Bioactive Norsesterterpene 1,2-Dioxanes from a Thai Sponge, *Mycale* sp.

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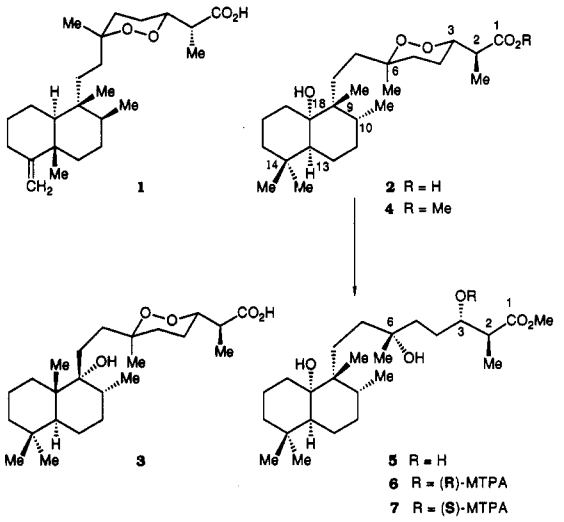
Two new norsesterterpene 1,2-dioxanes, mycaperoxides A (2) and B (3), have been isolated from a Thai sponge of the genus *Mycale*. Their relative structures were determined by X-ray and spectroscopic methods and their absolute configurations assigned by applying the Kusumi and Kakisawa modification of Mosher's method. Both compounds showed significant cytotoxicity and in vitro antiviral activity.

Since the first report on muqubilin by Kashman[2] in 1979, more than a dozen norsesterterpene cyclic peroxides have been reported from several species of marine sponge. These compounds are characterized by a 2-substituted propionic acid or methyl propionate group attached to a 1,2-dioxane ring at the C3 position as exemplified by sigmosceptrellin A (1)[3,4] (Scheme I). The norsesterterpenes may be acyclic,[5] monocyclic,[2,5,6] or bicyclic.[3,5,7-11] Many have antimicrobial activity.[9-12] In addition, muqubilin has been reported to inhibit the cell division of fertilized sea urchin eggs,[6] while the sigmosceptrellins display ichthyotoxicity.[3,7] In this paper, we describe the elucidation of the structures and absolute configurations of two new homologues, mycaperoxides A (2) and B (3), both of which exhibit significant cytotoxicity and antiviral activity.

A methanol extract of a frozen sample of *Mycale* sp. was subjected to chromatography over silica gel which by subsequent HPLC on a reversed-phase column furnished mycaperoxides A (2) and B (3), each in a yield of more than 4% from the organic extract (Scheme I). Mycaperoxide A (2), obtained as colorless crystals, has molecular formula $C_{24}H_{42}O_5$ as determined by HR EIMS. Comparison of its ^1H and ^{13}C NMR data with those of known norsesterterpene cyclic peroxides[7,8] suggested that it was a related compound. The IR and ^{13}C NMR spectra

Scheme I



indicated the presence of a carboxyl group (1715 cm^{-1}, δ 179.4 s), a hydroxyl group (3540 cm^{-1}, δ 80.0 s), and a 1,2-dioxane ring (δ 81.1 d, 80.9 s). Furthermore, the absence of olefinic signals in the NMR spectra and the unsaturation requirements of the formula pointed to two additional rings. The four methyl signals (δ 0.74 d, 0.77 s, 0.80 s, and 1.17 s) in the ^1H NMR spectrum are indicative of a drimane or a rearranged drimane skeleton, which is common in this class of sesterterpenes. The two other methyl signals (δ 1.13 d and 1.21 s) were assigned to the C2 and C6 substituents, respectively (Table I). Lastly, 2D NMR studies (COSY, HMQC, HMBC) confirmed that the overall structure of 2 had a rearranged drimane skeleton.

