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ORIGINAL ARTICLE

Total synthesis and absolute configuration of avenolide, extracellular factor in *Streptomyces avermitilis*

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The first total synthesis of extracellular factor, "Avenolide", in *Streptomyces avermitilis* has been achieved using a convergent approach. The stereogenic centers in two key segments were installed using Sharpless epoxidation and dihydroxylation. This synthetic study allowed the determination of the absolute configuration of avenolide as 4*S*,10*R*, and yielded important information on its structure–activity relationship.
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Keywords: absolute configuration; avenolide; first total synthesis

INTRODUCTION

In several streptomycete microorganisms, γ-butyrolactones have important, sometimes crucial, roles as extracellular factors in determining the onset of secondary metabolite production and morphological differentiation. Up to date, the structures of all extracellular factors are consisted of γ-butyrolactones.[1,2] A-factor is required for streptomycin biosynthesis and sporulation in *Streptomyces griseus*[1,2] and virginia butanolide appears to control virginiamycin biosynthesis in *Streptomyces virginiae*.[3–5] Other studies γ-butyrolactones include IM-2 elicits production of showdomycin and minimycin in *Streptomyces lavendulae* FRI-5[3–5] and SCB1 also elicits the precocious production of actinorhodin in *Streptomyces coelicolor* A3(2).[3–5]

Genome-sequenced *Streptomyces avermitilis* is an important industrial microorganism for the production of anthelmintic and insecticidal macrocyclic lactone, avermectin, which is used as antiparasitic agents in the medical, veterinary and agricultural fields. To identify the extracellular factor(s) controlling avermectin production in *Streptomyces avermitilis*, about 1 mg of the extracellular factor from 1000 l of culture filtrate was isolated and purified by chromatographic separation steps.[6] Since spectroscopic analyses of the extracellular factor purified have elucidated that the factor is a new butenolide structure including two stereogenic centers (Scheme 1) but not γ-butyrolactone structure, the extracellular factor is named as "avenolide." The absolute configuration could not be elucidated, however, due to the trace quantities isolated and the presence of contaminants. Only the configuration of C4 (*S*, avenolide numbering) was proposed, by comparison of the CD spectra of avenolide and similar butenolide compounds.[7,8]

To undertake further biochemical properties of avenolide, the determination of the absolute structure, access to an adequate supply of avenolide and the elucidation of the structure–activity relationship are required. We describe here the first total synthesis of avenolide, which represents significant progress towards these objectives.

RESULTS AND DISCUSSION

We first embarked on the synthesis of (4*S*,10*R*)-avenolide (**1**). As outlined in Scheme 1, our retrosynthetic strategy is convergent. We speculated that **1** could be derived from epoxide **2** via ring closing metathesis, which itself could be constructed by the coupling of aldehyde **3** and iodide **4**. This route should provide a concise route amenable to large-scale synthesis of avenolide as well as various avenolide analogs, including stereoisomers.

The synthesis of **1** commenced with esterification of a commercially available β-methallyl alcohol with *p*-anisic acid (Scheme 2). Subsequent Sharpless dihydroxylation[9,10] using $(DHQ)_2PHAL$ as a chiral ligand afforded **5** (93% ee).[11–13] (The enantiomeric excess was determined by chiral HPLC analysis (condition: DAICEL CHIRALPAK AS-3 (0.46 cm ϕ×25 cm), 3:1 hexanes/2-propanol mobile phase (flow rate at 1.0 ml per min) and detection at 254 nm (room temperature, rt)). Tosylation of the primary alcohol followed by a substitution reaction with $Me_2CuCNLi_2$ and methanolysis gave diol **6**. After acetal formation, DIBAL reduction and Parikh–Doering oxidation[14] furnished the desired aldehyde **3**.

