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Synthesis and biological activity of mycalolide analogs

Kiyotake Suenaga,*,† Tomoyuki Kimura, Takeshi Kuroda, Keita Matsui, Saori Miya, Satomi Kuribayashi, Akira Sakakura and Hideo Kigoshi*

Department of Chemistry, University of Tsukuba, 1-1-1 Tennoudai, Tsukuba, Ibaraki 305-8571, Japan

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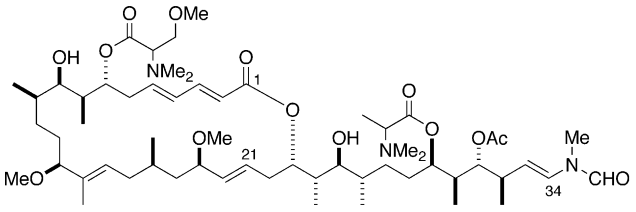
Abstract—Mycalolide analog **4**, consisting only of the side chain of mycalolide B (**2**), a trisoxazole macrolide of marine origin, was stereoselectively synthesized using Roush crotylboration, an Evans aldol reaction, and a Paterson aldol reaction as key steps. The analog **4** was found to have strong actin-depolymerizing activity.

1. Introduction

Actin-disrupting marine natural products are of interest to natural products chemists and pharmacologists.[1] These natural products consist of macrolides, cyclic peptides, and cyclodepsipeptides. Aplyronine A (**1**), an antitumor macrolide isolated from *Aplysia kurodai*,[2] interacts with actin, the major protein in cytoskeleton. Actin regulates various cell functions such as muscle contraction, cell motility, and cell division. Actin exists as a dynamic equilibrium mixture of two forms; one is polymeric F-actin and the other is monomeric G-actin. Aplyronine A (**1**) not only inhibits polymerization of actin by sequestering G-actin and forming a 1:1 complex, but also depolymerizes F-actin to G-actin by severing.[3] We achieved the total synthesis of **1** and investigated the structure–activity relationships of aplyronine A (**1**) using natural and synthetic analogs: the side chain in **1** is essential to actin-depolymerizing activity, and analog **3**, which consists only of the side-chain moiety of **1**, exhibits strong activity.[4] We recently determined the crystal structure



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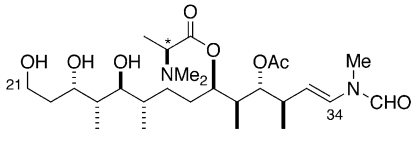
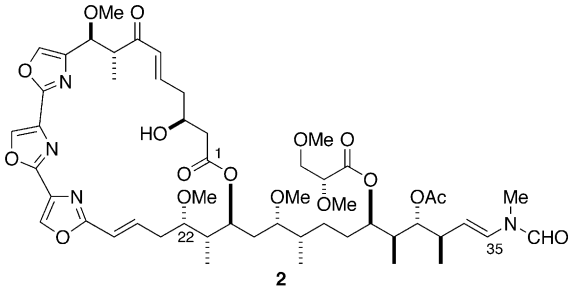


3 (* *S*/*R* = 9/1)



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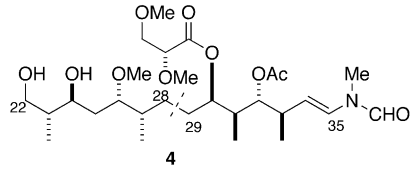

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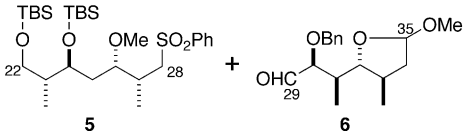

5 + **6**

* Corresponding authors. Tel./fax: +81 29 853 4313 (H.K.); e-mail: kigoshi@chem.tsukuba.ac.jp
† Present address: Department of Chemistry, Faculty of Science and Technology, Keio University, 3-14-1 Hiyoshi, Kohoku, Yokohama 223-8522, Japan.

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of actin–aplyronine A complex via synchrotron X-ray analysis[5] and obtained chemical evidence for the direct interaction between actin and the side-chain portion of aplyronine A by photoaffinity labeling experiments.[6] These results display the great importance of the side-chain moiety in the activity against actin.

Mycalolide B (**2**) is a cytotoxic and antifungal macrolide isolated from a sponge of the genus *Mycale* sp.[7] Mycalolide B (**2**) inhibits actomyosin Mg^{2+}-ATPase[8] and also interacts with actin in the same manner as **1**.[9] The total synthesis of mycalolide A, which lacks the 2,3-di-*O*-methyl-D-glyceroyl group at C30, has been achieved.[10] Recently, several crystal structures of trisoxazole macrolides with actin have been reported.[11] Since mycalolide B (**2**) possesses a similar side chain to that of **1**, analog **4** is expected to show actin-depolymerizing activity. We have previously reported the synthesis and actin-depolymerizing activity of analog **4**.[12] We describe herein details of the stereocontrolled synthesis of mycalolide analog **4** and its biological activities, including both its cytotoxicity and actin-depolymerizing activity, along with those of aplyronine analog **3**.

2. Results and discussion

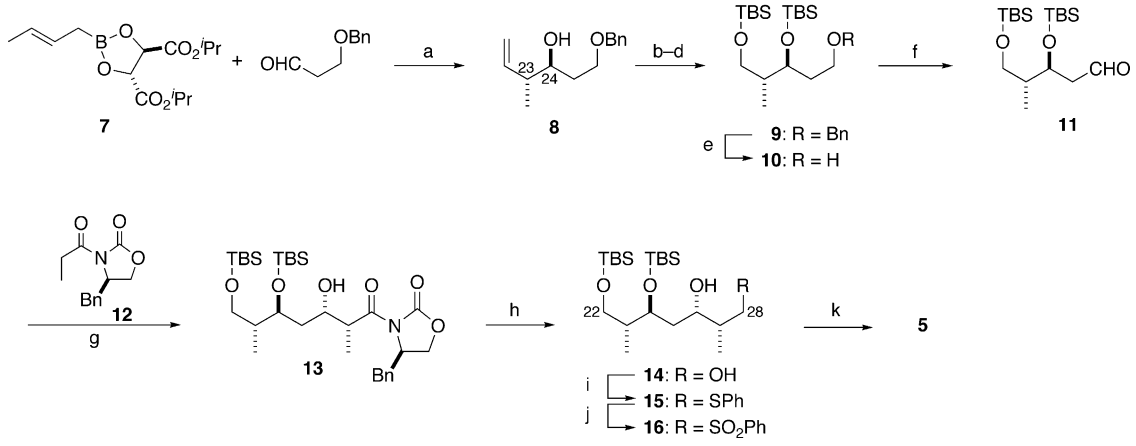
2.1. Chemical synthesis

The synthesis of mycalolide analog **4** has been carried out according to a convergent synthetic methodology connecting C22–C28 and C29–C35 segments, **5** and **6**.

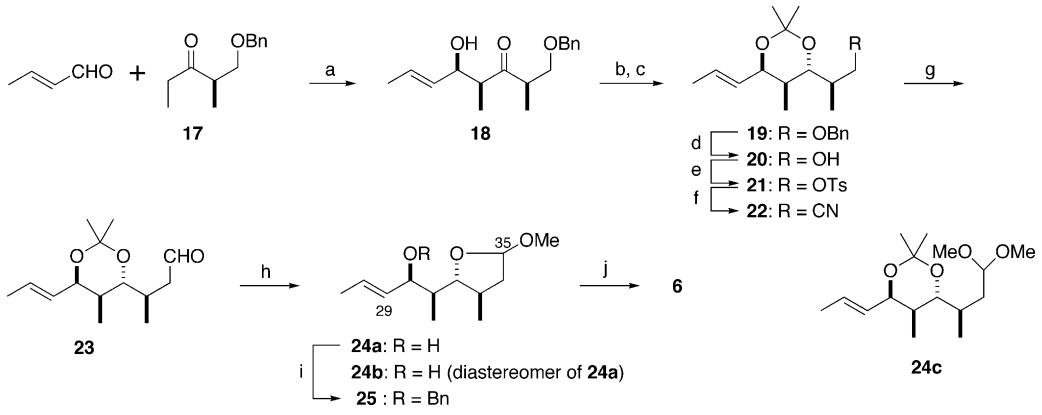
The synthesis of C22–C28 segment **5** is shown in Scheme 1. While *anti* stereocenters between C23 and C24 of **5** was previously constructed by using an anti-selective aldol reaction under Heathcock conditions,[13] the improved synthesis of **5** was developed by using Roush crotylboration[14] as the key step. Thus, the Roush crotylboration between boronate **7** and 3-benzyloxypropanal afforded homoallylic alcohol **8** (91%) as a single diastereomer (Scheme 1). Oxidative cleavage of the olefin moiety of **8**, reduction with $NaBH_4$, and silylation gave silyl ether **9**. The spectral data of **9** were identical to those of previously synthesized **9**,[12] this confirmed the stereochemistry. Cleavage of the benzyl-protecting group in **9** gave alcohol **10**, which was oxidized to aldehyde **11**. The Evans aldol reaction between aldehyde **11** and imide **12**[15] gave hydroxy imide **13** as a single diastereomer, which was converted into diol **14**. Diol **14** was transformed with $(PhS)_2$-Bu_3P[16] into sulfide **15**, which was oxidized with *m*-chloroperoxybenzoic acid to sulfone **16**. The secondary hydroxy group in **16** was methylated to afford C22–C28 segment **5** (57% from **7**).

The synthesis of C29–C35 segment **6** is shown in Scheme 2. While compound **6** with four contiguous *syn–anti–anti* stereocenters was previously prepared using the Evans aldol reaction and Sharpless epoxidation as the key steps,[4a–c] the improved synthesis of **6** was developed by using the Paterson aldol reaction[17] as the key step. Thus, the Paterson aldol reaction between ethyl ketone **17** and crotonaldehyde gave hydroxy ketone **18**.[17b] Stereoselective reduction of **18** with tetramethylammonium triacetoxyborohydride[18] afforded an *anti*-1,3-diol exclusively, which was transformed into acetonide **19**. Its stereochemistry was confirmed to be *anti* by the ^{13}C chemical shifts of two acetonide methyls (δ_C 25.8 and 23.7).[19] The benzyl-protecting group in **19** was removed with calcium in liquid ammonia to give alcohol **20**, which was converted into tosylate **21**. One carbon homologation with NaCN provided nitrile **22**, the reduction of which with DIBAL afforded aldehyde **23**. Aldehyde **23** was treated with PPTS in methanol to provide a separable mixture of diastereomeric acetals, **24a** and **24b**, and the dimethyl acetal **24c**.[20] After chromatographic separation, two minor products, **24b** and **24c**, were subjected to equilibration (PPTS in methanol) to afford a mixture of **24a**, **24b**, and **24c**, from which the major acetal **24a** was again obtained. By repeating this procedure, **24b** and **24c** could be transformed into **24a**. Protection of the hydroxy group in **24a** gave benzyl ether **25**, the double bond of which was cleaved oxidatively to afford the C29–C35 segment **6** (48% from **17**).

The Julia coupling reaction between **5** and **6** gave a hydroxy sulfone, which was converted into olefin **26** by reduction with sodium amalgam (Scheme 3). Removal of the benzyl-protecting group in **26** with calcium in liquid ammonia gave alcohol **27**, catalytic hydrogenation of which provided



Scheme 1. Reagents and conditions: (a) MS 4 Å, toluene, −78 °C, 91%; (b) OsO_4, NMO, THF–*t*-BuOH–H_2O, rt; then $NaIO_4$, rt; (c) $NaBH_4$, EtOH, rt; (d) TBSCl, imidazole, DMF, 50 °C, 79% (three steps); (e) H_2, 10% Pd–C, $NaHCO_3$, EtOAc, rt, 92%; (f) DMSO, $(COCl)_2$, CH_2Cl_2, −78 °C; Et_3N, −78 °C→0 °C, 89%; (g) **12**, Bu_2BOTf, Et_3N, CH_2Cl_2, −78 °C→0 °C, 100%; (h) $LiBH_4$, EtOH, Et_2O, −10 °C, 100%; (i) $(PhS)_2$, Bu_3P, DMF, rt, 96%; (j) *m*-CPBA, $NaHCO_3$, CH_2Cl_2, rt, 99%; (k) MeI, NaH, THF, rt, 94%.

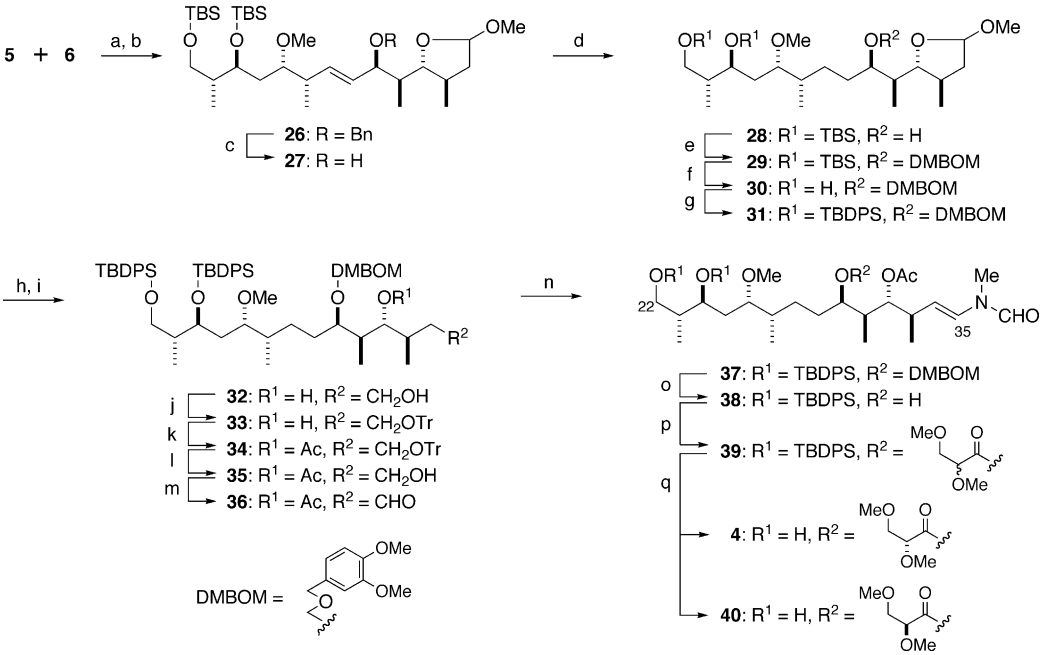


Scheme 2. Reagents and conditions: (a) $Sn(OTf)_2$, Et_3N, CH_2Cl_2, −78 °C→−60 °C, 85%; (b) $Me_4NBH(OAc)_3$, AcOH, MeCN, −25 °C; (c) $(MeO)_2CMe_2$, PPTS, acetone, rt, 84% (two steps); (d) Ca, liq. NH_3, *i*-PrOH, THF, −78 °C, 98%; (e) *p*-TsCl, pyridine, 0 °C, 100%; (f) NaCN, DMSO, 50 °C, 98%; (g) DIBAL, CH_2Cl_2, hexane, −78 °C, 95%; (h) PPTS, MeOH, rt, 82%; (i) BnBr, NaH, DMF, rt, 95%; (j) OsO_4, NMO, H_2O, acetone, rt; then $NaIO_4$, rt, 99%.

