Cat/A/B/C means the mixture of Cat and A was stirred for 20 min at first, then stirred for the same time in the presence of B, and finally reacted with C.

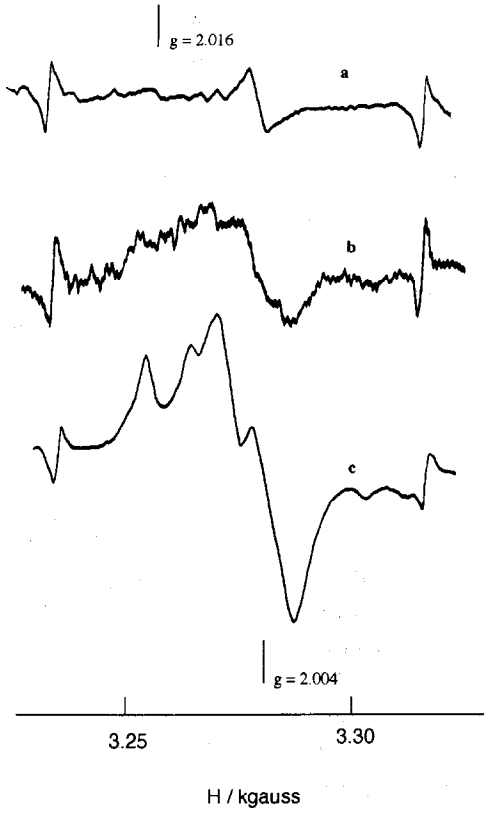


Fig. 6. ESR spectra of (a) $Mn^{II}TPP/N$-MeIm/O_2, (b) $Mn^{II}TPP/N$-MeIm/3-MeIn/O_2, and (c) $Mn^{II}TPP$/3-MeIn/O_2 in CH_2Cl_2 at 77 K; $[Mn^{II}TPP] = 10.0$ mmol dm^{-3} and $[N\text{-MeIm}] = [3\text{-MeIn}] = 1.0$ mol dm^{-3}.

THF (or SH)–Mn^{II}–S · /HO_2 · through the deprotonation and ligand-exchange reactions.

On the other hand, the Cat/His′/O_2/SH system ([His′]/[Cat] = 10–20) enhances the yield and selectivity of 2′-FAAP remarkably as compared with the Cat/O_2/SH system. The highest yield (50.0%) and selectivity (53.2%) of 2′-FAAP were obtained under the condition of [His′]/[Cat] = 20. The excess amount of His′ ([His′]/[Cat] = 60) in the Cat/His′/O_2/SH system, however, decreased the 2′-FAAP formation considerably. In the $Mn^{II}TPP$/His′/O_2 system the ESR signal of the ternary His′–$Mn^{III}TPP$–O_2^- complex was at $g = 2.004$ (O_2^-), similarly to SH(3-MeIn)–$Mn^{III}TPP$–O_2^- (see Fig. 6). The high concentration of His′ (or SH) with respect to [$Mn^{II}TPP$] ([His′ or SH]/[$Mn^{II}TPP$] = 100) was found to result in the low intensity of the ESR signal for the ternary His′–$Mn^{III}TPP$–O_2^- complex, probably because the ligand exchange from $Mn^{II}TPP$–$(His')_2/O_2$ (or $Mn^{II}(SH)_2$ in the absence of His′) to His′–$Mn^{III}TPP$–O_2^- (or SH–$Mn^{III}TPP$–O_2^- in the absence of His′) is depressed in the high concentration range of His′ (or SH). In fact, the $Mn^{II}TPP$/His′/SH (3-MeIn)/O_2 system (addition of SH before O_2 purge) considerably decreased the yield of indole-ring cleaved 2′-FAAP from 50.0% (in the former reaction system) to 7.9% one (see Table 3). The presence of the excess amount of such nucleophilic ligands (L) as SH (or His′) might depress the ternary complex formation through the ligand exchange from $Mn^{II}TPP(L)_2/O_2$ to L–Mn^{III}–O_2^-/L. Although the coordination strengths of His′ and SH (3-MeIn) and the ligand exchange by O_2 are discussed below, the formation of the ternary His′–$Mn^{III}TPP$–O_2^- complex did not suffer remarkably from the relatively high concentration ratio ([His′]/[Cat] = 20) and promoted the reaction between the ternary complex and SH (3-MeIn) to form the ring-opening product (2′-FAAP). It is necessary here to notice that His′ is able to form the ternary His′–$Mn^{III}TPP$–O_2^- complex easily in the presence of SH under the present reaction conditions ([His′]/[SH] = 2–6 and [His′]/ Cat] = 10–60).

