Table I. NMR Chemical Shifts[a] of Some Dioxiranes $^1R^2RCO_2$

compd	^1R	^2R	temp, °C	^1H NMR	$^{13}C\{^1H\}$ NMR	^{19}F NMR [^{17}O NMR]	ref
1b	CH_3	CH_3	0	1.65 (s)	22.6 (CH_3)		1, 3, 4
					102.2 (>CO_2)	[302 (s)][b]	
1d	CF_3	CF_3	-		-	-76.8 (s)[c]	17
1f	CH_3	CF_3	-20	1.97 (s)	14.51 (CH_3)[d]		this
					97.32 (>CO_2)[d,e]	[297 (s)][g]	work[h]
					122.2 (CF_3)[d,f]	-81.5 (s)	

[a] Chemical shifts (δ) in parts per million are relative to Me_4Si for ^1H NMR (200 MHz) and for ^{13}C NMR (50.309 MHz, unless noted otherwise); in ^{17}O NMR (27.120 MHz, unless noted otherwise), chemical shifts were measured from external $Me_2C{=}O$ and referred to H_2O (cf. ref 3), while ^{19}F NMR (188.220 MHz) δ are relative to internal $CFCl_3$; a Varian XL200 instrument was employed. [b] $\Delta\nu_{1/2}$ = 113 Hz. [c] Compare $(CF_3)_2C{=}O$: ^{19}F NMR (MeCN, $CFCl_3$) δ -76.1 (Redwood, M. E.; Willis, C. J. *Can. J. Chem.* **1967**, *45*, 389-395). [d] At 100.577 MHz (Bruker AM400). [e] Quartet, J_{CCF} = 40.2 Hz. [f] Quartet, J_{CF} = 280.7 Hz. [g] At 54.227 MHz (Bruker AM400); $\Delta\nu_{1/2}$ = 270 Hz. [h] In all of the NMR experiments, it was verified that the resonance signals attributed to the dioxirane instantly disappear upon quenching of the reactive peroxide in solution with $PhSCH_3$ (cf. ref 4).

The screening of the reactivity presented by **1f** in selective oxidations of organic substrates has begun. We find that this dioxirane is able to perform the effective oxygen transfer reactions reported for dimethyldioxirane,[1] *but at a much higher rate.* For instance, phenanthrene could be converted by **1f** into the corresponding 9,10-oxide[1] efficiently (80% conversion, ≥93% yield) within just 5 min at -20 °C; by way of comparison, we observed that 50% conversion of the said arene into the same epoxide requires about 20 h at 22 °C by using **1b**.[21] As for alkene epoxidations, dioxirane **1f** reacts rapidly (ca. 1 min, at -20 °C) and in a completely stereospecific manner with *cis*-2-octene and *trans*-β-methylstyrene to form *cis*-2,3-epoxyoctane[22] and *trans*-β-methylstyrene oxide,[23] respectively (yield ≥90%).[21] Product isolation is attractively simple since trifluoroacetone (**3**) (the solvent and the reduction product of **1f**) is quite volatile. Efficient oxidations of saturated carbon C-H bonds can also be performed; as an example, cyclohexane reacts readily with dioxirane **1f** solutions at -20 °C by a 1:2 stoichiometry, yielding cyclohexanone (≥95% yield, 30 min).[21] Under similar conditions, during ca. 45 min, *n*-heptane is oxidized (30% conversion, ~95% yield) by **1f** to yield a mixture of 4-heptanone (20%), 3-heptanone (40%), and 2-heptanone (40%).[21]

We shall soon report in detail on more cases which will further illustrate the finding that, in performing selective oxidations, dioxirane **1f** is remarkably more reactive than dimethyldioxirane (**1b**). It is unlikely that this is due solely to solvent effects (i.e., trifluoroacetone (**3**) vs acetone). Rather, the considerable difference in acid strength existing between $(CF_3)(CH_3)C{=}O^+H$ (pK = -14.9) and $(CH_3)_2C{=}O^+H$ (pK = -7.6)[24] suggests that it should be ascribed to the better leaving-group ability of CF_3COCH_3 with respect to acetone (and to incipient charge separation in the transition state) during nucleophilic displacement at the dioxirane O-O bond (electrophilic oxidation).[6a] Then, it is perhaps not surprising that doxirane **1f** should be so "ripe"[25] for oxygen transfer to organic substrates.