alcohol **28**.[4a] The hydroxyl group was protected to give 3,4-dimethoxybenzyloxymethyl ether **29**. At this stage, the TBS-protecting groups in **29** were changed to TBDPS groups, because TBS group was sensitive to acidic conditions at the later stage of the synthesis. Thus, deprotection of two TBS-protecting groups in **29** afforded diol **30**, which was converted into TBDPS ether **31**. The cyclic acetal moiety of **31** was hydrolyzed under acidic conditions to afford a hemiacetal, which was reduced with $NaBH_4$ to give diol **32**. Selective protection of the primary hydroxyl group in **32** provided trityl ether **33**, the secondary hydroxyl group of which was acetylated to afford acetate **34**. The trityl group of **34** was removed with formic acid to give alcohol **35**, which was oxidized to aldehyde **36**. Condensation between **36** and *N*-methylformamide under acidic conditions provided enamide **37**. Deprotection of the 3,4-dimethoxybenzyloxymethyl group of **37** gave alcohol **38**, which was esterified with 2,3-di-*O*-methyl-D-glyceric acid under Yamaguchi conditions to afford a mixture of diastereomeric esters **39**, which resulted from the racemization of 2,3-di-*O*-methyl-D-glyceric acid. After removal of the silyl groups in **39**, HPLC separation of the diastereomers provided analogs **4** and **40**.[21]

2.2. Biological activities

The actin-depolymerizing activity and cytotoxicity against HeLa S_3 cells of aplyronine A (**1**), mycalolide B (**2**), and their analogs **3**, **4**, and **40** are shown in Table 1. The mycalolide analog **3** exhibited strong activity comparable to that of



Scheme 3. Reagents and conditions: (a) BuLi, THF–hexane, −78 °C; (b) 5% Na–Hg, NaH_2PO_4, MeOH, 0 °C, 72% (two steps); (c) Ca, liq. NH_3, *i*-PrOH, THF, −78 °C, 89%; (d) H_2, 5% Pd–C, $NaHCO_3$, EtOH, 55 °C, 95%; (e) 3,4-dimethoxybenzyloxymethyl chloride, *i*-Pr_2NEt, CH_2Cl_2, rt, 84%; (f) Bu_4NF, THF, rt, 99%; (g) TBDPSCl, imidazole, DMF, rt, 75%; (h) 1 M HCl, DME, rt; (i) $NaBH_4$, EtOH, rt, 70% (two steps); (j) TrCl, pyridine, 50 °C, 95%; (k) Ac_2O, pyridine, DMAP, rt, 100%; (l) HCO_2H, Et_2O, rt, 77%; (m) Dess–Martin periodinane, pyridine, CH_2Cl_2, rt, 91%; (n) MeNHCHO, PPTS, hydroquinone, MS 3 Å, benzene, reflux, 55%; (o) DDQ, 1 M phosphate buffer (pH 6), *t*-BuOH, CH_2Cl_2, rt, 90%; (p) 2,3-di-*O*-methyl-D-glyceric acid, 2,4,6-trichlorobenzoyl chloride, Et_3N, DMAP, CH_2Cl_2, rt, 74%; (q) HF·pyridine, pyridine, THF, rt; separation by HPLC, (**4**) 55%, (**40**) 34%.

Table 1. Cytotoxicity and actin-depolymerizing activity of mycalolide B, aplyronine A, and their analogs

Compounds	Actin-depolymerizing activity[a]		Cytotoxicity against HeLa S_3 cells	
	IC_{50} (μM)[b]	Relative potency[c]	IC_{50} (μg/mL)	Relative potency[c]
Aplyronine A (**1**)	1.6	100	0.00048[e]	100
Mycalolide B (**2**)[d]	nd	nd	0.0035	14
3	7.9	20	>10	<0.01
4	2.7	59	>10	<0.01
40	4.4	36	>10	<0.01

[a] Activity was monitored by measuring the fluorescent intensity of pyrenyl actin. For the conditions of assay, see Section 4.2.
[b] IC_{50} indicates the concentration required to depolymerize F-actin (3.7 μM) to 50% of its control amplitude.
[c] The relative potencies were calculated from the IC_{50} values of the compound (aplyronine A=100).
[d] Mycalolide B was purchased from Wako Pure Chemical Industries, Inc.
[e] Ref. 4c.

aplyronine A (**1**). This result revealed that the side-chain portion in mycalolide B (**2**) is responsible for the potent activity of **2**, as is the case with aplyronine A (**1**). Comparison of the activities of **3**, **4**, and **40** revealed that both the structure and stereochemistry of the acyl group influenced activity. In contrast, analogs **3**, **4**, and **40** showed no cytotoxicity at 10 μg/mL, thus indicating that the presence of the macrolide ring is essential to the strong cytotoxicity of **1** and **2**, and that actin-depolymerization is not directly related to the cytotoxicity.

3. Conclusion

The stereocontrolled synthesis of mycalolide analog **4**, consisting only of the side chain of mycalolide B (**2**), was carried out. The mycalolide analog **4** and aplyronine analog **3** were found to exhibit strong actin-depolymerizing activity. In contrast, the analogs did not show cytotoxicity against HeLa S_3 cells. These results clearly indicated that the side-chain portions of aplyronine A and mycalolide B are essential to the actin-depolymerizing activity and that the combination of the side-chain portion and the macrolactone portion is responsible for the cytotoxicity.

4. Experimental

4.1. General

Melting points are uncorrected. Optical rotations were measured with a JASCO DIP-370 polarimeter or a JASCO DIP-1000 polarimeter. ^{1}H NMR spectra were recorded on a JEOL JNM-EX270 (270 MHz) or a Bruker AVANCE-400M (400 MHz) instrument. Chemical shifts are reported in parts per million from internal standards [tetramethylsilane (0.00 ppm) for $CDCl_3$ and C_6D_5H (7.16 ppm) for C_6D_6] and J values are in hertz. ^{13}C NMR spectra were recorded on a JEOL JNM-EX270 instrument (67.8 MHz) using $CDCl_3$ as a solvent. Chemical shifts are reported in parts per million from the solvent peak (77.0 ppm). FAB mass spectra were recorded on a JEOL SX-102 instrument. ESI mass spectra were recorded on a QStar/Pulsar *i* spectrometer (Applied Biosystems). Both TLC analysis and preparative TLC were conducted on E. Merck precoated silica gel 60 F_{254} (0.25 mm layer thickness). Fuji Silysia silica gel BW-820 MH and FL-60D were used for column chromatography unless otherwise noted. Organic solvents for moisture-sensitive reactions were distilled from the following drying agents: THF and ether (Na-benzophenone ketyl), benzene (Na), acetonitrile and triethylamine (calcium hydride), DMSO (calcium hydride under reduced pressure), CH_2Cl_2 (P_2O_5), acetone (anhydrous K_2CO_3), and MeOH (Mg). All moisture-sensitive reactions were performed under an atmosphere of nitrogen, and the starting materials were azeotropically dried with benzene before use. All new compounds were determined to be >95% pure by ^{1}H NMR unless otherwise noted.

4.1.1. Homoallylic alcohol 8. Preparation of crotylboronate **7**: To a stirred solution of potassium *tert*-butylalkoxide (8.4 g, 75 mmol) in THF (60 mL) cooled at −78 °C was added liquefied *trans*-2-butene (7.2 mL, 78 mmol) cooled at −78 °C and then 1.56 M solution of BuLi in hexane (48 mL, 75 mmol) so as to maintain the reaction temperature below −65 °C for 2 h. The mixture was stirred at −50 °C for 15 min and re-cooled to −78 °C, triisopropyl borate (17.2 mL, 75.0 mmol) was added so as to keep the reaction temperature below −65 °C for 1 h. The mixture was stirred for 15 min and poured into 1 M aqueous HCl saturated with NaCl (220 mL). To the mixture D-(−)-diisopropyl tartrate (17.5 g, 74.7 mmol) was added, and the mixture was extracted with ether (4×50 mL). The combined organic layers were dried with $MgSO_4$ and concentrated to give boronate **7** (25.4 g) as a colorless oil. Crotylboration: To a stirred mixture of boronate **7** (1.2 g, 9.7 mmol) and MS 4 Å (50 mg) in toluene (10 mL) cooled at −78 °C was added a solution of 3-benzyloxypropanal (517 mg, 3.15 mmol) in toluene (6 mL, 2×2 mL rinse). The mixture was stirred at −78 °C for 1.5 h and diluted with 2 M aqueous NaOH (50 mL). The mixture was stirred at 0 °C for 30 min and extracted with EtOAc (3×10 mL). The extracts were dried with $MgSO_4$ and concentrated. The residual oil was purified by column chromatography on silica gel (12 g, hexane–EtOAc 10:1) to give **8** (630 mg, 91%) as a colorless oil: TLC R_f 0.46 (hexane–EtOAc 3:1); $[\alpha]_D^{20}$ 3.38 (c 1.00, $CHCl_3$) ; ^{1}H NMR (270 MHz, $CDCl_3$) δ 7.38–7.24 (m, 5H), 5.81 (m, 1H), 5.11 (m, 1H), 5.05 (m, 1H), 4.56 (s, 2H), 3.77–3.61 (m, 3H), 2.47 (br s, 1H, OH), 2.24 (m, 1H), 1.78–1.71 (m, 2H), 1.05 (d, J=7.0 Hz, 3H); ^{13}C NMR (67.8 MHz, $CDCl_3$) δ 140.2, 137.8, 128.2, 127.5, 115.3, 74.0, 73.2, 69.1, 44.0, 33.6, 15.8.

4.1.2. Silyl ether 9. To a stirred solution of homoallylic alcohol **8** (5.00 g, 22.7 mmol) in THF (65 mL) were added a solution of *N*-methylmorpholine-*N*-oxide (4.05 g, 34.6 mmol) in H_2O (13 mL) and a 0.078 M solution of osmium tetroxide in *tert*-butyl alcohol (15 mL, 1.2 mmol). After being stirred at room temperature for 2.5 h, sodium periodate (10.5 g, 49.1 mmol) was added. The reaction mixture was stirred at room temperature for 1.5 h, diluted with saturated aqueous $Na_2S_2O_3$ (50 mL), and extracted with EtOAc (3×50 mL). The combined extracts were washed with saturated aqueous $Na_2S_2O_3$ (3×20 mL) and brine (30 mL), dried (Na_2SO_4), and concentrated to give a crude aldehyde (8.0 g).

To a stirred solution of the crude aldehyde (8.0 g) in EtOH (230 mL) was added sodium borohydride (1.06 g, 28.0

mmol), and the mixture was stirred at room temperature for 20 min. The reaction was quenched by addition of acetone (50 mL) and the resulting mixture was stirred at room temperature for 10 min and concentrated. The mixture was diluted with H_2O (30 mL) and extracted with EtOAc (5×20 mL). The extracts were combined, washed with brine (30 mL), dried (Na_2SO_4), and concentrated. The residual oil was purified by column chromatography on silica gel (100 g, hexane–EtOAc 1:1) to give a crude diol (4.77 g).

To a stirred solution of the crude diol (4.77 g) and imidazole (12.7 g, 186 mmol) in DMF (10 mL) cooled at 0 °C was added *tert*-butyldimethylsilyl chloride (14.1 g, 93.6 mmol). The resulting solution was stirred at 50 °C for 11 h, cooled to room temperature, and diluted with cold water (50 mL). The mixture was extracted with EtOAc (3×50 mL). The combined extracts were washed with H_2O (3×30 mL) and brine (30 mL), dried (Na_2SO_4), and concentrated. The residual oil was purified by column chromatography on silica gel (FL60D 100 g, hexane–EtOAc 50:1) to give **9** (8.15 g, 79% in three steps) as a colorless oil : TLC, R_f 0.57 (hexane–EtOAc 4:1); $[\alpha]_D^{26}$ −7.2 (*c* 1.02, $CHCl_3$); IR (neat) 1471, 1255, 1092, 837 cm^{-1}; ^1H NMR (270 MHz, $CDCl_3$) δ 7.34–7.27 (m, 5H), 4.49 (s, 2H), 3.91 (dt, *J*=4.5, 7.3 Hz, 1H), 3.59–3.52 (m, 2H), 3.52 (dd, *J*=7.0, 9.9 Hz, 1H), 3.41 (dd, *J*=6.4, 9.9 Hz, 1H), 1.88–1.78 (m, 2H), 1.71 (m, 1H), 0.88 (s, 9H), 0.87 (s, 9H), 0.85 (d, *J*=7.0 Hz, 3H), 0.04 (s, 3H), 0.03 (s, 3H), 0.02 (s, 6H); HRMS (ESI) calcd for $C_{25}H_{49}O_3Si_2$ $(M+H)^+$ 453.3220, found 453.3204.

4.1.3. Alcohol 10. A mixture of silyl ether **9** (7.95 g, 17.6 mmol), $NaHCO_3$ (1.79 g, 21.3 mmol), and 10% Pd on carbon (1.32 g) in EtOAc (176 mL) was stirred under a hydrogen atmosphere at room temperature for 11 h. The mixture was filtered through a pad of Celite, and the residue was washed with EtOAc. The filtrate and the washings were combined and concentrated. The residual oil was purified by column chromatography on silica gel (100 g, hexane–EtOAc 20:1) to give **10** (5.88 g, 92%) as a colorless oil: TLC R_f 0.55 (hexane–EtOAc 5:1); $[\alpha]_D^{28}$ −8.7 (*c* 0.992, $CHCl_3$); IR ($CHCl_3$) 3425, 1473, 1255 cm^{-1}; ^1H NMR (270 MHz, $CDCl_3$) δ 3.95 (dt, *J*=5.3, 6.3 Hz, 1H), 3.72 (t, *J*=5.6 Hz, 2H), 3.42 (dd, *J*=6.6, 9.9 Hz, 1H), 3.24 (dd, *J*=6.0, 9.9 Hz, 1H), 2.29 (br, 1H, OH), 1.98–1.82 (m, 2H), 1.66 (m, 1H), 0.87 (s, 9H), 0.86 (s, 9H), 0.82 (d, *J*=6.9 Hz, 3H), 0.06 (s, 3H), 0.05 (s, 3H), 0.01 (s, 6H); ^{13}C NMR (67.8 MHz, $CDCl_3$) δ 72.4, 65.1, 60.7, 41.0, 33.8, 26.0, 25.9, 18.3, 18.1, 11.7, −4.4, −4.5, −5.3, −5.4; HRMS (ESI) calcd for $C_{18}H_{43}O_3Si_2$ $(M+H)^+$ 363.2751, found 363.2751.

