cis-Dihydroxylation of Olefins by a Non-Heme Iron Catalyst: A Functional Model for Rieske Dioxygenases**

Kui Chen and Lawrence Que, Jr.*

In memory of Sir Derek H. R. Barton

The biodegradation of aromatic hydrocarbons and related environmental pollutants is initiated by a family of bacterial enzymes called Rieske dioxygenases.[1] These enzymes catalyze the NADH-mediated enantiospecific *cis*-dihydroxylation of arene and alkene double bonds with the incorporation of both atoms of dioxygen into *cis*-diol products.[1–4] The recently reported crystal structure of naphthalene 1,2-dioxygenase shows that the active site contains a mononuclear non-heme Fe^{II} center in close proximity to a Rieske Fe_2S_2 center.[5] The mononuclear Fe^{II} center is coordinated by two histidine residues and a bidentate aspartate group with two *cis* sites available for exogenous ligands (**A**, Figure 1). By analogy to

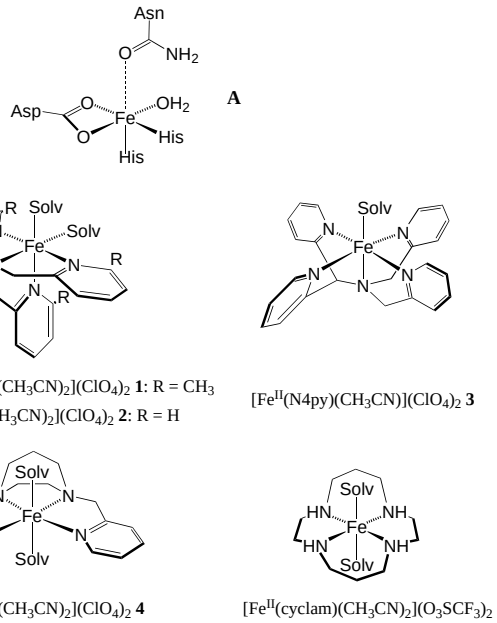

Figure 1. Structures of the mononuclear Fe^{II} center of naphthalene 1,2-dioxygenase (**A**) and synthetic non-heme iron complexes.[5, 10, 12, 15] Solv = solvent.

the mechanism of cytochrome P450,[6] it is proposed that the Fe^{II} center binds O_2 and accepts an electron from the Rieske cluster to give an Fe^{III} peroxo species that is responsible for oxidation of the substrate.[2, 7, 8] Since the only synthetic reagents capable of *cis*-dihydroxylation of olefins are OsO_4 and related high-valent compounds with *cis*-dioxo groups,[9] we sought a precedent among biomimetic iron complexes for the proposed enzyme mechanism. However, no combination of an iron complex with O_2 or H_2O_2 has thus far been shown to perform *cis*-dihydroxylation of olefins.[10–14] Herein we report the first example of a non-heme iron complex that catalyzes olefin *cis*-dihydroxylation as a functional model for Rieske dioxygenases.

The mononuclear non-heme iron complex $[Fe^{II}(6\text{-}Me_3\text{-}tpa)(CH_3CN)_2](ClO_4)_2$ (**1**, Figure 1) consists of an Fe^{II} center with a tetradentate ligand and two *cis*-coordinated solvent ligands.[15] Treatment of cyclooctene with 0.7 mM **1** and 10 equiv of H_2O_2 affords 4.9 turnover numbers (TN) of *cis*-cyclooctane-1,2-diol (Table 1, entry 1), a result unaffected by

Table 1. Catalysis of olefin oxidation by mononuclear iron complexes in combination with H_2O_2.[a]

