cis-Dihydroxylation of Alkenes by a Non-Heme Iron Enzyme Mimic

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Dedicated to Professor Sir Jack Baldwin, on the occasion of his 70th birthday

Abstract: Using the non-heme iron oxidase active site as a template, a peptidomimetic ligand has been designed, synthesized, and used to effect the dihydroxylation of alkene substrates. Fenton-type radical pathways are also observed.

Key words: alkenes, bioorganic chemistry, dihydroxylations, iron, oxygenations

Nature uses a diverse range of iron-based enzyme systems to effect the oxidation of hydrocarbon substrates with remarkable efficiency and exquisite control.[1–3] These include the porphyrin-based cytochrome P450s,[1] the soluble di-iron mono-oxygenases[3] and non-heme iron oxidases[2] such as naphthalene dioxygenase,[4] and isopenicillin N synthase.[5] These biological systems have served as the starting point for wide-ranging inquiry in search of biomimetic iron-based oxidizing systems for application in organic synthesis.[6–9] Amongst the most noteworthy are the Gif systems pioneered by Sir Derek Barton,[6] terpyridylamine-based dihydroxylating agents and other systems developed by Que et al.,[7,9,10] and the bis(pyridinyl/pyrrolidinyl) systems applied by the White group to execute an impressive range of selective C–H activation chemistry.[11] As part of our endeavor to develop simple amino acid based systems as the basis of iron-centered catalysts for hydrocarbon oxidation,[12] we report here the synthesis and application of a tetradentate ligand based on pyridine-2,6-dimethanol and (*S*)-mandelic acid that combines with iron(II) acetate to effect *cis*-dihydroxylation of alkene substrates.

The active site of non-heme iron oxidase enzymes (NHIOs) is highly conserved and contains an iron(II) center coordinated by two histidine residues and an aspartate or glutamate residue (referred to as the '2-His-1-carboxylic acid' facial triad)[13] and one or more water ligands (Figure 1).

We envisaged a biomimetic complex **2**, formed between iron(II) and the peptidomimetic ligand **3** to mimic the structure and function of the NHIO active site. The ligand **3** is tetradentate and incorporates two oxygen and two nitrogen donors to replicate the iron-binding environment created by the 2-His-1-carboxylate triad and one water molecule.

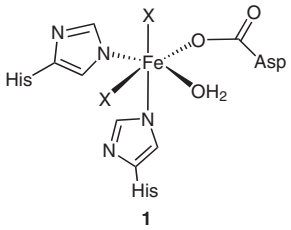
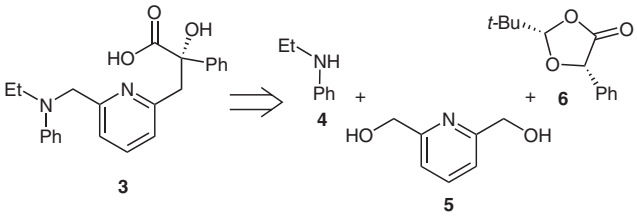


Figure 1 The generalized active-site environment of non-heme iron oxidase enzymes **1**, incorporating the '2-His-1-carboxylate' facial triad of protein-derived ligands, and two vacant sites for substrate and/or oxygen binding; and the small-molecule iron(II) complex **2** designed to mimic the NHIO active site in structure and function



Scheme 1 The tetradentate ligand **3** required to generate complex **2**, and its retrosynthesis to *N*-ethylaniline **4**, pyridine-2,6-dimethanol **5**, and the Seebach dioxolonone **6**

The complex **2** is designed to incorporate two vacant *cis* sites at the iron center for dioxygen/peroxide binding: Que has proposed that the presence of two vacant sites *cis* to each other is an absolute requirement for small-molecule iron complexes to effect *cis*-dihydroxylation using H_2O_2 as the oxidant.[7] Pyridine and tertiary amine nitrogen atoms model the histidine ligands, while mandelic acid mimics both the carboxylate and a water ligand. This design incorporates two aromatic groups to increase steric density around the metal and counter the propensity of iron complexes to give bridged products via competing intermolecular oxidation reactions [e.g., Fe(III)–O–Fe(III) species].