4.1.4. Aldehyde 11. To a stirred solution of oxalyl chloride (0.44 mL, 5.0 mmol) in CH_2Cl_2 (12 mL) cooled at −78 °C was added a solution of DMSO (0.65 mL, 9.2 mmol) in CH_2Cl_2 (1.5 mL) dropwise. The resulting solution was stirred at −78 °C for 30 min, and a solution of alcohol **10** (1.22 g, 3.35 mmol) in CH_2Cl_2 (1.5 mL, 3×0.5 mL rinse) was added dropwise. The mixture was stirred at −78 °C for 40 min, and triethylamine (2.4 mL, 17.2 mmol) was added. The resulting mixture was stirred at −78 °C for 1 h, warmed to 0 °C, and stirred for 1 h. The mixture was diluted with H_2O (30 mL) and extracted with EtOAc (3×30 mL). The combined extracts were washed with H_2O (20 mL), saturated aqueous $NaHCO_3$ (20 mL), and brine (20 mL), successively, dried (Na_2SO_4), and concentrated. The residual oil was purified by column chromatography on silica gel (50 g, hexane–ether 50:1→25:1) to give **11** (1.07 g, 89%) as a colorless oil: TLC, R_f 0.55 (hexane–EtOAc 5:1); $[\alpha]_D^{21}$ −4.56 (*c* 1.00, $CHCl_3$); IR (neat) 2854, 1732, 1257, 1086, 837 cm^{-1}; ^1H NMR (270 MHz, $CDCl_3$) δ 9.81 (t, *J*=2.6 Hz, 1H), 4.35 (dt, *J*=4.9, 5.6 Hz, 1H), 3.48 (dd, *J*=5.9, 10.0 Hz, 1H), 3.43 (dd, *J*=7.0, 10.0 Hz, 1H), 2.50 (dd, *J*=2.6, 5.9 Hz, 2H), 1.92 (m, 1H), 0.89 (s, 9H), 0.87 (s, 9H), 0.86 (d, *J*=7.0 Hz, 3H), 0.07 (s, 3H), 0.05 (s, 3H), 0.04 (s, 3H), 0.04 (s, 3H); ^{13}C NMR (67.8 MHz, $CDCl_3$) δ 202.4, 68.9, 64.8, 47.8, 41.7, 26.0, 25.9, 18.3, 18.1, 11.6, −4.4, −4.5, −5.3, −5.4; HRMS (ESI) calcd for $C_{18}H_{40}NaO_3Si_2$ $(M+Na)^+$ 383.2414, found 383.2367.

4.1.5. Hydroxy imide 13. To a stirred solution of imide **12** (226 mg, 0.970 mmol) in CH_2Cl_2 (1.4 mL) cooled at 0 °C were added 1 M solution of dibutylboron triflate in CH_2Cl_2 (0.97 mL, 0.97 mmol) and triethylamine (0.19 mL, 1.3 mmol), successively. The reaction mixture was stirred at 0 °C for 40 min and cooled to −78 °C. A solution of aldehyde **11** (212 mg, 0.587 mmol) in CH_2Cl_2 (0.3 mL, 2×0.2 mL rinse) was added, and the reaction mixture was stirred at −78 °C for 2 h and at 0 °C for 20 min. After the reaction was quenched by addition of 0.5 M phosphate buffer (pH 7, 2 mL) and MeOH (3 mL), 30% aqueous hydrogen peroxide (1.5 mL) in MeOH (3 mL) was added slowly, and the resulting solution was stirred at 0 °C for 1 h. The organic solvents were evaporated, and the mixture was cooled to 0 °C. Saturated aqueous $Na_2S_2O_3$ (3 mL) was added slowly, and the mixture was extracted with ether (3×5 mL). The extracts were combined, washed with saturated aqueous $NaHCO_3$ (5 mL) and brine (5 mL), dried (Na_2SO_4), and concentrated. The residual oil was purified by column chromatography on silica gel (60 g, hexane–EtOAc 15:1→10:1→5:1) to give **13** (358 mg, 100%) as a colorless oil along with recovered **12** (64 mg). Compound **13**: TLC, R_f 0.40 (hexane–EtOAc 5:1); $[\alpha]_D^{22}$ −31.9 (*c* 1.00, $CHCl_3$); IR (neat) 3525, 1783, 1697, 1387, 1251, 1209, 1091 cm^{-1}; ^1H NMR (270 MHz, $CDCl_3$) δ 7.37–7.15 (m, 5H), 4.69 (m, 1H), 4.26–4.14 (m, 3H), 3.98 (dt, *J*=4.0, 6.6 Hz, 1H), 3.75 (dq, *J*=4.0, 7.0 Hz, 1H), 3.61 (dd, *J*=6.0, 10.0 Hz, 1H), 3.53 (br s, 1H, OH), 3.47 (dd, *J*=6.0, 10.0 Hz, 1H), 3.28 (dd, *J*=3.3, 13.5 Hz, 1H), 2.77 (dd, *J*=9.6, 13.5 Hz, 1H), 1.96 (dt, *J*=6.0, 6.6 Hz, 1H), 1.67 (ddd, *J*=4.0, 10.0, 14.0 Hz, 1H), 1.55 (ddd, *J*=2.6, 6.6, 14.0 Hz, 1H), 1.27 (d, *J*=7.0 Hz, 3H), 0.89 (s, 18H), 0.88 (d, *J*=7.0 Hz, 3H), 0.10 (s, 3H), 0.08 (s, 3H), 0.04 (s, 6H); ^{13}C NMR (67.8 MHz, $CDCl_3$) δ 176.2, 153.0, 135.1, 129.3, 128.9, 127.3, 68.6, 66.1, 65.9, 65.0, 55.3, 43.4, 40.7, 37.9, 36.2, 26.0, 26.9, 18.4, 18.1, 12.6, 11.3, −4.3, −4.6, −5.2, −5.3; HRMS (ESI) calcd for $C_{31}H_{55}NaO_6Si_2$ $(M+Na)^+$ 616.3466, found 616.3466.

4.1.6. Alcohol 14. To a stirred solution of hydroxy imide **13** (878 mg, 1.48 mmol) in ether (26 mL) in the presence of anhydrous ethanol (0.10 mL, 1.8 mmol) cooled at −10 °C was added a 2.0 M solution of lithium borohydride in THF (0.90 mL, 1.8 mmol), and the solution was stirred at −10 °C for 30 min. The reaction was quenched by addition of 1 M aqueous NaOH (4 mL), and the mixture was stirred at 0 °C for 15 min. The mixture was diluted with saturated aqueous $Na_2S_2O_3$ (40 mL) and extracted with

ether (4×20 mL). The combined extracts were washed with brine (20 mL), dried (Na_2SO_4), and concentrated. The residual oil was purified by column chromatography on silica gel (60 g, hexane–EtOAc 3:1→1:1→1:3) to give **14** (628 mg, 100%) as a colorless oil: TLC, R_f 0.54 (hexane–ether 1:3); $[\alpha]_D^{19}$ +7.9 (*c* 1.00, $CHCl_3$); IR (neat) 3392, 1471, 1255, 1064, 836 cm^{-1}; ^1H NMR (270 MHz, $CDCl_3$) δ 4.07 (ddd, *J*=2.4, 2.4, 10.5 Hz, 1H), 3.89 (dt, *J*=4.6, 7.3 Hz, 1H), 3.70–3.55 (m, 2H), 3.61 (dd, *J*=5.4, 9.7 Hz, 1H), 3.45 (dd, *J*=5.4, 9.7 Hz, 1H), 3.14 (br s, 1H, OH), 1.94 (m, 1H), 1.77 (m, 1H), 1.72 (ddd, *J*=4.6, 10.5, 12.4 Hz, 1H), 1.50 (ddd, *J*=2.4, 4.6, 12.4 Hz, 1H), 0.89 (d, *J*=7.0 Hz, 3H), 0.84 (s, 9H), 0.83 (s, 9H), 0.80 (d, *J*=7.0 Hz, 3H), 0.07 (s, 3H), 0.03 (s, 3H), 0.00 (s, 3H), −0.01 (s, 3H); ^{13}C NMR (67.8 MHz, $CDCl_3$) δ 72.6, 72.0, 66.8, 65.9, 64.9, 39.9, 39.8, 35.1, 26.0, 25.4, 18.4, 18.1, 13.4, 11.2, −4.1, −4.7, −5.2, −5.3; HRMS (ESI) calcd for $C_{21}H_{48}NaO_4Si_2$ $(M+Na)^+$ 443.2989, found 443.2995.

4.1.7. Sulfide 15. To a stirred solution of alcohol **14** (215 mg, 0.511 mmol) and diphenyl disulfide (197 mg, 0.904 mmol) in DMF (2 mL) cooled at 0 °C was added tributylphosphine (0.25 mL, 1.0 mmol), and the resulting solution was stirred at room temperature for 11 h. Water (0.5 mL) was added, and the resulting mixture was concentrated. The residual oil was purified by column chromatography on silica gel (60 g, hexane–ether 10:1) to give **15** (254 mg, 99%) as a colorless oil: TLC, R_f 0.46 (hexane–ether 5:1); $[\alpha]_D^{22}$ +11.1 (*c* 1.00, $CHCl_3$); IR (neat) 3496, 1471, 1254, 1063, 837 cm^{-1}; ^1H NMR (270 MHz, $CDCl_3$) δ 7.35–7.11 (m, 5H), 4.05 (m, 1H), 3.93 (m, 1H), 3.63 (dd, *J*=5.8, 9.8 Hz, 1H), 3.50 (dd, *J*=5.4, 9.8 Hz, 1H), 3.18 (dd, *J*=6.1, 12.7 Hz, 1H), 2.78 (dd, *J*=7.8, 12.7 Hz, 1H), 2.01 (m, 1H), 1.73 (m, 1H), 1.68 (ddd, *J*=3.6, 10.8, 14.7 Hz, 1H), 1.51 (ddd, *J*=1.9, 5.4, 14.7 Hz, 1H), 1.02 (d, *J*=6.8 Hz, 3H), 0.91 (s, 9H), 0.90 (s, 9H), 0.85 (d, *J*=7.0 Hz, 3H), 0.11 (s, 3H), 0.08 (s, 3H), 0.05 (s, 6H); ^{13}C NMR (67.8 MHz, $CDCl_3$) δ 137.1, 129.2, 128.9, 128.7, 128.5, 125.4, 72.3, 70.0, 64.9, 40.0, 38.9, 37.2, 36.3, 26.0, 25.9, 18.4, 18.1, 13.7, 13.2, −4.2, −4.7, −5.2, −5.3; HRMS (ESI) calcd for $C_{27}H_{52}NaO_3SSi_2$ $(M+Na)^+$ 535.3073, found 535.3054.

4.1.8. Sulfone 16. To a stirred solution of sulfide **15** (1.36 g, 2.66 mmol) in CH_2Cl_2 (26 mL) cooled at 0 °C were added $NaHCO_3$ (1.67 g, 19.9 mmol) and 77% *m*-chloroperoxybenzoic acid (1.61 g, 7.18 mmol). After 5 min, the mixture was warmed to room temperature and stirred at room temperature for 15 min. The reaction mixture was diluted with saturated aqueous $Na_2S_2O_3$ (10 mL) and H_2O (30 mL), stirred at room temperature for 30 min, and extracted with ether (3×50 mL). The combined extracts were washed with saturated aqueous $NaHCO_3$ (30 mL) and brine (30 mL), dried (Na_2SO_4), and concentrated. The residual oil was purified by column chromatography on silica gel (43 g, hexane–EtOAc 6:1) to give **16** (1.44 g, 99%) as a colorless oil: TLC, R_f 0.4 (hexane–ether 1:1); $[\alpha]_D^{22}$ +7.4 (*c* 1.00, $CHCl_3$); IR (neat) 3519, 1471, 1306, 1255, 1147, 1086, 837 cm^{-1}; ^1H NMR (270 MHz, $CDCl_3$) δ 7.97–7.94 (m, 2H), 7.70–7.55 (m, 3H), 4.00–3.90 (m, 2H), 3.67 (dd, *J*=5.7, 9.7 Hz, 1H), 3.52 (dd, *J*=5.7, 9.7 Hz, 1H), 3.52 (br s, 1H, OH), 3.45 (dd, *J*=4.1, 14.0 Hz, 1H), 3.01 (dd, *J*=7.8, 14.0 Hz, 1H), 2.25 (m, 1H), 1.99 (m, 1H), 1.64 (ddd, *J*=4.9, 10.3, 14.3 Hz, 1H), 1.55 (ddd, *J*=2.7, 4.9, 14.3 Hz, 1H), 1.11 (d, *J*=7.0 Hz, 3H), 0.98 (s, 9H), 0.94 (s, 9H), 0.88 (d, *J*=7.0 Hz, 3H), 0.18 (s, 3H), 0.15 (s, 3H), 0.12 (s, 3H), 0.12 (s, 3H); ^{13}C NMR (67.8 MHz, $CDCl_3$) δ 140.0, 133.4, 129.1, 127.7, 72.3, 70.4, 64.8, 59.4, 39.7, 35.7, 34.5, 26.0, 25.9, 18.4, 18.0, 14.1, 13.4, −4.2, −4.7, −5.2, −5.4; HRMS (ESI) calcd for $C_{27}H_{52}NaO_5Si_2$ $(M+Na)^+$ 567.2972, found 567.2980.

4.1.9. C22–C28 segment 5. To a stirred solution of sulfone **16** (1.44 g, 2.64 mmol) in THF (26 mL) cooled at 0 °C were added methyl iodide (0.84 mL, 13.5 mmol) and NaH (401 mg of 60% dispersion in mineral oil, 10.0 mmol), successively. The mixture was stirred at room temperature for 16 h, and the reaction was quenched by addition of ice (2 g) and saturated aqueous NH_4Cl (30 mL). The mixture was extracted with ether (3×30 mL). The combined extracts were washed with saturated aqueous $Na_2S_2O_3$ (30 mL) and brine (30 mL), dried (Na_2SO_4), and concentrated. The residual oil was purified by column chromatography on silica gel (75 g, benzene–ether 100:1→50:1) to give **5** (1.39 g, 95%) as a colorless oil: TLC, R_f 0.58 (benzene–ether 10:1); $[\alpha]_D^{26}$ −39.3 (*c* 1.00, $CHCl_3$); IR (neat) 1471, 1306, 1255, 1149, 1088, 837 cm^{-1}; ^1H NMR (270 MHz, $CDCl_3$) δ 7.97–7.94 (m, 2H), 7.70–7.55 (m, 3H), 3.87 (ddd, *J*=1.9, 3.5, 5.1 Hz, 1H), 3.38 (d, *J*=7.0 Hz, 2H), 3.35 (dd, *J*=1.5, 14.0 Hz, 1H), 3.23 (ddd, *J*=1.9, 3.8, 10.0 Hz, 1H), 3.09 (s, 3H), 2.78 (dd, *J*=10.3, 14.0 Hz, 1H), 2.53 (m, 1H), 1.89 (dtq, *J*=3.5, 7.0, 7.0 Hz, 1H), 1.31 (ddd, *J*=1.9, 10.0, 14.0 Hz, 1H), 1.08 (d, *J*=7.0 Hz, 3H), 1.07 (m, 1H), 0.89 (s, 9H), 0.88 (s, 9H), 0.80 (d, *J*=7.0 Hz, 3H), 0.08 (s, 3H), 0.07 (s, 3H), 0.04 (s, 6H); ^{13}C NMR (67.8 MHz, $CDCl_3$) δ 139.9, 133.5, 129.2, 128.0, 80.3, 69.5, 65.2, 57.3, 56.3, 42.2, 31.8, 29.0, 26.1, 26.0, 18.3, 16.3, 10.6, −3.8, −4.4, −5.2, −5.3; HRMS (ESI) calcd for $C_{28}H_{54}NaO_5SSi_2$ $(M+Na)^+$ 581.3128, found 581.3095; Anal. Calcd for $C_{28}H_{54}O_5SSi_2$: C, 60.17; H, 9.74. Found: C, 60.05; H, 9.68.