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(20) The decomposition kinetics of **1f** (iodometric titer) at 0 °C appears to follow a mixed first-order and second-order rate law; this suggests that the decomposition might also involve some oxygen production, i.e., $2\,\mathbf{1f} \rightarrow 2\,(CH_3)(CF_3)C{=}O + O_2$ (cf. ref 6).

(21) Reactions were monitored and product yields determined by GC (OV 101 or SE 30, 30 m × 0.25 μm i.d. capillary column, internal standard $CFCl_2CFCl_2$) or HPLC (reverse-phase, 10 μm C-18, 25 cm × 4.6 mm i.d. analytical column, MeCN/water or MeOH/water); the reaction products were identified upon comparison of their ^1H NMR and/or GC/MS (Hewlett-Packard 5970) spectra with those of authentic samples.

(22) Bp 70-72 °C (18 mmHg); ^1H NMR ($CDCl_3$, Me_4Si) δ 0.88 (t, 3 H, $CH_3(CH_2)_4$, J = 6 Hz), 1.25 (d, 3 H, $OCHCH_3$, J = 5.5 Hz), 1.26-1.48 (m, 8 H, $(CH_2)_4$), 2.87 (complex m, 1 H, $C_5H_{11}CHO$), 3.02 (d of q, 1 H, $CHOCHCH_3$, J_{HCH_3} = 5.5, J_{HH}(cis) = 4.3 Hz).

(23) Audier, H. E.; Dupin, J. F.; Jullien, J. *Bull. Soc. Chim. Fr.* **1966**, 2811-2816.

(24) Levy, G. C.; Cargioli, J. D.; Racela, W. *J. Am. Chem. Soc.* **1970**, *92*, 6238-6246.

(25) Our group has given this reactive species the laboratory name of Mello ("mellow") dioxirane.

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Enzymic Regioselectivity in the Hydroxylation of Cholesterol Catalyzed by a Membrane-Spanning Metalloporphyrin

Summary: The hydroxylation of simple alkanes and the selective C-25 hydroxylation of cholesterol have been achieved with a membrane-spanning Mn(III) porphyrin positioned in a synthetic bilayer assembly by appended steroidal substituents.

Sir: Cytochrome P-450 enzymes occur widely in nature and function as the monooxygenation catalysts of many lipophilic compounds. Among the oxidations this enzyme catalyzes, perhaps the most unique is the hydroxylation of saturated hydrocarbons. This reaction is important in the conversion of natural substrates such as cholesterol to corticosteroids and in the hydrophilization of foreign compounds such as petroleum products.[1] It has been shown in recent years that synthetic iron(III) and manganese(III) porphyrins are capable of mimicking the hydroxylation activity of the natural enzyme.[2,3] Although

(1) See: *Cytochrome P-450*; Ortiz de Montellano, P. R., Ed.; Plenum: New York, 1986.

(2) Groves, J.T.; Nemo, T. E. *J. Am. Chem. Soc.* **1983**, *105*, 5786, 6243.

(3) (a) De Poorter, B.; Ricci, M.; Meunier, B. *Tetrahedron Lett.* **1985**, *26*, 4429. (b) Battioni, P.; Renaud, J.-P.; Bartoli, J.-F.; Mansuy, D. *J. Chem. Soc., Chem. Commun.* **1986**, 341.

