Compound 5 showed marginal cytotoxicity to murine leukemia cells (P388), ED50 = 3.5 μg/mL, but the other compounds in the group were inactive.

Experimental Section

The sponge, a *Spongia* sp., was collected at Dalton Reef, Queensland, Australia, in 1983 and immediately frozen. The specimens were freeze dried (246 g), cut into small pieces, and extracted three times with methylene chloride at room temperature assisted by agitation in a sonication bath. The combined extracts were concentrated on a rotary evaporator, and the residue (6.0 g) was chromatographed on a column of silica gel (180 g, 230–400 mesh) beginning with $CHCl_3$ and then using $CHCl_3$ with increasing amounts of CH_3OH.

3β,17,19-Trihydroxyspongia-13(16),14-dien-2-one (1). Fraction 10 from above, eluted with $CHCl_3$ (~400 mg), was rechromatographed over silica gel (10 g) using 1.5% MeOH in $CHCl_3$ to give four fractions. Purification of fraction 2 by HPLC using a reversed-phase column (C-18, 5 μm) with 40% aqueous CH_3OH afforded 1 (18 mg) and 3 (9 mg). For 1: mp 157–159 °C (lit.[1] mp 194.7–197 °C);[12] IR (KBr) 3500, 3430, 3390, 1695, and 1030 cm^{-1}; mass spectrum (12 eV, low resolution), *m/e* (relative intensity) 348 (M^+, 4), 318 (25), 317 (12), 288 (17), 287 (100), 269 (23), 251 (3), 241 (7), 213 (10), 199 (5), 187 (7), 185 (7), 161 (7), 159 (5), 147 (42), 135 (25), 133 (33), 121 (8), 105 (15), and 91 (21).

2β,3β,17,19-Tetrahydroxyspongia-13(16),14-diene (4). Fraction 15 of the initial chromatography, eluted with 2% MeOH–$CHCl_3$, on purification by HPLC using a reversed-phase column (C-18, 5 μm) with 40% H_2O–MeOH yielded a crude polar fraction and compound 2 as a gum (5.1 mg): IR (KBr) 3400 (br) cm^{-1}; mass spectrum (12 eV, low resolution), *m/e* 350 (M^+, 36), 320 (24), 319 (100), 301 (32), 287 (24), 283 (35), 271 (14), 265 (9), 253 (11), 227 (5), 225 (6), 187 (5), 185 (6), 173 (7), 161 (8), 149 (12), 147 (39), 135 (20), 132 (9), 121 (12), 119 (19), and 95 (4).

2-Oxa-17,19-dihydroxyspongia-13(16),14-dien-3-one (5): colorless gum; IR ($CHCl_3$) 3480, 1702 cm^{-1}; mass spectrum (12 eV, low resolution), *m/e* 334 (M^+, 1), 305 (6), 304 (28), 303 (4), 285 (1), 274 (19), 273 (100), 255 (1), 227 (2), 200 (3), 199 (14), 187 (2), 147 (21), 132 (5), and 121 (2); high-resolution mass spectrum, observed *m/z* (composition, calculated millimass) 334.17997 ($C_{19}H_{26}O_5$, 334.17803).

(12) In ref 1, the 3α-epimer of 1 is reported to have mp 157.5–160.5 °C. Since our ^{13}C data clearly matches that of 1 in ref 1, we presume an error was made in reporting the melting points of these epimers initially.

17-Acetoxy-4-epispongialactone A Methyl Ester (7). The crude polar fraction obtained in the isolation of 2 was purified by HPLC using a reversed-phase (C-18, 5 μm) HPLC column with 70% H_2O–CH_3OH to give impure 6 (13 mg). Impure 6 (13 mg) was dissolved in 16 mL of H_2O–CH_3OH–diethyl ether (1:2:5) and methylated at 5 °C with excess CH_2N_2 to give a methyl ester which was then acetylated with pyridine–acetic anhydride (9:1, 5 mL) at room temperature overnight. The usual workup, afforded crude ester 7, which was purified using a SiO_2 Sep-Pak column with chloroform elution to give pure 7: gum (6 mg); IR ($CHCl_3$) 1732, 1720 (sh) cm^{-1}; mass spectrum (12 eV, low resolution), *m/z* 418 (M^+, 22), 360 (2), 359 (4), 358 (7), 347 (16), 346 (64), 345 (100), 328 (2), 327 (4), 313 (5), 304 (13), 303 (29), 299 (4), 286 (4), 285 (12), 271 (6), 258 (5), 249 (3), 225 (2), 199 (2), 185 (3), 169 (4), 149 (1), and 84 (1); high-resolution mass spectrum, observed *m/e* (composition, calculated millimass) 418.20338 ($C_{23}H_{30}O_7$, M^+, 418.19916).

19-Nor-3-hydroxyspongia-3,13(16),14-trien-2-one (8). Fraction 4 that eluted with $CHCl_3$ (~30 mg) on the first open column chromatography was further resolved by HPLC using a reversed-phase column (C-18, 5 μm) and 20% aqueous CH_3OH as eluent to give 8 as white crystals (6 mg): mp 145–147 °C ($CHCl_3$); $[\alpha]_D$ = 3.0° (*c* 0.1, $CHCl_3$); IR ($CHCl_3$) 3460, 1665, 1640, cm^{-1}; mass spectrum (12 eV, low resolution), *m/e* 300 (M^+, 100), 285 (19), 282 (11), 267 (7), 229 (7), 215 (5), 201 (10), 200 (13), 185 (13), 161 (7), 151 (7), 149 (8), 148 (8), 147 (10), 135 (8), and 121 (8); high-resolution mass spectrum, observed *m/z* (composition, calculated millimass) 300.17024 ($C_{19}H_{24}O_3$, M^+, 300.17255).

Acknowledgment. This work was supported by Department of Commerce, NOAA, Office of Sea Grant Project NA 883AA-D-00011. We thank Dr. E. O. Pordesimo for executing several NMR analyses, Drs. Pomponi, Harbor Branch Research Foundation, Ft. Pierce, FL, and P. Bergquist, University of Auckland, New Zealand, for sponge identification, and Dr. John Coll for inviting F.J.S. to participate in a cruise of the R. V. Kirby, James Cook University, Townsville, Queensland, Australia, during which the sponge was collected. We gratefully acknowledge NSF Grant CHE 8113507 and the University of Oklahoma Research Associates Fund for funds to purchase a high-field NMR spectrometer.

Registry No. 4, 130611-11-9; 5, 130574-79-7; 6, 130574-80-0; 8, 130611-12-0.

Epoxidation of Styrene with Aqueous Hypochlorite Catalyzed by a Manganese(III) Porphyrin Bound to Colloidal Anion-Exchange Particles

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Received April 13, 1990

Epoxidation of styrene in aqueous sodium hypochlorite solution was catalyzed by the tetrasodium salt of 5,10,15,20-tetrakis(2,6-dichloro-3-sulfonatophenyl)porphinatomanganese(III) chloride (1). Manganese porphyrin 1 was more active bound to 60 nm diameter colloidal anion exchange particles than in aqueous solution: 0.12 mol % of 1 bound to four different types of particles gave 71–81% conversion of 1.2 mmol of styrene to styrene oxide with 1.5 mmol of hypochlorite in 1 h at room temperature. Although the activity of the catalyst decreased with time due to oxidative degradation of the porphyrin, 620 mol of styrene oxide per mol of catalyst were produced in the presence of excess oxidant. The conditions were highly selective for epoxidation of styrene: substituted styrenes epoxidized more slowly, and aliphatic alkenes did not react. Visible spectra indicated that oxidized forms of 1 were present in the hypochlorite solutions, and that the form of 1 in solution was different from that in the particles.