Entry	Ligand[b]	Labile sites	Substrate	H_2O_2	*cis*-Diol[c]	Epoxide[c]
1	6-Me_3-tpa	two *cis*	cyclooctene	10	4.9(6)	0.7(2)
2	6-Me_3-tpa	two *cis*		20	10(2)	0.8(1)
3	6-Me_3-tpa	two *cis*		40	22(1)	1.6(1)
4	6-Me_3-tpa	two *cis*	*cis*-2-hexene[d]	10	5.2(6)	0.3(1)
5	6-Me_3-tpa	two *cis*	*trans*-2-hexene[d]	10	4.0(7)	0.3(1)
6	tpa	two *cis*	cyclooctene	10	2.6(3)	2.3(2)
7	N4py	one	cyclooctene	10	0	0.6(2)
8	bph	two *trans*	cyclooctene	10	0	2.5(2)
9	cyclam[10]	two *trans*	cyclohexene	50	0	20
10	tmp[e][11]	two *trans*	cyclooctene	200	0	4
11	F_{20}tpp[e][11]	two *trans*	cyclooctene	200	0	172

[a] In entries 1–8, 0.7 mM of iron complex was used with a ratio of iron catalyst to substrate of 1:1000. [b] See Figure 1. [c] Turnover numbers (TN, moles of product per mole of catalyst) reported here are based on the average of at least three runs. [d] Less than 0.1 TN of *trans*-diol was detected. [e] F_{20}tpp = *meso*-tetrakis(pentafluorophenyl)porphinato dianion, tmp = tetramesitylporphinato dianion.

the presence of O_2. The *cis*-diol product was unequivocally identified by its characteristic GC retention time and ^{1}H NMR spectrum, which are readily distinguished from those of the *trans* isomer.[16] The iron catalyst is robust, as the 50% conversion efficiency into the *cis*-diol is maintained with additional aliquots of H_2O_2 (entries 2 and 3). Complex **1** also catalyzes the oxidation of *cis*- and *trans*-2-hexene to the corresponding *cis*-diol products with formation of only traces of the *trans* isomers (entries 4 and 5). The fact that neither cyclooctene oxide nor *cis*-2-hexene oxide is transformed into diol products under these reaction conditions suggests that epoxides are not the precursors of the *cis*-diols. The reaction with **1** as catalyst is thus quite remarkable, as the combination of Fe^{II}/H_2O_2 often generates HO$^\bullet$ and gives rise to non-stereospecific oxidation of substrates.[17] There are only a few non-heme iron catalysts that are capable of stereospecific alkene epoxidation in combination with O_2 or H_2O_2,[10, 13, 14] but **1** is the first catalyst for *cis*-dihydroxylation of alkenes.

Labeling experiments with ^{18}O in the *cis*-dihydroxylation of olefins by **1**/H_2O_2 further show the similarity of the reactions catalyzed by the model and the enzymes. When the oxidation of cyclooctene is carried out in air with 10 equiv of $H_2{}^{18}O_2$ and 1000 equiv of $H_2{}^{16}O$, 95(1)% of the *cis*-diol is doubly labeled and 4(1)% is singly labeled. The complementary experiment in the presence of 10 equiv of $H_2{}^{16}O_2$ and 1000 equiv of $H_2{}^{18}O$ leads to only 1(1)% incorporation of a single ^{18}O label into

[*] Prof. L. Que, Jr., K. Chen
Department of Chemistry and Center for Metals in Biocatalysis
University of Minnesota
Minneapolis, MN 55455 (USA)
Fax: (+1) 612-624-7029
E-mail: que@chem.umn.edu

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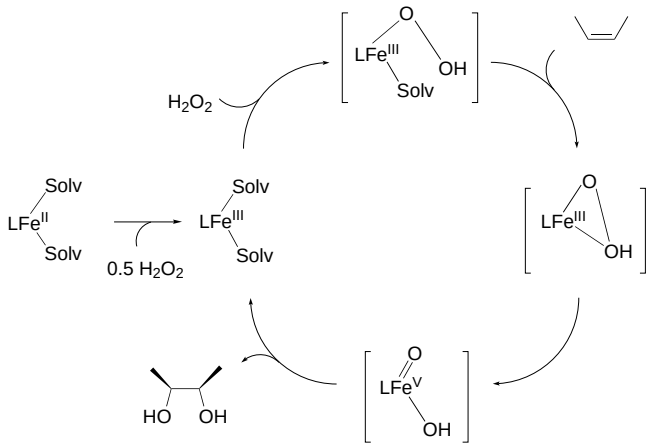
the *cis*-diol and no doubly labeled product. Hence, the oxygen atoms in the *cis*-diol product are derived exclusively from H_2O_2, not from H_2O or O_2. Furthermore, when a mixture of 3.8 equiv of $H_2{}^{18}O_2$ and 6.2 equiv of $H_2{}^{16}O_2$ is used, the resulting *cis*-diol is 40(1)% doubly labeled and 1(1)% singly labeled, and this excludes the possibility that the two oxygen atoms of the *cis*-diol product are derived from two different molecules of H_2O_2. Therefore, like Rieske dioxygenases,[2, 4] **1** catalyzes *cis*-dihydroxylation of olefins with incorporation of both oxygen atoms of one molecule of the oxidant.