Our retrosynthesis of **3** (Scheme 1) leads to *N*-ethylaniline **4**, pyridine-2,6-dimethanol **5**, and the dioxolonone **6** [derived from (*S*)-mandelic acid and pivaldehyde as reported by Seebach].[14] Ligand **3** was prepared from these starting materials in seven steps and an overall yield of 27% (Scheme 2).[12,15]

Thus diol **5** was first monoprotected using sodium hydride and *tert*-butyldimethylsilyl chloride in moderate yield (57%). The unprotected alcohol was brominated in excellent yield (94%) using carbon tetrabromide and triphe-

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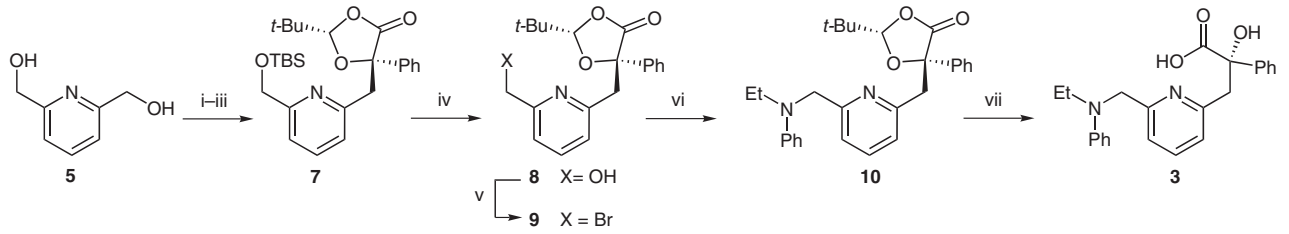
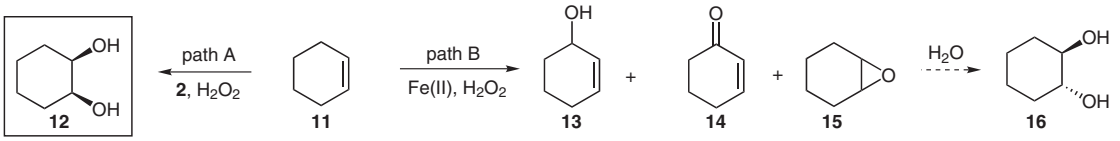


Scheme 2 Synthesis of ligand **3**. Reaction conditions: i. NaH, TBSCl, CH_2Cl_2, r.t., 5 h, 57%; ii. CBr_4, PPh_3, CH_2Cl_2, r.t., 2 h, 94%; iii. LDA-**6** (premixed), THF, –78 °C, 6 h, 91%; iv. TBAF, THF, r.t., 90 min, 88%; v. CBr_4, PPh_3, CH_2Cl_2, r.t., 2 h, 77%; vi. EtNHPh, *n*-BuLi, DMPU (premixed), THF, –40 °C, 5 h, 85%; vii 1 M LiOH, THF, reflux, 17 h then 1 M HCl, 95%.[15]



Scheme 3 Potential products from the reaction of cyclohexene **11** under the turnover conditions used: the desired *cis*-dihydroxylation reaction mediated by the iron complex **2** (path A), and competing Fenton-type reactivity via the allylic radical, to the alcohol **13**, ketone **14**, and epoxide **15** products (path B); the *trans*-diol product **16** could arise from subsequent hydrolysis of **15**

nylphosphine, then the newly established bromide was displaced using the lithium enolate of **6**.[14] Removal of the silyl ether and bromination of the unmasked alcohol both proceeded smoothly (88% and 77%) to give **9**. Nucleophilic attack on **9** by the lithium amide derived from *N*-ethylaniline **4** introduced the second nitrogen, and alkaline hydrolysis to open the dioxolonone revealed the target ligand **3** in excellent conversion (95%). The complex **2** was prepared in situ by mixing equimolar amounts of ligand **3** (as its sodium salt) and iron(II) acetate in methanol, then used directly in oxidation reactions with five aliphatic alkenes using hydrogen peroxide as the oxidant (Table 1).[16] Diol products were observed for all five of these substrates and epoxides for four of them. In all cases yields of diol are greater than of epoxide. In control experiments carried out in the absence of ligand **3** no diol products were observed. Thus ligand **3** and iron(II) acetate combine to mediate an alkene dihydroxylation reaction not observed without the ligand: using a 'two-nitrogen-two-oxygen' peptidomimetic ligand to mimic the NHIO active site we have effected alkene *cis*-dihydroxylation.