The stereochemistry of the substituents attached to the 1,2-dioxane ring was deduced by applying the empirical rules of Capon and MacLeod.[5] The value of the ^{13}C shift of the C6 methyl group (δ 20.2) suggests that the two side chains are attached to the 1,2-dioxane ring in a trans diequatorial arrangement (Table I). However, the low value of the ^{13}C signal of the C7 atom (δ 35.1) creates an ambiguity, since it is characteristic of an axial methylene

(1) (a) Department of Pharmacognosy. (b) Department of Chemistry.
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(3) Albericci, M.; Collart-Lempereur, M.; Braekman, J. C.; Daloze, D.; Tursch, B.; Declercq, J. P.; Germain, G.; Van Meerssche, M. *Tetrahedron Lett.* **1979**, 2687.
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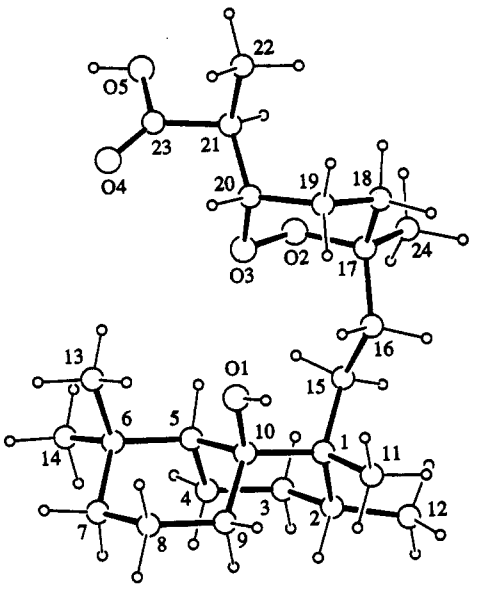

Figure 1. Perspective drawing of the X-ray structure of mycaperoxide A (2). (Numbering is that used in the supplementary material.)

Table I. NMR Data (δ, J in Hz) for Mycaperoxides A (2) and B (3) in $CDCl_3$

	2		3	
C no.	^{13}C	^1H	^{13}C	^1H
1	179.2 s		178.7 s	
2	42.1 d	2.75 m, J = 7.2, 7.2	42.6 d	2.56 dq
2Me	12.9 q	1.13 d, J = 7.3	12.5 q	1.12 d, J = 7.2
3	81.1 d	4.19 ddd, J = 7.9, 7.9, 3.6	81.3 d	4.20 m
4	21.7 t	1.78 m, 1.69 m	22.5 t	1.66 m, 1.66 m
5	32.4 t	1.54 m, 1.64 m	32.4 t	1.60 m, 1.38 m
6	80.9 s		80.4 s	
6Me	20.2 q	1.21 s	20.1 q	1.26 s
7	35.1 t	not assigned	36.2 t	1.54 m, 1.41 m
8	20.6 t	1.66 m, 1.24 m	27.0 t	1.60 m, 1.42 m
9	43.3 s		77.0 s	
9Me	18.1 q	0.80 s		
10	39.2 d	1.46 m	36.4 d	1.69 m
10Me	16.7 q	0.74 d, J = 6.9	16.2 q	0.80 d, J = 6.7
11	29.6 t	1.23 m, 1.13 m	31.2 t	1.40 m, 1.22 m
12	25.8 m	1.52 m, 1.19 m	21.6 t	1.47 m, 1.23 m
13	46.7 d	1.51 m	46.2 d	1.36 dd, J = 12.0, 2.4
14	34.0 s		33.2 s	
14Me	32.1 q	0.77 s	21.9 q	0.78 s
	30.2 q	1.17 s	33.7 q	0.82 s
15	34.0 t	1.29 m, 1.08 m	41.7 t	1.25 m, 10.8 ddd, J = 13.0, 13.0, 3.6
16	18.5 t	1.76 m, 1.36 m	18.5 t	1.52 m, 1.40 m
17	26.3 t	1.54 m, 1.42 m	31.8 t	1.39 m, 1.39 m
18	80.0 s		43.2 s	
18Me			16.1 q	0.87 s

group. The stereochemistry of the C2–C3 fragment appears to be erythro if the chemical shift of the C2 methyl group (δ 1.13) is taken as a guide. On the other hand, the splitting pattern displayed by the C3 proton (δ 4.19), which is part of an ABX system, cannot be interpreted with confidence in favor of the erythro or the threo stereochemistry. Consequently, in light of these uncertainties, recourse was made to X-ray analysis to determine the structure of 2. It is immediately seen that the erythro configuration is, in fact, correct (Figure 1). The two side chains have the trans conformation, but, in the solid state, they are diaxial with respect to the 1,2-dioxane ring which adopts a chair conformation. The other six-membered rings are also disposed as chairs.