4.1.10. Hydroxy ketone 18. To a mixture of $Sn(OTf)_2$ (5.8 g, 14 mmol) and triethylamine (2.4 mL, 17 mmol) in CH_2Cl_2 (130 mL) cooled at −78 °C was added a solution of ethylketone **17** (2.2 g, 11 mmol), and the mixture was stirred at −78 °C for 2 h. A 2.3 M solution of crotonaldehyde in CH_2Cl_2 (6.6 mL, 16 mmol) was added, and the reaction mixture was stirred at −78 °C for 2 h and −60 °C for 1 h. The mixture was warmed to room temperature and diluted with 0.5 M phosphate buffer (pH 7.0, 130 mL). The organic layer was separated, and the aqueous layer was extracted with ether (3×130 mL). The organic layer and the extracts were combined, washed with 0.5 M phosphate buffer (pH 7.0, 130 mL) and brine (130 mL), dried (Na_2SO_4), and concentrated. The residual oil was purified twice by column chromatography on silica gel (100 g, hexane–ether 5:1→4:1→3:1) and (FL60D 100 g, hexane–ether 5:1→4:1→3:1) to give **18** (2.5 g, 85%) as a colorless oil: TLC, R_f 0.41 (hexane–ether 1:1); $[\alpha]_D^{26}$ −0.07 (*c* 1.00, $CHCl_3$); IR (neat) 3446 (br), 1701, 1655 cm^{-1}; ^1H NMR (270 MHz, $CDCl_3$) δ 7.36–7.24 (m, 5H), 5.65 (ddq, *J*=1.1, 15.1, 6.5 Hz, 1H), 5.41 (ddq, *J*=6.2, 15.1, 1.4 Hz, 1H), 4.49 (d, *J*=11.9 Hz, 1H), 4.43 (d, *J*=11.9 Hz, 1H), 4.42 (m, 1H), 3.64 (dd, *J*=8.6, 8.6 Hz, 1H), 3.45 (dd, *J*=5.1, 8.6 Hz, 1H), 3.15 (m, 1H), 2.84 (dq, *J*=3.5, 7.0 Hz, 1H), 2.60 (br, 1H, OH), 1.67 (dd, *J*=1.4, 6.5 Hz, 3H), 1.08

(d, J=7.3 Hz, 3H), 1.03 (d, J=7.0 Hz, 3H); ^{13}C NMR (67.8 MHz, $CDCl_3$) δ 217.4, 137.5, 130.3, 128.3, 127.6, 127.5, 127.5, 73.4, 72.9, 72.3, 51.3, 45.4, 17.8, 15.6, 10.2; HRMS (ESI) calcd for $C_{17}H_{24}NaO_3$ $(M+Na)^+$ 299.1623, found 299.1625.

4.1.11. Acetonide 19. To a solution of tetramethylammonium triacetoxyborohydride (24.8 g, 94.2 mmol) in acetonitrile (88 mL) and acetic acid (93 mL) cooled at –25 °C was added a solution of hydroxy ketone **18** (5.14 g, 18.6 mmol) in acetonitrile (3 mL, 2×1 mL rinse). The reaction mixture was stirred at −25 °C for 1 h and at −15 °C for 34 h. The mixture was diluted with 0.5 M aqueous Na/K tartrate (350 mL) and vigorously stirred at room temperature for 1 h. The organic layer was separated, and the aqueous layer was extracted with EtOAc (3×50 mL). The organic layer and the extracts were combined, washed with H_2O (200 mL), saturated aqueous $NaHCO_3$ (3×200 mL), and brine (200 mL), respectively, dried (Na_2SO_4), and concentrated to give a crude diol (5.73 g).

To a solution of the diol (5.73 g) in acetone (77 mL) and 2,2-dimethoxypropane (77 mL) was added pyridinium *p*-toluenesulfonate (487 mg, 1.94 mmol). The mixture was stirred at room temperature for 1.5 h and diluted with saturated aqueous $NaHCO_3$ (150 mL). The organic layer was separated, and the aqueous layer was extracted with ether (3×100 mL). The organic layer and the extracts were combined, washed with brine (100 mL), dried (Na_2SO_4), and concentrated. The residual oil was purified by column chromatography on silica gel (100 g, hexane–ether 10:1) to give **19** (4.97 g, 84%) as a colorless oil: TLC, R_f 0.89 (hexane–ether 1:1); $[\alpha]_D^{26}$ −10.9 (c 0.862, $CHCl_3$); IR (neat) 1222, 735, 698 cm^{-1}; ^{1}H NMR (270 MHz, $CDCl_3$) δ 7.34–7.24 (m, 5H), 5.67 (ddq, J=0.8, 15.4, 5.9 Hz, 1H), 5.45 (ddq, J=7.0, 15.4, 1.6 Hz, 1H), 4.52 (d, J=11.9 Hz, 1H), 4.47 (d, J=11.9 Hz, 1H), 4.28 (m, 1H), 3.60 (dd, J=4.9, 9.2 Hz, 1H), 3.36 (dd, J=7.0, 9.2 Hz, 1H), 3.28 (dd, J=5.1, 7.3 Hz, 1H), 1.96 (m, 1H), 1.89 (m, 1H), 1.71 (dd, J=1.6, 5.9 Hz, 3H), 1.32 (s, 6H), 1.02 (d, J=7.0 Hz, 3H), 0.87 (d, J=7.0 Hz, 3H); ^{13}C NMR (67.8 MHz, $CDCl_3$) δ 138.7, 128.6, 128.2, 127.5, 127.4, 127.3, 100.3, 76.2, 73.1, 72.2, 70.9, 38.0, 37.7, 25.8, 23.7, 18.0, 14.4, 13.4; MS (FAB) m/z 341 $(M+Na)^+$; HRMS (ESI) calcd for $C_{20}H_{30}NaO_3$ $(M+Na)^+$ 341.2093, found 341.2094.

4.1.12. Alcohol 20. Calcium (1.37 g, 34.2 mmol) was added to a stirred solution of acetonide **19** (4.96 g, 15.6 mmol) in THF (250 mL), isopropyl alcohol (85 mL), and liquid NH_3 (170 mL) cooled at −78 °C. After the mixture was stirred at −78 °C for 2 h, NH_4Cl (14.3 g) and $Fe(NO_3)_3\cdot 9H_2O$ (2.5 g) were added. The mixture was stirred at −78 °C for 1 h and allowed to warm to room temperature. The residue was diluted with H_2O (400 mL), and the mixture was stirred at room temperature for 1 h and extracted with EtOAc (5×100 mL). The combined extracts were washed with brine (100 mL), dried (Na_2SO_4), and concentrated. The residual oil was purified by column chromatography on silica gel (150 g, hexane–ether 3:1→2:1) to give **20** (3.49 g, 98%) as a colorless oil: TLC, R_f 0.40 (hexane–ether 1:1); $[\alpha]_D^{32}$ −33.2 (c 1.25, $CHCl_3$); IR (neat) 3455 (br), 1678, 1225 cm^{-1}; ^{1}H NMR (270 MHz, $CDCl_3$) δ 5.64 (m, 1H), 5.39 (ddq, J=7.3, 15.4, 1.6 Hz, 1H), 4.31 (m, 1H), 3.69 (dd, J=3.2, 11.1 Hz, 1H), 3.52 (dd, J=5.9, 11.1 Hz, 1H), 3.26 (dd, J=5.7, 7.6 Hz, 1H), 2.68 (br, 1H), 1.82 (dd, J=2.2, 6.9 Hz, 1H), 1.75 (m, 1H), 1.65 (dd, J=1.6, 6.2 Hz, 1H), 1.32 (s, 3H), 1.31 (s, 3H), 0.96 (d, J=6.9 Hz, 3H), 0.83 (d, J=6.9 Hz, 3H); MS (FAB) m/z 251 $(M+Na)^+$; HRMS (ESI) calcd for $C_{13}H_{24}NaO_3$ $(M+Na)^+$ 251.1623, found 251.1607.

4.1.13. Tosylate 21. To a stirred solution of alcohol **20** (1.00 g, 4.38 mmol) in pyridine (2.2 mL) cooled at 0 °C was added *p*-toluenesulfonyl chloride (1.50 g, 7.87 mmol), and the mixture was stirred at 0 °C for 6 h. The mixture was diluted with H_2O (15 mL), stirred at room temperature for 30 min, and extracted with ether (3×15 mL). The combined extracts were washed with brine (15 mL), dried (Na_2SO_4), and concentrated. The residual oil was purified by column chromatography on silica gel (50 g, hexane–ether 5:1→3:1) to give **21** (1.67 g, 100%) as a colorless oil: TLC, R_f 0.60 (hexane–ether 1:1); $[\alpha]_D^{26}$ −3.87 (c 1.08, $CHCl_3$); IR (neat) 1598, 1495, 1224, 794, 706 cm^{-1}; ^{1}H NMR (270 MHz, $CDCl_3$) δ 7.79 (d, J=7.9 Hz, 2H), 7.34 (d, J=7.9 Hz, 2H), 5.65 (ddq, J=0.8, 15.1, 6.2 Hz, 1H), 5.40 (ddq, J=7.0, 15.1, 1.6 Hz, 1H), 4.23 (m, 1H), 4.15 (dd, J=9.5, 4.0 Hz, 1H), 3.98 (dd, J=6.8, 9.5 Hz, 1H), 3.16 (dd, J=6.2, 7.6 Hz, 1H), 2.45 (s, 3H), 1.92 (m, 1H), 1.74 (m, 1H), 1.70 (dd, J=1.6, 6.2 Hz, 3H), 1.24 (s, 6H), 0.97 (d, J=6.8 Hz, 3H), 0.84 (d, J=6.8 Hz, 3H); MS (FAB) m/z 405 $(M+Na)^+$; HRMS (ESI) calcd for $C_{20}H_{30}NaO_5S$ $(M+Na)^+$ 405.1712, found 405.1720.

4.1.14. Nitrile 22. To a stirred solution of tosylate **21** (1.04 g, 2.72 mmol) in DMSO (15 mL) was added sodium cyanide (656 mg, 13.4 mmol) at room temperature, and the reaction mixture was stirred at 50 °C for 2 h. After cooling, the mixture was diluted with H_2O (38 mL) and extracted with ether (4×40 mL). The combined extracts were washed with brine (30 mL), dried (Na_2SO_4), and concentrated. The residual oil was purified by column chromatography on silica gel (25 g, hexane–ether 10:1→5:1) to give **22** (633 mg, 98%) as a colorless oil: TLC, R_f 0.63 (hexane–ether 1:1); $[\alpha]_D^{26}$ −19.5 (c 0.91, $CHCl_3$); IR (neat) 2246, 1224 cm^{-1}; ^{1}H NMR (270 MHz, $CDCl_3$) δ 5.69 (m, 1H), 5.44 (ddq, J=7.0, 15.4, 1.2 Hz, 1H), 4.30 (m, 1H), 3.15 (dd, J=7.0, 7.0 Hz, 1H), 2.45 (m, 2H), 1.95 (m, 1H), 1.74 (m, 1H), 1.71 (dd, J=1.2, 6.7 Hz, 3H), 1.35 (s, 3H), 1.33 (s, 3H), 1.14 (d, J=6.7 Hz, 3H), 0.92 (d, J=6.8 Hz, 3H); ^{13}C NMR (67.8 MHz, $CDCl_3$) δ 127.9, 127.9, 118.9, 100.6, 76.8, 70.5, 39.1, 35.1, 25.6, 23.6, 20.6, 18.0, 16.2, 13.5; MS (FAB) m/z 260 $(M+Na)^+$; HRMS (ESI) calcd for $C_{14}H_{23}NaO_2$ $(M+Na)^+$ 260.1626, found 260.1605.

4.1.15. Aldehyde 23. To a stirred solution of nitrile **22** (456 mg, 1.92 mmol) in CH_2Cl_2 (7.7 mL) cooled at −78 °C was added a 1.0 M solution of diisobutylaluminum hydride in hexane (2.4 mL, 2.4 mmol). The solution was stirred at −78 °C for 70 min, and the reaction was quenched by addition of MeOH (3 mL). After the mixture was warmed to room temperature, 0.5 M aqueous Na/K tartrate (25 mL) was added. The resulting mixture was vigorously stirred at room temperature for 1 h, and the organic layer was separated. The aqueous layer was extracted with ether (3×30 mL). The organic layer and the extracts were combined, washed with brine (10 mL), dried (Na_2SO_4), and

concentrated. The residual oil was purified by column chromatography on silica gel (FL60D 15 g, benzene–ether 100:1→80:1→20:1) to give **23** (439 mg, 95%) as a colorless oil: TLC, R_f 0.52 (benzene–ether 20:1); $[\alpha]_D^{26}$ −26.8 (*c* 0.81, $CHCl_3$); IR (neat) 2725, 1726, 1225 cm^{-1}; ^1H NMR (270 MHz, $CDCl_3$) δ 9.74 (dd, *J*=2.2, 2.2 Hz, 1H), 5.69 (ddq, *J*=1.0, 15.4, 6.3 Hz, 1H), 5.42 (ddq, *J*=7.0, 15.4, 0.8 Hz, 1H), 4.30 (m, 1H), 3.11 (dd, *J*=5.9, 7.0 Hz, 1H), 2.52 (ddd, *J*=2.2, 5.4, 15.9 Hz, 1H), 2.31 (ddd, *J*=2.2, 6.8, 15.9 Hz, 1H), 2.21 (m, 1H), 1.75 (m, 1H), 1.71 (dd, *J*=0.8, 6.3 Hz, 3H), 1.31 (s, 6H), 1.03 (d, *J*=6.8 Hz, 3H), 0.90 (d, *J*=7.0 Hz, 3H); ^{13}C NMR (67.8 MHz, $CDCl_3$) δ 202.2, 128.2, 127.7, 100.5, 78.4, 70.7, 47.7, 39.1, 33.2, 25.6, 23.6, 18.0, 17.1, 13.5; MS (FAB) *m/z* 263 $(M+Na)^+$; HRMS (ESI) calcd for $C_{14}H_{25}O_3$ $(M+H)^+$ 241.1804, found 241.1817.

