Chloroperoxidase as Enantioselective Epoxidation Catalyst: An Efficient Synthesis of (*R*)-(−)-Mevalonolactone†

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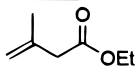
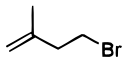
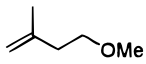
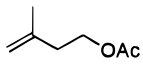
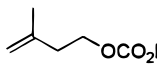
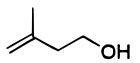
As an important intermediate in biosynthetic pathways leading to sterols, terpenes, corotenoids, and other pentanoid compounds,[1] (*R*)-(−)-mevalonic acid or especially its lactone form (**4**) has been a synthetic target of considerable interest. A number of chemical methodologies have been described, the most popular of which involves asymmetric induction provided by Sharpless epoxidation of a suitable allylic alcohol.[2] Chiral pool chemistry has been employed, either as chiral auxiliary[3] or as reagent[4] or involving Seebach's "self-reproduction of stereogenic centers".[5] Other interesting methodologies include the use of a chiral sulfoxide[6] and axially dissymmetric binaphthyldiamines,[7] although in these two cases the resulting ee was relatively low.

Enzymes have been quite useful for the preparation of (*R*)-mevalonolactone. Lipase-catalyzed kinetic resolutions[8] and esterase hydrolysis of a prochiral diester[9] have proven effective. Attempts to oxidize a triol with either *Gluconobacter sp.*[10] or horse liver alcohol dehydrogenase[11] have generated the (*S*)-lactone.

Considering the complexity and probable expense of many of these procedures, and given the high enantioselectivities observed with chloroperoxidase-catalyzed epoxidation of methallyl-type substrates,[12] we envisioned a concise route to (*R*)-mevalonolactone (**4**) as illustrated below.

Table 1. Substrates Screened for Enantioselectivity[a]

entry	substrate	%ee of epoxide[b]
1.		93 (*R*)[c]
2.		85
3.		76
4.		44
5.		43
6.		—[d]

[a] Substrate (25 μL), *t*-BuOOH (54 μL), CPO (1.6 mg), 3 mL of 0.01 M Na citrate, 1 h. [b] Determined by chiral GLC. [c] Absolute configuration was proven by conversion to (*R*)-(−)-mevalonolactone. Other compounds were not determined but results so far have been consistently *R* for methallyl-type substrates (see ref 12). [d] No epoxide; complete recovery of starting material.

Table 1 lists 3-methyl-3-butenol and five derivatives. Each was tested for enantioselective epoxidation using chloroperoxidase[13] and *tert*-butyl hydroperoxide as terminal oxidant. The alcohol (entry 6) was recovered unchanged.[14] Moderate facioselectivity observed with the acetate (entry 4) was surprising when compared to far higher selectivity as seen with the reversed ester (entry 1) and with methallyl propionate[12] (94% ee). Each posseses essentially the same molecular length, though the distance between the olefinic and carbonyl moieties is different. How this relates to enantiomer distribution in the active site of chloroperoxidase is currently being investigated in our laboratories.

The substrate providing superior enantioselectivity (entry 1) was epoxidized at a 3.0 mmol scale using 2.0 equiv of *tert*-butyl hydroperoxide and 0.014 mol % chloroperoxidase in 45 mL citrate buffer at rt (Scheme 1). Epoxide **1** was isolated in 67% distilled yield[15] and having an ee of 93%. Hydrocyanation of **1** occurred in 90% yield with no evidence of the other regioisomer. Chemoselective reduction of the cyano ester **2** with sodium borohydride in ethanol[16] produced mevalononitrile (**3**, 98%) which was hydrolyzed and lactonized (81%) to (*R*)-mevalonolactone (**4**).[17] The entire sequence was repeated but without purification of any intermediates providing (*R*)-mevalonolactone in 57% overall yield and identical purity.

† Dedicated to Professor Emeritus Nelson J. Leonard on the occasion of his 80th birthday.

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(13) Chloroperoxidase (EC 1.11.1.10) is commercially available from Sigma Chemical Co. or Chirazyme Laboratories. From the latter source, the 0.44 mmol required for the preparation described herein would currently cost approximately $90.

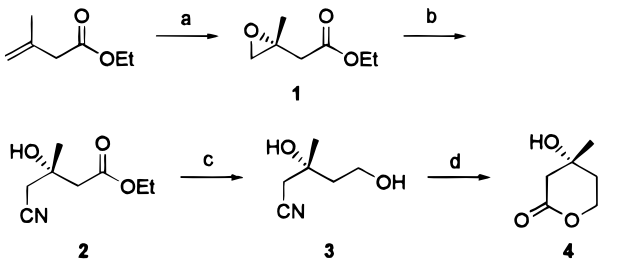
(14) If the alcohol is left to incubate with chloroperoxidase and *t*-BuOOH for 0.5 h, and a known substrate is then added, epoxidation proceeds smoothly with the known substrate. Therefore, we believe 3-methyl-3-butenol does not inactivate the enzyme, but merely fails to act as a substrate. This is consistent with other polar alkenes that we have tested (unpublished results). Preliminary observations of the active site as revealed by an X-ray crystal structure of chloroperoxidase (Sundaramoorthy, M.; Terner, J; Poulos, T. L. *Structure* **1995**, *3*, 1367−77) suggest that two phenylalanine residues flanking the active site may inhibit proper binding orientation through hydrophobic-hydrophilic repulsion. Molecular modeling studies currently underway are directed at testing this hypothesis.