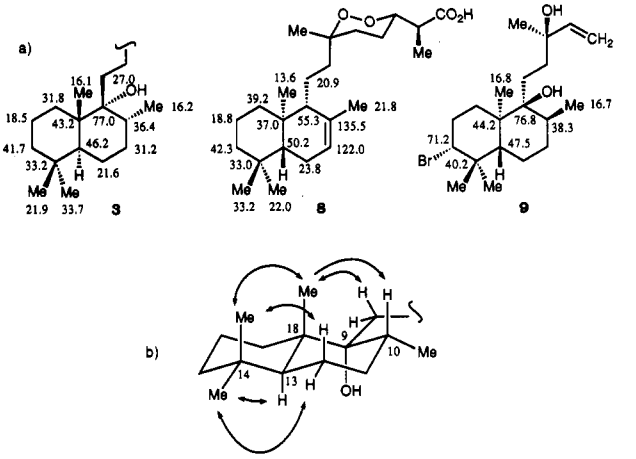

Figure 2. (a) Comparison of ^{13}C NMR data for the bicyclic portion of mycaperoxide B (3) with those of the related peroxide 8 and concinndiol (9). (b) NOE results for 3. Observed NOE is indicated by ↔ for phase-sensitive NOESY and → for difference NOE.

Table II. ^1H NMR Chemical Shifts (δ) and Their Differences ($\Delta\delta$) for Selected Protons of (*R*)- and (*S*)-MTPA Esters

	δ			δ		
C no.	6	7	$\Delta\delta$	17	18	$\Delta\delta$
CO_2Me	3.577	3.634	−0.057	3.581	3.639	−0.058
2-H	2.837	2.846	−0.009	2.853	2.865	−0.012
2-Me	1.116	1.179	−0.063	1.112	1.177	−0.065
6-Me	1.210	1.201	+0.009	1.125	1.049	+0.076
9-Me	1.125	1.103	+0.022			
10-Me	0.791	0.783	+0.008	0.844	0.829	+0.015
18-Me				0.906	0.894	+0.012

The absolute configuration of 2 was assigned by applying the Kusumi and Kakisawa modification of the method developed initially by Mosher.[13] The method is based on the induced chemical shift differences of diastereomeric esters of a secondary alcohol. Thus, the methyl ester 4, prepared from 2, was converted to the triol 5 by catalytic hydrogenation (Scheme I). Treatment of 5 with (+)-(*R*)- and (−)-(*S*)-α-methoxy-α-(trifluoromethyl)phenylacetyl (MTPA) chloride and pyridine gave the corresponding (*R*)-MTPA (6) and (*S*)-MTPA esters (7). Comparison of the ^1H NMR spectra showed that the signals for the ester methoxy and C(2)HMe protons of 7 appeared at lower fields than the corresponding signals of 6 (Table II). This difference specifies the 3*S* configuration. Thus, mycaperoxide A (2) has the absolute configuration 2*S*,3*S*,6*R*,9*R*,10*R*,13*S*,18*S* (Figure 1).

Mycaperoxide B (3), obtained as a gum, has the same molecular formula $C_{24}H_{42}O_5$ and structural features as 2, namely, a carboxyl group (1715 cm^{-1}, δ 178.7 s), a tertiary hydroxyl group (3540 cm^{-1}, δ 77.0 s), a 1,2-dioxane moiety (δ 81.3 d, 80.4 s), and six methyl groups. 2D NMR studies (COSY, HMQC, HMBC) confirmed the norsesterterpene drimane structure (3) and permitted the assignment of all the NMR resonances (Table I). The relative configuration of the bicyclic portion was secured by comparing the ^{13}C NMR signals with those of related molecules and by NOE experiments. The ^{13}C NMR data of ring A of 3 correlate well with those of the known sesterterpene (8)[8] while the data for ring B agree with those of concinndiol (9) (Figure

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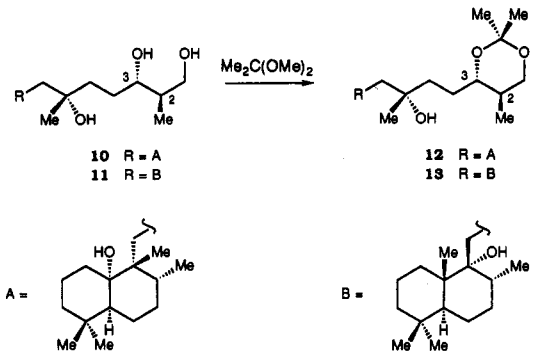
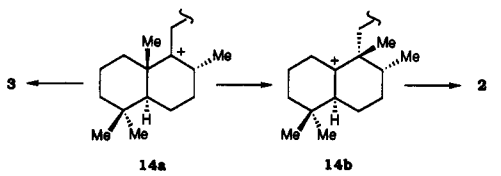
Scheme II



Scheme III



2a).[14] The results of NOESY and difference NOE studies corroborate those obtained from the NMR comparison (Figure 2b).