4.1.16. Methyl acetal 24a. Aldehyde **23** (1.06 g, 4.41 mmol) was dissolved in a 0.009 M solution of pyridinium *p*-toluenesulfonate (16.2 mL, 0.146 mmol) in MeOH, and the solution was stirred at room temperature for 30 min. The reaction was quenched by addition of triethylamine (3 mL), and the resulting mixture was concentrated. The residual oil was purified by column chromatography on silica gel (FL60D 100 g, hexane–ether 10:1→7:1→6:1→5:1→4:1) to give **24a** (409 mg, 43%), **24b** (319 mg), and dimethyl acetal **24c** (199 mg) as a colorless oil, respectively. Acetals **24b** (319 mg) and **24c** (199 mg) were dissolved in a 0.009 M solution of pyridinium *p*-toluenesulfonate (8.0 mL, 0.072 mmol) in MeOH. After the mixture was stirred at room temperature for 1 h, triethylamine (1.5 mL) was added, and the resulting mixture was concentrated. The residual oil was purified by column chromatography on silica gel (FL60D 50 g, hexane–ether 7:1→6:1→5:1→4:1) to give **24a** (225 mg, 24%), **24b** (167 mg) and **24c** (46.7 mg) as colorless oil, respectively. Further, from acetals **24b** (167 mg) and **24c** (46.7 mg), acetals **24a** (89 mg, 9%), **24b** (84 mg), and **24c** (19 mg, 2%) were obtained by repeating the procedure described above. Further from **24b** (84 mg) and **24c** (19 mg), acetals **24a** (49 mg, 5%), **24b** (38 mg, 4%), and **24c** (4 mg, 0.3%) were obtained again. In total, methyl acetal **24a** (772 mg, 82%) was obtained from aldehyde **23** (1.06 g). Compound **24a**: TLC, R_f 0.37 (hexane–ether 1:1); $[\alpha]_D^{26}$ +77.2 (*c* 0.97, $CHCl_3$); IR ($CHCl_3$) 3490 (br), 1670, 1455, 1380, 1235 cm^{-1}; ^1H NMR (270 MHz, $CDCl_3$) δ 5.67 (ddq, *J*=1.1, 15.1, 6.5 Hz, 1H), 5.52 (ddq, *J*=6.2, 15.1, 1.6 Hz, 1H), 4.90 (d, *J*=4.9 Hz, 1H), 4.25 (br, 1H), 3.54 (dd, *J*=8.1, 8.1 Hz, 1H), 3.31 (s, 3H), 3.21 (m, 1H), 2.29 (m, 1H), 2.05 (dd, *J*=7.0, 12.7 Hz, 1H), 1.79 (m, 1H), 1.70 (dd, *J*=1.6, 6.5 Hz, 3H), 1.57 (ddd, *J*=4.9, 10.8, 12.7 Hz, 1H), 1.04 (d, *J*=6.8 Hz, 3H), 0.87 (d, *J*=7.3 Hz, 3H); ^{13}C NMR (67.8 MHz, $CDCl_3$) δ 131.4, 126.3, 104.9, 89.0, 74.4, 55.0, 44.0, 41.8, 35.8, 18.7, 17.9, 12.0; MS (FAB) *m/z* 237 $(M+Na)^+$; HRMS (ESI) calcd for $C_{12}H_{23}O_3$ $(M+H)^+$ 215.1647, found 215.1651. Compound **24b**: TLC, R_f 0.43 (hexane–ether 1:1); $[\alpha]_D^{29}$ +112 (*c* 0.995, $CHCl_3$); IR ($CHCl_3$) 3490 (br), 1675, 1450, 1380, 1230 cm^{-1}; ^1H NMR (270 MHz, $CDCl_3$) δ 5.70 (ddq, *J*=1.0, 15.5, 6.3 Hz, 1H), 5.55 (ddq, *J*=6.3, 15.5, 1.3 Hz, 1H), 4.99 (d, *J*=2.3, 5.6 Hz, 1H), 4.15 (m, 1H), 3.60 (dd, *J*=7.3, 7.3 Hz, 1H), 3.48 (br m, 1H), 3.34 (s, 3H), 2.30 (m, 1H), 2.04 (m, 1H), 1.81 (m, 1H), 1.73 (dd, *J*=1.3, 6.3 Hz, 3H), 1.51 (ddd, *J*=2.3, 5.6, 13.5 Hz, 1H), 1.10 (d, *J*=6.6 Hz, 3H), 0.95 (d, *J*=7.2 Hz, 3H); MS (FAB) *m/z* 237 $(M+Na)^+$. Compound **24c**: TLC, R_f 0.76 (hexane–ether 1:1); $[\alpha]_D^{28}$ −17 (*c* 0.34, $CHCl_3$); IR ($CHCl_3$) 1675, 1455, 1380, 1225 cm^{-1}; ^1H NMR (270 MHz, $CDCl_3$) δ 5.67 (ddq, *J*=0.7, 15.5, 6.3 Hz, 1H), 5.45 (ddq, *J*=7.3, 15.5, 1.3 Hz, 1H), 4.48 (dd, *J*=4.3, 7.6 Hz, 1H), 4.29 (ddd, *J*=0.7, 5.0, 7.3 Hz, 1H), 3.34 (s, 3H), 3.30 (s, 3H), 3.13 (dd, *J*=4.6, 7.3 Hz, 1H), 1.89 (ddd, *J*=3.3, 7.6, 14.2 Hz, 1H), 1.81–1.70 (m, 2H), 1.71 (dd, *J*=1.3, 6.3 Hz, 3H), 1.40 (ddd, *J*=4.3, 9.6, 14.2 Hz, 1H), 1.32 (s, 6H), 0.99 (d, *J*=6.6 Hz, 3H), 0.87 (d, *J*=7.2 Hz, 3H); MS (FAB) *m/z* 309 $(M+Na)^+$; HRMS (ESI) calcd for $C_{16}H_{30}NaO_4$ $(M+Na)^+$ 309.2042, found 309.2048.

4.1.17. Benzyl ether 25. To a stirred solution of methyl acetal **24a** (771 mg, 3.60 mmol) in DMF (11 mL) cooled at 0 °C were added benzyl bromide (1.3 mL, 11 mmol) and sodium hydride (438 mg of 60% dispersion in mineral oil, 11 mmol), successively. The mixture was stirred at room temperature for 3 h, cooled to 0 °C, and diluted with H_2O (20 mL) and saturated aqueous NH_4Cl (40 mL). The mixture was extracted with ether (3×30 mL), and the combined extracts were washed with saturated aqueous $NaHCO_3$ (2×30 mL) and brine (30 mL), dried (Na_2SO_4), and concentrated. The residual oil was purified by column chromatography on silica gel (FL60D 50 g, hexane–ether 40:1→20:1) to give **25** (1.04 g, 95%) as a colorless oil: TLC, R_f 0.56 (hexane–ether 5:1); $[\alpha]_D^{26}$ −26.8 (*c* 0.81, $CHCl_3$); IR (neat) 2933, 1207, 734 cm^{-1}; ^1H NMR (270 MHz, $CDCl_3$) δ 7.27–7.11 (m, 5H), 5.57 (m, 1H), 5.42 (ddq, *J*=7.3, 15.1, 1.1 Hz, 1H), 4.81 (d, *J*=4.9 Hz, 1H), 4.49 (d, *J*=11.9 Hz, 1H), 4.21 (d, *J*=11.9 Hz, 1H), 4.13 (m, 1H), 3.60 (dd, *J*=6.8, 9.4 Hz, 1H), 3.21 (s, 3H), 2.14 (m, 1H), 1.99 (m, 1H), 1.64 (dd, *J*=1.1, 6.2 Hz, 3H), 1.56 (m, 1H), 1.52 (m, 1H), 0.99 (d, *J*=6.5 Hz, 3H), 0.85 (d, *J*=7.0 Hz, 3H); ^{13}C NMR (67.8 MHz, $CDCl_3$) δ 139.5, 131.0, 128.0, 127.6, 127.1, 126.9, 104.6, 87.3, 80.1, 70.4, 54.3, 46.5, 42.5, 35.5, 19.9, 17.9, 9.6; MS (FAB) *m/z* 327 $(M+Na)^+$; HRMS (ESI) calcd for $C_{19}H_{28}NaO_3$ $(M+Na)^+$ 327.1936, found 327.1935.

4.1.18. C29–C35 segment 6. To a stirred solution of benzyl ether **25** (69.2 mg, 0.227 mmol) in acetone (1.5 mL) and H_2O (0.5 mL) were added *N*-methylmorpholine-*N*-oxide (40.0 mg, 0.341 mmol) and a 2.4% solution of osmium tetroxide in *tert*-butyl alcohol (0.15 mL, 0.011 mmol). After being stirred at room temperature for 2 h, sodium periodate (133 mg, 0.622 mmol) was added. The reaction mixture was stirred at room temperature for 30 min, diluted with saturated aqueous $Na_2S_2O_3$ (10 mL), and extracted with EtOAc (3×13 mL). The combined extracts were washed with saturated aqueous $Na_2S_2O_3$ (13 mL) and brine (13 mL), dried (Na_2SO_4), and concentrated. The residual oil was purified by column chromatography on silica gel (3 g, hexane–ether 3:1→1:1) to give **6** (66.9 mg, 99%) as a colorless oil: TLC, R_f 0.51 (hexane–ether 1:1); $[\alpha]_D^{26}$ +24.7 (*c* 1.34, $CHCl_3$); IR (neat) 2829, 2698, 1732, 1207, 739, 698 cm^{-1}; ^1H NMR (270 MHz, $CDCl_3$) δ 9.73 (d, *J*=1.3 Hz, 1H), 7.41–7.25 (m, 5H), 4.92 (d, *J*=4.9 Hz, 1H), 4.72 (d, *J*=11.3 Hz, 1H), 4.61 (d, *J*=11.3 Hz, 1H), 4.23 (dd, *J*=1.3, 3.0 Hz, 1H), 3.60 (dd, *J*=6.8, 10.0 Hz, 1H), 3.31 (s, 3H), 2.28 (m, 1H), 2.12 (dd, *J*=6.8, 12.7 Hz, 1H), 2.10 (m, 1H), 1.66 (ddd, *J*=4.9, 9.9, 12.7 Hz, 1H), 1.11 (d, *J*=6.5 Hz, 3H), 0.92 (d, *J*=7.0 Hz, 3H); ^{13}C NMR (67.8 MHz, $CDCl_3$) δ 204.4,

137.8, 128.3, 127.7, 127.7, 104.9, 86.4, 85.2, 73.3, 54.8, 42.9, 42.4, 35.9, 20.0, 10.7; MS (FAB) *m/z* 315 $(M+Na)^+$; HRMS (FAB) calcd for $C_{17}H_{24}NaO_4$ $[(M+Na)^+]$ 315.1572, found 315.1593; Anal. Calcd for $C_{17}H_{24}O_4$: C, 69.84; H, 8.27. Found: C, 69.43; H, 8.31.

4.1.19. Olefin 26. To a stirred solution of C22–C28 segment **5** (744 mg, 1.30 mmol) in THF (4.3 mL) cooled at −78 °C was added a 1.52 M solution of BuLi in hexane (0.73 mL, 1.7 mmol) dropwise. The mixture was stirred at −78 °C for 30 min, and then a solution of C29–C35 segment **6** (115.3 mg, 0.394 mmol) in THF (4 mL) was added dropwise, and the resulting mixture was stirred at −78 °C for 3 h. The reaction was quenched by addition of saturated aqueous NH_4Cl (5 mL), and the mixture was extracted with ether (3×5 mL). The combined extracts were washed with brine (5 mL), dried (Na_2SO_4), and concentrated. The residual oil was purified by column chromatography on silica gel (20 g, hexane–ether 10:1→4:1→2:1→1:1) to give a diastereomeric mixture of hydroxy sulfones (343 mg) as a colorless oil along with recovered **5** (498 mg, 67%). The hydroxy sulfones were employed in the next experiment without separation of the diastereomers. To a vigorously stirred solution of the diastereomeric mixture of hydroxy sulfones (343 mg) in MeOH (13 mL) cooled at 0 °C were added Na_2HPO_4 (965 mg, 6.80 mmol) and 5% sodium amalgam (2.2 g, 4.8 mmol). The mixture was stirred at 0 °C for 2 h, diluted with saturated aqueous NH_4Cl (10 mL), then stirred at room temperature for 30 min, and extracted with ether (3×20 mL). The combined extracts were washed with brine (10 mL), dried (Na_2SO_4), and concentrated. The residual oil was purified by column chromatography on silica gel (6 g, hexane–ether 10:1→5:1) to give olefin **26** (196 mg, 72% from **6**) as a colorless oil: TLC, R_f 0.51 (hexane–ether 4:1); $[\alpha]_D^{25}$ −19.6 (*c* 1.00, $CHCl_3$); IR (neat) 1470, 1254, 1092 cm^{-1}; ^1H NMR (270 MHz, $CDCl_3$) δ 7.36–7.20 (m, 5H), 5.62 (dd, *J*=7.0, 16.0 Hz, 1H), 5.46 (dd, *J*=7.0, 16.0 Hz, 1H), 4.88 (d, *J*=4.9 Hz, 1H), 4.57 (d, *J*=11.9 Hz, 1H), 4.35 (d, *J*=11.9 Hz, 1H), 4.20 (dd, *J*=2.7, 7.0 Hz, 1H), 4.01 (dt, *J*=9.5, 2.7 Hz, 1H), 3.67 (dd, *J*=7.0, 8.9 Hz, 1H), 3.47–3.24 (m, 3H), 3.32 (s, 3H), 3.28 (s, 3H), 2.54 (m, 1H), 2.21 (m, 1H), 2.06 (dd, *J*=7.6, 12.4 Hz, 1H), 1.88 (m, 1H), 1.67–1.57 (m, 2H), 1.42–1.24 (m, 2H), 1.07 (d, *J*=7.0 Hz, 3H), 0.97 (d, *J*=7.0 Hz, 3H), 0.92 (d, *J*=7.0 Hz, 3H), 0.88 (s, 9H), 0.86 (s, 9H), 0.77 (d, *J*=7.0 Hz, 3H), 0.06 (s, 3H), 0.05 (s, 3H), 0.00 (s, 6H); ^{13}C NMR (67.8 MHz, $CDCl_3$) δ 139.4, 135.2, 129.5, 128.0, 127.2, 127.7, 104.5, 87.3, 31.2, 80.3, 70.5, 69.8, 65.3, 56.6, 54.3, 46.4, 42.6, 42.4, 38.8, 35.4, 33.7, 26.1, 26.0, 19.9, 18.3, 18.2, 16.0, 10.7, 9.7, −3.8, −4.4, −5.3, −5.4; HRMS (ESI) calcd for $C_{39}H_{72}NaO_6Si_2$ $(M+Na)^+$ 715.4765, found 715.4727.

4.1.20. Alcohol 27. Calcium (1.13 g, 28.1 mmol) was added to a stirred solution of olefin **26** (616 mg, 0.956 mmol) in THF (30 mL), isopropyl alcohol (10 mL), and liquid NH_3 (20 mL) cooled at −78 °C. After the mixture was stirred at −78 °C for 1.5 h, NH_4Cl (6.0 g) and $Fe(NO_3)_3\cdot 9H_2O$ (1.3 g) were added. The mixture was stirred at −78 °C for 1 h and allowed to warm to room temperature. The residue was diluted with H_2O (70 mL), and the mixture was stirred at room temperature for 1.5 h and extracted with EtOAc (3×50 mL). The combined extracts were washed with brine (50 mL), dried (Na_2SO_4), and concentrated. The residual oil was purified by column chromatography on silica gel (30 g, hexane–ether 5:1→1:1) to give **27** (512 mg, 89%) as a colorless oil: TLC, R_f 0.08 (hexane–ether 4:1); $[\alpha]_D^{28}$ −9.8 (*c* 1.00, $CHCl_3$); IR (neat) 3460 (br), 1458, 1253, 1089 cm^{-1}; ^1H NMR (270 MHz, $CDCl_3$) δ 5.67 (dd, *J*=6.5, 15.7 Hz, 1H), 5.48 (dd, *J*=5.7, 15.7 Hz, 1H), 4.90 (d, *J*=5.1 Hz, 1H), 4.29 (m, 1H), 3.98 (dt, *J*=7.0, 3.2 Hz, 1H), 3.55 (t, *J*=8.1 Hz, 1H), 3.44–3.20 (m, 3H), 3.32 (s, 3H), 3.31 (s, 3H), 2.57 (m, 1H), 2.26 (m, 1H), 2.05 (dd, *J*=7.0, 12.4 Hz, 1H), 1.93–1.73 (m, 2H), 1.68 (br s, OH), 1.57 (ddd, *J*=5.1, 11.1, 12.4 Hz, 1H), 1.31 (ddd, *J*=2.4, 9.5, 13.8 Hz, 1H), 1.25 (ddd, *J*=2.4, 9.7, 13.8 Hz, 1H), 1.04 (d, *J*=6.5 Hz, 3H), 0.97 (d, *J*=7.0 Hz, 3H), 0.89 (s, 9H), 0.87 (d, *J*=7.0 Hz, 3H), 0.86 (s, 9H), 0.77 (d, *J*=6.8 Hz, 3H), 0.05 (s, 3H), 0.04 (s, 3H), 0.00 (s, 6H); ^{13}C NMR (67.8 MHz, $CDCl_3$) δ 133.1, 130.3, 105.1, 89.1, 81.4, 74.8, 69.8, 65.5, 56.6, 55.2, 44.3, 42.5, 41.9, 38.1, 36.0, 33.6, 26.2, 26.1, 18.9, 18.4, 18.3, 15.9, 12.3, 10.9, −3.7, −4.2, −5.1, −5.2; HRMS (ESI) calcd for $C_{32}H_{66}NaO_6Si_2$ $(M+Na)^+$ 625.4296, found 625.4288.