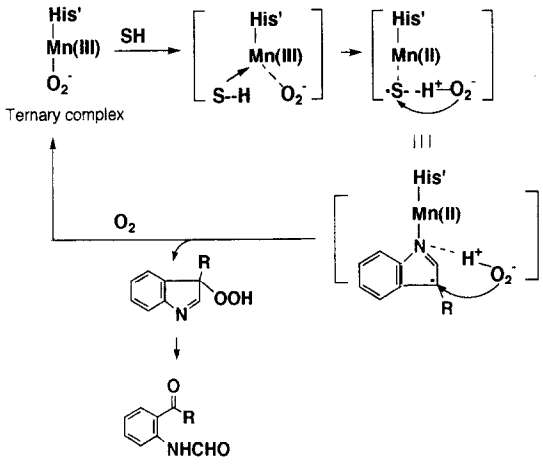
The relative coordination strengths of His′ and SH (3-MeIn) as a ligand of $Mn^{II}TPP$ were estimated from the equilibrium constant K_b of the complex formation between the His′ or SH ligand and $Zn^{II}TPP$ which does not require the sixth ligand [3]. The K_b (mol^{-1} dm^3) values of 8.69×10^{12} (His′) and 6.09×10^3 (SH) indicated the considerably higher coordination

[3] The K_b values between a fifth ligand (L) and $Zn^{II}TPP$ (Zn) were determined from the following linear relation obtained by means of UV spectrophotometric measurements: $[Zn]_0/(Abs - \epsilon_{Zn}[Zn]_0 - \epsilon_L[L]_0) = 1/K_b[L](\epsilon_{Zn-L} - \epsilon_{Zn} - \epsilon_L)) + 1(\epsilon_{Zn-L} - \epsilon_{Zn} - \epsilon_L)$, where Abs = absorbance, [Zn or $L]_0$ = initial concentration of Zn or L, and ϵ_c(c = Zn–L, Zn, or L) = molecular extinction coefficient of c.

strength of His′ as compared with that of SH. In the reaction system of Cat/His′/O_2/SH, the His′ (*N*-MeIm) ligand predominantly coordinates to $Mn^{II}TPP$ as compared with SH (3-MeIn). The preliminary mixing of Cat ($Mn^{II}TPP$) and His′ under [His′]/[$Mn^{II}TPP$] = 10–20 converted ca. 95% of $Mn^{II}TPP$ to $Mn^{II}TPP(His')_2$ [4], but the exchange of the sixth His′ ligand by O_2 is possible when ones take notice of the equilibrium constant K ($K = 8.695 \times 10^7$ mol^{-1} dm^3 $Torr^{-1}$ at −78°C in toluene) for the ligand exchange reaction from $Mn^{II}TPP$–His′/O_2 to $Mn^{II}TPP$–O_2/His′ (His′ = *N*-MeIm) [25]. Therefore, the formation of ternary His′–$Mn^{III}TPP$–O_2^- complex detected by ESR at $g = 2.004$ becomes possible, even in the $Mn^{II}TPP$/His′/SH (3-MeIn)/O_2 system (see Fig. 6).