We propose a mechanism for the *cis*-dihydroxylation of alkenes by **1**/H_2O_2 that involves an intermediate Fe^{III}(OOH) species.[12, 14, 18] Although such an intermediate was not observed during the reaction of **1** and H_2O_2, $Fe^{III}(\eta^1$-OOH) intermediates were observed for closely related iron complexes such as $[Fe^{II}(tpa)(CH_3CN)_2](ClO_4)_2$ (**2**) and $[Fe^{II}(N4\text{-}py)(CH_3CN)](ClO_4)_2$ (**3**, Figure 1).[12, 14, 15, 18] Interestingly, the reactivities of **2** and **3** towards cyclooctene differ in spite of the similar spectroscopic properties of the Fe^{III}(OOH) intermediates.[12, 14, 18] With 10 equiv of H_2O_2, **2** gives rise to 2.6 TN of *cis*-diol and 2.3 TN of epoxide, while **3** yields no *cis*-diol and only 0.6 TN of epoxide (Table 1, entries 6 and 7, respectively).[19] The inability of **3** to catalyze *cis*-dihydroxylation of alkenes in the presence of H_2O_2 suggests that the ability to form an $Fe^{III}(\eta^1$-OOH) intermediate alone is insufficient to elicit *cis*-dihydroxylation activity.[18] We note that both **1** and **2** contain two *cis* solvent-occupied sites, a feature absent in **3**, which contains a pentadentate ligand.[12, 15] Thus, the two labile sites in *cis* postions may play an important role in *cis*-dihydroxylation of alkenes by **1** and **2**. This hypothesis is supported by the reactivity of another related iron complex, namely, $[Fe^{II}(bph)(CH_3CN)_2](ClO_4)_2$ (**4**, Figure 1), which catalyzes only alkene epoxidation (Table 1, entry 8). Complex **4** has two labile sites *trans* to one another because the tetradentate ligand bph occupies four equatorial sites of the metal center. Similarly configured catalysts such as $[Fe^{II}(cyclam)(CH_3CN)_2](O_3SCF_3)_2$ (Figure 1) and iron porphyrins also exhibit the same reactivity pattern (entries 9–11).[10, 11] Hence, two labile sites in *cis* positions on the iron center are required for *cis*-dihydroxylation activity of the iron catalyst in combination with H_2O_2.[20]

Why are two labile sites in *cis* positions required for *cis* dihydroxylation of alkenes? An attractive hypothesis is the involvement of an η^2-peroxo intermediate, which can only be derived from an iron complex with two labile sites *cis* to one another (Scheme 1). The few known $Fe^{III}(\eta^2\text{-}O_2{}^{2-})$ species are unreactive as electrophiles[21, 22] and are said to be activated by protonation.[21, 23] We thus suggest the participation of an $Fe^{III}(\eta^2$-OOH) intermediate in the reaction of **1** with H_2O_2. This intermediate could attack the alkene directly or via a transient high-valent oxo iron species that resembles the *cis*-dioxo metal moieties of OsO_4, $MnO_4{}^-$, and RuO_4 that effect the *cis*-dihydroxylation of alkenes.[9] The two C–O bonds of the *cis*-diol could then be formed concertedly, so that both oxygen atoms of H_2O_2 are incorporated into the product. Further studies are in progress to test this hypothesis.