4.1.21. Alcohol 28. A mixture of alcohol **27** (369 mg, 0.612 mmol), $NaHCO_3$ (206 mg, 2.45 mmol), and 5% Pd on carbon (121 mg) in ethanol (6 mL) was stirred under a hydrogen atmosphere at room temperature for 18 h. The mixture was filtered through a pad of Celite, and the residue was washed with EtOAc. The filtrate and the washings were combined and concentrated. The residual oil was purified by column chromatography on silica gel (20 g, hexane–ether 5:1→1:1) to give **28** (353 mg, 95%) as a colorless oil: TLC R_f 0.63 (hexane–ether 1:1); $[\alpha]_D^{26}$ +1.8 (*c* 1.00, $CHCl_3$); IR ($CHCl_3$) 3481, 1462, 1255, 1095 cm^{-1}; ^1H NMR (270 MHz, $CDCl_3$) δ 4.88 (d, *J*=5.1 Hz, 1H), 3.96 (m, 1H), 3.83 (m, 1H), 3.54 (t, *J*=7.8 Hz, 1H), 3.45 (dd, *J*=7.6, 10.3 Hz, 1H), 3.33 (dd, *J*=6.5, 10,3 Hz, 1H), 3.21 (m, 1H), 3.30 (s, 3H), 3.28 (s, 3H), 2.24 (m, 1H), 2.05 (dd, *J*=7.0, 12.3 Hz, 1H), 1.90–1.35 (m, 8H), 1.33–1.27 (m, 2H), 1.03 (d, *J*=6.5 Hz, 3H), 0.93 (d, *J*=7.0 Hz, 3H), 0.86 (s, 9H), 0.86 (s, 9H), 0.83 (d, *J*=7.0 Hz, 3H), 0.79 (d, *J*=6.8 Hz, 3H), 0.04 (s, 3H), 0.04 (s, 3H), 0.00 (s, 6H); HRMS (ESI) calcd for $C_{32}H_{69}O_6Si_2$ $(M+H)^+$ 605.4633, found 605.4649.

4.1.22. 3,4-Dimethoxybenzyloxymethyl ether 29. To a stirred solution of alcohol **28** (607 mg, 1.00 mmol) in CH_2Cl_2 (8 mL) cooled at 0 °C were added diisopropylethylamine (8.8 mL, 52 mmol) and the 1 M solution of (3,4-dimethoxybenzyloxy)methyl chloride in CH_2Cl_2 (13 mL, 13 mmol) prepared from 3,4-dimethoxybenzyl (methylthio)methyl ether according to Ref. 4c. The mixture was stirred at room temperature for 3 h, and the reaction was quenched by addition of MeOH (40 mL) and $NaHCO_3$ (500 mg). The resulting mixture was stirred at room temperature for 1.5 h, and H_2O (20 mL) was added. The organic layer was separated, and the aqueous layer was extracted with hexane (5×20 mL). The organic layer and the extracts were combined, washed with saturated aqueous $NaHCO_3$ (20 mL), H_2O (20 mL), and brine (20 mL), successively, dried (Na_2SO_4), and concentrated. The residual oil was purified by column chromatography on alumina (50 g, hexane–EtOAc 2:1) and silica gel (FL60D 25 g, benzene–acetone 100:1→50:1→20:1) to give **29** (663 mg, 84%) as a colorless oil: TLC, R_f 0.59 (hexane–EtOAc 3:1); $[\alpha]_D^{25}$ +8.7 (*c* 1.00,

$CHCl_3$); IR (neat) 1516, 1463, 1380, 1257, 1097, 1032 cm^{-1}; ^1H NMR (270 MHz, $CDCl_3$) δ 6.86–6.73 (m, 3H), 4.81 (d, J=4.6 Hz, 1H), 4.75 (s, 2H), 4.52 (s, 2H), 4.01 (m, 1H), 3.94 (m, 1H), 3.81 (s, 3H), 3.80 (s, 3H), 3.50 (dd, J=6.5, 9.7 Hz, 1H), 3.45–3.10 (m, 3H), 3.17 (s, 3H), 3.17 (s, 3H), 2.16 (m, 1H), 2.02 (dd, J=7.6, 12.7 Hz, 1H), 1.81 (m, 1H), 1.66 (m, 1H), 1.63–1.33 (m, 6H), 1.27–1.15 (m, 2H), 1.03 (d, J=6.5 Hz, 3H), 0.83 (d, J=7.0 Hz, 3H), 0.81 (s, 9H), 0.80 (s, 9H), 0.77 (d, J=7.0 Hz, 3H), 0.74 (d, J=7.0 Hz, 3H), 0.01 (s, 3H), 0.00 (s, 3H), −0.01 (s, 6H); ^{13}C NMR (67.8 MHz, $CDCl_3$) δ 148.9, 148.5, 130.8, 120.6, 111.4, 110.9, 104.7, 94.5, 87.3, 81.8, 78.6, 70.2, 69.5, 65.4, 57.0, 56.1, 56.0, 54.6, 43.6, 42.7, 42.5, 36.1, 35.5, 33.1, 30.8, 27.7, 26.3, 26.2, 26.1, 20.4, 18.4, 18.4, 15.6, 11.1, 9.1, −3.7, −4.2, −5.1, −5.2; HRMS (ESI) calcd for $C_{42}H_{80}NaO_9Si_2$ $(M+Na)^+$ 807.5239, found 807.5221.

4.1.23. Diol 30. To a stirred solution of 3,4-dimethoxybenzyloxymethyl ether **29** (664 mg, 0.846 mmol) in THF (42 mL) was added a 1.0 M solution of tetrabutylammonium fluoride in THF (2.0 mL, 2.0 mmol). The mixture was stirred at room temperature for 2 h, diluted with saturated aqueous NH_4Cl (9 mL), and extracted with ether (3×30 mL). The combined extracts were washed with brine (10 mL), dried (Na_2SO_4), and concentrated. The residual oil was purified by column chromatography on silica gel (20 g, hexane–EtOAc 1:1→EtOAc) to give **30** (466 mg, 99%) as a colorless oil: TLC, R_f 0.5 (EtOAc); $[\alpha]_D^{26}$ +6.9 (c 1.07, $CHCl_3$); IR (neat) 3428, 1516, 1265, 1095, 1029 cm^{-1}; ^1H NMR (270 MHz, $CDCl_3$) δ 6.83–6.71 (m, 3H), 4.78 (d, J=4.6 Hz, 1H), 4.75 (d, J=9.2 Hz, 1H), 4.72 (d, J=9.2 Hz, 1H), 4.49 (s, 2H), 3.95 (m, 1H), 3.78 (s, 3H), 3.77 (s, 3H), 3.87–3.39 (m, 5H), 3.32 (m, 1H), 3.26 (s, 3H), 3.19 (s, 3H), 2.12 (m, 1H), 1.99 (m, 1H), 1.85–1.27 (m, 10H), 1.00 (d, J=6.5 Hz, 3H), 0.82 (d, J=6.8 Hz, 3H), 0.78 (d, J=7.3 Hz, 3H), 0.71 (d, J=6.8 Hz, 3H); ^{13}C NMR (67.8 MHz, $CDCl_3$) δ 148.9, 148.5, 130.7, 120.5, 111.3, 110.9, 104.8, 94.6, 87.2, 83.6, 78.4, 75.2, 69.5, 68.1, 57.5, 56.3, 56.0, 55.9, 54.7, 43.6, 42.5, 40.1, 36.1, 35.0, 30.3, 27.5, 20.4, 15.9, 13.9, 9.0; HRMS (ESI) calcd for $C_{30}H_{52}NaO_9$ $(M+Na)^+$ 579.3509, found 579.3496.

4.1.24. TBDPS ether 31. To a stirred solution of diol **30** (80.8 mg, 0.145 mmol) in DMF (1.5 mL) cooled at 0 °C were added imidazole (749 mg, 8.71 mmol) and *tert*-butyldiphenylsilyl chloride (1.1 mL, 4.4 mmol). The mixture was stirred at room temperature for 5 h, diluted with cold water (1 mL), and extracted with ether (3×5 mL). The combined extracts were washed with brine (5 mL), dried (Na_2SO_4), and concentrated. The residual oil was purified by column chromatography on silica gel (10 g, benzene–ether 50:1→20:1) to give **31** (113 mg, 75%) as a colorless oil: TLC, R_f 0.7 (hexane–EtOAc 3:1); $[\alpha]_D^{26}$ +6.9 (c 1.00, $CHCl_3$); IR (neat) 1516, 1462, 1427, 1109, 1028 cm^{-1}; ^1H NMR (270 MHz, $CDCl_3$) δ 7.70–7.46 (m, 8H), 7.45–7.24 (m, 12H), 6.82–6.68 (m, 3H), 4.87 (d, J=4.6 Hz, 1H), 4.84 (d, J=7.0 Hz, 1H), 4.81 (d, J=7.0 Hz, 1H), 4.59 (s, 2H), 4.16 (m, 1H), 3.92 (m, 1H), 3.76 (s, 3H), 3.76 (s, 3H), 3.47 (dd, J=6.5, 10.0 Hz, 1H), 3.36 (dd, J=7.8, 10.0 Hz, 1H), 3.25 (dd, J=6.7, 10.0 Hz, 1H), 3.07 (s, 3H), 2.92 (m, 1H), 2.76 (s, 3H), 2.11 (m, 1H), 2.00 (m, 1H), 1.97 (dd, J=7.0, 12.2 Hz, 1H), 1.56–1.15 (m, 9H), 1.11 (d, J=6.5 Hz, 3H), 1.06 (s, 9H), 1.00 (s, 9H), 0.90 (d, J=7.0 Hz, 3H), 0.88 (d, J=7.0 Hz, 3H), 0.75 (d, J=7.0 Hz, 3H); ^{13}C NMR (150 MHz, $CDCl_3$) δ 149.1, 148.6, 136.3, 136.2, 135.8, 135.7, 134.7, 134.6, 134.0, 133.8, 130.9, 129.6, 129.6, 129.5, 127.7, 127.7, 127.5, 120.7, 111.4, 111.0, 104.7, 94.5, 87.3, 81.5, 78.6, 72.0, 69.5, 66.1, 56.8, 56.1, 55.9, 54.5, 43.5, 42.7, 42.0, 36.0, 35.6, 33.9, 30.7, 27.7, 27.4, 27.3, 26.9, 20.3, 19.7, 19.3, 15.1, 11.3, 8.9; HRMS (ESI) calcd for $C_{62}H_{88}NaO_9Si_2$ $(M+Na)^+$ 1055.5865, found 1055.5845; Anal. Calcd for $C_{62}H_{88}O_9Si_2$: C, 72.05; H, 8.58. Found: C, 72.53; H, 9.01.

4.1.25. Diol 32. To a stirred solution of TBDPS ether **31** (178 mg, 0.198 mmol) in 1,2-dimethoxyethane (5.0 mL) was added 1 M aqueous HCl (1.2 mL). The solution was stirred at room temperature for 7 h, cooled to 0 °C, and diluted with ether (10 mL) and saturated aqueous $NaHCO_3$ (10 mL). The organic layer was separated, and the aqueous layer was extracted with ether (3×10 mL). The organic layer and the extracts were combined, washed with brine (10 mL), dried (Na_2SO_4), and concentrated. The residual oil was purified by column chromatography on silica gel (6 g, hexane–EtOAc 5:1→4:1→2:1) to give a diastereomeric mixture of hemiacetals (143 mg) as a colorless oil along with recovered **31** (54 mg, 29%). Using the same procedure as described above, a diastereomeric mixture of hemiacetals (399 mg) was obtained from **31** (626 mg, 0.606 mmol). To a stirred solution of the hemiacetals (542 mg) in ethanol (10 mL) was added sodium borohydride (71.7 mg, 1.89 mmol). The mixture was stirred at room temperature for 1.5 h, diluted with saturated aqueous NH_4Cl (10 mL), and extracted with ether (5×20 mL). The combined extracts were washed with brine (10 mL), dried (Na_2SO_4), and concentrated. The residual oil was purified by column chromatography on silica gel (20 g, hexane–EtOAc 4:1→2:1→1:1→EtOAc) to give **32** (590 mg, 70%) as a colorless oil: TLC, R_f 0.2 (hexane–EtOAc 1:1); $[\alpha]_D^{28}$ −19.6 (c 1.00, $CHCl_3$); IR (neat) 3434, 1516, 1462, 1427, 1109, 1028 cm^{-1}; ^1H NMR (270 MHz, $CDCl_3$) δ 7.72–7.56 (m, 8H), 7.45–7.26 (m, 12H), 6.88–6.78 (m, 3H), 4.78 (d, J=7.0 Hz, 1H), 4.71 (d, J=7.0 Hz, 1H), 4.62 (d, J=11.6 Hz, 1H), 4.50 (d, J=11.6 Hz, 1H), 4.26 (m, 1H), 3.91 (m, 1H), 3.87 (s, 3H), 3.86 (s, 3H), 3.72 (m, 1H), 3.60–3.32 (m, 5H), 2.80 (s, 3H), 2.12 (m, 1H), 1.99–1.82 (m, 2H), 1.76–1.36 (m, 9H), 1.06 (s, 9H), 1.01 (d, J=7.0 Hz, 3H), 0.98 (s, 9H), 0.90 (d, J=7.0 Hz, 3H), 0.85 (d, J=6.5 Hz, 3H), 0.74 (d, J=6.2 Hz, 3H); ^{13}C NMR (67.8 MHz, $CDCl_3$) δ 149.1, 148.8, 136.1, 136.1, 135.7, 135.6, 134.5, 134.5, 133.8, 133.8, 129.8, 129.5, 129.5, 129.4, 127.6, 127.6, 127.5, 127.5, 120.5, 111.1, 111.0, 94.4, 81.1, 81.1, 77.4, 72.1, 70.4, 66.1, 59.7, 56.9, 56.1, 56.0, 42.0, 37.8, 35.4, 33.3, 32.7, 29.9, 29.7, 28.1, 27.4, 27.0, 19.8, 19.4, 17.4, 15.4, 11.6, 11.4; HRMS (ESI) calcd for $C_{61}H_{88}NaO_9Si_2$ $(M+Na)^+$ 1043.5865, found 1043.5869.