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Table I. Hydroxylation of Saturated Hydrocarbons and Cholesterol

substrate	method[a]	mol of product/mol of $Mn^{III}ChP(Cl)$	product
cyclooctane	I	3.4	cyclooctanone
cyclooctane	II	6.2	12/1 cyclooctanol/ cyclooctanone
adamantane	I	1.6	adamantan-1-ol
ethylbenzene	I	6.0	acetophenone
ethylbenzene	II	8.8	8/1 1-phenylethanol/ acetophenone
fluorene	I	2.3	9-fluorenone
cholesterol	I	0.8	25-hydroxycholesterol

[a] Method I: Vesicles containing 10 μmol of DMPC, 2.0 μmol of substrate, and 0.05 μmol of Mn(CHP)Cl were formed in 2 mL of Tris pH 8.6 buffer. The solution was purged with oxygen, and 100 μmol of ascorbic acid was added. The reaction time was 15 h. Method II: Vesicles containing 10 μmol of DMPC, 2.0 μmol of substrate, 0.05 μmol of Mn(ChP)Cl, and 0.25 μmol of 1-pentylimidazole were formed in 2 mL of phosphate buffer at pH 7.4. Sodium periodate (10 μmol) was added as oxidant after 15 min. The reaction time was 3 h.

liver microsomal forms of cytochrome P-450 are not found to be very selective, numerous specific steroidal transformations are mediated by this enzyme. Especially striking is the C-26 hydroxylation of cholesterol by liver mitochondrial cytochrome P-450.[4] Thus, a number of stereoselective[5] and regioselective[6] synthetic metalloporphyrin catalytic systems have been developed for the epoxidation of olefins[2] and the hydroxylation of alkanes.[2,7]

There has been a growing interest in performing reactions in synthetic vesicles.[8,9] Among the potential advantages of such media are the possibility of using lipophilic materials in an aqueous milieu and the prospect of taking advantage of the ordered environment of the membrane. We have recently reported the synthesis of a membrane-spanning metalloporphyrin which, because of its four steroidal appendages, is organized in a synthetic phospholipid bilayer. This system was found to selectively epoxidize the lipophilic end of unsaturated fatty acids and steroidal dienes.[10] In this paper we describe a steroidal manganese(III) porphyrin bilayer assembly for the hydroxylation of saturated hydrocarbons. *Applied to cholesterol, this system supports a selective hydroxylation at C-25.*

The steroidal manganese porphyrin, chloro[α,β,α,β-*meso*-tetrakis[*o*-(3β-hydroxy-5-cholenamido)phenyl]-porphyrinato]manganese(III) [Mn^{III}(ChP)Cl], was synthesized by condensation of α,β,α,β-*meso*-tetrakis(*o*-aminophenyl)porphyrin with 3β-hydroxy-5-cholenic acid through formation of an amide bond as previously described.[10] The free-base porphyrin was then metalated with manganese(II) acetate tetrahydrate in refluxing THF.