The NMR data for the peroxide ring portion of **3** were similar to those of **2** except for the chemical shift of the C2 proton (δ 2.56 dq) which was slightly different (δ 2.75 m). This discrepancy could be due to a difference in conformation or configuration at the C2 atom. In order to clarify this point, both **2** and **3** were converted through tetrols **10** and **11** to the 1,3-dioxanes **12** and **13** which were then compared by NMR spectroscopy (Scheme II). It was found that the NMR data for the dioxane rings of both compounds were nearly identical, thereby indicating that the C2–C3 portions had the same erythro configuration. The absolute configuration of the C2 center in **3** was established by using the previous procedure. The methyl ester **15** was obtained from **3** by methylation. Catalytic hydrogenation of **15** gave the triol **16** which was converted into the (*R*)- and (*S*)-MTPA esters **17** and **18**. Comparison of their NMR data (Table II) revealed that the C2 center has the *S* configuration. Therefore, the peroxide ring portions of both compounds have the same absolute stereochemistry. Obviously, mycaperoxides A and B stem from common Wagner–Meerwein-related biosynthetic intermediates (e.g., **14a** and **14b**) (Scheme III). Consequently, it can be assumed that the nonrearranged chiral centers in the drimane skeleton of **3** have the same configurations as those in **2**. Thus, the absolute configuration of **3** must be 2*S*,3*S*,6*S*,9*R*,10*R*,13*S*,18*S*.

Mycaperoxides A (**2**) and B (**3**) showed significant cytotoxicity (IC_{50} 0.5–1.0 μg/mL) against the cell lines of P-388, A-549, and HT-29 and displayed antiviral activity (IC_{50} 0.25–1.0 μg/mL) against vesicular stomatitis virus and herpes simplex virus type-1. Both compounds also inhibited the growth of the gram positive bacteria *Bacillus subtilis* and *Staphylococcus aureus*.

Experimental Section

Collection, Extraction, and Isolation. The blue mucous sponge *Mycale* sp. was collected by using SCUBA at a depth of 5 m at Kang Ta Sin of Sichang Island, Thailand, in March, 1991, and was kept frozen until it was extracted. The sample (1.4 kg) was steeped in 1.5 L of distilled CH_3OH, and the extract was filtered and concentrated to give an aqueous residue which was further extracted with CH_2Cl_2. The organic layer was concentrated to give 3.38 g of an oil. Most of the oil (3.1 g) was chromatographed on silica gel by using a step gradient of hexane/EtOAc. Fractions eluted with 10:1 to 3:1 hexane/EtOAc were collected to give 768 mg of an oil which was then separated by HPLC using a reversed-phase column (Cosmosil 5C_{18}-AR) with aqueous CH_3OH to give 135 mg of mycaperoxide A (**2**) and 130 mg of B (**3**).

Mycaperoxide A (2): mp 158–159.5 °C (acetone); $[\alpha]^{30}_D$ −41.0° (*c* 1.28, acetone); IR (CCl_4) 3540, 1715 cm^{-1}; ^{1}H and ^{13}C NMR (Table I); EIMS *m/z* 410 (M^+, 2), 395 (2), 281 (6), 111 (87), 95 (77), 69 (99), 43 (100 rel); HR EIMS obsd 410.3005, calcd for $C_{24}H_{42}O_5$ 410.3032.

Crystallographic Data. Slow evaporation of an acetone solution yielded colorless crystals. Orthorhombic, $P2_12_12_1$, a = 9.595(1) Å, b = 12.369(2) Å, c = 20.027(2) Å, V = 2376.8(5) Å^3, Z = 4, D_{calcd} = 1.15 g·cm^{-3}, μ = 0.073 mm^{-1}, F_{000} = 904. Cell dimensions and intensities were measured at room temperature with graphite-monochromated Mo Kα radiation; two reference reflections were measured every 60 min and showed a decrease of about 6%. All reflections were corrected for this drift. The structure was solved by direct methods (MULTAN-80) and refined by full matrix least-squares with the X-TAL 3.0 program.[15] All the coordinates of the H atoms were calculated except those of the OH groups which were observed from a difference electron density map. The final value of R was 0.058 for 1460 contributing reflections ($|F_o| > 4\sigma(F_o)$). The molecular packing is fixed by intermolecular hydrogen bonds: O(1)···O(5) $1-x, y-{}^1/_2, {}^3/_2-z$ = 2.683(6) Å.