4.1.26. Trityl ether 33. To a stirred solution of diol **32** (63.5 mg, 0.0614 mmol) in pyridine (0.7 mL) was added trityl chloride (86.6 mg, 0.307 mmol) at room temperature. The solution was stirred at 50 °C for 12 h and was cooled to room temperature. Saturated aqueous $NaHCO_3$ (3 mL) was added, and the mixture was stirred at room temperature for 2 h and extracted with ether (3×5 mL). The combined extracts were washed with brine (5 mL), dried (Na_2SO_4), and concentrated. The residual oil was purified by column

chromatography on silica gel (2 g, hexane–ether–Et_3N 5:1:0.1→2:1:0→1:1:0) to give **33** (73.5 mg, 95%) as a colorless oil: TLC, R_f 0.8 (hexane–EtOAc 3:1); $[\alpha]_D^{28}$ −2.3 (*c* 1.00, $CHCl_3$); IR (neat) 3500, 1516, 1462, 1427, 1109, 1029 cm^{-1}; ^1H NMR (270 MHz, $CDCl_3$) δ 7.75–7.53 (m, 8H), 7.48–7.15 (m, 27H), 6.86–6.75 (m, 3H), 4.78 (d, *J*=7.0 Hz, 1H), 4.71 (d, *J*=7.0 Hz, 1H), 4.60 (d, *J*=11.6 Hz, 1H), 4.49 (d, *J*=11.6 Hz, 1H), 4.26 (m, 1H), 3.84 (s, 3H), 3.83 (s, 3H), 3.82 (m, 1H), 3.55–3.30 (m, 4H), 3.21 (m, 1H), 3.03 (m, 1H), 2.85 (s, 3H), 2.11 (m, 1H), 1.97–1.70 (m, 2H), 1.66–1.31 (m, 6H), 1.05 (s, 9H), 0.97 (s, 9H), 1.13–0.88 (m, 3H), 0.89 (d, *J*=6.8 Hz, 3H), 0.86 (d, *J*=8.1 Hz, 3H), 0.84 (d, *J*=6.5 Hz, 3H), 0.75 (d, *J*=6.8 Hz, 3H); ^{13}C NMR (150 MHz, $CDCl_3$) δ 149.1, 148.8, 144.6, 136.3, 136.2, 135.8, 135.7, 134.6, 134.6, 133.9, 133.8, 130.0, 129.6, 129.6, 129.5, 128.8, 127.8, 127.7, 127.7, 127.6, 127.0, 120.6, 111.2, 111.0, 94.6, 86.6, 81.8, 81.1, 77.6, 72.1, 70.3, 66.0, 62.0, 56.9, 56.0, 55.9, 41.9, 38.0, 35.4, 34.1, 31.9, 29.5, 29.4, 28.2, 27.3, 26.9, 19.7, 19.3, 17.5, 15.3, 11.7, 11.3; HRMS (ESI) calcd for $C_{80}H_{102}NaO_9Si_2$ $(M+Na)^+$ 1285.6960, found 1285.6974.

4.1.27. Acetate 34. To a stirred solution of trityl ether **33** (688 mg, 0.545 mmol) in pyridine (4.0 mL) were added acetic anhydride (2.0 mL) and 4-(dimethylamino)pyridine (30.0 mg, 0.246 mmol). The mixture was stirred at room temperature for 12 h and concentrated. The residual oil was purified by column chromatography on silica gel (10 g, hexane–ether 4:1→2:1→1:1) to give **34** (725 mg, 100%) as a colorless oil: TLC, R_f 0.8 (hexane–EtOAc 3:1); $[\alpha]_D^{28}$ +5.1 (*c* 1.00, $CHCl_3$); IR (neat) 1732, 1516, 1462, 1427, 1240, 1109, 1031 cm^{-1}; ^1H NMR (270 MHz, $CDCl_3$) δ 7.75–7.55 (m, 8H), 7.48–7.18 (m, 27H), 6.87–6.77 (m, 3H), 4.99 (dd, *J*=2.7, 10.0 Hz, 1H), 4.75 (d, *J*=7.0 Hz, 1H), 4.67 (d, *J*=7.0 Hz, 1H), 4.60 (d, *J*=11.9 Hz, 1H), 4.46 (d, *J*=11.9 Hz, 1H), 4.26 (m, 1H), 3.85 (s, 6H), 3.48 (dd, *J*=7.6, 10.3 Hz, 1H), 3.43–3.34 (m, 2H), 3.20 (m, 1H), 3.13–2.92 (m, 2H), 2.81 (s, 3H), 2.36–1.75 (m, 4H), 1.97 (s, 3H), 1.70–1.20 (m, 5H), 1.06 (s, 9H), 0.97 (s, 9H), 1.13–0.80 (m, 3H), 0.93 (d, *J*=7.0 Hz, 3H), 0.89 (d, *J*=7.0 Hz, 3H), 0.75 (d, *J*=6.6 Hz, 3H), 0.70 (d, *J*=6.6 Hz, 3H); ^{13}C NMR (67.8 MHz, $CDCl_3$) δ 170.8, 148.9, 148.5, 144.3, 136.2, 136.1, 135.7, 135.6, 134.6, 134.6, 133.8, 133.7, 130.8, 129.5, 128.7, 127.8, 127.6, 127.6, 127.5, 126.9, 120.4, 111.3, 110.9, 95.4, 86.6, 81.5, 78.8, 77.4, 72.2, 69.7, 66.1, 61.5, 56.6, 56.1, 56.0, 42.0, 37.0, 34.9, 33.7, 31.2, 30.8, 29.8, 27.4, 27.1, 27.0, 21.3, 19.8, 19.4, 17.2, 15.3, 11.5, 9.8; HRMS (ESI) calcd for $C_{82}H_{104}NaO_{10}Si_2$ $(M+Na)^+$ 1327.7066, found 1327.7075.

4.1.28. Alcohol 35. To a stirred solution of acetate **34** (52.4 mg, 0.0401 mmol) in ether (0.6 mL) was added formic acid (0.4 mL), and the mixture was stirred at room temperature (23 °C) for 15 min. The mixture was poured into saturated aqueous $NaHCO_3$ (4 mL) cooled at 0 °C, and the resulting mixture was extracted with ether (3×3 mL). The combined extracts were washed with brine (3 mL), dried (Na_2SO_4), and concentrated. The residual oil was purified by column chromatography on silica gel (2 g, hexane–ether 2:1→1:1→1:2→1:5) to give **35** (33.0 mg, 77%) as a colorless oil: TLC, R_f 0.4 (hexane–ether 1:5); $[\alpha]_D^{28}$ −1.1 (*c* 1.00, $CHCl_3$); IR (neat) 3497, 1731, 1516, 1240, 1109, 1031 cm^{-1}; ^1H NMR (270 MHz, $CDCl_3$) δ 7.69–7.58 (m, 8H), 7.43–7.27 (m, 12H), 6.88–6.78 (m, 3H), 4.98 (dd, *J*=2.7, 9.4 Hz, 1H), 4.74 (d, *J*=7.0 Hz, 1H), 4.65 (d, *J*=7.0 Hz, 1H), 4.59 (d, *J*=11.6 Hz, 1H), 4.46 (d, *J*=11.6 Hz, 1H), 4.25 (m, 1H), 3.86 (s, 3H), 3.86 (s, 3H), 3.86 (m, 1H), 3.74 (m, 1H), 3.67–3.32 (m, 3H), 3.02 (m, 1H), 2.80 (s, 3H), 2.18–1.90 (m, 2H), 2.04 (s, 3H), 1.86 (m, 1H), 1.80–1.15 (m, 6H), 1.05 (s, 9H), 0.97 (s, 9H), 1.03–0.75 (m, 3H), 0.91 (d, *J*=7.0 Hz, 3H), 0.89 (d, *J*=6.8 Hz, 3H), 0.88 (d, *J*=7.3 Hz, 3H), 0.77 (d, *J*=6.8 Hz, 3H); ^{13}C NMR (150 MHz, $CDCl_3$) δ 170.9, 148.9, 148.5, 136.1, 136.1, 135.7, 135.6, 134.6, 134.4, 133.7, 133.7, 130.6, 129.5, 129.4, 127.6, 127.5, 127.4, 120.4, 111.1, 110.7, 95.1, 81.3, 78.3, 78.2, 71.8, 69.5, 65.9, 60.7, 56.4, 55.9, 55.8, 41.7, 36.4, 34.4, 32.7, 30.9, 30.3, 29.7, 27.1, 26.7, 26.5, 21.1, 19.6, 19.1, 16.8, 15.1, 11.1, 9.4; HRMS (ESI) calcd for $C_{63}H_{90}NaO_{10}Si_2$ $(M+Na)^+$ 1085.5970, found 1085.5970.

4.1.29. Aldehyde 36. To a stirred solution of alcohol **35** (19.0 mg, 0.0178 mmol) in CH_2Cl_2 (0.2 mL) were added pyridine (0.02 mL, 0.178 mmol) and the Dess–Martin periodinane (10.3 mg, 0.0243 mmol). The mixture was stirred at room temperature for 30 min and diluted with saturated aqueous $Na_2S_2O_3$ (2 mL) and saturated aqueous $NaHCO_3$ (1 mL). The resulting mixture was stirred at room temperature for 30 min and extracted with ether (3×2 mL). The combined extracts were washed with H_2O (2 mL) and brine (2 mL), dried (Na_2SO_4), and concentrated. The residual oil was purified by column chromatography on silica gel (0.3 g, hexane–ether 1:1→1:2) to give **36** (17.3 mg, 91%) as a colorless oil: TLC, R_f 0.8 (hexane–EtOAc 1:1); $[\alpha]_D^{28}$ −2.3 (*c* 1.00, $CHCl_3$); IR (neat) 2719, 1730, 1516, 1238, 1169, 1031 cm^{-1}; ^1H NMR (270 MHz, $CDCl_3$) δ 9.73 (br s, 1H), 7.76–7.55 (m, 8H), 7.48–7.22 (m, 12H), 6.89–6.79 (m, 3H), 5.01 (dd, *J*=3.0, 9.5 Hz, 1H), 4.76 (d, *J*=7.0 Hz, 1H), 4.66 (d, *J*=7.0 Hz, 1H), 4.61 (d, *J*=11.9 Hz, 1H), 4.47 (d, *J*=11.9 Hz, 1H), 4.27 (m, 1H), 3.88 (s, 6H), 3.52–3.35 (m, 3H), 3.03 (m, 1H), 2.80 (s, 3H), 2.48 (m, 1H), 2.43 (m, 1H), 2.39–1.97 (m, 3H), 1.77 (m, 1H), 1.70–1.23 (m, 6H), 1.07 (s, 9H), 0.97 (s, 9H), 0.96 (d, *J*=6.8 Hz, 3H), 0.91 (d, *J*=6.8 Hz, 3H), 0.89 (d, *J*=7.3 Hz, 3H), 0.75 (d, *J*=6.8 Hz, 3H); ^{13}C NMR (150 MHz, $CDCl_3$) δ 201.8, 170.7, 149.1, 148.7, 136.3, 136.2, 135.8, 135.7, 134.7, 134.6, 133.9, 133.9, 130.8, 129.6, 129.6, 129.6, 129.5, 127.7, 127.7, 127.6, 120.5, 111.3, 111.0, 95.2, 81.5, 78.4, 77.8, 72.1, 69.7, 66.1, 56.6, 56.1, 55.9, 45.0, 41.9, 37.0, 34.7, 33.4, 30.7, 29.9, 29.7, 27.3, 26.9, 21.1, 19.7, 19.3, 18.2, 15.3, 11.3, 9.8; HRMS (ESI) calcd for $C_{68}H_{92}NaO_{10}Si_2$ $(M+Na)^+$ 1083.5814, found 1083.5833.

4.1.30. Enamide 37. A solution of aldehyde **36** (112 mg, 0.106 mmol), *N*-methylformamide (1.1 mL, 19 mmol), hydroquinone (46.3 mg, 0.421 mmol), and pyridinium *p*-toluenesulfonate (79.4 mg, 0.316 mmol) in benzene (50 mL) was heated to reflux for 5 h under a stream of nitrogen with continuous removal of water using molecular sieves of 3 Å. The mixture was cooled to room temperature, diluted with triethylamine (2 mL) and saturated aqueous $NaHCO_3$ (5 mL), and extracted with ether (3×10 mL). The combined extracts were washed with brine (10 mL), dried (Na_2SO_4), and concentrated. The residual oil was purified by column chromatography on silica gel (20 g, hexane–EtOAc 4:1→2:1→1:1→1:2) to give **37** (64.6 mg, 55%) as a colorless

oil: TLC, R_f 0.5 (hexane–EtOAc 1:1); $[\alpha]_D^{28}$ −10.3 (*c* 1.00, $CHCl_3$); IR (neat) 1733, 1654, 1515, 1238, 1109, 1030 cm^{-1}; ^1H NMR (270 MHz, $CDCl_3$) δ 8.24 [7.92] (s, 1H), 7.72–7.54 (m, 8H), 7.45–7.22 (m, 12H), 6.45 [7.14] (d, *J*=14.0 Hz, 1H), 6.88–6.78 (m, 3H), 4.99 (dd, *J*=9.2, 14.0 Hz, 1H), 4.75 (d, *J*=7.0 Hz, 1H), 4.75 (m, 1H), 4.64 [4.65] (d, *J*=7.0 Hz, 1H), 4.69 (d, *J*=11.9 Hz, 1H), 4.46 [4.48] (d, *J*=11.9 Hz, 1H), 4.29 (m, 1H), 3.89 (s, 3H), 3.83 (s, 3H), 3.52–3.32 (m, 3H), 3.01 (m, 1H), 2.87 (s, 3H), 2.80 [2.81] (s, 3H), 2.76 (m, 1H), 2.06 (s, 3H), 2.19–1.97 (m, 2H), 2.84–1.22 (m, 4H), 1.06 (s, 9H), 0.97 (s, 9H), 1.16–0.78 (m, 3H), 0.88 (d, *J*=6.8 Hz, 3H), 0.86 (d, *J*=8.1 Hz, 3H), 0.74 (d, *J*=6.8 Hz, 3H). The minor counterparts of doubled signals in the ratio of 2:1 are in brackets; HRMS (ESI) calcd for $C_{65}H_{91}NNaO_{10}Si_2$ $(M+Na)^+$ 1124.6079, found 1124.6060.

4.1.31. Alcohol 38. To a stirred solution of enamide **37** (30.0 mg, 0.0272 mmol) in CH_2Cl_2 (7.6 mL), *tert*-butyl alcohol (0.4 mL), and 1 M phosphate buffer (pH 6, 0.4 mL) cooled at 0 °C was added 2,3-dichloro-5,6-dicyano-*p*-benzoquinone (DDQ) (8.3 mg, 0.030 mmol). The mixture was warmed to room temperature and stirred at room temperature for 30 min. To the mixture cooled at 0 °C was added DDQ (7.0 mg, 0.025 mmol), and the mixture was stirred at room temperature for 40 min. Further, the mixture was cooled to 0 °C and DDQ (8.0 mg, 0.029 mmol) was added. After the mixture was stirred at room temperature for 40 min, DDQ (7.5 mg, 0.027 mmol) was added to the mixture cooled at 0 °C. The mixture was stirred at room temperature for 20 min and diluted with 1 M phosphate buffer (pH 6, 5 mL) was added. The mixture was stirred at room temperature for 1 h and extracted with ether (3×5 mL). The combined extracts were washed with brine (5 mL), dried (Na_2SO_4), and concentrated. The residual oil was purified by column chromatography on silica gel (3 g, hexane–ether–acetone 15:15:1) to give **38** (22.7 mg, 90%) as a colorless oil: TLC, R_f 0.4 (benzene–ether–acetone 10:1:1); $[\alpha]_D^{21}$ +28.3 (*c* 0.97, $CHCl_3$); IR (neat) 3514, 1696, 1657, 1427, 1249, 1109 cm^{-1}; ^1H NMR (270 MHz, $CDCl_3$) δ 8.29 [8.02] (s, 1H), 7.78–7.54 (m, 8H), 7.47–7.22 (m, 12H), 6.50 [7.17] (d, *J*=14.3 Hz, 1H), 5.00 [5.01] (dd, *J*=9.5, 14.3 Hz, 1H), 4.83 (m, 1H), 4.28 (m, 1H), 3.51–3.33 (m, 3H), 3.04 (m, 1H), 2.98 [2.96] (s, 3H), 2.81 (s, 3H), 2.57 (m, 1H), 2.50 (m, 1H), 2.10 (m, 1H), 2.15 (s, 3H), 1.44–1.17 (m, 4H), 1.06 (s, 9H), 1.05 (d, *J*=6.5 Hz, 3H), 0.97 (s, 9H), 1.16–0.78 (m, 3H), 0.88 (d, *J*=6.8 Hz, 3H), 0.85 (d, *J*=6.0 Hz, 3H), 0.73 (d, *J*=6.5 Hz, 3H). The minor counterparts of doubled signals in the ratio of 2:1 are in brackets; HRMS (ESI) calcd for $C_{55}H_{79}NNaO_7Si_2$ $(M+Na)^+$ 944.5293, found 944.5297.