Introduction

Colloidal polymer particles in water, also known as latexes, are produced by emulsion polymerization in large amounts for synthetic rubber and for paints. They also are promising catalyst supports because of their high surface areas and their ability to concentrate organic reactants in the active catalyst phase by absorption from water.[1] In the absence of organic solvent aqueous colloidal

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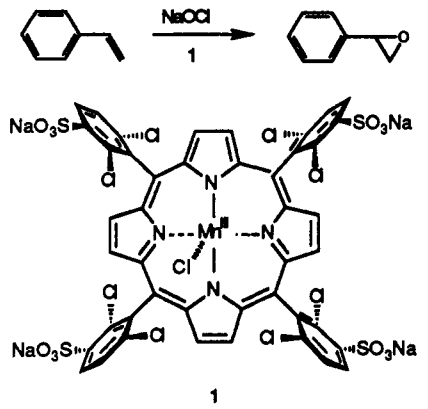
0022-3263/91/1956-1253$02.50/0

catalysts are active for oxidations of organic compounds, such as cobalt-catalyzed autoxidation of tetralin,[2] cobalt phthalocyanine catalyzed autoxidation of 1-decanethiol,[3] cobalt phthalocyanine catalyzed autoxidation of 2,6-di-*tert*-butylphenol,[4] and copper-catalyzed autoxidation of ascorbic acid.[5] When one reactant is an anion, colloidal particles charge-stabilized with quaternary ammonium ions serve both as binding sites for transition-metal complexes and as phase-transfer catalysts. The activities of the metal complexes bound to colloidal polymers exceed their activities in aqueous solution for oxidations of organic substrates that have only low solubility in water.[2-4] Such catalysts could be used for the oxidative destruction of organic contaminants in water and for solvent-free organic chemical processing. We report here a Mn(III) porphyrin catalyst bound to cationic colloidal particles for epoxidation of styrene by aqueous hypochlorite.

Synthetic metalloporphyrins are of great interest as oxidation catalysts because their structures resemble those of the cytochromes P-450, nature's catalysts for oxidation of foreign organic compounds in our bodies. Although the cytochromes P-450 use dioxygen and an electron-transfer reducing agent, most of the synthetic metalloporphyrin catalysts are used with oxidizing agents such as iodosylbenzene, alkyl hydroperoxides, amine *N*-oxides, peroxycarboxylic acids, hydrogen peroxide, aqueous persulfate, and aqueous hypochlorite that do not require a reducing agent.[6] Most synthetic P-450 analogues are soluble only in organic solvents, and their activities with water-soluble oxidizing agents depend upon phase-transfer catalysis. The organic solutions in which most P-450 mimics are studied do not resemble physiological conditions.

Most metalloporphyrins are unstable under the conditions of hydrocarbon oxidation. Two methods have been used in attempts to improve their stability. (1) Reactions have been carried out with the oxidizing agent as the limiting reagent and a large excess of substrate, and yields and turnovers have been reported after low conversion of substrate. Such experiments have been vital to elucidation of the mechanisms of catalysis and structures of the metalloporphyrins, but they are synthetically impractical. (2) Tetraarylporphyrins have been modified with electron-withdrawing groups to decrease reactivity toward electrophiles, and with bulky substituents in the ortho positions of the phenyl rings to retard attack of electrophiles at the nitrogens and at the meso positions of the porphyrin ring. Here we report use of the latter method to improve the oxidative stability of the water-soluble porphyrin catalyst **1** for styrene epoxidation by hypochlorite ion, an inexpensive and convenient oxidizing agent with the potential for application to large-scale synthetic processes. Hypochlorite ion with manganese(III) porphyrin catalysts was found by Munier to epoxidize alkenes in two-phase aqueous–organic mixtures containing a phase-transfer catalyst at or below room temperature, and Mn(III) hypochlorite systems have been studied extensively.[7] A



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polymer support for the catalyst enhances the rates of epoxidation.[8]

Results and Discussion

Preparation of the Catalyst. As a water-soluble catalyst we chose the tetrasodium salt of 5,10,15,20-tetrakis(2,6-dichloro-3-sulfonatophenyl)porphinatomanganese(III) chloride (**1**) because the eight ortho chloro substituents stabilize the porphyrin toward oxidation by both electron withdrawing and steric effects. 5,10,15,20-Tetrakis(2,6-dichlorophenyl)porphyrin (**2**) was prepared from 2,6-dichlorobenzaldehyde and pyrrole by the method of Lindsey[9-11] and was sulfonated with concentrated sulfuric acid at 185 °C to give 5,10,15,20-tetrakis(2,6-dichloro-3-sulfonatophenyl)porphyrin (**3**). Elemental and ^{1}H NMR analyses showed 3.4–3.5 sulfonate groups per porphyrin ring, and ^{1}H NMR analysis showed sulfonation as 75% at the 3-position and 25% at the 4-position. Metalation with $MnCl_2$ was carried out at pH 6.0–9.5, and the Mn porphyrin **1** was purified by cation exchange chromatography. Elemental analyses of two batches of **1** showed 65% and 85% incorporation of manganese, and the presence of 11 and 1 molar equiv of sodium chloride per porphyrin, using respectively the H^+ and Na^+ forms of the ion-exchange resin.

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Table I. Epoxidation of Styrene with NaOCl Catalyzed by Soluble and Latex-Bound Porphyrin 1[a]

polymer	mol % DVB	mol % N^+Cl^-	mol % CH_2Cl	av diam	% styrene oxide[b]
none[c]					32
L-6	4.8	81	10	55 nm	73
L-8[d]	1.0	85	13	59 nm	81
L-10	1.0	76	22	67 nm	71
L-12	1.0	75	23	81 nm	78
IRA-420C[e]		ca. 65		ca. 0.5 mm	11

[a] The typical conditions at the beginning of the Results and Discussion were used with 40–41 mg of latex and magnetic stirring except where noted otherwise. [b] Estimated error ±5%. [c] Porphyrin 1 in the absence of latex. [d] Porphyrin 1 = 0.39 mM, 48 mg of latex, [NaOCl] = 0.185 M, [NaOH] = 0.57 M, pyridine = 0.0185 M, volume 6.7 mL. [e] 3.40×10^{-3} mmol of 1 bound to 40 mg of Amberlite IRA-420C.

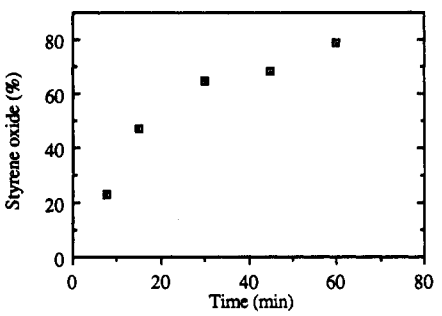

Figure 1. Epoxidation of styrene using porphyrin 1 bound to Latex L-10 and shaking agitation of a standard reaction mixture (see text and footnotes to Table I).