In summary, we have discovered the first example of the *cis*-dihydroxylation of olefin by an iron catalyst **1** in combination with H_2O_2. Both oxygen atoms of the oxidant are incorporated into the *cis*-diol product, so **1** serves as an excellent functional model for Rieske dioxygenases. Our studies show that two labile sites in *cis* positions are required for the reaction, consistent with the active-site geometry of naphthalene 1,2-dioxygenase (Figure 1).[5] We propose that these two sites activate H_2O_2 via an $Fe^{III}(\eta^2$-OOH) intermediate. The similarities between the synthetic catalyst and the enzymes strengthen the hypothesis that an Fe^{III} peroxo species is involved in the *cis*-dihydroxylation reactions of Rieske dioxygenases.[2, 7, 8]



Scheme 1. Proposed mechanism for *cis*-dihydroxylation of alkenes by **1**/H_2O_2.

Experimental Section

All reagents were purchased from Aldrich and used as received unless noted otherwise. $H_2{}^{18}O$ (96.5% ^{18}O-enriched) and $H_2{}^{18}O_2$ (90% ^{18}O-enriched, 2% solution in $H_2{}^{16}O$) were obtained from ICON. CH_3CN was pretreated by refluxing over CaH_2. All substrates were purified by distillation. In a typical reaction, 0.3 mL of a 70 mM H_2O_2 solution (21 μmol, diluted from 35% H_2O_2 solution in H_2O) in CH_3CN was delivered by syringe pump over 30 min at 25 °C under air to a vigorously stirred solution in CH_3CN (2.7 mL) containing 0.7 mM **1** (2.1 μmol) and 0.7 M cyclooctene (2.1 mmol). The reaction was quenched by addition of 0.1 mL of 1-methylimidazole and 1 mL of acetic anhydride to esterify the diol product[24] for GC (AT-1701, FID) or GC/CI-MS analysis (HP 5898 with DB-5, Finnigan MAT 95 mass detector, NH_3 as ionization gas).

Caution: Complexes with organic ligands and perchlorate anions are potentially explosive.

bph: An aqueous solution of NaOH (0.400 g, 10.0 mmol, 5 mL) was added dropwise to an aqueous solution of picolyl chloride hydrochloride (0.823 g, 4.92 mmol, 8 mL) at 0 °C. To this mixture was added an aqueous solution of homopiperazine (hpz, 0.251 g, 2.46 mmol, 5 mL) over 15 min, and the mixture was stirred for 3 d. The reaction mixture was extracted with $CHCl_3$ (4×20 mL), and the organic layer was washed (satd $NaHCO_3$, 2×20 mL) and dried (Na_2SO_4). Removal of the solvent gave a yellow oil (83% yield). ^{1}H NMR (300 MHz, $CDCl_3$, 25 °C): $\delta = 8.53$ (2H, α-py), 7.65, 7.14 (4H, β-py), 7.48 (2H, γ-py), 3.82 (4H, CH_2-py), 2.81 (4H, 5-, 7-CH_2 of hpz), 2.77 (4H, 2-, 3-CH_2 of hpz), 1.84 (2H, 6-CH_2 of hpz).

3: Equimolar amounts of BPH and $Fe(ClO_4)_2 \cdot 6H_2O$ were mixed in CH_3CN under Ar. The complex was precipitated by vapor diffusion of diethyl ether into the clear red solution. ^{1}H NMR (300 MHz, CD_3CN, 25 °C): $\delta = 244.5$ (2H, $T_1 = 2.9$ ms, α-py), 168.6, 114.1 (4H, $T_1 = 2.8$, 1.7 ms; 2-, 3-CH_2 of hpz), 102.2 (4H, $T_1 = 1.2$ ms; 5-, 7-CH_2 of hpz), 89.5, 17.2 (4H, $T_1 = 0.7$, 0.8 ms; CH_2-py), 54.7, 43.3 (4H, $T_1 = 14.5$, 17.0 ms; β-py), 0.2 (2H, $T_1 = 34.4$ ms, γ-py), −27.4, −34.2 (2H, $T_1 = 5.1$, 4.5 ms; 6-CH_2 of hpz).