Mycaperoxide B (3): gum; $[\alpha]^{30}_D$ −41.3° (*c* 1.27, acetone); IR (CCl_4) 3540, 1715 cm^{-1}; ^{1}H and ^{13}C NMR (see Table I); EIMS *m/z* 410 (M^+, 11), 348 (8), 280 (4), 209 (42), 69 (100), 43 (88 rel); HR EIMS obsd 410.3042, calcd for $C_{24}H_{42}O_5$ 410.3032.

Mycaperoxide A Methyl Ester (4). To a solution of 14.3 mg of **2** in 1 mL of CH_3OH was added dropwise a hexane solution of (trimethylsilyl)diazomethane ($TMSCHN_2$) until the solution stayed yellow. After standing at room temperature for 10 min, the solution was concentrated to give 14.0 mg (95%) of methyl ester **4**: gum; $[\alpha]^{21}_D$ +21° (*c* 0.25, $CHCl_3$); ^{13}C NMR ($CDCl_3$) δ 174.6 s, 81.6 d, 81.1 s, 80.0 s, 52.0 q, 46.8 d, 43.4 s, 42.3 d, 39.3 d, 34.9 t, 34.0 t, 33.9 s, 32.4 t, 32.2 q, 30.4 q, 29.6 t, 26.2 t, 25.8 t, 21.9 t, 20.8 q, 20.2 t, 18.6 t, 18.2 q, 16.8 q, 13.3 q; EIMS *m/z* 424 (M^+, 17), 409 (5), 171 (70), 69 (100 rel); HR EIMS obsd 424.3180, calcd for $C_{25}H_{44}O_5$ 424.3189.

Hydrogenation of 4 to Triol 5. A mixture of 13.8 mg of **4**, 5 mg of 10% Pd/C, and 4 mL of EtOAc was stirred under H_2 overnight. The mixture was filtered and the filtrate concentrated to furnish a residue which was separated by preparative TLC (silica gel, 4:1 CH_2Cl_2/EtOAc) to give 4.0 mg (29%) of pure triol **5** and 1.3 mg and 6.5 mg of two unidentified compounds. Triol **5**: gum; $[\alpha]^{20}_D$ +9.6° (*c* 0.090, $CHCl_3$); ^{1}H NMR ($CDCl_3$) δ 3.72 (3 H, s), 3.68 (1 H, brdd, J = 7.0, 7.0 Hz), 2.55 (1 H, dq, J = 7.0, 7.0 Hz), 1.21 (3 H, d, J = 6.9 Hz), 1.21 (3 H, s), 1.17 (3 H, s), 0.85 (3 H, s), 0.83 (3 H, s), 0.80 (3 H, d, J = 7.3 Hz); EIMS *m/z* 426 (M^+, 0.5), 408 (35), 393 (23), 245 (49), 96 (100 rel); HR EIMS obsd 408.3248, calcd for $C_{25}H_{44}O_4$ ($M - H_2O$) 408.3240.

Treatment of Triol 5 with (+)-(*R*)-α-Methoxy-α-(trifluoromethyl)phenylacetyl (MTPA) Chloride. A mixture of 1.50 mg of triol **5**, 15 μL of (+)-(*R*)-MTPA chloride, and 50 μL of dry pyridine was allowed to stand under N_2 at room temperature for 1 h. After the consumption of starting material was confirmed by TLC, a drop of H_2O and two drops of CH_2Cl_2 and CH_3OH were added. The mixture was then separated by preparative TLC (silica gel, 5:1 CH_2Cl_2/EtOAc) to give 1.25 mg (68%) of (*R*)-MTPA ester **6**: gum; $[\alpha]^{22}_D$ +23° (*c* 0.075, $CHCl_3$); ^{1}H NMR ($CDCl_3$) δ 7.536 (2 H, m), 7.402 (3 H, m), 5.376 (1 H, ddd, J =

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7.1, 7.1, 3.8 Hz), 3.577 (3 H, s), 3.515 (3 H, d, J = 1.0 Hz), 2.837 (1 H, dq, J = 7.1, 7.1 Hz), 1.210 (3 H, s), 1.125 (3 H, s), 1.116 (3 H, d, J = 6.9 Hz), 0.831 (6 H, s), 0.791 (3 H, d, J = 6.6 Hz); EIMS m/z 642 (M^+, 0.8), 96 (100 rel); HR EIMS obsd 642.3737, calcd for $C_{35}H_{53}O_7F_3$ 642.3744.