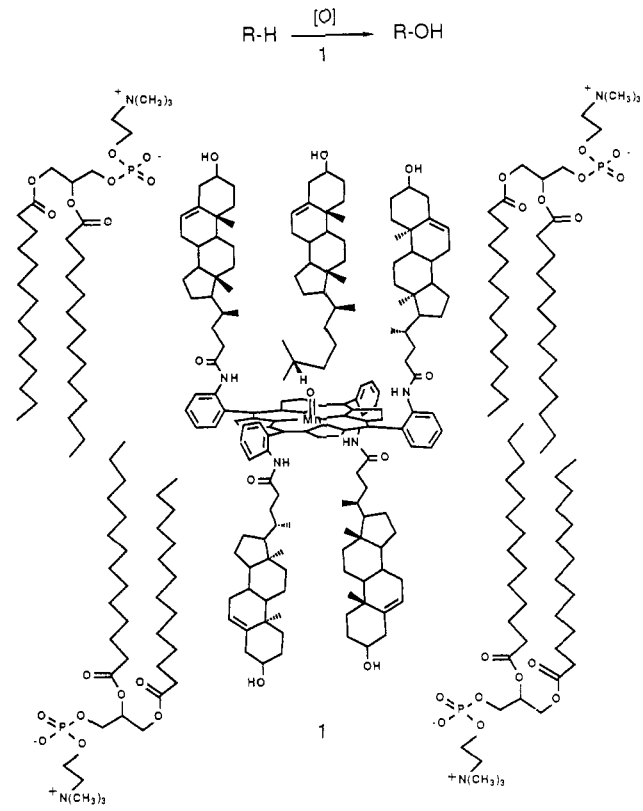

Figure 1. Idealized molecular bilayer assembly of oxo[α,β,α,β-*meso*-tetrakis[*o*-(3β-hydroxy-5-cholenamido)phenyl]-porphyrinato]manganese(IV) [Mn^{IV}ChP(O), 1], cholesterol, and dimeristoylphosphocholine.

Formation of the manganese(III) complex was confirmed by observation of the visible spectrum (λ_{max} in benzene 373, 402, 420 (sh), 480, 582, and 617 nm) and the FAB-MS on a *m*-nitrobenzyl alcohol matrix which gave the M − Cl ion at *m/z* 2153. Hydroxylations were carried out under aerobic conditions with ascorbic acid as the reducing agent[11] (method I) or by using sodium periodate as the primary oxidant[12] (method II).

The metalloporphyrin–bilayer assembly was prepared by sonicating a thin film of 10 μmol of dimyristoylphosphocholine (DMPC), 2.0 μmol of substrate, and 0.05 μmol of Mn^{III}(ChP)Cl in 2 mL of aqueous solution. The unilamellar vesicles obtained were equilibrated at room temperature for 15 min, and the reaction was initiated by addition of ascorbic acid or sodium periodate. At the completion of the reaction, ether was added and the phases were vigorously shaken to destroy the vesicles. Analysis was performed by GC using authentic samples as references. The results for typical alkanes as presented in Table I show that the metalloporphyrin–bilayer assembly is a viable catalytic hydroxylation system for a variety of substrates. The aerobic system oxidized alcohols to ketones when such a transformation was possible. The analogous Fe^{III}(ChP)Cl porphyrin was inactive under these conditions. *Most significantly, a similar hydroxylation of cholesterol afforded only 25-hydroxycholesterol.* In comparison with adamantane, this selective hydroxylation is surprisingly efficient on a per hydrogen basis. Other more active positions on the molecule such as the Δ^5 double bond were left unchanged. The identity of the product

(4) (a) Pederson, J. I.; Björkhem, I.; Gustafsson, J. *J. Biol. Chem.* **1979**, *254*, 6464. (b) Pederson, J. I.; Saarem, K. *J. Steroid Biochem.* **1978**, *9*, 1165.

(5) (a) Groves, J. T.; Myers, R. S. *J. Am. Chem. Soc.* **1983**, *105*, 5791. (b) Mansuy, D.; Battioni, P.; Renaud, J.-P.; Guerin, P. *J. Chem. Soc., Chem. Commun.* **1985**, 155.

(6) (a) Tabushi, I.; Morimitsu, K. *J. Am. Chem. Soc.* **1984**, *106*, 6871. (b) De Carvalho, M. E.; Meunier, B. *Nouv. J. Chim.* **1986**, *10*, 223.

(7) (a) Suslick, K.; Cook, B.; Fox, M. *J. Chem. Soc., Chem. Commun.* **1985**, 580. (b) Cook, B. R.; Reinart, T. J.; Suslick, K. S. *J. Am. Chem. Soc.* **1986**, *108*, 7281.

(8) van Esch, J.; Roks, M. F. M.; Nolte, R. J. M. *J. Am. Chem. Soc.* **1986**, *108*, 6093.

(9) (a) Czarniecki, M. F.; Breslow, R. *J. Am. Chem. Soc.* **1979**, *101*, 3675. (b) Moss, R. A.; Schreck, R. P. *J. Am. Chem. Soc.* **1985**, *107*, 6634. (c) Fuhrhop, J. H.; Mathieu, J. *Angew. Chem., Int. Ed. Engl.* **1984**, *23*, 100.

