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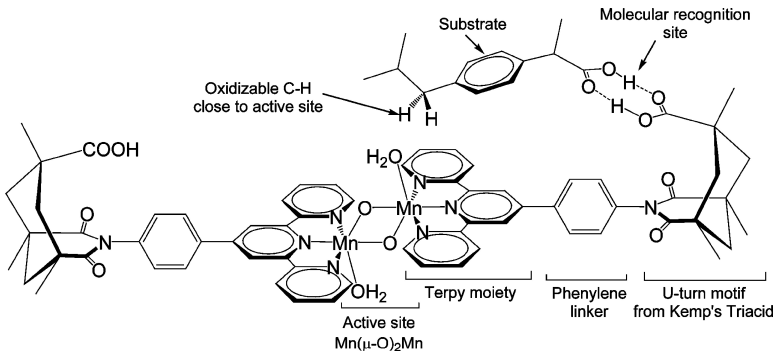

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High Turnover Remote Catalytic Oxygenation of Alkyl Groups: How Steric Exclusion of Unbound Substrate Contributes to High Molecular Recognition Selectivity

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Abstract: H-bonding mediated molecular recognition between substrate and ligand −COOH groups orients the substrate so that remote, catalyzed oxygenation of an alkyl C−H bond by a Mn-oxo active site can occur with very high (>98%) regio- and stereoselectivity. This paper identifies steric exclusion—exclusion of non H-bonded substrate molecules from the active site—as one requirement for high selectivity, along with the entropic advantage of intramolecularity. If unbound substrate molecules were able to reach the active site, they would react unselectively, degrading the observed selectivity. Both of the faces of the catalyst are blocked by two ligand molecules each with a −COOH group. The acid *p*-^{t}BuC$_6$H$_4$COOH binds to the ligand −COOH recognition site but is not oxidized and merely blocks approach of the substrate therefore acting as an effective inhibitor for ibuprofen oxidation in both free acid and ibuprofen ester form. Dixon plots show that inhibition is competitive for the free acid ibuprofen substrate, no doubt because this substrate can compete with the inhibitor for binding to the recognition site. In contrast, inhibition is uncompetitive for the ibuprofen-ester substrate, consistent with this ester substrate no longer being able to bind to the recognition site. Inhibition can be reversed with MeCOOH, an acid that can competitively bind to the recognition site but, being sterically small, no longer blocks access to the active site.

1. Introduction

Selective and controlled oxidation of alkyl groups with synthetic catalysts, especially regio- and stereoselective oxidation of saturated C−H bonds remote from an existing functional group, is a long-standing challenge.[1,2] In contrast, enzymes routinely achieve highly selective functionalization of saturated C−H bonds. These include the cytochrome P450 dependent monooxygenases that are highly specific in both alkane and alkene oxygenation in numerous biosynthetic processes.[3] The fatty acid desaturases, with their nonheme carboxylate bridged diiron center, catalyze O_2 dependent dehydrogenation of saturated fatty acyl CoA with high stereo- and regiospecificity.[4−6]

Soon after the first crystal structure of cytochrome P450 was reported, the Groves group reported the first porphyrin-based model system, in which the substrate was covalently attached to the porphyrin via an ester bond.[7] Thus, although regioselectivity was attained in oxygenation of saturated C−H bonds, no catalytic turnover could occur. Similarly, the Greico and the Sames groups reported some non-porphyrin Mn-based synthetic model complexes[8,9] where selectivity was attained by covalent attachment of the substrate but again only with single turnover chemistry.

Weak noncovalent molecular recognition forces can lead to reversible binding of substrates and are, thus, important elements in our design strategy for multiple turnovers. An enzyme anchors and orients the substrate via H-bonding, hydrophobic interactions and π-stacking. These are thought to bring the substrate reactive site close to the catalyst active site in a near attack conformer (NAC)[10,11] that facilitates reaction by controlling the approach of the substrate to the catalyst. In typical synthetic catalysts, reversible attachment of substrates via a molecular recognition functional group is a promising strategy. With the right conditions, molecular recognition should be tight enough to hold the substrate properly but still be weak enough to allow catalytic turnover.

In classic work, Breslow and co-workers reported a series of porphyrin-based Mn catalysts, where cyclodextrin groups are attached to the porphyrin ring.[12−15] Cholesterol substrates having

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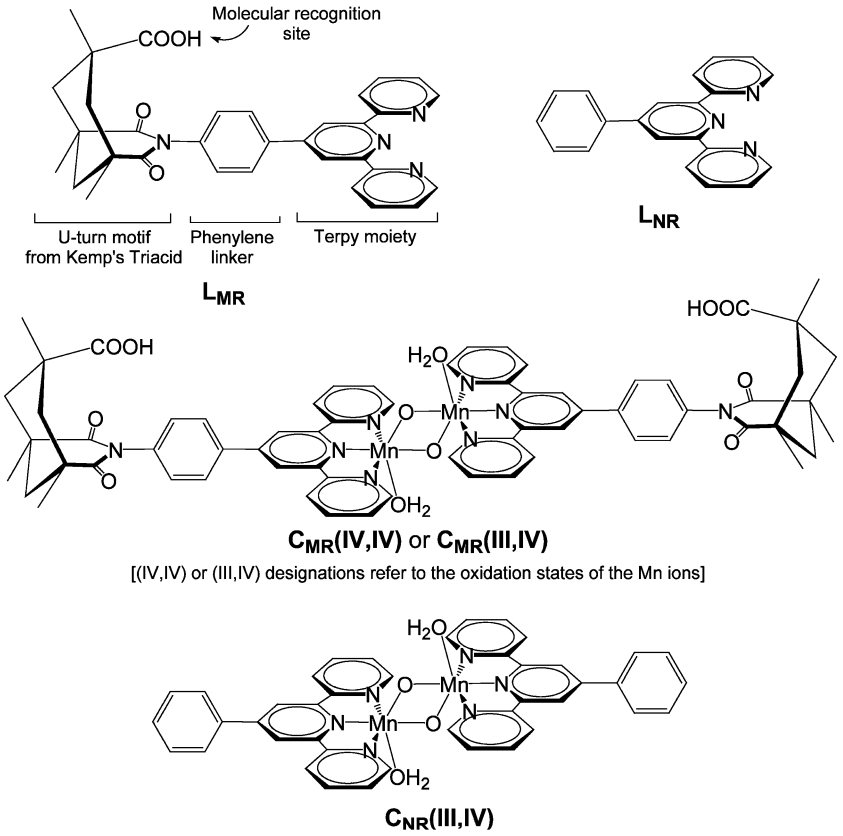
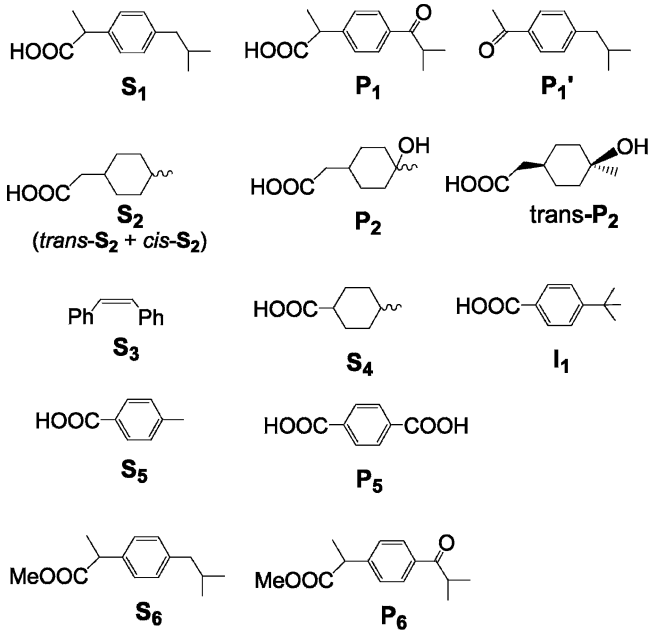
Scheme 1.