Anyway, the $Mn^{II}TPP$/His′/O_2/SH system facilitated the formation of the ring-opening product in the highest extent under [His′]/[$Mn^{II}TPP$] = 20 rather than the $Mn^{II}TPP$/His′/SH/O_2 system through the ternary His′–$Mn^{III}TPP$–O_2^- complex formation. Therefore, the predominant coordination of SH to form His′–Mn^{II}–SH before that of O_2 (to form His′–$Mn^{III}TPP$–O_2^-) is not an appropriate process for the present TDO-like reaction. In this sense, an assumption can be made for the TDO reaction process expressed roughly in Eq. (2). Namely, the native TDO enzyme traps the substrate SH by the binding site of the reaction cavity in which the catalytic $HemeFe^{II}$ site forms the ternary complex of (the proximal His)–$HemeFe^{III}$–O_2^- As a matter of fact, SH should be located closely toward the O_2 coordination site.

The ligand exchange reaction from His′–$Mn^{III}TPP$–O_2^-/SH to His′–$Mn^{III}TPP$–SH/O_2^- leads to the deprotonation reaction from SH by



Scheme 4.

O_2^- to generate His′–$Mn^{II}TPP$–S · /HO_2 · with the electron back-donation from the coordinated SH (not from O_2^-) to Mn^{III} in the complex, as mentioned in the process B in Scheme 3. The HO_2 radical attacks the indole-ring 3-C radical position of the S · species in the His′–$Mn^{II}TPP$–S · complex in concert (Scheme 4).

Thus, the His′ ligand was found to behave like the proximal His ligand of $HemeFe^{II}$ in TDO, and the addition of SH after the formation of the ternary His′–$Mn^{III}TPP$–O_2^- complex formation in the $Mn^{II}TPP$/His′/O_2/SH system rather than the addition of O_2 after the formation of His′–$Mn^{II}TPP$–SH in $Mn^{II}TPP$/His′/SH/O_2 was very important to obtain the ring-opening product (via the hydroperoxide (SOOH)) in the high yield and high selectivity in the present mimetic TDO-reaction.

3.7. Stereoselective dioxygenolysis of L (or D)-Trp-OMe with optically active manganese(II) porphyrins

In order to confirm the present TDO-like catalysis of Mn^{II} porphyrins for the indole-ring opening of L-tryptophan via the process B expressed in Scheme 3, the stereoselective dioxygenolysis of L (or D)-Trp-OMe was carried out with $Mn^{II}T_{men}PP$ (equatorial), $Mn^{II}T_{ben}PP$(equatorial), $Mn^{II}T_{cam}PP(\alpha,\alpha,\alpha,\alpha)$,

[4] The use of equilibrium constant ($\beta_2 = 8530$ mol^{-2} dm^6) for the formation of $Mn^{II}TPP(N\text{-MeIm})_2$ from $Mn^{II}TPP$ and *N*-MeIm is described in Ref. [24]. $Mn^{II}TPP$/*N*-MeIm turns into $Mn^{II}TPP(N\text{-MeIm})_2$ in ca. 95% of $Mn^{II}TPP$.

Table 4
The stereoselective ring-opening dioxygenolysis of methyl *N*-acetyl-L (or D)-tryptophanate (L (or D)-Trp-OMe) with manganese(II) porphyrins [a]

Catalyst	Substrate			*R*-(−)- or *S*-(+)-2′-FABP			
	Conv./%	e.e./%	Config.	Yield/%	Selectivity/%	e.e./%	Config.
$Mn^{II}T_{men}PP$(equatorial)	29	17	L	2.4	8	15	*S*-(+)
$Mn^{II}T_{ben}PP$(equatorial)	45	3	D	5.9	13	10	*R*-(−)
$Mn^{II}T_{cam}PP(\alpha,\alpha,\alpha,\alpha)$	9.5	9	D	6.1	64	23	*R*-(−)
$Mn^{II}T_{cam}PP(\alpha,\alpha,\alpha,\beta)$	15	8	D	8.5	57	1	*R*-(−)
$Mn^{II}T_{cam}PP(\alpha,\alpha,\beta,\beta)$	13	7	D	10.4	80	6	*R*-(−)

[a] [Catalyst] = 2.0 mmol dm^{-3} and [substrate] = 20.0 mmol dm^{-3} in THF at 298 K for 24 h under atmospheric O_2.