(10) Groves, J. T.; Neumann, R. *J. Am. Chem. Soc.* **1987**, *109*, 5045.

(11) Mansuy, D.; Fontecave, M.; Bartoli, J.-F. *J. Chem. Soc., Chem. Commun.* **1983**, 253.

(12) (a) Groves, J. T.; McMurry, T. J. Presented at the 187th National Meeting of the American Chemical Society, St. Louis, MO, April 1983. (b) Groves, J. T.; Stern, M. K. *J. Am. Chem. Soc.*, submitted.

was confirmed by GC and GC-MS using an authentic sample (Research Plus) as reference (SPB-1 column, 30 m, 0.32-mm i.d. 0.25-μm coating at 260 °C; retention time 26.4 min). The MS displayed peaks at m/z (relative intensity) 402 (24), 384 (100), 369 (47), 367 (13), 351 (36), 300 (28), 299 (33), 273 (38), 271 (69), and lower m/z steroidal clusters. Similar oxidation of cholesterol in a homogeneous solution gave small amounts of many products.

We have shown that a steroidal manganese(III) porphyrin catalyst intercalated in a synthetic bilayer is capable of the hydroxylation of hydrocarbons in moderate yields. This assembly also selectively hydroxylated cholesterol at carbon 25. The O_2/ascorbic acid/manganese porphyrin system has been shown in other studies to generate an oxomanganese(IV) porphyrin as the reactive intermediate.[12b,13] The selectivity observed here must be due to the enforced proximity of the tertiary hydrogen at C-25 to the incipient manganyl group of the oxidized catalyst (1) at the catalytic center (Figure 1), precisely the motif that has been revealed in the X-ray crystal structure of the enzyme-substrate complex of cytochrome P-450$_{cam}$.[14]

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(13) Groves, J. T.; Stern, M. K. *J. Am. Chem. Soc.* **1987**, *109*, 3812.
(14) Poulos, T. In ref 1, pp 505 ff.

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Use of the Brønsted Relationship To Detect a Mechanistic Shift for Reaction of 2-(Methylthio)ethyl Chloride with Thiophenoxide Anions

Summary: The linear Brønsted plot for reaction of thiophenoxide ions with 2-(methylthio)ethyl chloride in DMSO changes slope for highly deactivated thiophenoxides, thus suggesting a mechanism change. Attack of a neutral nucleophile on an anionic electrophile may be involved.

Sir: We recently demonstrated that *p*-aminothiophenoxide reacts with 2-(methylthio)ethyl chloride (1) or 2-(phenylthio)ethyl brosylate in dimethyl sulfoxide (DMSO) by a direct displacement S_N2 mechanism.[1] On the other hand, solvolysis of these derivatives in DMSO proceeds by neighboring sulfur participation to give a cyclic sulfonium ion (a k_Δ mechanism).[1] In this paper we report that a series of seven highly nucleophilic thiophenoxide anions undergo reaction with 1 in DMSO to provide second-order rate constants, Table I, which give a linear Brønsted plot,[2]

(1) Sedaghat-Herati, M. R.; Harris, J. M.; McManus, S. P. *J. Org. Chem.* **1988**, *53*, 2539.
(2) Bordwell, F. G.; Hughes, D. L. *J. Org. Chem.* **1982**, *47*, 3223. See also: Bordwell, F. G.; Cripe, T. A.; Hughes, D. L. In *Nucleophilicity*; Harris, J. M., McManus, S. P., Eds.; Advances in Chemistry 215; American Chemical Society, Washington, DC, 1987; Chapter 9.