hydrophobic tails bound to the hydrophobic cyclodextrin cavity gave multiturnover oxidation at specific C−H bonds. In another classic example, Groves et al. reported a "membrane spanning" iron-porphyrin catalyst.[16] Steroid groups on the porphyrin enabled the catalyst to intercalate in a lipid bilayer to place the Fe active site at midmembrane. Remarkable regioselectivity was seen in epoxidation of several sterol substrates that were oriented by the bilayer. Suslick and co-workers saw high shape selectivity in the epoxidation of alkenes[17−20] by bis-pocket Mn porphyrin and dendrimer Mn porphyrin catalysts. None of these systems use attractive, labile molecular recognition interactions, however.

In porphyrin-based synthetic catalysts, efficiency tends to be low,[12] so we have turned to a simpler and more highly catalytic system based on a dimanganese-bis-μ-oxo terpyridine complex[21,22] with oxone (HSO_5^-) as the primary oxidant that prior studies suggest does not operate via freely diffusing radicals.[23] Our modified terpyridine ligand (**L_{MR}**) (Scheme 1), containing a molecular recognition group capable of binding carboxylic acid substrates, gives the molecular recognition (MR) catalyst,

Scheme 2.



C_{MR}(IV,IV), $[H_2O(\mathbf{L_{MR}})Mn(\mu\text{-}O)_2Mn(\mathbf{L_{MR}})H_2O](NO_3)_4$. With oxone as the primary oxidant, this catalyst gives highly regio- and stereoselective oxygenation of saturated C−H bonds with multi-hundred turnovers. Control experiments with a similar but nonrecognition (NR) catalyst, **C_{NR}(III,IV)**, $[H_2O(\mathbf{L_{NR}})Mn(\mu\text{-}O)_2Mn(\mathbf{L_{NR}})H_2O](ClO_4)_3$ (**L_{NR}**: 4′-phenyl-2,2′:6′,2″-terpyridine), which lacks the key −COOH group, do not show any selectivity.[22] The (IV,IV) or (III,IV) designations refer to the oxidation states of the Mn ions in the catalyst precursor. The

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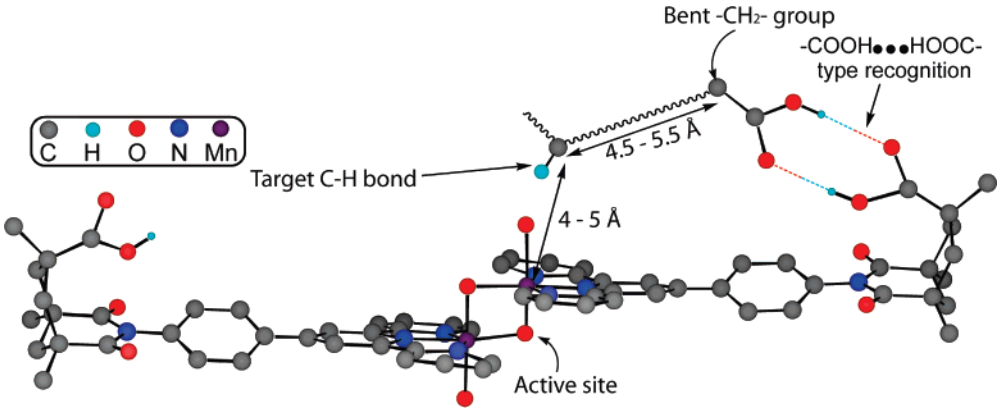


Figure 1. Molecular model (Chem 3D) of catalyst $\mathbf{C_{MR}(IV,IV)}$ docked to a cartoon substrate. Due to the ~32° angle between the KTA −COOH group and the plane containing the imide group, the substrate needs a bent $-CH_2-$ group after the −COOH group for the rest of the substrate to come into proximity with the active site (ideally 4−5 Å between target C-atom and Mn).

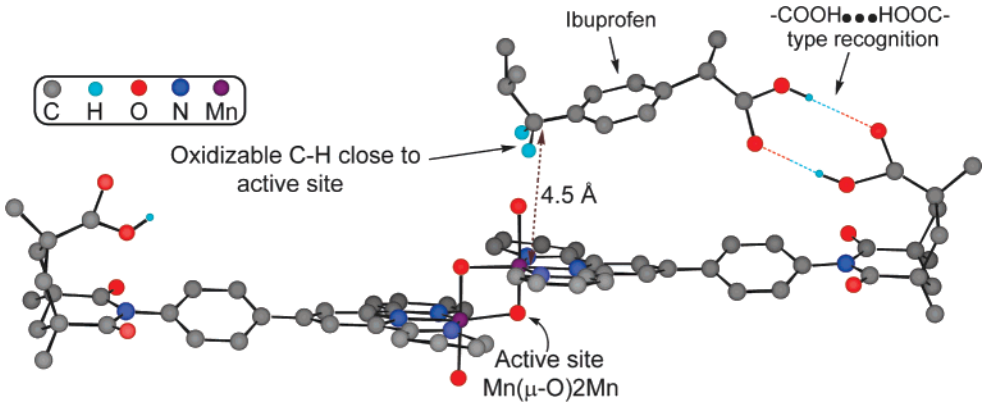


Figure 2. Molecular model (Chem 3D) of $\mathbf{S_1}$ docked to the catalyst $\mathbf{C_{MR}(IV,IV)}$ via −COOH···HOOC− type H-bonding. The distance between the target C-atom and the manganese ion is ~4.5 Å.