However, the yields and turnover numbers (TON) of diol products are generally low, and competing pathways are evidently in operation. Further investigation into the reaction of cyclohexene **11** revealed that as well as the desired *cis*-diol **12**, significant amounts of the allylic oxidation products **13** and **14** are also formed (Scheme 3). This product profile evinces competing reactions via Fenton-type pathways,[17] by which iron(II) and hydrogen peroxide combine to unleash hydroxyl radicals that effect allylic C–H abstraction and give rise to products **13** and **14**. These radical processes may also give rise to the epoxide product **15** and thence the *trans*-diol **16** via epoxide opening.

Thus while we have observed iron-mediated dihydroxylation of alkene substrates using a biomimetic ligand, yields are generally low and turnover numbers show that

Table 1 Turnover Products from Reaction of Alkene Substrates with **3**, $Fe(OAc)_2$, and H_2O_2 in Methanol

Substrate	Epoxide[a]		*cis*-Diol[a]		*trans*-Diol[a]		Diol/epoxide
	μmol	TON[b]	μmol	TON[b]	μmol	TON[b]	Ratio[c]
Cyclopentene	n.d.[d,e]		0.78	0.08	n.d.[d]		n.a.
Cyclohexene	0.10	0.010	0.13	0.01	0.13	0.01	1.3:1
Cyclooctene	0.18	0.019	0.21	0.02	n.d.[d]		1.2:1
1-Heptene	0.01	0.001	1.48	0.15	n.a.		148:1
1-Octene	0.08	0.008	0.92	0.10	n.a.		11.5:1

[a] Results are the average of at least two runs. Yields were determined by gas chromatography, using the single point internal standard method.
[b] Turnover number (TON) represents the amount of product (μmol) per amount Fe complex (μmol). Actual yields may be higher as complex was synthesized in situ and used immediately.
[c] Ratio *cis*-diol/epoxide.
[d] n.d. = none detected.
[e] The epoxide product was not detected for the reaction of cyclopentene, however, it is possible that this product was formed in the reaction but lost during workup (cyclopentene oxide, bp 102 °C).

the reaction is far from catalytic. This is due primarily to competing oxidative chemistry mediated by hydroxyl radicals, which diverts a significant amount of the oxidizing power and gives rise to C–H abstraction and allylic oxidation. Hydroxyl radical intermediates are generated from the hydrogen peroxide oxidant either by reaction with uncomplexed iron(II) in solution (the traditional Fenton reaction) or by reaction with an iron-ligand species on a 'Fenton-type' path. Either way, the ligand **3** does not afford sufficient control over the chemistry of iron(II) and hydrogen peroxide. Work is under way to prepare improved ligand architectures which combine more effectively with iron(II) to suppress these competing Fenton pathways.