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[1] D. T. Gibson, V. Subramanian in *Microbial Degradation of Aromatic Hydrocarbons* (Ed.: D. T. Gibson), Marcel Dekker, New York, **1984**, pp. 181 – 251.

[2] P. Wende, F.-H. Bernhardt, K. Pfleger, *Eur. J. Biochem.* **1989**, *181*, 189 – 197.

[3] K. Lee, D. T. Gibson, *J. Bacteriol.* **1996**, *178*, 3353 – 3356.

[4] a) S. Beil, B. Happe, K. N. Timmis, D. H. Pieper, *Eur. J. Biochem.* **1997**, *247*, 190 – 199; b) C. C. Lange, L. P. Wackett, *J. Bacteriol.* **1997**, *179*, 3858 – 3865.

[5] B. Kauppi, K. Lee, E. Carredano, R. E. Parales, D. T. Gibson, H. Eklund, S. Ramaswamy, *Structure* **1998**, *6*, 571 – 586.

[6] P. R. Ortiz de Montellano, *Cytochrome P-450 Structure, Mechanism and Biochemistry*, Plenum, New York, **1995**.

[7] D. Ballou, C. Batie, *Oxidases and Related Redox Systems*, Alan R. Liss, New York, **1988**, pp. 211 – 226.

[8] L. Que, Jr., R. Y. N. Ho, *Chem. Rev.* **1996**, *96*, 2607 – 2624.

[9] a) M. Schröder, *Chem. Rev.* **1980**, *80*, 187 – 213; b) K. B. Wiberg, *Oxidation in Organic Chemistry, Vol. 5-A*, Academic Press, New York, **1965**, pp. 1 – 68; c) T. K. M. Shing, E. K. W. Tam, V. W.-F. Tai, I. H. F. Chung, Q. Jiang, *Chem. Eur. J.* **1996**, *2*, 50 – 57.

[10] W. Nam, R. Y. N. Ho, J. S. Valentine, *J. Am. Chem. Soc.* **1991**, *113*, 7052 – 7054.

[11] T. G. Traylor, S. Tsuchiya, Y.-S. Byun, C. Kim, *J. Am. Chem. Soc.* **1993**, *115*, 2775 – 2781.

[12] M. Lubben, A. Meetsma, E. C. Wilkinson, B. Feringa, L. Que, Jr., *Angew. Chem.* **1995**, *107*, 1610 – 1612; *Angew. Chem. Int. Ed. Engl.* **1995**, *34*, 1512 – 1514.

[13] a) A. Suga, T. Sugiyama, M. Otsuka, M. Ohno, *Tetrahedron* **1991**, *47*, 1191 – 1204; b) R. J. Guajardo, S. E. Hudson, S. J. Brown, P. K. Mascharak, *J. Am. Chem. Soc.* **1993**, *115*, 7971 – 7977; c) E. H. Ha, R. Y. N. Ho, J. F. Kisiel, J. S. Valentine, *Inorg. Chem.* **1995**, *34*, 2265 – 2266.

[14] C. Kim, K. Chen, J. Kim, L. Que, Jr., *J. Am. Chem. Soc.* **1997**, *119*, 5964 – 5965.

[15] Y. Zang, J. Kim, Y. Dong, E. C. Wilkinson, E. H. Appelman, L. Que, Jr., *J. Am. Chem. Soc.* **1997**, *119*, 4197 – 4205.

[16] *cis*-Cyclooctane-1,2-diol has a ^{1}H chemical shift of $\delta = 3.88$ for the C*H*OH protons versus $\delta = 3.57$ for the *trans* isomer.

[17] C. Walling, *Acc. Chem. Res.* **1975**, *8*, 125 – 131.

[18] R. Y. N. Ho, G. Roelfes, B. L. Feringa, L. Que, Jr., *J. Am. Chem. Soc.* **1999**, *121*, 264 – 265.

[19] The *cis*-diol product was overlooked in the previous experiments on olefin oxidation by **2**/H_2O_2 due to its adsorption onto the silica gel used to remove the iron catalyst from the organic product in the original workup procedure.[14]

[20] In contrast, alkene epoxidation does not require an iron catalyst with two labile sites in *cis* positions (see Table 1). Detailed studies on the mechanism of alkene epoxidation are in progress.