Another key intermediate **4** was derived from a commercially available 1,5-pentanediol. Mono-TBS protection, TEMPO oxidation, Wittig olefination with $Ph_3P{=}CHCO_2Et$ and DIBAL reduction

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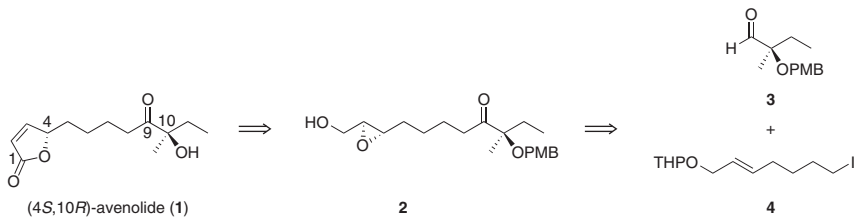
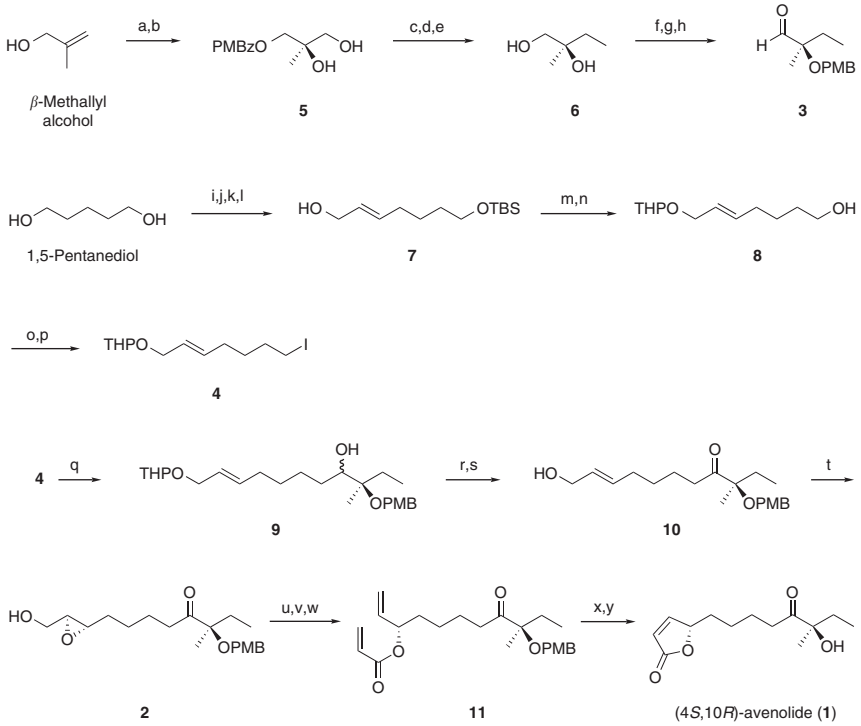


Scheme 1 Retrosynthetic analysis of (4*S*,10*R*)-avenolide (**1**).



Scheme 2 Reagents and conditions: (a) *p*-anisic acid, WSC, DMAP, CH_2Cl_2, rt, 99%; (b) $K_3Fe(CN)_6$, K_2CO_3, $(DHQ)_2PHAL$, $K_2OsO_4(OH)_4$, *t*-BuOH:H_2O=1:1, 0 °C, quant., 93% ee; (c) *p*-TsCl, Et_3N, Me_3N·HCl, CH_2Cl_2, rt, 99%; (d) CuCN, MeLi, THF, 0 °C; (e) K_2CO_3, MeOH, rt, 90% (two steps); (f) *p*-methoxybenzaldehyde dimethylacetal, PPTS, rt, CH_2Cl_2, quant.; (g) DIBAL, CH_2Cl_2, 0 °C, 87%; (h) SO_3·Py, DMSO, Et_3N, CH_2Cl_2, rt, 89%; (i) NaH, TBSCl, THF, rt, 80%; (j) $PhI(OAc)_2$, TEMPO, CH_2Cl_2, rt, quant.; (k) Ph_3P=$CHCO_2Et$, benzene, 80 °C, quant.; (l) DIBAL, CH_2Cl_2, 0 °C, quant.; (m) PPTS, DHP, CH_2Cl_2, 0 °C, quant.; (n) TBAF, THF, rt, quant.; (o) MsCl, Et_3N, Me_3N·HCl, CH_2Cl_2, rt, 93%; (p) NaI, acetone, reflux, quant.; (q) *t*-BuLi, pentane, Et_2O, −78 °C, then **3**, −78 °C to 0 °C, 96%; (r) TPAP, NMO, CH_2Cl_2, rt, 97%; (s) PPTS, MeOH, rt, 99%; (t) (+)-DET, Ti(O*i*-Pr)$_4$, 4 Å molecular sieves, *t*-BuOOH, CH_2Cl_2, −20 °C, 86%; (u) PPh_3, I_2, imidazole, THF:MeCN=4:1, rt, 80%; (v) NaI, Zn, MeOH, 90 °C, quant.; (w) acryloyl chloride, DMAP, Et_3N, CH_2Cl_2, rt, 90%; (x) DDQ, CH_2Cl_2:H_2O=2:1, rt, quant.; (y) Grubbs second-generation catalyst, CH_2Cl_2, 40 °C, quant.