$Mn^{II}T_{cam}PP(\alpha,\alpha,\alpha,\beta)$, or $Mn^{II}T_{cam}PP(\alpha,\alpha,\beta,\beta)$. The experimental results summarized in Table 4 indicates that the picket fence-type $Mn^{II}T_{cam}PP(\alpha,\alpha,\alpha,\alpha)$ complex consumed D-Trp-OMe predominantly in some extent (enantiomeric excess (e.e.) of 8.6%) and produced *R*-(−)-2′-FABP {*R*-(−)-2-acetamido-3-(2′-formamidobenzyl)propanoate}predominantly from D-Trp-OMe in the highest selectivity of 64.2% (23.3% e.e.).

As Scheme 5 indicates, the L (or D)-Trp-OMe substrate (L (or D)-SH) coordinates to $Mn^{II}T_{cam}PP(\alpha,\alpha,\alpha,\alpha)$ from the other side of the picket fence-type ligand and forms the ternary L (or D)-SH–Mn^{III}–O_2^- complex {Mn^{III} = $Mn^{III}T_{cam}PP(\alpha,\alpha,\alpha,\alpha)$} in the presence of O_2. The substrate coordination to the ternary complex from the picket fence-type chiral ligand side (viz. the coordinated O_2^- side) resulted in the predominant exchange from L (or D)-SH–Mn^{III}–O_2^-/L (or D)-SH to L (or D)-SH–Mn^{III}–D-SH/O_2^-, so as to produce *R*-(−)-2′-FABP through the same reaction steps depicted in Scheme 5.

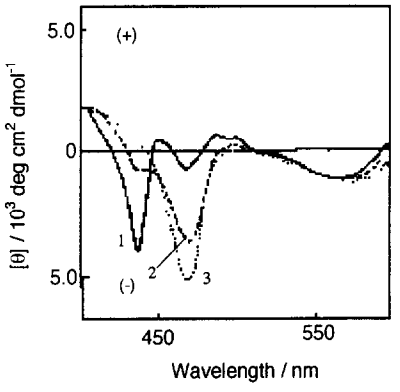
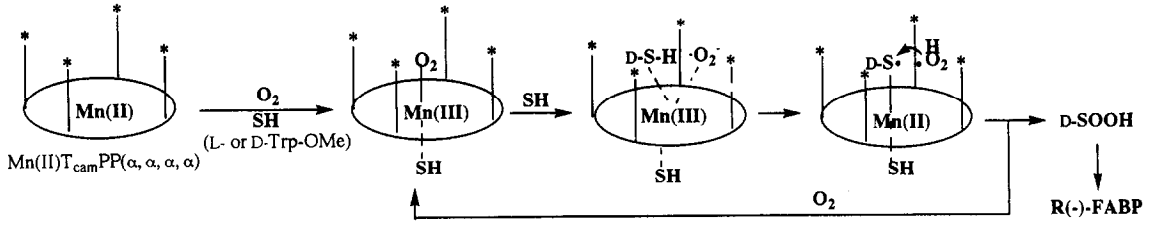


Fig. 7. CD spectra of the Soret band from (1) $Mn^{II}T_{cam}PP(\alpha,\alpha,\alpha,\alpha)$, (2) $Mn^{II}T_{cam}PP(\alpha,\alpha,\alpha,\beta)$, and (3) $Mn^{II}T_{cam}PP(\alpha,\alpha,\beta,\beta)$ in MeOH.

In this stereoselective dioxygenolysis of D-Trp-OMe by $Mn^{II}T_{cam}PP(\alpha,\alpha,\alpha,\alpha)$, the picket fence-type chiral ligand played an important role by inducing the asymmetric field on the porphyrin ring of $Mn^{II}T_{cam}PP(\alpha,\alpha,\alpha,\alpha)$ (see Fig. 7). In this regard, the native TDO has such an asymmetric field in its reaction cavity for the stereospecific binding of the L-Trp substrate



Scheme 5.

and for the production of *S*-(+)-FK (formylkynurenine).

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