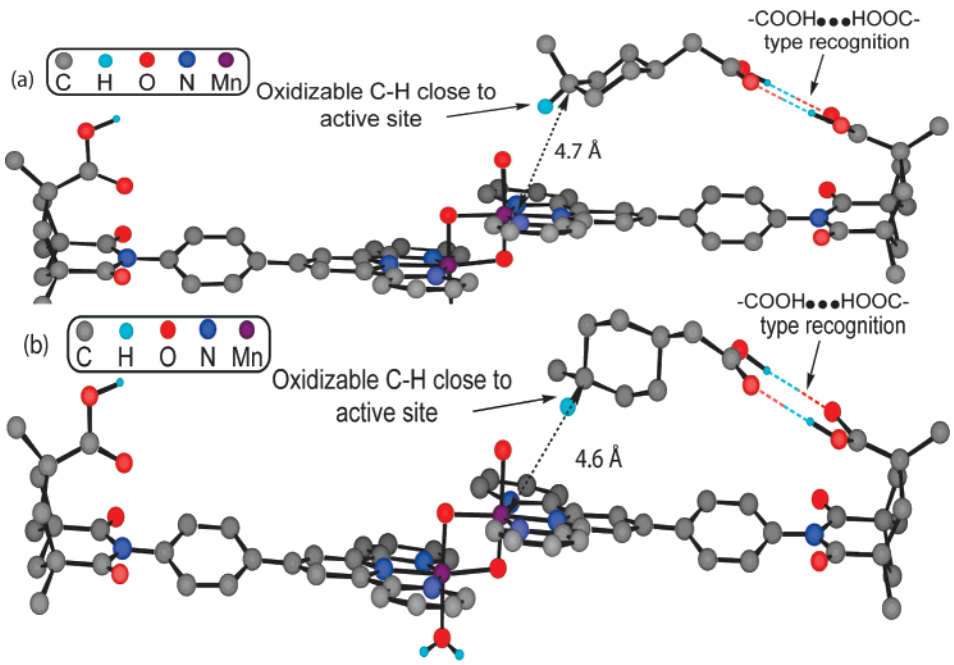
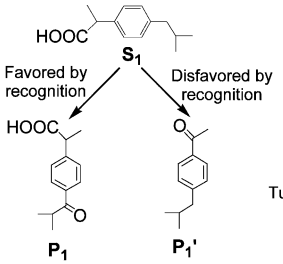


Figure 3. Molecular models (Chem 3D) of the catalyst $\mathbf{C_{MR}(IV,IV)}$ docked to (a) *cis*-$\mathbf{S_2}$ and (b) *trans*-$\mathbf{S_2}$ via H-bonding. The distance between the target C-atom and the manganese ion is 4.7 and 4.6 Å for *cis* and *trans* isomers, respectively. In both cases, the isomers are drawn in a way so that C, H, and O atoms are approximately colinear to facilitate NAC formation.

difference in oxidation state reflects differences in the ease of isolation between the MR and NR catalysts: every indication suggests they both take part in exactly the same catalytic cycle. Selectivity can also be degraded by exposing the MR system to excess MeCOOH, which successfully competes for the recognition site.[22]

Table 1. Product Distribution from Ibuprofen (S_1) Oxidation (by ^1H NMR Spectroscopy)[d]



Temperature	Catalyst	Conversion	P_1	P_1'	Total Turnovers
20°C	C_{MR}(IV,IV)	50%	97.5%	2.5%	50
	C_{NR}(III,IV)	53%	77%	23%	53
0°C	C_{MR}(IV,IV)	53%	98.5%	1.5%	53
	C_{NR}(III,IV)	54%	78%	22%	54
-20°C	C_{MR}(IV,IV)	53%	98.5%	1.5%	53
	C_{NR}(III,IV)	54%	77%	23%	54
0°C	C_{MR}(IV,IV)[a]	58%	98.5%	1.5%	580
20°C	C_{MR}(IV,IV)[a,b]	71%	96.5%	3.5%	710
20°C	C_{MR}(IV,IV)[a,c]	68%	98%	2%	680

[a] Subtrate/catalyst/oxidant = 100:0.1:500. [b]Solvent/CD_3CN. [c]Solvent/CH_3NO_2. [d]Total Turnovers = mol of products per mol of catalyst; oxidant: TBAO; subtrate/catalyst/oxidant = 100:1:500, solvent: CH_3CN, if not mentioned otherwise.

In this paper, we show how the substrate that is bound to the recognition group blocks the reactive site and prevents unbound substrate from reacting unselectively. This steric exclusion effect is a contributing feature in obtaining molecular recognition-induced selectivity. For this work, we have been able to isolate the mixed-valent Mn(III,IV) di-μ-oxo dimanganese complex (**C_{MR}(III,IV)**) derived from **L_{MR}**. The complex, **C_{MR}(III,IV)**, found to be more soluble than **C_{MR}(IV,IV),** gives essentially the same selectivity and activity suggesting that the active species is the same. With **C_{MR}(III,IV)**, we have been able to select a suitable reversible inhibitor, *p-tert*-butylbenzoic acid (**I_1**), that is not itself oxidized but almost entirely inhibits the catalyst by binding to the molecular recognition site and thus blocking the active site.

Schemes 1 and 2 show the structures of all the substrates, inhibitors, and catalysts used in this study.

2. Discussion of Our Molecular Recognition Catalyst C_{MR}(IV,IV)

There is no reason to think that the usual proposed mechanism,[24−34] H atom abstraction from the substrate C−H bond followed by a fast "rebound" of the resulting metal bound OH group to form the C−OH bond of the product, does not also apply to this system.

The terpy ligand is not only more available than porphyrins but also more oxidation resistant.[35,36] Porphyrin-based catalysts without halogen or nitro substituents are oxidized after only ca. 10 turnovers even for a reactive benzylic substrate,[12,13] while our catalysts give as many as 500 turnovers without any special protective substitution.

Incorporation of the MR group is relatively straightforward. In ligand **L_{MR}** (Scheme 1), we have attached a *p*-phenylene linker and a Kemp's triacid (KTA) "U"-turn unit to the terpy ligand to provide a rigid[37] −COOH group projecting toward the active site. Ligand **L_{MR}**, thus, lacks any oxidation-prone component.

3. Synthesis of Catalysts

The ligands **L_{MR}** and **L_{NR}** and the corresponding catalysts **C_{MR}(IV,IV)** and **C_{NR}(III,IV)** were synthesized according to standard methods as described in our prior report and summarized in Schemes S1 and S2 of the Supporting Information.[22] Catalyst **C_{MR}(III,IV)** was synthesized from Mn(**L_{MR}**)Cl_2 using a limited amount of oxone.[22]

4. Standardization of Catalytic Conditions

With a −COOH···HOOC− H-bonding motif for molecular recognition, we needed an oxidation resistant solvent that is polar enough to dissolve the catalyst and promote catalysis but not so polar as to interfere with H-bonding. Acetonitrile with 0.5% water proved suitable. Acetonitrile is not completely oxidation resistant, however. When CH_3CN was replaced with CD_3CN, the turnover was found to improve significantly without affecting the selectivity, presumably due to C−D bonds being more oxidation resistant than C−H bonds. We have also used CH_3NO_2. However the catalyst was insufficiently soluble in benzonitrile or nitrobenzene. Catalytic runs were performed with 1 eq of substrate, 0.001−0.005 eq of catalyst and 5 eq of oxone in minimum amounts of CH_3CN or CH_3NO_2 as solvents.