Colloidal anion exchange resins were prepared by emulsion polymerization of chloromethylstyrenes with divinylbenzene followed by quaternization with trimethylamine.[4] The 60 nm diameter dry particles contained 75–85% of ionic repeat units. Porphyrin 1 bound quantitatively to the ion-exchange latexes, for visible spectrophotometric analyses of the ultrafiltrates obtained after mixing 1 with latexes detected <0.1% of the added porphyrin.

Epoxidation of Styrene. Typical oxidations were carried out by stirring or shaking at room temperature for 1 h 6 mL of a mixture containing 0.38 M sodium hydroxide, 1.20 mmol of styrene, commercial laundry bleach containing 1.50 mmol of hypochlorite, and 2.2 μmol of catalyst 1 either dissolved in the water or bound to colloidal particles. Thus in an experiment using 1 which contained 65% of the stoichiometric amount of manganese, the relative molar amounts of hypochlorite/styrene/porphyrin/manganese were 1040/830/1.54/1.0. Organic components of the product mixture were extracted into diethyl ether, and the solution was analyzed by gas chromatography to contain >99% of mixtures of styrene and styrene oxide, and in some cases <1% of benzaldehyde. Styrene oxide was stable under the reaction conditions.

Results are shown in Table I from four different batches of latex catalyst, which differ in the degree of functionalization with quaternary ammonium ions and in the degree of cross-linking of the latex. Manganese porphyrin catalyst 1 was more active bound to latexes than in aqueous solution. All of the latexes had about the same activity. In the absence of 1, hypochlorite did not react with styrene with or without latex. Porphyrin 1 bound to a commercial anion-exchange resin, Amberlite IRA-420C, had much lower activity, which shows the advantage of the small size of the latex particle supports. A time course of epoxidation in Figure 1 shows >60% conversion in 30 min. *Our typical 75% conversion of styrene to styrene oxide with latex catalyst corresponds with 620 turnovers (moles of product per mole of catalyst) in the presence of excess sodium hypochlorite. Most previous reports of metalloporphyrin-catalyzed epoxidations have used a large excess of alkene and reported yields based on the limiting amount of oxidant.*

Table II. Effect of the Latex Concentration[a]

mg of latex	% styrene oxide	mg of latex	% styrene oxide
40.4	71	10.1	39
20.2	63	none[b]	32

[a] Typical conditions were employed with latex L-10 (Table I, footnote *a*). [b] Porphyrin 1 but no latex.

Table III. Effect of Concentration of Porphyrin 1 Bound to Latex[a]

$[Mn(III)] \times 10^4$ M	% styrene oxide
2.41	71
1.32	55
0.60	26

[a] Typical conditions with 40.4 mg of latex L-10 (Table I, footnote *a*).

Table IV. Effect of the Mixing Method[a]

catalyst type	mixing method	reaction vessel	% styrene oxide
latex-bound	shaking[b]	20-mL test tube	79
latex-bound	stirring	25-mL flask	71
latex-bound[c]	stirring	25-mL flask	25
latex-bound	rotating[d]	12-mL centrifuge tube	61
latex-bound	sonication	25-mL flask	6
soluble	shaking[b]	20-mL test tube	46
soluble	stirring	25-mL flask	32
soluble[e]	osc shaking[f]	25-mL flask	31

[a] Typical conditions were employed with 40.4 mg of latex L-10. [b] A wrist-action shaker with 5-cm amplitude and a frequency of 160 per min was used. [c] 0.15 volume fraction methanol in water. [d] The test tube was clamped in the middle to a stirrer which was positioned at 45° angle relative to the bench, and the tube was rotated end over end at about 400 rpm. [e] [NaOCl] = 0.21 M and [NaOH] = 0.63 M. [f] An oscillating table shaker was used.

The conversion to styrene oxide increased as the amount of latex in the reaction mixture increased, with all other amounts kept constant, including the 2.2 μmol of catalyst 1 (Table II). When the amount of 1 bound to a constant amount of latex increased, conversion to styrene oxide also increased (Table III).

Since the solubility of styrene in water is only 0.07 g/L (7×10^{-4} M) at room temperature,[12] and it should be even less soluble in sodium hypochlorite solution because of the high electrolyte concentration, we suspected that rates of epoxidation might be affected by rates of transfer of styrene from a separate liquid phase through the water to the catalyst particles. Therefore several different techniques were used to agitate the reaction mixtures and thereby vary the mass transfer rate (see Table IV). More efficient mixing gives more efficient mass transfer.[13] Because wrist-action shaking of a test tube and magnetic stirring in a round-bottom flask gave highest activities of the latex-bound catalyst, those methods were used in all other experiments to minimize the influence of mass-transfer rates on the results of experiments in which we studied

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Table V. Effects of pH and Source of Hypochlorite[a]

ClO^- source	$[OH^-]$, M[b]	[NaCl], M[c]	% styrene oxide
Clorox	3.2×10^{-5} [d]	0.65[d]	50
Clorox	6.5×10^{-4}	0.63	61
Clorox	0.01	0.62	54
Clorox	0.06	0.57	74
fresh[e]	0.25	0.16	63
fresh[f]	0.29	0.19	72
Clorox	0.38	0.25	79
Clorox	0.81	0.25	69

[a] Standard conditions were employed with 40.4 mg of latex L-10 and shaking of 6.0 mL of reaction mixture. [b] $[OH^-] = 6.5 \times 10^{-4}$ M from bleach solution of pH 11.3 plus added NaOH. [c] [NaCl] = 0.25 M from bleach solution (estimated from the reaction of 2 mol of NaOH with 1 mol of Cl_2, which gives 1 mol of NaOCl, NaCl, and H_2O each) plus added NaCl. [d] Porphyrin 1 = 0.505 mM [85% of porphyrin rings contain Mn(III); pH = 9.5; [NaCl] = 0.25 M from bleach solution plus added NaCl (1.35 mmol) and HCl (0.91 mmol). [e] 9.2 mL of reaction mixture. [f] 7.9 mL of reaction mixture.

other variables. Two other attempts to improve styrene mass transfer did not increase the observed oxidation rates: An ultrasonic water bath was ineffective, perhaps due to coagulation of colloidal particles.[14] Addition of 0.15 volume fraction of methanol to increase the solubility of styrene in the aqueous phase reduced the rate of epoxidation.

Pyridines and *N*-alkylimidazoles often have improved catalytic activity of metalloporphyrin oxidation catalysts.[7,8] A strong donor ligand on the metal increases its reactivity with electrophilic oxidizing agents. Pyridine and 1-methylimidazole, present in 50-fold molar excess over soluble catalyst 1 in 0.7 M NaOH reaction mixtures, did not affect the rate of epoxidation, probably because oxide (O^-) already serves the purpose of a strong donor ligand to the manganese.

Table V reports experiments carried out at various hydroxide ion concentrations. Since the commercial laundry bleach also contained 0.25 M NaCl, most of the experiments were carried out at constant ionic strength with [NaOH] + [NaCl] = 0.63 M. Highest yields were attained at 0.06–0.38 M NaOH, due to greater stability of the porphyrin in highly alkaline solutions. Since the composition of NaOCl solutions may change over long periods of time, experiments were performed using NaOCl freshly prepared from equimolar amounts of NaOH and chlorine. There was no major difference between the activity of a freshly prepared NaOCl solution and Clorox of uncertain age.