4.1.32. Ester 39. To a stirred solution of alcohol **38** (3.2 mg, 0.0035 mmol) and 2,3-di-*O*-methyl-D-glyceric acid (12.8 mg, 0.0955 mmol) were added triethylamine (0.027 mL, 0.019 mmol), 2,4,6-trichlorobenzoyl chloride (0.023 mL, 0.014 mmol), and 4-(dimethylamino)pyridine (0.7 mg, 0.006 mmol). The mixture was stirred at room temperature for 1.5 h, diluted with 10% citric acid (5 mL), and extracted with ether (5×5 mL). The combined extracts were washed with brine (5 mL), dried (Na_2SO_4), and concentrated. The residual oil was purified by column chromatography on silica gel (2 g, benzene–acetone 30:1→20:1→15:1→10:1) to give a diastereomeric mixture of **39** (2.7 mg, 74%) as a colorless oil, which was employed in the next experiment without separation of the diastereomers: TLC, R_f 0.3 (benzene–acetone 8:1); ^1H NMR (400 MHz, $CDCl_3$) δ 8.26 [7.95, 8.25, 7.93][a] (s, 1H), 7.69–7.56 (m, 8H), 7.43–7.27 (m, 12H), 6.47 [7.14][b] (d, *J*=14.0 Hz, 1H), 5.07 [5.05][c] (m, 1H), 4.94 (dd, *J*=6.2, 14.0 Hz, 1H), 4.76 [4.80][c] (m, 1H), 4.25 (m, 1H), 3.92 (m, 1H), 3.80–3.58 (m, 2H), 3.42 [3.43][b] (s, 3H), 3.38 [3.38, 3.35][d] (s, 3H), 3.51–3.36 (m, 2H), 3.00 (m, 1H), 2.91 [2.86, 2.89, 2.63][a] (s, 3H), 2.54 (m, 1H), 2.07 [2.08][c] (s, 3H), 1.67–1.18 (m, 9H), 1.04 (s, 9H), 1.00 [0.99][c] (d, *J*=6.8 Hz, 3H), 0.96 (s, 9H), 0.94 [0.93, 0.91, 0.90][a] (d, *J*=7.3 Hz, 3H), 0.86 [0.86][b] (d, *J*=7.3 Hz, 3H), 0.72 (d, *J*=6.8 Hz, 3H). The minor counterparts of doubled signals in the ratios of 9:6:3:2 (superscript a), 3:2 (superscript b), 2:1 (superscript c), and 3:4:2 (superscript d) are in brackets.

4.1.33. Analogs 4 and 40. A solution of esters **39** (2.7 mg, 0.0026 mmol) in a 5:3:8 mixture of HF·pyridine, pyridine, and THF (0.32 mL) was stirred at room temperature for 7 h. The mixture was diluted with EtOAc (5 mL) and poured into saturated aqueous $NaHCO_3$ (5 mL) cooled at 0 °C, and the resulting mixture was extracted with EtOAc (3×5 mL). The combined extracts were washed with brine (5 mL), dried (Na_2SO_4), and concentrated. The residual oil was purified by column chromatography on silica gel (FL60D 1 g, benzene–acetone 2:1→1:1) followed by reversed-phase HPLC [Develosil Ph-UG-5 (20×250 mm), 50% aqueous MeOH, 5.0 mL/min, detection at UV 215 nm] to give **4** (t_R=91 min, 0.8 mg, 55%) and **40** (t_R=97 min, 0.5 mg, 34%) as a colorless oil, respectively. Compound **4**: TLC, R_f 0.5 (benzene–acetone 1:1); $[\alpha]_D^{28}$ +88.5 (*c* 0.067, $CHCl_3$); IR (neat) 3675, 1575, 1488, 1237, 1202, 1043 cm^{-1}; ^1H NMR (400 MHz, $CDCl_3$) δ 8.30 [8.08] (s, 1H), 6.49 [7.17] (d, *J*=14.0 Hz, 1H), 5.11 (m, 1H), 4.97 [4.99] (dd, *J*=9.6, 14.0 Hz, 1H), 4.80 (dd, *J*=2.8, 10.0 Hz, 1H), 3.93 (m, 1H), 3.79–3.29 (m, 5H), 3.50 (s, 3H), 3.40 (s, 3H), 3.37 (s, 3H), 3.20 (m, 1H), 3.03 [3.07] (s, 3H), 2.51 (m, 1H), 2.09 [2.08] (s, 3H), 1.90–1.72 (m, 2H), 1.76 (m, 1H), 1.69–1.36 (m, 6H), 1.02 [1.01] (d, *J*=6.8 Hz, 3H), 0.98 (d, *J*=6.8 Hz, 3H), 0.89 (d, *J*=6.8 Hz, 3H), 0.82 (d, *J*=6.8 Hz, 3H). The minor counterparts of doubled signals in the ratio of 2:1 are in brackets; HRMS (ESI) calcd for $C_{28}H_{51}NNaO_{10}$ $(M+Na)^+$ 584.3411, found 584.3398. Compound **40**: TLC, R_f 0.5 (benzene–acetone 1:1); $[\alpha]_D^{28}$ +103 (*c* 0.042, $CHCl_3$); IR (neat) 3648, 1575, 1488, 1237, 1202, 1043 cm^{-1}; ^1H NMR (400 MHz, $CDCl_3$) δ 8.29 [8.08] (s, 1H), 6.49 [7.16] (d, *J*=14.0 Hz, 1H), 5.13 (m, 1H), 4.98 [4.99] (m, 1H), 4.82 (dd, *J*=2.8, 10.8 Hz, 1H), 3.94 (m, 1H), 3.77–3.09 (m, 6H), 3.49 (s, 3H), 3.38 (s, 3H), 3.37 (s, 3H), 3.03 [3.07] (s, 3H), 2.54 (m, 1H), 2.17 [2.09] (s, 3H), 1.90–1.39 (m, 9H), 1.02 [1.01] (d, *J*=7.2 Hz, 3H), 0.95 (d, *J*=6.8 Hz, 3H), 0.89 [0.89] (d, *J*=6.8 Hz, 3H), 0.83 [0.83] (d, *J*=7.2 Hz, 3H). The minor counterparts of doubled signals in the ratio of 2:1 are in brackets; HRMS (ESI) calcd for $C_{28}H_{51}NNaO_{10}$ $(M+Na)^+$ 584.3411, found 584.3398.

4.2. Actin-depolymerizing activity

The actin-depolymerizing activity of the compounds was measured as previously described.[3,6] To the 3.7 μM solution of actin (0.30 mL, containing 10% pyrenyl actin) in G-buffer

was added a 0.15 M solution of $MgCl_2$ (2.0 μL), and the mixture was stirred at room temperature for 1 h to polymerize G-actin to F-actin. To the solution of F-actin was added various concentrations of compounds in dimethyl sulfoxide (2.0 μL) with stirring, and the time course of depolymerization was continuously monitored by measuring fluorescence of pyrenyl actin (10% of total actin) with a fluorometer (HITACHI, F-4000, equipped with a magnetic stirrer) at 25 °C at 365-nm excitation and 407-nm emission wavelengths. The IC_{50} values were the concentrations required to depolymerize F-actin to 50% of its control amplitude.

4.3. Cytotoxicity

Growing cells of HeLa S_3 were suspended in Eagle's minimal essential medium containing 10% fetal bovine serum, penicillin (0.1 units/mL), streptomycin (0.1 mg/mL), and amphotericin B (0.25 μg/mL) at 3×10^4 cells/mL, and samples dissolved in DMSO were added. The mixture was incubated at 37 °C for 4 days in a CO_2 incubator with a humidified atmosphere containing 5% CO_2. The number of viable cells was assessed using an MTT (3-[4,5-dimethylthiazol-2-yl]-2,5-diphenyl-tetrazolium bromide) assay.[22] The IC_{50} value (concentration required for 50% inhibition of cell growth) was determined using a growth curve.

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References and notes

1. (a) Fenteany, G.; Shoutian, Z. *Curr. Top. Med. Chem.* **2003**, *3*, 593–616; (b) Yeung, K.-S.; Paterson, I. *Angew. Chem., Int. Ed.* **2002**, *41*, 4632–4653.
2. (a) Yamada, K.; Ojika, M.; Ishigaki, T.; Yoshida, Y.; Ekimoto, H.; Arakawa, M. *J. Am. Chem. Soc.* **1993**, *115*, 11020–11021; (b) Ojika, M.; Kigoshi, H.; Ishigaki, T.; Yamada, K. *Tetrahedron Lett.* **1993**, *34*, 8501–8504; (c) Ojika, M.; Kigoshi, H.; Ishigaki, T.; Nisiwaki, M.; Tsukada, I.; Mizuta, K.; Yamada, K. *Tetrahedron Lett.* **1993**, *34*, 8505–8508; (d) Ojika, M.; Kigoshi, H.; Ishigaki, T.; Tsukada, I.; Tsuboi, T.; Ogawa, T.; Yamada, K. *J. Am. Chem. Soc.* **1994**, *116*, 7441–7442.
3. Saito, S.; Watabe, S.; Ozaki, H.; Kigoshi, H.; Yamada, K.; Fusetani, N.; Karaki, H. *J. Biochem.* **1996**, *120*, 552–555.
4. (a) Kigoshi, H.; Ojika, M.; Suenaga, K.; Mutou, T.; Hirano, J.; Sakakura, A.; Ogawa, T.; Nisiwaki, M.; Yamada, K. *Tetrahedron Lett.* **1994**, *35*, 1247–1250; (b) Kigoshi, H.; Ojika, M.; Ishigaki, T.; Suenaga, K.; Mutou, T.; Sakakura, A.; Ogawa, T.; Yamada, K. *J. Am. Chem. Soc.* **1994**, *116*, 7443–7444; (c) Kigoshi, H.; Suenaga, K.; Mutou, T.; Ishigaki, T.; Atsumi, T.; Ishiwata, H.; Sakakura, A.; Ogawa, T.; Ojika, M.; Yamada, K. *J. Org. Chem.* **1996**, *61*, 5326–5351; (d) Suenaga, K.; Kamei, N.; Okugawa, Y.; Takagi, M.; Akao, A.; Kigoshi, K.; Yamada, K. *Bioorg. Med. Chem. Lett.* **1997**, *7*, 269–274; (e) Kigoshi, H.; Suenaga, K.; Takagi, M.; Akao, A.; Kanematsu, K.; Kamei, N.; Okugawa, Y.; Yamada, K. *Tetrahedron* **2002**, *58*, 1075–1102.
5. Hirata, K.; Muraoka, S.; Suenaga, K.; Kuroda, T.; Kato, K.; Tanaka, H.; Yamamoto, M.; Takata, M.; Yamada, K.; Kigoshi, H. *J. Mol. Biol.* **2006**, *356*, 945–954.
6. Kuroda, T.; Suenaga, K.; Sakakura, A.; Handa, T.; Okamoto, K.; Kigoshi, H. *Bioconjugate Chem.* **2006**, *17*, 524–529.
7. (a) Fusetani, N.; Yasumuro, K.; Matsunaga, S.; Hashimoto, K. *Tetrahedron Lett.* **1989**, *30*, 2809–2812; (b) Matsunaga, S.; Liu, P.; Celatka, C. A.; Panek, J. S.; Fusetani, N. *J. Am. Chem. Soc.* **1999**, *121*, 5605–5606.
8. Hori, M.; Saito, S.; Shin, Y.; Ozaki, H.; Fusetani, N.; Karaki, H. *FEBS Lett.* **1993**, *322*, 151–154.
9. Saito, S.; Watabe, S.; Ozaki, H.; Fusetani, N.; Karaki, H. *J. Biol. Chem.* **1994**, *269*, 29710–29714.
10. (a) Liu, P.; Panek, J. S. *J. Am. Chem. Soc.* **2000**, *122*, 1235–1236; (b) Panek, J. S.; Liu, P. *J. Am. Chem. Soc.* **2000**, *122*, 11090–11097.
11. (a) Klenchin, V. A.; Allingham, J. S.; King, R.; Tanaka, J.; Marriott, G.; Rayment, I. *Nature Struct. Biol.* **2003**, *10*, 1058–1063; (b) Allingham, J. S.; Tanaka, J.; Marriott, G.; Rayment, I. *Org. Lett.* **2004**, *6*, 597–599.
12. Suenaga, K.; Miya, S.; Kuroda, T.; Handa, T.; Kanematsu, K.; Sakakura, A.; Kigoshi, H. *Tetrahedron Lett.* **2004**, *45*, 5383–5386.
13. (a) Walker, M. A.; Heathcock, C. H. *J. Org. Chem.* **1991**, *56*, 5747–5750; (b) Raimundo, B. C.; Heathcock, C. H. *Synlett* **1995**, 1213–1214.
14. Roush, W. R.; Ando, K.; Powers, D. B.; Palkowitz, A. D.; Halterman, R. L. *J. Am. Chem. Soc.* **1990**, *112*, 6339–6348.
15. Evans, D. A.; Bartroli, J.; Shih, T. L. *J. Am. Chem. Soc.* **1981**, *103*, 2127–2129.
16. Nakagawa, I.; Hata, T. *Tetrahedron Lett.* **1975**, 1409–1412.
17. (a) Paterson, I.; Tiller, R. D. *Tetrahedron Lett.* **1991**, *32*, 1749–1752; (b) Paterson, I.; Lister, M. A.; Ryan, G. R. *Tetrahedron Lett.* **1992**, *33*, 4233–4236.
18. Evans, D. A.; Chapman, K. T.; Carreira, E. M. *J. Am. Chem. Soc.* **1988**, *110*, 3560–3578.
19. (a) Rychnovsky, S. D.; Skalitzky, D. J. *Tetrahedron Lett.* **1990**, *31*, 945–948; (b) Evans, D. A.; Rieger, D. L. *Tetrahedron Lett.* **1990**, *31*, 7099–7100; (c) Rychnovsky, S. D.; Rogers, B.; Yang, G. *J. Org. Chem.* **1993**, *58*, 3511–3515.
20. The stereochemistry at C35 of acetals **24a** and **24b** was not determined.
21. The stereochemistry of **4** and **26** concerning the 2,3-di-*O*-methylglyceroyl group was determined by degradation and chiral HPLC analyses in the same manner as that used for mycalolide B.[7b]
22. Alley, M. C.; Scudiero, D. A.; Monks, A.; Hursey, M. L.; Czerwinski, M. J.; Fine, D. L.; Abbott, B. J.; Mayo, J. G.; Shoemaker, R. H.; Boyd, M. R. *Cancer Res.* **1988**, *48*, 589–601.