To maintain −COOH···HOOC− H-bonding, all the carboxylic acid groups should remain protonated. The primary oxidant, tetrabutylammonium oxone, has a pH of ∼1.5 in H_2O consistent with this requirement. Deprotonation destroys the selectivity.[22]

5. Results and Discussion

5.1. Molecular Modeling. Molecular models of catalyst **C_{MR}(IV,IV)** were constructed by importing crystal structure parameters of ligand **L_{MR}**[22] and the Mn(μ-O)$_2$Mn core[35] followed by energy minimization (MM2, CAChe 5). The crystal structure of **L_{MR}** showed that the −COOH group is bent at an angle of ∼32° with respect to the plane of the imide group. In a *p*-substituted benzoic acid substrate, the remote C−H bonds would, therefore, be too far from the active site for reaction; these substrates proved to be useful inhibitors, however. This led us to incorporate a $-CH_2-$ unit next to the −COOH group

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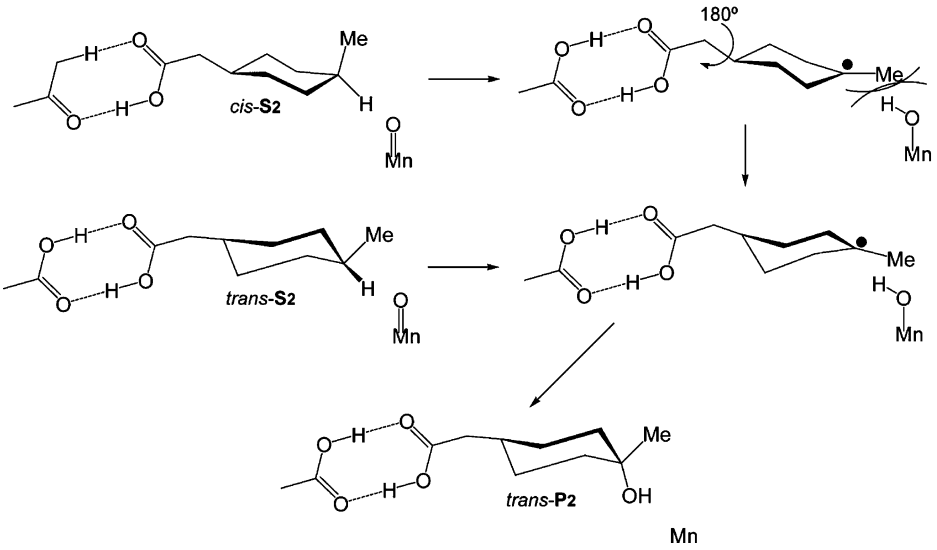
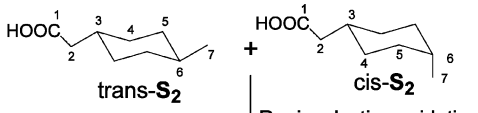


Figure 4. Both the *cis* and *trans* isomers of $\mathbf{S_2}$ give the same carbon-centered radical, which after rebound of the OH group from Mn−OH gives *trans*-$\mathbf{P_2}$. *cis*-$\mathbf{S_2}$ after H-atom abstraction gives a C-centered radical, which to avoid steric hindrance rotates 180°.

Table 2. Product Distribution from $\mathbf{S_2}$ (*cis*-$\mathbf{S_2}$ + *trans*-$\mathbf{S_2}$) (by ^{1}H NMR Spectroscopy)[c]



Temperature	Catalyst	Conversion	trans-$\mathbf{P_2}$ OH	cis-$\mathbf{P_2}$ OH	Other products	Total Turnovers
20°C	$\mathbf{C_{MR}(IV,IV)}$	13%	>99%	<1%	<1%	130
	$\mathbf{C_{NR}(III,IV)}$	19%	~30%	~30%	~40%	190
	$\mathbf{C_{MR}(IV,IV)}$[a]	18%	>99%	<1%	<1%	180
	$\mathbf{C_{MR}(IV,IV)}$[b]	19%	>99%	<1%	<1%	190

[a] Solvent: CD_3CN. [b]Solvent: CH_3NO_2. [c]Total Turnovers = mol of products per mol of catalyst; oxidant: TBAO; subtrate/catalyst/oxidant = 100:0.1:500, solvent: CH_3CN, if not mentioned otherwise.

to give the substrates a bent structure that was expected to bring the remote C−H bonds close to the active site. From crystal structures of cytochrome P450−substrate complexes, it was proposed that the carbon atom to be oxygenated needs to be within an ~4−5 Å distance from the metal.[3] To impart some rigidity to the substrate, we incorporated a benzene ring or a cyclohexyl group. Molecular modeling suggested that the C-6 or C-7 C−H bonds would then come close to the active site (Figure 1).

Ibuprofen [2-(4-isobutyl-phenyl)-propionic acid] ($\mathbf{S_1}$) and 4-methyl-cyclohexyl acetic acid ($\mathbf{S_2}$) were found to be the most suitable substrates. These molecules have relatively rigid rings, either planar C_6H_4 or chair C_6H_{10}, and have at least two alternate sites of attack. We expected selective oxidation at the remote benzylic and tertiary C−H bonds in $\mathbf{S_1}$ and $\mathbf{S_2}$, respectively, with the molecular recognition catalysts $\mathbf{C_{MR}(IV,IV)}$ and $\mathbf{C_{MR}(III,IV)}$, as shown in Figures 2 and 3.

5.2. Regioselective Oxygenation of Ibuprofen ($\mathbf{S_1}$). For ibuprofen, selective oxidation occurs at the remote benzylic CH_2 group to give ketone $\mathbf{P_1}$ with the molecular recognition catalyst (Table 1). Otherwise, a competitive oxidation pathway also occurs leading to decarboxylation of the −COOH group to give product $\mathbf{P_1'}$ (Table 1).[38] Control catalyst $\mathbf{C_{NR}(III,IV)}$, lacking molecular recognition, gave both $\mathbf{P_1}$ and $\mathbf{P_1'}$ (3:1), but with the molecular recognition catalyst $\mathbf{C_{MR}(IV,IV)}$, essentially only $\mathbf{P_1}$ is formed (~98%) (Table 1).

5.3. Regio- and Stereoselective Oxygenation of Cyclohexane Acetic Acid ($\mathbf{S_2}$). 4-Methylcyclohexane acetic acid, substrate $\mathbf{S_2}$, was employed to investigate alkyl CH hydroxylation. Our sample was a 5:4 mixture of *cis* and *trans* isomers (*cis*-$\mathbf{S_2}$ and *trans*-$\mathbf{S_2}$). Both isomers react (cis > trans), but only a single product, trans-$\mathbf{P_2}$, was formed (Table 2). Control catalysis with $\mathbf{C_{NR}(III,IV)}$ gave both isomers and many other unidentified products. The methyl ester of the product from the molecular recognition catalyst was isolated by preparative thin layer chromatography and characterized as *trans*-$\mathbf{P_2}$ based on ^{1}H