Further experiments were carried out to test activity over longer reaction time and during repeated use of the catalyst. Figure 1 shows diminished catalytic activity after 60 min, and in a separate experiment the yield of styrene oxide was unchanged after 120 min. When new portions of styrene and NaOCl solution were added to a reaction mixture after 60 min, the yield of styrene oxide from the second portion of styrene was 48% (63% overall yield from both portions of styrene). In another longer test of catalyst activity, a reaction was run with excess styrene (19.2 mmol), and 1.50-mmol portions of hypochlorite were added at 11-min intervals. During the first 100 min, 2.3 mmol of styrene oxide was produced, but only another 0.2 mmol was formed during the next 240 min, and after that time the color of the mixture was yellow rather than the red-brown color of the original porphyrin.

Other manganese compounds were catalytically inactive: $MnCl_2$, $Mn(OAc)_2$, and 5,10,15,20-tetrakis(4-sulfonatophenyl)porphinatomanganese(III) acetate (4). The green-brown color of the latter bleached in seconds under the reaction conditions.

Table VI. Epoxidations of Substituted Styrenes[a]

substrate	% epoxide,[b]	k_{rel}[c]
α-methylstyrene	37	0.46
4-chlorostyrene	22	0.27
4-methylstyrene	18	0.38
β-methylstyrene	<5[d]	–

[a] Standard conditions were used with porphyrin 1 = 3.01×10^{-3} mmol [85% of porphyrin rings contain Mn(III)], 40.4 mg of latex L-10, and shaking of 6.0 mL of reaction mixture. [b] Determined by GLC analysis. [c] Ratio of rate of epoxidation of substituted styrene to rate of epoxidation of styrene from 1.20 mmol each of the two styrenes and 1.50 mmol of hypochlorite. [d] Product was not identified.

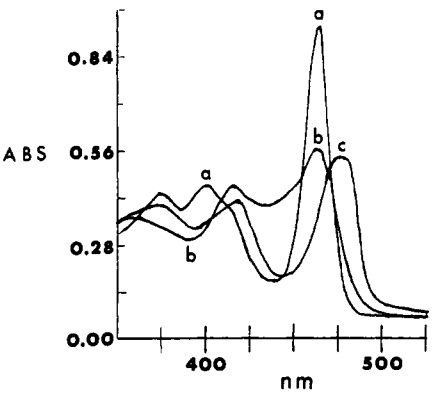

Figure 2. Absorption spectra of 9.90×10^{-5} M porphyrin 1 (a) at pH 7.0 and (b) in 0.38 M NaOH, and (c) 1.30×10^{-4} M 1 at pH 9.0 with 8.5 mg of latex L-10 per milliliter. The absorbance of c is 1.43 times that shown.

Substituted styrenes were epoxidized slower than styrene. Table VI reports both single substrate and competition experiments. Since the lesser reactivity of both 4-chlorostyrene and 4-methylstyrene cannot be explained by substituent effects in any one reaction mechanism, we suspect that lower solubility of the substituted styrenes in the latex is responsible.

Attempts to epoxidize other alkenes, under the standard conditions that produced 75–79% of styrene oxide, produced no detectable epoxide from 1-decene and only traces of epoxides from 1-octene, cyclooctene, and *cis*-stilbene. Attempts to improve the yield of cyclooctene oxide by addition of 50% methanol, 50% acetonitrile, and 22% acetonitrile as cosolvents gave yields of less than 1%, 4%, and 6%, respectively.

Visible Spectra of Porphyrin 1. Carnieri, Harriman, and Porter analyzed equilibria of water-soluble 5,10,15,20-tetrakis(aryl)manganese(III) porphyrins (aryl = 4-carboxyphenyl, 4-sulfonatophenyl, 4-pyridyl, and *N*-methyl-4-pyridyl) in the range $9 < pH < 14$ by visible spectrophotometry, oxidation with many oxidizing agents, electrochemical oxidation–reduction potentials, and magnetic moments.[15] Many of their results are similar, and their conclusions are applicable to our observations.

Spectra of 1 were obtained with the sample that contained 0.85 M Mn per porphyrin. Spectra in pH 7.0 and in 0.38 M sodium hydroxide solutions are shown in Figure 2. An absorbance maximum at 472 nm is typical of Mn(III) porphyrins, and the spectra resemble closely that of 5,10,15,20-tetrakis(4-carboxyphenyl)porphinatomanganese(III) at pH 14.[15] Spectra of 1 with varied pH showed that the species present in neutral solutions

(14) Doktycz, S. J.; Suslick, K. S. *Science* **1990**, *247*, 1067.

(15) Carnieri, N.; Harriman, A.; Porter, G. *J. Chem. Soc., Dalton Trans.* **1982**, 931.

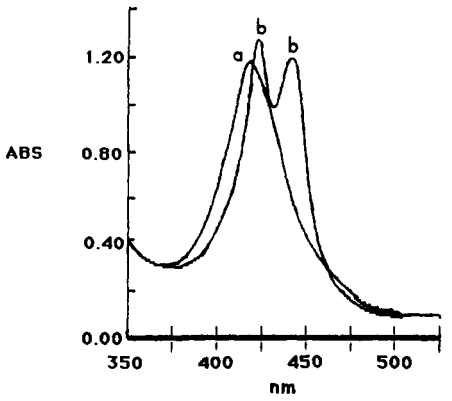

Figure 3. Absorption spectra of 1.30×10^{-4} M porphyrin 1 in 0.065 M NaOCl and 0.38 M NaOH in the presence of (a) no added latex and (b) 8.5 mg of latex L-10 per milliliter.

changed at pH 12–13 to the form present in strongly basic solutions. The spectrum in strong base may be attributed to deprotonation of two molecules of water ligated to manganese.[15,16] When latex was added to a solution of 1 at pH 9, the 464-nm peak shifted to longer wavelength and broadened, and the band at 402 nm also shifted to longer wavelength, as shown in Figure 2c. The changes in the spectrum of 1 at pH 9 in the latex could be due to changes in ligation or oxidation state of Mn or to the polymer environment. In contrast to the spectra of the Mn porphyrin 1, the spectra of the unmetallated porphyrin (3) had a maximum at 414 nm ($\epsilon = 2.6 \times 10^5$) independent of pH that shifted to 422 nm ($\epsilon = 2.1 \times 10^5$) upon addition of latex.

In a basic sodium hypochlorite solution such as that used for epoxidations in the absence of latex, 1 has a broad absorption band at 416 nm (Figure 3a) that may be attributed to a Mn(V)=O species. Carnieri, Harriman, and Porter[15] found that at pH 14 oxidation of 5,10,15,20-tetrakis(4-carboxyphenyl)porphinatomanganese(III) with 3 equiv of hypochlorite produced a stable Mn(V)=O species with an absorption maximum at 420 nm, and surprisingly, the oxidized Mn porphyrin failed to react with added Mn(III) porphyrin. Although a long-lasting monomeric Mn(V)=O species is formed in basic aqueous hypochlorite, Mn(V) species are not usually observed under catalytic conditions. A μ-oxo dimer, Mn(IV)–O–Mn(IV), is formed with milder aqueous oxidants,[15] and tetramesitylporphinatomanganese(III) reacts rapidly with the corresponding Mn(V)=O species in organic solvent to produce the μ-oxo dimer.[17]

Addition of latex to a basic hypochlorite solution of 1 gave a band at 423 nm (Figure 3b) which we assign to the same Mn(V)=O species in the latex that is present in aqueous solution with a small contribution from the 15% of unmetalated porphyrin (3), and a band at 441 nm which we assign to a new oxidized Mn species formed only in the latex. During epoxidation of styrene in the presence of latex the two bands at about 423 and 441 nm persisted for about 30 min, and then quickly the mixture changed color from brown-red to green as a new absorption band at 472 nm appeared in place of the band at 441 nm (Figure 4). Since the catalytic activity of 1 also decreased markedly at about this time, the new absorption band is probably due to an inactive porphyrin derivative. The inactive species formed during hypochlorite epoxidations catalyzed by hindered Fe(III) porphyrins have been identified as N-alkylated hemins,[18] but we have not tried to identify any product from 1.