Table I. Second-Order Rates for Reactions of ArS$^-$ Ions with MeSCH$_2$CH$_2$Cl (1) and *n*-Butyl Chloride in DMSO at 25 °C

		10^3k_2 (M^{-1} s^{-1})	
ArS$^-$, Ar =	pK_a[a]	1[b]	*n*-BuCl[c]
4-MeOC$_6$H$_4$	11.2	95.4 ± 2.4	105 ± 20 (100)
C$_6$H$_5$	10.3	41.8 ± 0.2	44.2 (42.6)
2-naphthyl	9.5	26.6 ± 1.5	25.6 ± 0.6 (25.4)
4-BrC$_6$H$_4$	9.0	18.3 ± 2.8	(13.3)
3-ClC$_6$H$_4$	8.57	12.1 ± 1.1	
2-ClC$_6$H$_4$	8.5	13.7 ± 0.5	9.01 (8.77)
3-CF$_3$C$_6$H$_4$	8.1	9.86 ± 0.20	7.54 ± 0.60 (5.78)
2,4,5-Cl$_3$C$_6$H$_2$	6.00	8.62 ± 0.86	1.04 ± 0.18 (0.852)
4-NO$_2$C$_6$H$_4$	5.5	22.9 ± 3.1	0.313 ± 0.029 (0.279)
C$_6$Cl$_5$	3.3	23.8 ± 6.7	0.099 ± 0.0045 (0.0581)

[a] From ref 2. [b] Rates were run as pseudo first order by following the disappearance of the thiophenoxide by UV as described in ref 1. [c] The values in parentheses are from ref 2.

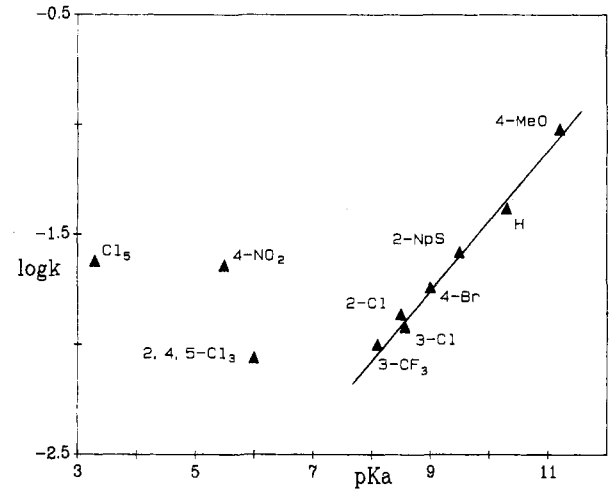

Figure 1. Brønsted plot for reaction of 2-(methylthio)ethyl chloride (1) with thiophenoxide anions in DMSO.

Figure 1. The log k values for reaction of the same seven nucleophiles also plot linearly against log k values for their S_N2 reaction with *n*-butyl chloride, Table I.[2] With the β-thioethyl substrates, we anticipated a break from the linear relationship with weakly nucleophilic thiophenoxide ions because at some point the first-order k_Δ process should be faster than the second-order S_N2 reactions. We did indeed find a break in the linear relationship but, surprisingly, the weakest thiophenoxide nucleophiles react *more* rapidly, indicating that some mechanism other than k_Δ is involved, Figure 1.

The thiophenoxides with pK_a values less than 8.1 still follow second-order kinetics. The calculated pseudo-first-order rates for the least reactive anions are still more than an order of magnitude greater than the measured first-order rate for 1 in DMSO in the absence of any thiophenoxide ion ($k = 8.5 \times 10^{-5}$ s^{-1}). The rate *increase* observed for the 4-nitro- and pentachlorothiophenoxide ions, Figure 1, is not predicted by either the normal S_N2 process or by the neighboring sulfur-assisted process. With regard to the mechanism, it is instructive to note that the deviating substrates follow second-order kinetics and *give substitution products expected for an S_N2 process.*

The most logical explanation for the break in the Brønsted plot, Figure 1, is that there has been a change in mechanism to some bimolecular substitution process other than that followed by the seven more nucleophilic thiophenoxide anions. The mechanism must account for a nucleophilic reactivity order which is the inverse of the intrinsic nucleophilicity of the thiophenoxide ions! Since

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