Treatment of Triol 5 with (−)-(*S*)-MTPA Chloride. A mixture of 1.40 mg of triol **5**, 15 μL of (−)-(*S*)-MTPA chloride, and 50 μL of dry pyridine was treated in the same manner as above to furnish 1.38 mg (65%) of (*S*)-MTPA ester **7**: gum; $[\alpha]^{22}_D$ −4.5° (c 0.069, $CHCl_3$); ^1H NMR ($CDCl_3$) δ 7.541 (2 H, m), 7.400 (3 H, m), 5.366 (1 H, ddd, J = 7.3, 7.3, 4.0 Hz), 3.634 (3 H, s), 3.530 (3 H, d, J = 1.0 Hz), 2.846 (1 H, dq, J = 7.3, 7.3 Hz), 1.201 (3 H, s), 1.179 (3 H, d, J = 7.3 Hz), 1.103 (3 H, s), 0.826 (3 H, s), 0.815 (3 H, s), 0.783 (3 H, d, J = 6.6 Hz); EIMS m/z 642 (M^+, 0.5), 96 (100 rel); HR EIMS obsd 642.3765, calcd for $C_{35}H_{53}O_7F_3$ 642.3744.

$LiAlH_4$ Reduction of Mycaperoxide A (2). To a stirred suspension of 90 mg of $LiAlH_4$ in 0.7 mL of ether was added dropwise a solution of 3.7 mg of **2** in 0.4 mL of ether. The reaction mixture was stirred under N_2 at room temperature for 30 min. After the reagent was quenched with EtOAc and dilute aqueous HCl, the mixture was partitioned between the organic and aqueous layers. The organic layer was taken and dried (Na_2SO_4). Removal of solvent after filtration gave 3.6 mg (100%) of tetrol **10**: gum; $[\alpha]^{20}_D$ −17° (c 0.039, $CHCl_3$); ^1H NMR ($CDCl_3$) δ 3.77 (1 H, dd, J = 10.9, 3.8 Hz), 3.62 (1 H, dd, J = 10.9, 7.6 Hz), 3.56 (1 H, m), 1.20 (3 H, s), 1.18 (3 H, s), 0.88 (3 H, d, J = 6.9 Hz), 0.85 (3 H, s), 0.83 (3 H, s), 0.80 (3 H, d, J = 6.6 Hz); EIMS m/z 398 (M^+, 0.4), 380 (24), 365 (18), 303 (17), 245 (42), 191 (43), 96 (100 rel); HR EIMS obsd 380.3270, calcd for $C_{24}H_{44}O_3$ ($M - H_2O$) 380.3291.

1,3-Dioxane 12. A mixture of 3.2 mg of tetrol **10**, 0.7 mL of 2,2-dimethoxypropane, and a catalytic amount of *p*-toluenesulfonic acid monohydrate was stirred under N_2 at room temperature for 45 min. The mixture was partitioned between EtOAc and saturated aqueous $NaHCO_3$. The organic layer was dried (Na_2SO_4). Evaporation of solvent gave 3.3 mg of a crude product which was purified by preparative TLC (silica gel, 4:1 CH_2Cl_2/EtOAc) to give 2.2 mg (63%) of **12**: gum; $[\alpha]^{21}_D$ −31° (c 0.068, $CHCl_3$); ^1H NMR ($CDCl_3$) δ 3.67 (1 H, dd, J = 11.6, 5.2 Hz, 1e-H), 3.49 (1 H, dd, J = 11.6, 11.6 Hz, 1a-H), 3.43 (1 H, ddd, J = 9.6, 9.6, 2.2 Hz, 3-H), 1.76 (1 H, m, 4-H), 1.68 (1 H, m, 2-H), 1.54 (1 H, m, 4-H), 1.41 (3 H, s, $O_2C(Me)_2$), 1.39 (3 H, s, $O_2C(Me)_2$), 1.23 (3 H, s), 1.14 (3 H, s), 0.86 (3 H, s), 0.82 (3 H, s), 0.80 (3 H, d, J = 6.7 Hz), 0.76 (3 H, d, J = 6.7 Hz, 2-Me); ^{13}C NMR ($CDCl_3$) δ 98.6 s, 79.9 s, 76.8 d, 72.7 s, 66.0 t, 46.7 d, 43.5 s, 40.5 t, 39.6 d, 37.0 t, 34.1 2 t, 34.1 s, 32.3 d, 30.8 q, 29.8t, 29.7 q 27.3 t, 26.3 t, 26.0 t, 25.2 q, 20.7 t, 19.2 q, 19.0 q, 18.6 t, 16.8 q, 13.0 q; EIMS m/z 438 (M^+, 0.4), 420 (3), 405 (13), 362 (32), 245 (25), 191 (78), 43 (100 rel); HR EIMS obsd 438.3714, calcd for $C_{27}H_{50}O_4$ 438.3709.