(15) GLC yield using decane as internal standard indicated complete conversion and 95% yield. The reaction currently stands at approximately 6800 turnovers and has not been optimized.

(16) Another example of such chemoselectivity using $NaBH_4$/ethanol: Mauger, J.; Robert, A. *Tetrahedron* **1988**, *44*, 2493−502.

S0022-3263(96)00074-6 CCC: $12.00

Scheme 1[a]



[a] (a) *t*-BuOOH (2.0 equiv), CPO (0.014 equiv), 0.01 M Na citrate, pH = 5.5; (b) KCN (3.0 equiv), F_3CCO_2H (2.0 equiv), EtOH; (c) $NaBH_4$, EtOH; (d) i. NaOH, ii. 10% HCl, MeOH, iii. CH_3SO_3H, THF.

Ethyl 3-methyl-3-butenoate may be prepared by a number of methods, including deconjugation of ethyl 3-methyl-2-butenoate,[18] palladium-catalyzed alkoxycarbonylation of methallyl acetate,[19] and Fischer esterification of the corresponding carboxylic acid.[20] The acid may be obtained by carboxylation of either methallyllithium[20] or methallylmagnesium bromide,[21] or by Jones oxidation of 3-methyl-3-butenol.[22] Strategies involving the introduction of labeled atoms into an intermediate leading to mevalonate[1,5] can be deduced based upon our synthesis combined with the variety of approaches to the starting ester.

To our knowledge, this represents the first multistep synthesis featuring enantioselective epoxidation moderated by chloroperoxidase. Because of chloroperoxidase's ready availability, ease of use, and potential for high selectivity, its use in the synthesis of intermediates difficult to obtain by other methods is a continuing investigation in our laboratories.

Experimental Section

General. Chirazyme Laboratories provided *Caldariomyces fumago* cultures from which chloroperoxidase was isolated by standard methods as a solution in Na acetate buffer with R_z = 1.45 and an enzyme concentration that varied typically from 30−75 mg/mL. Flash chromatography was performed using silica gel (60 Å, 230−400 mesh).

^{1}H and ^{13}C nuclear magnetic resonance (NMR) spectra were recorded at 300 MHz and 75.5 MHz frequency, respectively, with $CDCl_3$ as solvent, unless otherwise indicated. Chiral GLC analyses were obtained with an Astec Chiraldex G-TA column (40 m × 0.25 mm).

Ethyl 3-Methyl-3-butenoate. 3-Methyl-3-butenoic acid[22] (5.0 g, 0.05 mol) was dissolved in 30 mL of ethanol. Methansulfonic acid (1.0 mL) was added, and the mixture was heated to 70 °C for 2 h. After cooling, the solution was poured into pentane (60 mL) and washed twice with water and once with brine. Drying ($MgSO_4$), evaporation of solvent, and distillation (56−57 °C, 30 mmHg) provided 5.3 g of product as a clear, colorless oil (83%): ^{1}H NMR δ 1.25 (t, J = 6.9 Hz, 3H), 1.79 (s, 3H), 3.00 (s, 2H), 4.13 (q, J = 6.9 Hz, 2H), 4.85 (d, J = 18.3 Hz, 2H); ^{13}C NMR δ 14.2, 22.4, 43.5, 60.6, 114.6, 138.6, 171.4; FTIR (thin film) 1738 cm^{-1}.

(*R*)-Ethyl 2-Methyloxiraneacetate (1). To 45 mL of 0.01 M sodium citrate buffer pH = 5.5 were added sequentially and with rapid stirring ethyl 3-methyl-3-butenoate (385 mg, 3.0 mmol), *tert*-butyl hydroperoxide (0.81 mL, 70% aqueous, 6.0 mmol), and chloroperoxidase (250 μL, 0.44 μmol). After 1.5 h, excess oxidant was quenched with 2 mL of concentrated $Na_2S_2O_3$, and the reaction mixture was saturated with NaCl and extracted with ether (3 × 50 mL). Combined ether layers were dried ($MgSO_4$), and the solvent was evaporated. The residue was pressure filtered through a pad of silica gel eluting with 4:1 pentane/ether. Evaporation of solvent gave 423 mg of oil which was distilled in an Aldrich Quick-distill apparatus (20 mm Hg, bath temp = 85 °C) providing 290 mg of **1** as a clear, colorless oil (67%): chiral GLC indicated 93% ee; $[\alpha]^{26}_D = -27.1$ (c = 1.62, $CHCl_3$); ^{1}H NMR δ 1.21 (t, J = 6.8 Hz, 3H), 1.36 (s, 3H), 2.38 (d, J = 15.2 Hz, 1H), 2.61 (d, J = 4.9 Hz, 1H), 2.64 (d, J = 15.2 Hz, 1H), 2.69 (d, J = 4.9 Hz, 1H), 4.1 (q, J = 6.8 Hz, 1H); ^{13}C NMR δ 14.0, 21.1, 25.6, 42.4, 53.5, 60.6, 170.0; FTIR (thin film) 1737 cm^{-1}. Anal. Calcd for $C_7H_{12}O_3$: C, 58.32; H, 8.39. Found: C, 58.23; H, 8.70.