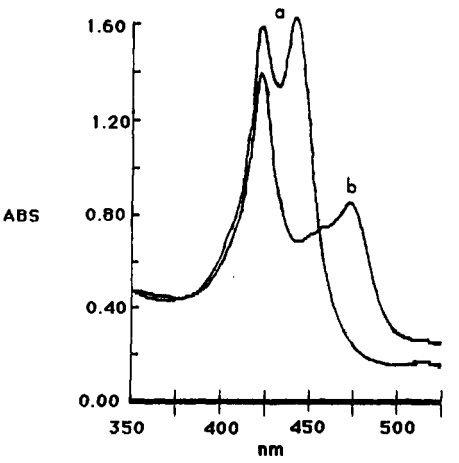

Figure 4. Decreasing absorption of 2.0×10^{-4} M porphyrin 1 bound to the latex in a reaction mixture containing 0.25 M NaOCl and 0.38 M NaOH at (a) 27 min and (b) 44 min after addition of styrene.

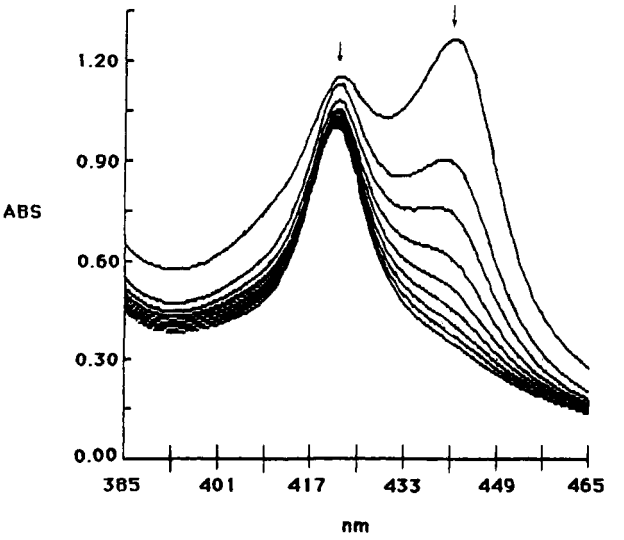

Figure 5. Decreasing absorption of 1.32×10^{-5} M porphyrin 1 bound to 0.18 mg of latex L-10 per milliliter in 0.25 M NaOCl and 0.38 M NaOH in 30-min intervals from 1 to 241 min after preparation.

Table VII. Half-Lives of Porphyrins 1 and 4[a]

porphyrin	conditions	$t_{1/2}$[b]
1	pH 10.8, no latex	57 ± 2 min
1	[NaOH] = 0.38 M, no latex	19.4 ± 1.1 h
1	pH 10.8, latex	22 ± 1 min
1	[NaOH] = 0.38 M, latex	13.5 ± 1.7 h[c]
1	[NaOH] = 0.38 M, latex	41 ± 3 min[d]
4	pH 10.8, no latex	<20 s
4	pH > 13, no latex	<60 s

[a] Determined from least-squares fit to first-order disappearance of the strongest visible absorption band relative to a base line drawn tangent to the minima at either side of the band. The samples contained $(1.16–1.33) \times 10^{-5}$ M 1, 18 mg of latex, and 0.25 M NaOCl, and the temperature was 30.0 °C. [b] Error limits are one standard deviation from least-squares value. [c] Band at 423 nm. [d] Band at 441 nm.

(16) Harriman, A.; Porter, G. *J. Chem. Soc., Faraday Trans. 2* **1979**, *75*, 1532.
(17) Lee, R. W.; Nakagaki, P. C.; Bruice, T. C. *J. Am. Chem. Soc.* **1989**, *111*, 1368.
(18) (a) Brothers, P. J.; Collman, J. P. *Acc. Chem. Res.* **1986**, *19*, 209. (b) Mashiko, T.; Dolphin, D.; Nakano, T.; Traylor, T. G. *J. Am. Chem. Soc.* **1985**, *107*, 3735. (c) Traylor, T. G.; Nakano, T.; Miksztal, A. R.; Dunlap, B. E. *J. Am. Chem. Soc.* **1987**, *109*, 3625. (d) Collman, J. P.; Hampton, P. D.; Brauman, J. I. *J. Am. Chem. Soc.* **1990**, *112*, 2977.

Table VIII. Concentrations under Catalysis Conditions in Solution and in Colloidal Particles

component	concn in solution, M	concn in latex, M[a]
porphyrin	3.7×10^{-4}	1.54×10^{-2}
manganese	2.4×10^{-4}	1.00×10^{-2}
initial ClO^-	0.25	0.62
HO^-	0.38	0.05
Cl^-	0.28	0.34
styrene	$<7 \times 10^{-4}$	est. $\gg 10^{-4}$

[a] Estimated as explained in text.

Decreases in intensities of the absorption maxima of the visible spectra of 1 in basic hypochlorite solutions were used to determine rates of disappearance of the species presumed to be a Mn(V)=O porphyrin. The half-lives reported in Table VII show a much longer lifetime of oxidized 1 in 0.38 M NaOH than at pH 10.8. In the presence of latex the two strongest absorption bands of 1 disappeared at much different rates, as reported in Table VII and illustrated in Figure 5. The 41-min half-life for disappearance of the 441-nm band is consistent with the time for loss of activity, whereas the 13.5-h half-life of the 423-nm band indicates that it is not sensitive to change of the catalytic species. The greater stability of 1 in strong base accounts for the higher conversions of styrene to styrene oxide in 0.38 M NaOH. The rapid disappearance of visible absorption of a basic hypochlorite solution of 5,10,15,20-tetrakis(4-sulfonatophenyl)porphinatomanganese(III) acetate (4) accounts for its lack of catalytic activity.

Mechanism of Catalysis. The simplest explanation for the higher activity of porphyrin 1 bound to ion-exchange latexes is that the latex serves to concentrate both reactants and catalyst into the polymer phase of the reaction mixture. Although the dry weight of polymer was only 40 mg per 6 mL of reaction mixture, the 1% cross-linked particles are expected to swell to about 4 times their dry volume in water (on the basis of similar degrees of swelling of ion exchange resins of the same cross-link density and quaternary ammonium ion content). Estimates of the concentrations of the reactants and catalyst in the active phase during the solution experiments and the latex experiments are listed in Table VIII. The latex has available 0.153 mmol of quaternary ammonium sites, of which 0.0076 mmol are occupied by porphyrin 1. Chloride, hydroxide, and hypochlorite compete for the remaining 0.145 mmol of binding sites. The selectivity ratio for binding of chloride vs hydroxide is about 10, and we estimate that the selectivity ratio for hypochlorite vs chloride is about 2 (equal to the selectivity ratio for bromide vs chloride).[19] Since the amounts of anions in the mixture are 1.50 mmol of hypochlorite, 2.28 mmol of hydroxide, and 1.65 mmol of chloride, the amounts bound are estimated to be 0.089 mmol of hypochlorite, 0.007 mmol of hydroxide, and 0.049 mmol of chloride. Assuming the density of the dry catalyst is equal to that of a commercial anion exchange resin in Cl^- form (1.11 g mL^{-1}), the concentration of hydroxide in the latex is about 0.047 M, lower than in the external solution. The concentration of hypochlorite in the latex is about 0.62 M, compared with 0.25 M in the external solution, which accounts for the lesser stability of the porphyrin in the latex.