Tetrol 11. Reduction of **3** (6.3 mg) with $LiAlH_4$ as previously described gave 3.3 mg (54%) of tetrol **11**: gum; $[\alpha]^{19}_D$ +22° (c 0.032, $CHCl_3$); ^1H NMR ($CDCl_3$) δ 3.76 (1 H, dd, J = 10.6, 3.5 Hz), 3.63 (1 H, dd, J = 10.6, 7.6 Hz), 3.53 (1 H, brdd, J = 7.6, 7.6 Hz), 1.19 (3 H, s), 0.93 (3 H, s), 0.88 (3 H, d, J = 7.0 Hz), 0.87 (3 H, d, J = 6.6 Hz), 0.87 (3 H, s), 0.83 (3 H, s); EIMS m/z 398 (M^+, 3), 380 (28), 303 (16), 241 (72), 143 (100 rel); HR EIMS obsd 398.3417, calcd for $C_{24}H_{46}O_4$ 398.3396.

1,3-Dioxane 13. The reaction of **11** (2.8 mg) with 2,2-dimethoxypropane gave 2.3 mg (74%) of **13**: gum; $[\alpha]^{20}_D$ −13° (c 0.064, $CHCl_3$); ^1H NMR ($CDCl_3$) δ 3.68 (1 H, dd, J = 11.6, 5.2 Hz, 1e-H), 3.49 (1 H, dd, J = 11.6, 11.6 Hz, 1a-H), 3.44 (1 H, brdd, J = 8.1, 8.1 Hz, 3-H), 1.42 (3 H, s, $O_2C(Me)_2$), 1.38 (3 H, s, $O_2C(Me)_2$), 1.15 (3 H, s), 0.93 (1 H, s), 0.87 (3 H, d, J = 7.0 Hz), 0.87 (3 H, s), 0.83 (3 H, s), 0.76 (3 H, d, J = 6.7 Hz, 2-Me); ^{13}C NMR ($CDCl_3$) δ 99.0 s, 77.8 s, 76.7 d, 72.6 s, 66.5 t, 54.0 s, 46.8 d, 42.4 t, 37.8 t, 37.7 d, 37.7 t, 34.4 d, 34.3 q, 33.9 s, 32.8 t, 32.1 t, 30.2 q, 28.4 t, 28.0 q, 27.8 t, 22.6 q, 22.3 t, 19.7 q, 19.3 t, 17.3 q, 16.7 q, 13.4 q; EIMS m/z 438 (M^+, 11), 420 (19), 405 (16), 380 (17), 362 (23), 223 (60), 143 (73), 43 (100 rel); HR EIMS obsd 438.3687, calcd for $C_{27}H_{50}O_4$ 438.3709.

Mycaperoxide B Methyl Ester (15). Methylation of **3** with $TMSCHN_2$ gave **15** as a gum: $[\alpha]^{21}_D$ +8.9° (c 0.025, $CHCl_3$); ^{13}C NMR ($CDCl_3$) δ 173.0, 81.7, 80.5, 77.2, 51.9, 46.3, 43.5, 42.7, 41.7, 36.5, 36.2, 33.7, 33.3, 32.4, 31.9, 31.3, 27.1, 22.7, 22.0, 21.6, 20.3, 18.6, 16.4, 16.2, 12.8; EIMS m/z 424 (M^+, 51), 406 (17), 285 (20), 171 (42), 69 (100 rel); HR EIMS obsd 424.3212, calcd for $C_{25}H_{44}O_5$ 424.3189.