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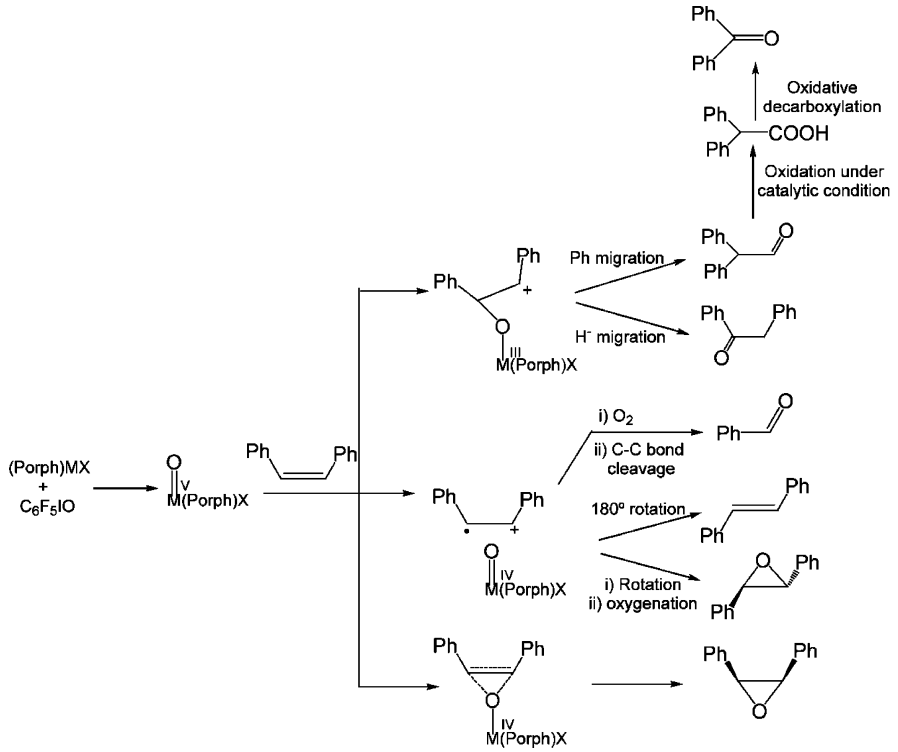

Figure 5. Possible products seen from *cis*-stilbene (**S$_3$**) oxidation with metalloporphyrins following several mechanisms.

Table 3. Mechanistic Investigations with **C$_{MR}$(IV, IV)**[b]

Substrates (eq.)	Catalyst eq.	Oxidant eq.	Catalytic conditions				Products (% of total product)	Yields
			Temp.	Time	Atmosphere	Additive (eq.)		
S$_1$ (1)	0.001	5	20°C	2 hours	N_2	NONE	**P$_1$** (98%) **P$_1$′** (2%)	61%
S$_3$	0.001	1	0°C	15 min	Air	NONE	*cis*-stilbene oxide, benzophenone, 1,2-diphenylethanone[a]	~95%
S$_1$ (1)	0.001	5	20°C	2 hours	Air	$CBrCl_3$ (5 eq.)	**P$_1$** (~98%) **P$_1$′** (~2%)	52%
S$_1$ (1)	0.001	5	20°C	2 hours	Air	NBS (5 eq.)	**P$_1$** (~98%) **P$_1$′** (~2%)	51%

[a] Based on GC-MS, quantitative analysis was not performed. [b]Yields are w.r.t. substrates; oxidant: TBAO.

NMR, ^{1}H NOE, ^{1}H COSY, ^{1}H homonuclear decoupling, and ^{13}C NMR data (Scheme 2). This suggests that both substrate isomers give a common intermediate radical[39] that has time to undergo bond rotation and accept the OH group exclusively from one side of the molecule in the "rebound" step (Figure 4).

5.4. Mechanism of C−H Bond Oxygenation. 5.4.1. Effect of Air. In C−H bond oxygenation, any long-lived carbon-centered radical intermediate typically gives radical chain products with air.[23] Our catalytic runs under air or N_2 gave the same product distribution (Table 3), so any radical intermediate is "caged" by the molecular recognition and is short-lived.

5.4.2. Epoxidation of *cis*-Stilbene. *cis*-Stilbene (**S$_3$**) is one of the substrates commonly used to investigate the radical vs cationic mechanism of oxygenation.[23,40−42] *cis*-Stilbene can give a variety of products (Figure 5).[23] With **C$_{MR}$(IV,IV)**, the major products were *cis*-stilbene oxide, benzophenone, and 1,2-diphenylethanone, but no *trans*-stilbene oxide (Table 3). These products can be explained by fast "rebound" or by a carbocation intermediate (Figure 5). The absence of any *trans*-stilbene oxide indicates the absence of any freely diffusing, long-lived carbon-radical intermediate (Table 3).

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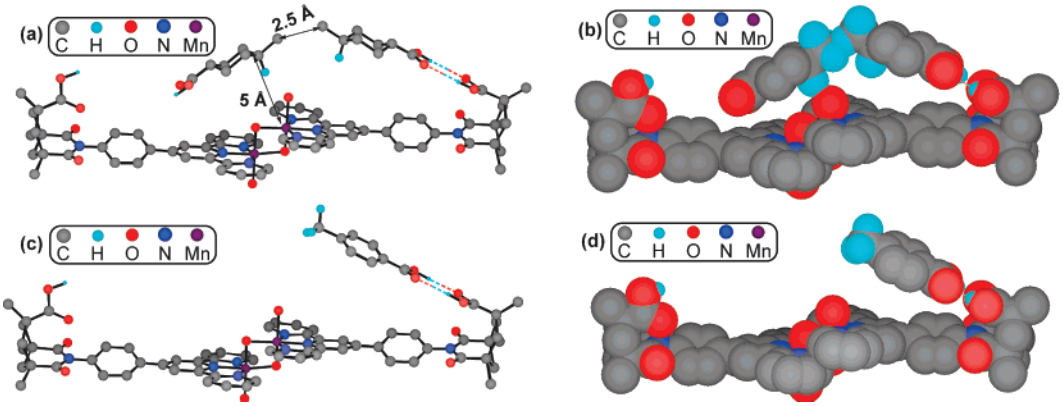

Figure 6. Molecular models (Chem 3D) of catalyst $\mathbf{C_{MR}(III,IV)}$ docked to $\mathbf{S_4}$ and $\mathbf{S_5}$. (a) Bound $\mathbf{S_4}$ blocks access of unbound $\mathbf{S_4}$. (b) The same in a space filling representation. (c) Bound $\mathbf{S_5}$ does not block the active site. (d) The same in a space filling representation.

Table 4. Investigation of Molecular Recognition Mechanism[a]

Substrates (eq.)	Catalysts (eq.)	Additives (eq.)	Temp.	Time	Products (% of total product)	Yields
$\mathbf{S_1}$ (1)	$\mathbf{C_{MR}(IV,IV)}$ (0.001)	CH_3COOH (4)	20°C	2 hours	$\mathbf{P_1}$ (75%), $\mathbf{P_1'}$ (25%)	56%
	$\mathbf{C_{NR}(III,IV)}$ (0.001)				$\mathbf{P_1}$ (77%), $\mathbf{P_1'}$ (23%)	58%
$\mathbf{S_4}$ (1)	$\mathbf{C_{MR}(III,IV)}$ (0.001)	NONE	0°C	6 hours	No Oxidation	0
	$\mathbf{C_{NR}(III,IV)}$ (0.001)				Several oxidized products	Not calculated
$\mathbf{S_5}$ (1)	$\mathbf{C_{MR}(III,IV)}$ (0.001)	NONE	0°C	2 hours	$\mathbf{P_5}$ (100%)	8%
	$\mathbf{C_{NR}(III,IV)}$ (0.001)				$\mathbf{P_5}$ (100%)	22%

[a] Yields are w.r.t. substrates; oxidant: TBAO (5 equiv of w.r.t. substrates).