[21] F. Neese, E. I. Solomon, *J. Am. Chem. Soc.* **1998**, *120*, 12829 – 12848.

[22] M. Selke, M. F. Sisemore, J. S. Valentine, *J. Am. Chem. Soc.* **1996**, *118*, 2008 – 2012.

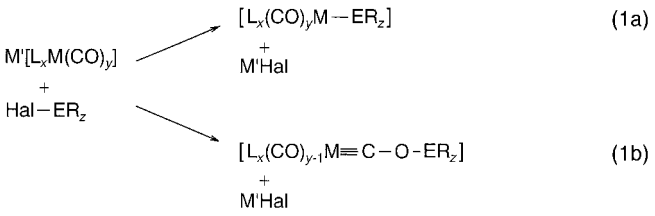
[23] D. L. Harris, G. H. Loew, *J. Am. Chem. Soc.* **1998**, *120*, 8941 – 8948.

[24] L. E. Elvebak, II, T. Schmitt, G. R. Gray, *Carbohydr. Res.* **1993**, *246*, 1 – 11.

Unique Rearrangement of an Oxycarbyne Complex: Synthesis and Structure of Novel Diborane(4)yl Complexes**

Holger Braunschweig,* Margot Koster, and Karl W. Klinkhammer

The salt elimination reaction between mononuclear anionic transition metal carbonyl complexes $M'[L_xM(CO)_y]$ and main group element halides $HalER_z$ is a fundamental reaction in transition metal chemistry, and has had a pivotal role in establishing complexes $[L_x(CO)_yM{-}ER_z]$ with bonds between main group elements and transition metals [Eq. (1a)].[1a,b] The general formation of M–E bonds suggests that the transition metal acts as the nucleophilic center in these reactions.[1b] There is, however, spectroscopic and experimental evidence that the carbonyl oxygen atom also displays some nucleophilic character in anionic complexes $M'[L_xM(CO)_y]$, especially towards hard and bulky Lewis acids.[2a–d] The addition of the carbonyl oxygen atom to the element E with salt elimination [Eq. (1b)] is expected to lead to the formation of transiton metal oxycarbyne complexes of the type $[L_x(CO)_{y-1}M{\equiv}C{-}O{-}ER_z]$; this alternative pathway to the common formation of $[L_x(CO)_yM{-}ER_z]$ [Eq. (1a)], however, has only been observed in one example.[3]



(1a)

(1b)

Over the last six years, salt elimination reactions have been very successfully employed in the synthesis of transition metal complexes of boron, especially for boryl and borylene complexes.[4a,b] Recently, we described the synthesis and characterization of the first diborane(4)yl complexes $[(\eta^5\text{-}C_5H_5)(CO)_nM\{B(NMe_2)B(NMe_2)Hal\}]$ ($M = Fe$, Ru, $n = 2$; $M = Mo$, W, $n = 3$; $Hal = Cl$, Br), which were obtained by this method from reactions of the corresponding anionic transition metal complexes and $B_2(NMe_2)_2Hal_2$.[5a,b]

In contrast to the known reactivity of $K[(\eta^5\text{-}C_5H_5)M(CO)_3]$ ($M = Mo$, W) towards 1,2-dibromo- and 1,2-dichlorodiboranes(4), the corresponding reactions with $B_2(NMe_2)_2I_2$ give the dinuclear oxycarbyne complexes **1a**, **b** (Scheme 1). These products were formed by a nucleophilic attack of a CO oxygen atom on each boron center with elimination of two equivalents of KI. Both products, which were isolated as

[*] Dr. H. Braunschweig, M. Koster
Institut für Anorganische Chemie der Technischen Hochschule
D-52056 Aachen (Germany)
Fax: (+49) 241-8888288
E-mail: holger.braunschweig@ac.rwth-aachen.de
Dr. K. W. Klinkhammer
Institut für Anorganische Chemie der Universität Stuttgart
Pfaffenwaldring 55, D-70569 Stuttgart (Germany)

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