Triol 16. Catalytic hydrogenation of **15** (12.0 mg) with Pd/C in the same manner as described for **4** gave 9.9 mg (83%) of triol **16** as a gum: $[\alpha]^{20}_D$ +14° (c 0.050, $CHCl_3$); ^1H NMR ($CDCl_3$) δ 3.71 (3 H, s), 3.67 (1 H, m), 2.55 (1 H, dq, J = 7.0, 7.0 Hz), 1.20 (3 H, d, J = 7.0 Hz), 1.16 (3 H, s), 0.93 (3 H, s), 0.86 (3 H, d, J = 6.7 Hz), 0.86 (3 H, s), 0.82 (3 H, s); EIMS m/z 426 (M^+, 4), 408 (34), 269 (75), 171 (100 rel); HR EIMS obsd 426.3344, calcd for $C_{25}H_{46}O_5$ 426.3345.

(*R*)-MTPA Ester 17. Treatment of **16** (1.50 mg) with (+)-(R)-MTPA chloride gave (*R*)-MTPA ester **17** (1.25 mg) as a gum: $[\alpha]^{20}_D$ +30° (c 0.257, $CHCl_3$); ^1H NMR ($CDCl_3$) δ 7.533 (2 H, m), 7.402 (3 H, m), 5.377 (1 H, ddd, J = 7.0, 7.0, 4.0 Hz, 3-H), 3.581 (3 H, s, CO_2Me), 3.509 (3 H, d, J = 1.0 Hz, OMe), 2.853 (1 H, dq, J = 7.0, 7.0 Hz, 2-H), 1.66–1.86 (2 H, m, 4-H), 1.125 (3 H, s, 6-Me), 1.112 (3 H, d, J = 6.9 Hz, 2-Me), 0.906 (3 H, s, 18-Me), 0.866 (3 H, s), 0.844 (3 H, d, J = 7.0 Hz, 10-Me), 0.829 (3 H, s); ^{13}C NMR ($CDCl_3$) δ 173.6, 166.0, 129.7, 128.5, 127.6, 77.7, 76.8, 72.4, 55.4, 51.9, 46.5, 43.6, 42.5, 41.8, 37.1, 37.0, 36.1, 33.8, 32.2, 31.5, 27.8, 27.3, 24.9, 22.1, 21.7, 18.7, 16.7, 16.2, 12.5; EIMS m/z 642 (M^+, 11), 251 (100 rel); HR EIMS obsd 642.3768, calcd for $C_{35}H_{53}O_7F_3$ 642.3744.

(*S*)-MTPA Ester 18. Treatment of **16** (1.40 mg) with (−)-(*S*)-MTPA chloride gave (*S*)-MTPA ester **18** (1.38 mg) as a gum: $[\alpha]^{21}_D$ +4.4° (c 0.285, $CHCl_3$); ^1H NMR ($CDCl_3$) δ 7.529 (2 H, m), 7.402 (3 H, m), 5.372 (1 H, ddd, J = 7.3, 7.3, 4.0 Hz, 3-H), 3.639 (3 H, s, CO_2Me), 3.533 (3 H, d, J = 1.0 Hz), 2.865 (1 H, dq, J = 7.2, 7.2 Hz, 2-H), 1.60–1.80 (2 H, m, 4-H), 1.177 (3 H, d, J = 6.9 Hz, 2-Me), 1.049 (3 H, s, 6-Me), 0.894 (3 H, s, 18-Me), 0.866 (3 H, s), 0.829 (3 H, d, J = 6.6 Hz, 10-Me), 0.829 (3 H, s); ^{13}C NMR ($CDCl_3$) δ 174.2, 166.5, 130.1, 128.9, 128.0, 77.8, 77.2, 72.8, 56.0, 52.5, 46.9, 44.0, 43.1, 42.3, 37.4, 37.4, 36.2, 34.3, 33.9, 32.7, 32.0, 28.3, 27.7, 25.2, 22.5, 22.2, 19.2, 17.2, 16.7, 13.3; EIMS m/z 642 (M^+, 8), 69 (100 rel); HR EIMS obsd 642.3748, calcd for $C_{35}H_{53}O_7F_3$ 642.3744.

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Supplementary Material Available: X-ray crystallographic data for **2** including atomic coordinates, anisotropic displacement parameters, bond lengths, bond angles, and torsional angles, ^1H and ^{13}C NMR spectra of **2**, **3**, **4**, **12**, **13**, **15**, **17**, and **18**, and ^1H NMR spectra of **5**, **6**, **7**, **10**, **11**, and **16** (22 pages). This material is contained in libraries on microfiche, immediately follows this article in the microfilm version of the journal, and can be ordered from the ACS; see any current masthead page for ordering information.