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(15) Conversion of **5** into **9** was achieved as detailed in ref. 12. Then a solution of ethylaniline **4** (0.19 mL, 1.49 mmol) in THF (10 mL) was cooled to 0 °C and *n*-BuLi (1.5 M, 1.19 mL) added via syringe. The yellow solution was stirred at 0 °C for 20 min, then DMPU (0.22 mL, 1.78 mmol) was added and the solution stirred for a further 20 min. The mixture was cooled to –40 °C and added via cannula to a solution of **9** (500 mg, 1.24 mmol) in THF (10 mL) also at –40 °C. The reaction was stirred at –40 °C for 5 h, then left to warm to r.t. overnight, The mixture was poured onto half-saturated NH_4Cl solution (4 mL), and the aqueous phase was extracted with Et_2O (3 × 8 mL). The organic extracts were combined, dried, and concentrated in vacuo. The crude product was purified by column chromatography (SiO_2, 10:1 cyclohexane–EtOAc) to give **10** as a yellow oil (470 mg, 85%).
Compound **10** (150 mg, 0.34 mmol) was dissolved in THF (0.30 mL); aq LiOH (1 M, 0.44 mL) was added. The mixture was heated at reflux for 17 h, then cooled to r.t., adjusted to pH 7 with 1 M HCl and extracted with CH_2Cl_2 (3 × 1 mL). The organic phases were combined, dried, and evaporated to a yellow oil which solidified under vacuum. Trituration with Et_2O (3 × 2 mL) gave **3** as an off-white solid (120 mg, 95%).
Characterization Data for (*S*)-3-{6-[(Ethyl-phenyl-amino)-methyl]-pyridin-2-yl}-2-hydroxy-2-phenyl-propionic Acid 3
R_f = 0.2 (CH_2Cl_2–MeOH, 10:1); $[\alpha]_D^{20}$ –92.6 ($CHCl_3$, *c* 0.38); mp 110–112 °C. IR (KBr): ν_{max} = 3355 (w, OH str), 1612 (s, C=O str), 1505 (m, C=C str), 1575 (m, C=C str), 807, 745 cm^{-1}. ^1H NMR (300 MHz, CD_3OD): δ = 1.24 (3 H, t, *J* = 7.0 Hz, NCH_2CH_3), 3.56 (2 H, q, *J* = 7.0 Hz, NCH_2CH_3), 3.64 (1 H, d, *J* = 14.5 Hz, 1 of $CH_2CC_6H_5$), 3.53 (1 H, d, *J* = 14.5 Hz, 1 of $CH_2CC_6H_5$), 4.57 (2 H, s, CH_2N), 6.65 (3 H, m, 3 NC_6H_5), 7.05 (1 H, d, *J* = 7.5 Hz, py-H_δ), 7.12–7.29 (6 H, m, 3 of CC_6H_5, 2 of NC_6H_5, py-H_β), 7.54 (1 H, t, *J* = 7.0 Hz, py-H_γ), 7.76 (2 H, m, 2 of CC_6H_5). ^{13}C NMR (75 MHz, $CDCl_3$): δ = 11.26 (NCH_2CH_3), 44.82 ($CH_2CC_6H_5$), 48.85 (NCH_2CH_3), 55.11 (CH_2N), 79.64 (CC_6H_5), 111.09 (CH of C_6H_5), 115.25 (py-C_β), 117.43 (py-C_δ), 121.94, 125.17, 125.35, 126.35, 128.15 (CH of C_6H_5), 136.61 (py-C_γ), 141.97 (C_{ipso} of CC_6H_5), 146.55 (C_{ipso} of NC_6H_5), 157.02 (py-C_α), 158.21 (py-C_ε), 176.53 (C=O). MS (ES^+): *m*/*z* (%) = 377 (100) $[MH]^+$, 359 (45) $[MH - H_2O]^+$. HRMS (ES^+): *m*/*z* calcd for $C_{23}H_{24}N_2O_3$: 377.1865; found: 377.1860 (100%) $[MH]^+$.
(16) **Turnover Procedure**
Methanol was distilled over CaH_2 and degassed in three freeze–thaw cycles before use. All substrates were distilled over CaH_2 and passed through activated alumina before use to remove any peroxides. All reactions were carried out under an atmosphere of argon. Ligand **3** (50 mg, 0.13 mmol) was dissolved in anhyd CH_2Cl_2 (0.75 mL) and treated with NaH (12 mg, 0.53 mmol). After stirring for 45 min at r.t. the solvent was removed in vacuo to give the sodium salt of **3** as a white powder. To a suspension of this salt (4.0 mg, 10 μmol) in MeOH (0.20 mL) was added $Fe(OAc)_2$ (1.7 mg, 10 μmol) in MeOH giving a yellow solution which was stirred at r.t. for 45 min. The solution was diluted with MeOH (15 mL), and the substrate alkene (10 mmol) was added. Hydrogen peroxide (100 μmol, 30% aq) diluted in MeOH (1 mL) was added to the stirring solution over 4 h, and the solution was stirred at r.t. for a further 12 h. The reaction was reduced in vacuo, diluted with EtOAc and filtered through SiO_2. *n*-Decane was added as an internal standard. Products **12**–**16** were analyzed by gas chromatography and GC-MS and identified unambiguously by comparison with authentic samples.
Gas chromatography was carried out on a Hewlett-Packard 5890 Series II gas chromatograph fitted with an HP-1ms column (30 m × 0.25 mm ID, 0.25 μm;S/N US2469051H), and (to distinguish *cis*- and *trans*-diols) a Hewlett-Packard 5890A gas chromatograph fitted with a BP-20 column (25 m × 0.22 mm ID, 0.25 μm) and ChemStation software. Both chromatographs were equipped with split/splitless capillary inlets and flame-ionisation detectors (FID).
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