The active epoxidizing agent in aqueous solution is likely the Mn(V)=O species observed spectrophotometrically. Such species have been proposed before to transfer an oxygen atom stereospecifically to *cis*-β-methylstyrene.[20] We were not able to test the stereospecificity with β-methylstyrene because it did not react. The low reactivity of styrene in aqueous solution is due to its low solubility. Two forms of the manganese porphyrin appeared to be present in latex particles under styrene epoxidation conditions, one with a visible absorption maximum at about 423 nm which may be the same Mn(V)=O species that exists in solution, and another with a maximum at about 441 nm that converts to a new species with a maximum at 472 nm concurrently with a marked decrease in catalytic activity. Previously the binding of (tetraphenylporphinato)manganese(III) to soluble poly(vinylpyridine) and to an insoluble polyisocyanide increased its activity for phase-transfer catalyzed oxidation of cyclohexene with aqueous hypochlorite.[8] The increased activity was attributed to suppression of μ-oxo dimerization of the Mn porphyrin. Our spectral results indicate that the 423-nm band, assigned to the Mn(V)=O species which absorbed at 416 nm in the absence of colloidal polymer, persisted long after catalytic activity ceased. The new unidentified species with the 441-nm absorbance maximum is likely the active catalyst.

Conclusions

(1) Manganese(III) porphyrin 1 is a more active catalyst for epoxidation of styrene with hypochlorite bound to colloidal anion-exchange particles than in aqueous solution because of higher concentration of styrene in the particles than in water, or because a different catalyst species in the particles is more active than the species present in water. (2) Aliphatic alkenes do not react under the conditions that give 71–81% conversion of styrene to styrene oxide, probably because the aliphatic alkenes are less soluble in the particles. (3) Typically 620 mol of styrene oxide are produced in the presence of excess oxidant, but further catalytic activity is limited by oxidative degradation of the porphyrin. (4) The porphyrin 1 is more stable in strongly basic 0.38 M NaOH solutions than in less basic sodium hypochlorite solutions.

Experimental Section

Materials. Deionized, glass-distilled water was used for all experiments. 4-Methylstyrene (Mobil Chemical Co.) was distilled under reduced pressure. 4-Chlorostyrene and α-methylstyrene (Aldrich) were passed through 10-cm alumina columns. Styrene was washed with 0.1 N NaOH and distilled under reduced pressure. Clorox bleach was obtained from a local supermarket, and its OCl^- content was determined by iodometric titration.[21] A freshly prepared NaOCl solution[22] was analyzed by iodometric titration to contain 2.09% NaOCl by weight (0.297 M). The tetrasodium salt of 5,10,15,20-tetrakis(4-sulfonatophenyl)-porphyrin (Strem Chemicals) was converted to the Mn(III) acetate (4) by the method 2 used below for preparation of 1. 1-Octene (Aldrich), 1-decene (Fluka), and cyclooctene (Fluka) were used as received. Silica gel (I.D. Grade 62, 60–200 mesh) was obtained from E. M. Science.

Analyses. Microanalyses of porphyrins dried under vacuum at 60 °C for 4 h were performed by Galbraith Laboratories, Knoxville, TN. Gas chromatography was performed with a 6 ft × $^1/_8$ in. Tenax packed column from Supelco Inc. and thermal conductivity detection. UV/visible spectra were obtained with 1 and 10 mm pathlength cells for 10^{-4} and 10^{-5} M porphyrin samples. The cell holder was thermostated, and temperature was measured in the reference cell, which contained water during

(19) Helfferich, F. *Ion Exchange*; McGraw-Hill: New York, 1962; p 175.

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(22) Audrieth, L. F.; Nickles, T. T. *Inorganic Syntheses*; Booth, H. S., Ed.; McGraw-Hill: 1939; Vol. I, p 90.

homogeneous solution studies and latex with no porphyrin during latex-bound Mn porphyrin studies.

5,10,15,20-Tetrakis(2,6-dichlorophenyl)porphyrin (2). This typical procedure is based on the method of Lindsey.[9-11] With minor modifications it was carried out 7 times with yields in the range 10–16%. Glassware was oven-dried at 130–135 °C, protected by a drying tube during cooling, and purged with argon. Pyrrole 99% (Aldrich) was distilled under reduced pressure at 98 °C and stored under argon over 4-Å molecular sieves at 5 °C protected from light. Boron trifluoride etherate (Alfa) was distilled under reduced pressure (1.5 mmHg/27 °C) and stored under argon over 4-Å molecular sieves at 5 °C. 2,6-Dichlorobenzaldehyde (99%), *p*-chloranil (99%), and manganese acetate tetrahydrate (all from Aldrich), and manganese chloride tetrahydrate (Mallinckrodt) were used as received. Amberlite IR-120 and Dowex 50-X8 were obtained from Rohm and Haas Co. and J. T. Baker Chemical Co. Reagent grade $CHCl_3$ (Fisher) was distilled from K_2CO_3 (50 g/L of $CHCl_3$) under argon over 4-Å molecular sieves and protected from light. (Recently it was found that ethanol in $CHCl_3$ was essential in the synthesis of 5,10,15,20-tetrakis(2,4,6-trimethylphenyl)porphyrin.[11] Simple distillation did not remove ethanol from $CHCl_3$.