5.4.3. Effect of Halogenating Agents. $CBrCl_3$ and *N*-bromosuccinimide (NBS) give rapid (ca. 10^8 L·mol^{-1} s^{-1})[43] Br atom transfer to alkyl radicals. With ibuprofen and $\mathbf{C_{MR}(IV,IV)}$, $CBrCl_3$ and NBS (0.125 M, 5 equiv vs substrate) did not yield any halogenated product (Table 3) suggesting that the half-life of the radical formed must be $\leq 10^{-7}$ s. This indicates the "rebound" happens at a rate $>10^8$ L·mol^{-1} s^{-1}.

5.5. Mechanism of Molecular Recognition. 5.5.1. H-bonding is required. The MR selectivity obtained with $\mathbf{C_{MR}(IV,IV)}$ disappears with control catalyst $\mathbf{C_{NR}(III,IV)}$ lacking the key −COOH group. With excess acetic acid (4 equiv vs substrate), the regioselectivity also disappeared (Table 4). Thus, acetic acid may disrupt the proposed recognition by binding to the ligand −COOH.

5.5.2. Inhibition of Reaction of Unbound Substrate is Required. In enzymes, there is only enough room for one substrate molecule to occupy the active site at any time. Some enzymes even undergo structural changes after binding of one substrate molecule, presumably in part to help prevent the entry of any further substrate molecules. Thus, a bound substrate can be seen as sterically excluding any other substrate molecules in the medium.

Synthetic catalysts generally do not have such a cavity, but in our case, the approach of unbound substrates must be blocked by the bound substrate in order to achieve the high regioselectivity we see. Where this is not the case, selectivity can be greatly reduced.[19]

With molecular recognition catalyst $\mathbf{C_{MR}(III,IV)}$ and 4-methylcyclohexane carboxylic acid ($\mathbf{S_4}$), no oxidation was seen. In contrast, for control catalyst $\mathbf{C_{NR}(III,IV)}$ lacking a molecular recognition function, several oxidized products were found (^{1}H NMR). A molecular model showed that the docked substrate, lacking the extra $-CH_2-$ group of the successful substrate, does not present an oxidizable C−H group to the active site (Figure 6a). Not only does oxygenation of the docked substrate not occur, but the docked substrate also prevents the approach of undocked substrate molecules. In contrast, the sterically smaller *p*-toluic acid ($\mathbf{S_5}$) yielded 1,4-benzenedicarboxylic acid ($\mathbf{P_5}$) with both molecular recognition catalyst $\mathbf{C_{MR}(III,IV)}$ and control catalyst $\mathbf{C_{NR}(III,IV)}$. A molecular model shows that when $\mathbf{S_5}$ is docked to $\mathbf{C_{MR}(III,IV)}$, its benzylic methyl group cannot approach the active site, but it also cannot prevent external substrate molecules from approaching (Figure 6c and Table 4). Substrates, $\mathbf{S_1}$ and $\mathbf{S_2}$, while docked to the catalyst $\mathbf{C_{MR}(III,IV)}$ result in significant steric bulk at the active site. These molecules, unlike $\mathbf{S_4}$, have reactive groups close to the active

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Table 5. Inhibition of Molecular Recognition Catalyst $\mathbf{C_{MR}(III,IV)}$[a]

Substrates (eq.)	Catalysts (eq.)	Inhibitors (eq.)	Temp.	Time	Products (% of total product)	Yields
$\mathbf{S_6}$ (1)	$\mathbf{C_{MR}(III,IV)}$ (0.005)	None	20°C	3 hours	$\mathbf{P_6}$ (~99%)	30%
		$\mathbf{S_4}$ (1)			$\mathbf{P_6}$ (~99%)	1.4%
		$\mathbf{I_1}$ (1)			$\mathbf{P_6}$ (~99%)	0.9%

[a] Yields are w.r.t. substrates; oxidant: TBAO (5 equiv of w.r.t. substrates).

Table 6. Inhibition and Reactivation of Molecular Recognition Catalyst $\mathbf{C_{MR}(III,IV)}$[e]

Reaction fractions	Substrate (eq.)	Catalyst (eq.)	Inhibitor (eq.)	Additive (eq.)	Temp.	Time	Products	Yield
1st [a]	$\mathbf{S_6}$ (1)	$\mathbf{C_{MR}(III,IV)}$ (0.005 eq)	$\mathbf{I_1}$ (0.3)	None	0°C	3 hours	$\mathbf{P_6}$	1.5%
2nd [b]				CH_3COOH (2.25 eq) [c]		3 hours[d]	$\mathbf{P_6}$	34%

[a] Aliquot consisting of half of the total catalytic solution was collected and worked up as the first fraction. [b]Second fraction: total catalytic solution minus first fraction. [c]Equivalents of acetic acid are w.r.t. the substrate in second fraction. [d]After first inactivation without acetic acid present (see text). [e]Yields are w.r.t. substrates; oxidant: TBAO (5 equiv of w.r.t. substrates).

site. Neither the target C−H bonds nor the other oxidation-prone C−H bonds in these molecules are terminal and unhindered.

As the substrate used in these catalyses is in large excess relative to the catalyst concentration (substrate/catalyst = 1000: 1), we propose that under the catalytic conditions all the catalytic recognition sites are saturated with docked substrates. The docked substrate thus covers the active site with a protective steric "umbrella" and makes it sterically inaccessible to unbound substrates, thereby preventing any nonregioselective catalysis. This steric exclusion of free substrates may well contribute to high regioselectivity in any systems like ours.

Although the substrates $\mathbf{S_1}$ and $\mathbf{S_2}$, while remaining bound to the MR group, could undergo bond rotations that would cause them to move far away from the active site and, thus, leave the Mn oxo site open to attack unbound substrate, this presumably does not occur because the observed regioselectivity is very high. Solvophobic and aromatic stacking forces between substrate and ligand may be responsible for this effect.

5.6. Inhibition and Reactivation of the Molecular Recognition Catalyst. 5.6.1. Inhibition. Since the steric exclusion effect of bound $\mathbf{S_4}$ prevents approach of any unbound $\mathbf{S_4}$ substrate to the active site of catalyst $\mathbf{C_{MR}(III,IV)}$, other bulky carboxylic acids should act similarly. Potential substrate $MeC_6H_{10}COOH$, $\mathbf{S_4}$, was found instead to act as an inhibitor with the methyl ester of ibuprofen ($\mathbf{S_6}$) as the substrate. $\mathbf{S_6}$, lacking any −COOH group, cannot bind to the catalyst $\mathbf{C_{MR}(III,IV)}$ but can only approach the active site by diffusion. Thus, $\mathbf{S_6}$ cannot compete with the inhibitor for binding at the molecular recognition site. In the absence of inhibitor $\mathbf{S_4}$, $\mathbf{S_6}$ is oxidized to give $\mathbf{P_6}$ as the major product with molecular recognition catalyst $\mathbf{C_{MR}(III,-IV)}$, giving ~30% yield after 3 h at 20 °C. But when $\mathbf{S_4}$ (1 equiv vs substrate) was added to the catalytic medium the yield of $\mathbf{P_6}$ decreased to a mere 1.4%. No oxidized product at all was obtained from $\mathbf{S_4}$ alone (Tables 4 and 5).