(*S*)-Ethyl 4-Cyano-3-hydroxy-3-methylbutanoate (2).[23] Potassium cyanide (330 mg, 5.1 mmol) was suspended in 3.0 mL of ice-cold ethanol, and trifluoroacetic acid (260 μL, 3.3 mmol) was added slowly, followed by epoxy ester **1** (237 mg, 1.6 mmol). The flask was hermetically sealed and heated to 50 °C for 14 h. The solution was cooled, concentrated, and triturated between 20 mL of ether and 10 mL of brine. Extraction of the aqueous phase with ether (3 × 20 mL), drying ($MgSO_4$), and evaporation of ether was followed by flash chromatography (25% EtOAc in hexanes) giving 253 mg (90%) of oil: $[\alpha]^{23}_D = -5.13$ (c = 1.13, $CHCl_3$); ^{1}H NMR δ 1.27 (t, J = 6.8 Hz, 3H), 2.6 (d, J = 16.8 Hz, 1H), 2.64 (s, 2H), 2.69 (d, J = 16.8 Hz, 1H), 4.19 (q, J = 6.8 Hz, 2H); ^{13}C NMR δ 14.1, 27.0, 30.7, 43.6, 61.3, 117.1, 172.0; FTIR (thin film) 2267, 1728 cm^{-1}. Anal. Calcd for $C_8H_{13}NO_3$: C, 56.13; H, 7.65; N, 8.18. Found: C, 55.99; H, 7.82; N, 7.90.

(*S*)-Mevalononitrile (3). In 4 mL of ethanol was dissolved cyano ester **2** (220 mg, 1.3 mmol). Sodium borohydride (110 mg, 2.9 mmol) was spooned in over three portions, 2 h apart. Stirring continued for 16 h following the final addition. Excess borohydride was quenched with dropwise addition of 1.1 mL of 10% HCl and 10 mL of brine. Extraction with 4 × 20 mL of ether, drying ($MgSO_4$), evaporation of solvent, and flash chromatography (EtOAc) gave 162 mg (98%) of a viscous oil. NMR and IR spectra were comparable to those previously reported[24] for racemic material: $[\alpha]^{24}_D = +1.62$ (c = 1.63, $CHCl_3$); ^{1}H NMR (D_2O) δ 1.34 (s, 3H), 1.87 (t, J = 6.9 Hz, 2H), 2.72 (s, 2H), 3.73 (t, J = 6.9 Hz, 2H); ^{13}C NMR δ 26.9, 31.3, 40.7, 59.2, 71.5, 117.8; FTIR (thin film) 2253 cm^{-1}.

(*R*)-(−)-Mevalonolactone (4). Cyano diol **3** (160 mg, 1.2 mmol) was heated to 100 °C with NaOH (710 μL, 2.0 N, 1.4 mmol) for 3 h in a sealed vial. The solvent was evaporated and replaced with 3.0 mL of methanol. While ice-cold, concd HCl was added until pH 2 (110 μL). The methanol was then stripped and the residue dissolved in 6.0 mL of THF. Methanesulfonic acid (60 μL) was added, and the mixture was filtered and then stirred at rt for 3 h. Triethylamine was added, and the solvent was evaporated and flash chromatogaphy (50% EtOAc/hexanes) provided product (126 mg, 81%) as a viscous oil with ^{1}H NMR, ^{13}C NMR, and IR spectra identical to racemic mevalonolactone purchased from Aldrich Chemical Co.: $[\alpha]^{25}_D$ −19.0 (c = 2.15, $CHCl_3$); ^{1}H NMR δ 1.38 (s, 3H), 1.90 (m, 2H), 2.50 (d, J = 17.2 Hz, 1H), 2.66 (d, J = 17.2 Hz, 1H), 4.34 (m, 1H), 4.60 (m, 1H); ^{13}C NMR δ 29.5, 35.7, 44.6, 66.4, 67.8, 171.4; FTIR (thin film) 1732, 1708 cm^{-1}.

(17) The specific rotation for "high purity" (*R*)-(−)-mevalonolactone varies considerably in the literature. See ref 4, Wilson *et al.* for a discussion. Chiral GLC of our sample indicated 93% ee, demonstrating no measurable racemization during the synthetic sequence.

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Supporting Information Available: Chiral GLC chromatograms (6 pages). This material is contained in libraries on microfiche, immediately follows this article in the microfilm version of the journal, and can be downloaded from the internet; see any current masthead page for ordering information and internet access instructions.

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