A 2-L flask fitted with a condenser, a gas inlet port, and an addition port was charged with 1 L of $CHCl_3$. With magnetic stirring pyrrole (680 mg, 10.1 mmol) was added via a syringe, 2,6-dichlorobenzaldehyde (1.768 g, 10.1 mmol) was added as a solid, and BF_3 etherate (561 mg, 4.0 mmol) was added via a syringe at about 5-min intervals. The mixture was stirred at 22 °C for 21 h with protection from light. Powdered *p*-chloranil (1.863 g, 7.6 mmol, 3 equiv per porphyrinogen, 0.75 equiv per pyrrole) was added all at once, and the mixture was refluxed for 3 h under air. After the mixture cooled, the BF_3 etherate was hydrolyzed with water, the aqueous phase was separated, and the organic phase was rotary evaporated to dryness. Methanol was mixed with the crude dry product until no big particles remained, and the slurry was filtered by gravity through Whatman 42 filter paper to collect a purple-bluish residue of **2**. (All components except **2** were soluble in methanol.) The filtrates were passed through the filter 3–4 times until no more purple residue could be collected. Each filtration took about 0.5 h. TLC on silica gel in 30% ethyl acetate–petroleum ether showed impurities at R_f = 0.32–0.69 along with the intense yellow-green spot of **2** at R_f = 0.0. When the TLC was run with dichloromethane, porphyrin **2** had R_f = 0.91, and the impurities had R_f = 0.28–0.0. The residue recovered from filtration was dissolved in CH_2Cl_2, adsorbed onto 5 g of silica gel, and poured onto a 60 × 2.5 cm column of silica gel packed in CH_2Cl_2. The column was washed with CH_2Cl_2 to elute the red-brown band of **2**. TLC analysis of the eluate showed a single yellow spot which moved with the solvent front in CH_2Cl_2 and stayed at the origin in 33% ethyl acetate–petroleum ether. Porphyrin **2** was recovered by evaporating the solvent and dried under vacuum at 65–70 °C for 12 h. The ^1H NMR spectrum of the product showed three small impurity peaks at 0.8–1.7 ppm along with expected peaks of **2**. The impure porphyrin was chromatographed again through silica gel with 50% ethyl acetate–petroleum ether and with ethyl acetate (in which the porphyrin is insoluble) to remove impurities, and with CH_2Cl_2 to recover **2**. Recrystallization from $CHCl_3$/heptane at 5 °C, gravity filtration, and drying yielded 16% of **2** based on 2,6-dichlorobenzaldehyde. ^1H NMR (300 MHz, $CDCl_3$, $(CH_3)_4Si$): δ 8.67 (s, 8 H, β-pyrrole-H), 7.79 (d, J = 8.4 Hz, 8 H, *m*-Ph-H), 7.70 (t, J = 8.0 Hz, 4 H, *p*-Ph-H), −2.53 (*N*-pyrrole-H), 1.56 (s, unidentified impurity). IR (KBr): 3420 (N—H stretch), 3300, 3080 (=C—H stretch), 1545 (C=C stretch), 1420, 1330, 1180, 960, 795, 770, 705 cm^{-1}. MS (FAB, DMSO/glycerol): m/e 891 (36), 855 (9, −Cl), 819 (5, −2Cl), 748 (7, −4Cl), 536 (6), 369 (17), 277 (100), 263 (29), 235 (26). Most abundant ion in isotopic cluster calcd for $C_{44}H_{22}N_4Cl_8$ 890.79. Anal. Calcd for $C_{44}H_{22}N_4Cl_8$ (FW 890.31): C, 59.36; H, 2.49; N, 6.29; Cl, 31.86. Found: C, 59.55; H, 2.55; N, 6.18; Cl, 32.12.

5,10,15,20-Tetrakis(2,6-dichloro-3-sulfonatophenyl)porphyrin (3). Using the sulfonation method of Fleischer[23] powdered porphyrin **2** (238 mg, 0.27 mmol) and concentrated H_2SO_4 (7 mL) in a 25-mL single-neck round-bottomed flask equipped with a condenser and an egg-shaped stirring bar were heated for 6 h with a 185 ± 5 °C oil bath. The mixture was cooled to room temperature, stirred for 24 h, and cautiously diluted with 40–45 mL of distilled water with stirring to precipitate the porphyrin. If more water was used, the precipitate dissolved to give a green solution. The green precipitate (the HSO_4^- salt of disulfonated porphyrin **2**[23]) was vacuum filtered in 2–3 h through a 60-mL 40F fritted funnel. The light green filtrate was discarded, and the hygroscopic solid **3**, which contained a small amount of water and H_2SO_4, was dissolved in methanol. The solvent was evaporated, and the residue was triturated three times with acetone, in which the sulfonated porphyrin has low solubility, to remove water and H_2SO_4. After drying, 291 mg of product **3** was obtained (87% yield based on porphyrin containing 3.5 SO_3^- and 4 H_2O molecules per porphyrin). ^1H NMR (CD_3OD, $Me_3Si(CH_2)_2CO_2^-Na^+$): δ 9.03 (m, 8 H, β-pyrrole-H), 8.63 (m, 4 H, *p*-Ph-H), 8.05 (m, 4.5–5 H, *m*-Ph-H), 5.87 (s, H_2O), 3.27 (s, CHD_2OD), 1.30 and 0.87 (impurities). Integration of the phenyl ring and pyrrole ring signals indicated 3.5 SO_3^- groups per porphyrin ring and substitution 75% at the 3-position and 25% at the 4-position of the phenyl rings. MS (FAB, dithiothreitol–dithioerythritol): m/e 1212 (6, −2Cl), 1134 (12), 1057 (15, $-C_6H_2Cl_2SO_3H$), 1050 (10), 891 (39, $-3.5SO_3H \times 4H_2O$), 759 (18), 697 (33), 565 (100).

Anal. Calcd for $C_{44}H_{18.5}N_4Cl_8(SO_3H)_{3.5}(H_2O)_4$ (FW 1242.6): C, 42.53; H, 2.43; N, 4.51; Cl, 22.83; S, 9.03. Found: C, 42.29; H, 2.63; N, 4.69; Cl, 22.71; S, 9.05.

Tetrasodium Salt of 5,10,15,20-Tetrakis(2,6-dichloro-3-sulfonatophenyl)porphinatomanganese(III) Chloride (1). Method 1. Porphyrin **3** in H^+ form (280.6 mg, 0.23 mmol) and $MnCl_2 \cdot 4H_2O$ (396.3 mg, 2.0 mmol) were dissolved in 20 mL of distilled water. The pH of the mixture was adjusted to 8.7 with 1 N NaOH solution. The mixture was heated with an 85 ± 2 °C oil bath. Every 7–8 h, the pH was measured, and more 1 N NaOH was added to raise the pH from 6.0–6.5 to 8.5–9.0. After 24 h the pH 8.7 mixture was evaporated to dryness, the porphyrin part of the residue was dissolved in methanol, and the mixture was filtered through a 60-mL 40F fritted funnel to remove insoluble salts. A 20 × 1 cm Dowex 50-X8 column was converted to Na^+ form with NaOH and washed with distilled water extensively until the pH of eluate dropped to 6.5. The methanol solution was passed through the column, and a mixture of the manganese porphyrin **1** and NaCl was eluted. After evaporation the residue was dried in vacuum at 65 °C, and was Soxhlet extracted twice with HPLC grade methanol. The yield of recovered, dried product was 355 mg (0.22 mmol, 96%). The elemental analysis corresponded with 85% manganese incorporation and 1 mol of NaCl per porphyrin. Anal. Calcd for $C_{44}H_{16.9}N_4Cl_{8.85}Mn_{0.85}(SO_3Na)_{3.4}(H_2O)_{12}(NaCl)$ (FW 1587.0): C, 33.30; H, 2.60; N, 3.53; S, 6.87; Mn, 2.94. Found: C, 32.83; H, 2.32; N, 3.05; S, 6.70; Mn, 2.95. The nonintegral numbers of H, Cl, and Mn in the calculated formula are due to partial metalation and sulfonation of the porphyrin.