Molecular modeling indicated that bulky 4-*tert*-butyl benzoic acid ($\mathbf{I_1}$) should be an even better inhibitor. Under catalytic conditions, the *tert*-butyl C−H bonds remain unaffected. When $\mathbf{I_1}$ is bound to the catalyst $\mathbf{C_{MR}(III,IV)}$, models suggest that this bulky *tert*-butyl group occupies and blocks the region close to the active site of the catalyst (Figure 7). With $\mathbf{I_1}$ (1 equiv vs substrate) present, the yield of oxidation product $\mathbf{P_6}$ decreased even further to 0.9% under the same conditions (Table 5).

We expected that a change of inhibitor concentration would change the extent of inhibition. Indeed, a decrease of the inhibitor ($\mathbf{I_1}$) concentration gave an increased yield of $\mathbf{P_6}$. A number of runs were performed varying the concentration of $\mathbf{I_1}$ and measuring the yields at low conversions where yield can be considered to be directly proportional to the velocity. A Dixon plot, obtained by plotting the reciprocal of rate (estimated from the yield at early reaction time) versus the inhibitor concentration, gave a straight line as found in enzyme inhibitor studies (Figure 8). By using three different concentrations of substrate ($\mathbf{S_6}$) for each concentration of the inhibitor, three different curves were obtained in the Dixon plot (Figure 8a). These were almost parallel, indicating that we have uncompetitive inhibition.[44] We verified that the system obeys saturation (Michaelis−Menten) kinetics, required for the Dixon approach to be valid, by noting that an increase in substrate concentration in the range of interest and in the absence of inhibitor does not cause any change in

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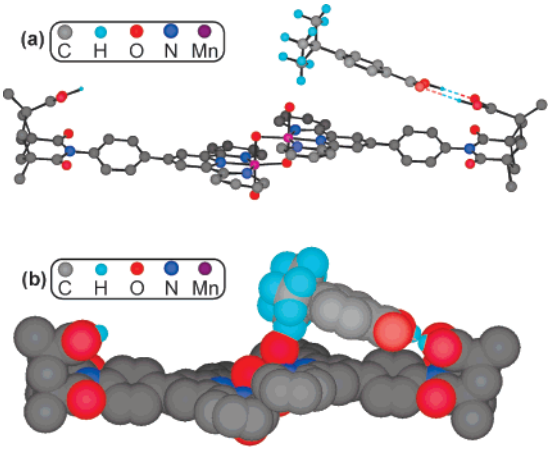

Figure 7. (a) Molecular model and (b) space filling model of $\mathbf{I_1}$ bound to $\mathbf{C_{MR}(III,IV)}$. The oxidation resistant *tert*-butyl group of $\mathbf{I_1}$ offers a significant steric bulk to block the active site.

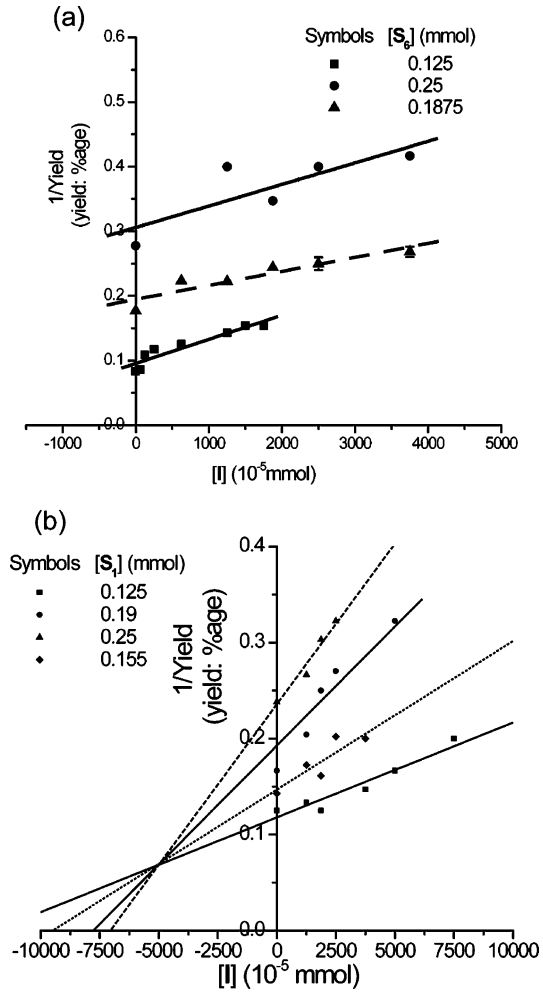

Figure 8. Dixon plots showing the inhibition of oxidation of (a) $\mathbf{S_6}$ and (b) $\mathbf{S_1}$ with molecular recognition catalyst $\mathbf{C_{MR}(III,IV)}$ and $\mathbf{I_1}$ as the inhibitor.

reaction rate indicating that all the measurements were done in the saturation regime.

Oxidation of ibuprofen ($\mathbf{S_1}$) is also inhibited by $\mathbf{I_1}$, although to a lesser extent than the case for for ibuprofen methyl ester, $\mathbf{S_6}$. The likely cause of the difference is competition for the molecular recognition site between the −COOH groups of substrate $\mathbf{S_1}$ and inhibitor $\mathbf{I_1}$, which is not possible for ester $\mathbf{S_6}$. The reciprocal of rate vs inhibitor concentration also gave a linear Dixon plot, but now having straight lines radiating from a point (Figure 8b) on the negative side of the *y*-axis, indicating a competitive inhibition in this case.[44] This is consistent with our hypothesis that both substrates and inhibitors bind to the molecular recognition site.

5.6.2. Reversal of Inhibition. Reactivation of the inhibited catalyst was found with acetic acid further confirming the reversibility of the binding of $\mathbf{C_{MR}(III,IV)}$ with $\mathbf{I_1}$. With $\mathbf{S_6}$ (1 equiv), $\mathbf{C_{MR}(III,IV)}$ (0.005 equiv), and $\mathbf{I_1}$ (0.3 equiv) as the inhibitor in CH_3CN, half of the catalytic solution was worked up after 3 h at 0 °C. Only 1.5% conversion of $\mathbf{S_6}$ to $\mathbf{P_6}$ occurred, indicating successful inhibition with $\mathbf{I_1}$ (Table 6). To the other half of the catalytic solution, excess acetic acid (2.25 equiv) was added, an amount too small to cause any significant change in the solvent polarity but enough to saturate all the catalytic molecular recognition sites by H-bonding. This addition of CH_3-COOH was expected to disrupt the binding of the inhibitor to the catalyst, reversing the inhibition. Indeed, after a further 3 h at 0 °C, $\mathbf{P_6}$ product was recovered with 34% conversion (Table 6) showing successful reversal of inhibition by $\mathbf{I_1}$ with CH_3-COOH. We are not aware of any similar reactivation in a synthetic catalyst.

5.7. Consequences of Steric Exclusion. The need for steric exclusion to block unselective reaction may explain why it has previously proved[21] difficult to obtain high selectivities via molecular recognition. A particular feature of our $\mathbf{C_{MR}}$ system is that it has two molecular recognition units, which can act together to block both faces of the catalyst at once. In order to do this, both −COOH sites of the ligand must be essentially fully occupied and steric repulsion between the two substrates or inhibitors must be sufficient to cause the two groups to occupy opposite faces of the catalyst.

Conclusion

The results reported here help confirm our earlier proposal that molecular recognition of the −COOH group in our substrates by the ligand −COOH group orients the substrate.[22] This allows remote catalytic oxygenation of an alkyl C−H bond by a Mn-oxo active site to occur with very high (>98%) regio- and stereoselectivity with oxone as the primary oxidant. Our work identifies steric exclusion—exclusion of free substrate molecules from the active site—as an important criterion for selectivity. If free substrates were able to reach the active site, they would react unselectively, degrading the selectivity. Both faces of the catalyst are protected by having two ligand −COOH groups. *p*-^{t}BuC$_6$H$_4$COOH ($\mathbf{I_1}$) was an effective inhibitor for our standard substrate, ibuprofen ($\mathbf{S_1}$), by binding to the ligand −COOH recognition site and blocking the approach of a substrate. Dixon plots show that inhibition is competitive for ibuprofen, suggesting that the substrate −COOH group competes with the inhibitor −COOH group for binding to the recognition site. In contrast, inhibition is uncompetitive for the alternate substrate, ibuprofen-ester ($\mathbf{S_6}$), consistent with this substrate no longer being able to bind to the recognition site. A much less bulky acid, MeCOOH, can reverse the inhibition caused by *p*-^{t}BuC$_6$H$_4$COOH ($\mathbf{I_1}$) by displacing it from the ligand −COOH recognition site and revealing the Mn-oxo active site.

6. Experimental Section

6.1. Materials. All solvents and reagents were obtained from commercial sources and were used without further purification. Oxone, used in all the catalytic runs, is the tetrabutylammonium salt of oxone. Tetrabutylammonium oxone (TBAO) and K[Oxone] were standardized using iodometric titration.

6.2. Physical Measurements. ^{1}H and ^{13}C NMR spectra were recorded in deuterated solvents (Cambridge Isotope Laboratories) at 25 °C on a Bruker 400 or Bruker 500 spectrometer at the Yale Department of Chemistry Instrument Center. Electrospray ionization-mass spectroscopy (ESI-MS) was done on a Waters ZQ LC-MS instrument. Gas chromatography−mass spectroscopy (GC-MS) was done on an Agilent 5973 GC-MS instrument.

6.3. Syntheses. L_{MR}, [(**L_{MR}**)$MnCl_2$], [(**L_{NR}**)$MnCl_2$], **C_{MR}(IV,IV)**, **C_{NR}(III,IV)** were synthesized following the literature method.[22]

6.3.1. [H_2O(L_{MR}**)Mn(μ-O)$_2$Mn(**L_{MR}**)H_2O](NO_3)$_3$ (C_{MR}(III,IV)).** [(**L_{MR}**)$MnCl_2$] (0.200 g, 0.3 mmol) was dissolved in a minimum amount of hot (almost boiling) H_2O-CH_3CN (1:1); the solution was cooled to 0 °C by stirring in an ice bath. 5 mL of ice-cold aq. K-Oxone (0.080 g, 0.2 mmol) were added dropwise very slowly over a time of 10 min. 10 mL of ice-cold aq. KNO_3 (4 g, 40 mmol, large excess) were added. The volume of the solution was minimized by stirring the solution under vacuo. The brown precipitate that formed was filtered and washed with ice-cold water (3 × 3 mL) and then with copious diethyl ether to give pure **C_{MR}(III,IV)**. Very slow addition of oxone and use of a smaller amount are important to obtain **C_{MR}(III,IV)** instead of **C_{MR}(IV,IV)**. Elemental analysis: ($C_{66}H_{64}Mn_2N_{11}O_{21}$) ($C_2H_5OH_5C_2$); Calcd. C, 54.89; H, 4.87; N, 10.07. Found: C, 54.96; H, 4.87; N, 10.07. A frozen solution of **C_{MR}(III,IV)** in CH_3CN gave a 16-line EPR spectrum (10 K), characteristic of a $Mn^{III}(\mu\text{-}O)_2Mn^{IV}$ unit.

6.3.2. Syntheses of Methyl Esters of Several Carboxylic Acids. The carboxylic acid was dissolved in a minimum amount of methanol−benzene solution (MeOH/PhH = 2:7) and cooled to 0 °C. An ice-cold solution of trimethylsilyldiazomethane in diethyl ether was added dropwise. The reaction was followed from the disappearance of the reddish color of trimethylsilyldiazomethane accompanied with the evolution of a gas. Once the reddish color persisted, the addition of trimethylsilyldiazomethane was stopped. The mixture was stirred at 0 °C for another 10 min and evaporated under vacuum, giving an oily product.

6.4. Catalytic Conditions. 6.4.1. Catalyzes with S_1, S_2, S_4, and S_5. 5×10^{-4} mmol of catalyst was dissolved in 50 μL of water and 2 mL of CH_3CN. This solution was added to 0.5 mmol of substrate. The solution was cooled to 0 °C. A solution of 2.5 mmol of TBAO in 8 mL of CH_3CN was prepared and was added to the cooled solution of catalyst and substrate. The resulting solution was stirred for 2 h at 20 °C, 8 h at 0 °C, and 2 days at −20 °C. The reaction was quenched by addition of 60 mL of a saturated aqueous solution of $NaHSO_3$. The solution was acidified with 0.1 N HCl. The unreacted substrate and products were extracted in ether, evaporated under vacuum, and dried in vacuo over P_2O_5 overnight. The resulting unreacted substrates and product mixtures were analyzed with ^{1}H NMR, ^{13}C NMR, and ESI-MS.

6.4.2. Oxygenation of S_6 in Presence of I_1. To a 2 mL acetonitrile solution of **S_6** (0.5 mmol) and **I_1** (concentration varies in different runs), a solution of **C_{MR}(III,IV)** (2.5×10^{-3} mmol) dissolved in a minimum amount of CH_3CN-H_2O (25 μL of H_2O and 1.0 mL of CH_3CN) was added and stirred in an ice bath to cool it down to 0 °C. A solution of TBAO (2.5 mmol) in 21 mL of acetonitrile was added to it and stirred for 3 h at 0 °C. The workup was done as above. The unreacted substrate and product mixture was analyzed with ^{1}H NMR.

6.5. Identification of Products. Identification of products obtained from oxidation of **S_1** and **S_2** with catalyst **C_{MR}(IV,IV)** has been discussed in our prior report as well as briefly in the Supporting Information.[22]

6.5.1. Oxidation of S_5. The product **P_5** was identified by comparing the ^{1}H NMR spectrum to that of the commercially available sample.

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Supporting Information Available: Experimental procedures and spectroscopic data. This material is available free of charge via the Internet at http://pubs.acs.org.

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