Method 2. A solution of **3** in H^+ form (221.7 mg, 0.18 mmol) and $MnCl_2 \cdot 4H_2O$ (307.4 mg, 1.55 mmol) in 20 mL of water was adjusted to pH 9.0 with 0.5 N NaOH solution, and heated for 24 h with an 85 ± 2 °C oil bath. During the reaction, the pH was raised three times to 9.0 with 0.5 N NaOH. After cooling 0.5 N NaOH solution was added to attain pH 11.5. The brown mixture was evaporated to dryness. The residue was dissolved in methanol and passed through a 40 × 1 cm column of Amberlite IR-120 (H^+ form). The eluate was collected in a receiver containing 0.62 mmol of NaOH solution to neutralize the sulfonic acid groups of the manganese porphyrin. The neutralized filtrate was evaporated to dryness, dissolved in methanol, and vacuum filtered through a 60-mL 40F fritted funnel. The filtrate was evaporated, and the residue was Soxhlet extracted twice with HPLC grade methanol. Evaporation followed by drying in vacuum at 65 °C left porphyrin **1**, with an analysis corresponding with 65% metal incorporation and 11 mol of NaCl per porphyrin. Anal. Calcd for $C_{44}H_{17.2}N_4Cl_{8.65}Mn_{0.65}(SO_3Na)_{3.5}(H_2O)_{12}(NaCl)_{11}$ (FW 2164.0): C, 24.42; H, 1.92; N, 2.59; Cl, 32.19; S, 5.19; Na, 15.40; Mn, 1.65. Found: C, 24.55; H, 2.08; N, 2.22; Cl, 32.31; S, 4.91; Na, 17.70; Mn, 1.65. The non-integral numbers of H, Cl, and Mn in the calculated

(23) Fleischer, E. B.; Palmer, J. M.; Srivastava, T. S.; Chatterjee, A. *J. Am. Chem. Soc.* **1971**, *93*, 3162.

formula are due to partial metalation and sulfonation of the porphyrin.

Binding of 1 to Colloidal Anion-Exchange Resins. Porphyrin 1 (1.25 mL, 2.22×10^{-3} mmol) solution was added slowly via a pipet to a latex (1.0 mL, 6 mg, $n_{R_4N^+Cl^-} = 2.53 \times 10^{-2}$ mequiv) while it was sonicated. The ratio of SO_3^- to R_4N^+ was 0.3 in the dispersion. After sonication for about 15 min the dispersion was ultrafiltered in an all glass system (Millipore) through a 0.1-μm cellulose acetate/nitrate membrane. The red-brown residue was washed with distilled water. The filtrate was diluted to 25 mL. Its visible spectrum showed 0.04% of the original porphyrin 1. However, there was about a 5-nm shift to $\lambda_{max} = 468$ nm in the spectrum relative to the $\lambda_{max} = 463$ nm spectrum of the original solution.

Binding of 1 to IRA-420C. Porphyrin 1 (4 mL of a 0.907 mM aqueous solution) and 83 mg of hydrated (40.4 mg dry) Amberlite IRA-420C were shaken for 100 h. The mixture was filtered, and from the visible absorption spectrum of the filtrate the amount of 1 bound was calculated to be 3.4 mmol.

Epoxidation of Styrene. A 25-mL single-neck round-bottomed flask was charged with pentadecane (internal standard, 60.9 mg) and latex (1.0 mL, 40.4 mg). With the flask in an ultrasonic water bath, an aqueous stock solution of 1.78×10^{-3} M porphyrin 1 (1.25 mL, 2.22×10^{-3} mmol), NaOH (0.76 mL, 2.30 mmol), and distilled water (0.90 mL) was added. (Without latex the sonication step was omitted.) The flask was equipped with a stirring bar and an adapter fitted with a septum and glass stopcock and was flushed with argon. Styrene (125 mg, 1.20 mmol) was added via a syringe. Argon-purged Clorox solution (1.95 mL, 1.50 mmol) was added via a syringe. The pH of Clorox was 11.3. The flask was protected from light by covering with a black sheet, and its contents were stirred for 1 h at ambient temperature. Alternatively the mixture was prepared in a 20-mL test tube, and the tube was sealed with a septum, wrapped with Teflon tape, and shaken with a wrist-action shaker. The reaction mixture was transferred to a 12-mL centrifuge tube with a screw cap. A few milliliters of diethyl ether was added, and the tube was shaken vigorously and centrifuged with a bench top centrifuge. The top organic phase was transferred to another flask with a disposable pipet. The extraction process was repeated 4-6 times until no styrene or product peak was seen on GLC chromatograms of the extract. The combined extract was analyzed quantitatively by GLC. Response factors of 0.99 and 0.91 relative to pentadecane were used for styrene and styrene oxide. The products were identified by ^{1}H NMR spectroscopy.

Epoxidations of substituted styrenes were performed by the same procedures. Only two components were detected from the reaction mixtures by GLC, and ^{1}H NMR spectra of the recovered product mixtures showed only the substituted styrene and its epoxide.

Spectrophotometry of Reacting Mixtures. Samples (200 μL) from test tube reaction mixtures were withdrawn with a syringe at timed intervals and diluted to 500 μL with water. The UV/visible spectrum of each sample was obtained immediately in a 1-mm path length cell. The samples were handled under air and light during preparation.

Acknowledgment. We thank the U.S. Army Research Office for support of this research and the government of Turkey for a scholarship supporting the Ph.D. study of Hayrettin Turk. The high-resolution mass spectrometer was supported in part by National Science Foundation grant BBS-8704089.

Registry No. 1, 131214-66-9; 3, 120644-24-8; $PhCH{=}CH_2$, 100-42-5; $PhC(CH_3){=}CH_2$, 98-83-9; p-$ClC_6H_4CH{=}CH_2$, 1073-67-2; p-$MeC_6H_4CH{=}CH_2$, 622-97-9; NaOCl, 7681-52-9; styrene oxide, 96-09-3; (chloromethylstyrene-divinylbenzene) copolymer-trimethylamine salt, 63453-89-4.

Assessment of the Active-Site Requirements of Lanosterol 14α-Demethylase: Evaluation of Novel Substrate Analogues as Competitive Inhibitors

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Received May 18, 1990

Lanosterol 14α-demethylase ($P450_{14DM}$), a cytochrome P450 enzyme, is responsible for the first stage in the biosynthesis of cholesterol (1) from lanosterol (2). Inhibitors of $P450_{14DM}$ may have therapeutic use in the treatment of familial hypercholesterolemia or as antifungal agents. The specificity of $P450_{14DM}$ has been investigated by using substrate analogues modified at the C-32 carbon. The hitherto undescribed 14α-ethyl and 14α-propyl analogues 15 and 13 of lanost-7-en-3β-ol, as well as the 14α-ethenyl and 14α-prop-2-enyl analogues 14 and 12, have been synthesized. These all proved to be good competitive inhibitors of the enzyme. A series of 32-oxiranes and 32-thiiranes was then synthesized and evaluated as inhibitors. Oxiranes 4 and 5 were excellent stereoselective competitive inhibitors of $P450_{14DM}$. The (2'*S*)-32-oxirane 4 had $K_i = 0.62$ μM, and the (2'*R*)-32-oxirane 5 showed $K_i = 2$ μM. The (2'*R*)-32-thiiranyl and (2'*S*)-32-thiiranyl compounds 10 and 11 were considerably less potent inhibitors. Comparison of the K_i values for analogues 12-15, also good competitive inhibitors of this enzyme, indicated the $P450_{14DM}$ active site to be relatively insensitive to the size and degree of unsaturation of C-14α alkyl substituents up to and including propyl.

Lanosterol 14α-demethylase ($P450_{14DM}$), a cytochrome P450 enzyme, is responsible for the first stage in the biosynthesis of cholesterol (1) from lanosterol (2) (Scheme I).[1] The 14α-demethylation involves three sequential steps, each requiring 1 mol of molecular oxygen and 1 mol of NADPH, with subsequent formation of 32-hydroxy and 32-oxo intermediates. The intermediate for the third step is still not established. There has been much recent interest in $P450_{14DM}$ since inhibitors of this enzyme may have therapeutic use in the treatment of familial hypercho-

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0022-3263/91/1956-1260$02.50/0