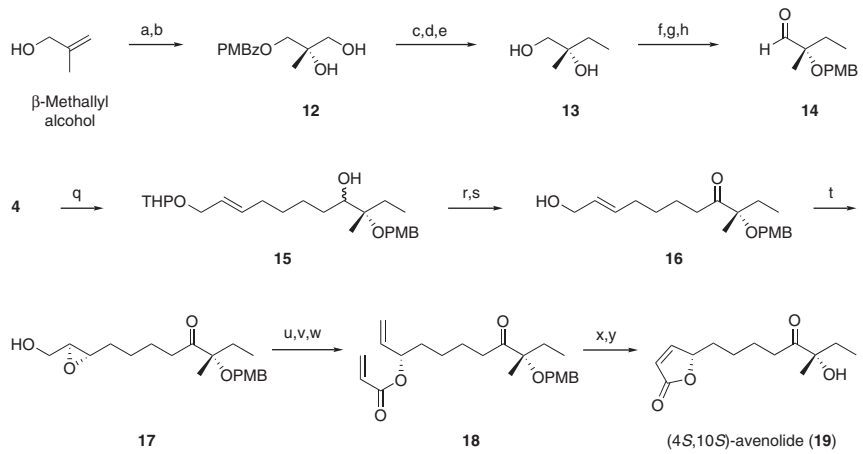
afforded *E*-allyl alcohol **7**.[15,16] This was then converted to **8** by a two-step sequence of protecting group manipulations. Mesylation of the primary alcohol of **8** followed by treatment with sodium iodide gave rise to **4**.

The aldehyde **3** was coupled with an alkyl lithium species derived from halogen–lithium exchange of iodide **4** with *t*-BuLi to furnish alcohol **9** as a diastereomeric mixture. TPAP oxidation and THP deprotection under acidic conditions afforded ketoalcohol **10**. Subsequent Sharpless asymmetric epoxidation[17] using (+)-DET as a chiral ligand gave the desired epoxyalcohol **2**. Transformation of the primary alcohol to iodide followed by treatment with zinc yielded the corresponding allyl alcohol, which was acylated with acryloyl chloride to provide **11**. Finally, deprotection of the PMB group by treatment with DDQ and ring closing metathesis[18–22] using Grubbs second-generation catalyst afforded **1**.

To determine the absolute configuration of **1**, we also synthesized (4*S*,10*S*)-avenolide (**19**), according to Scheme 2, with $(DHQD)_2PHAL$ used instead of $(DHQ)_2PHAL$ as the chiral ligand in Sharpless asymmetric dihydroxylation (Scheme 3).

We next analyzed the synthetic **1** and **19** with a chiral HPLC column (conditions: DAICEL CHIRALPAK IA-3 (0.46 cm ϕ×25 cm), EtOH mobile phase (flow rate of 0.3 ml per min) and detection at 200 nm (0 °C)). When a 1:1 mixture of **1** and **19** was injected onto the chiral HPLC system, the two diastereomers were completely resolved, as shown in Figure 1a. Figures 1b and c show the HPLC chromatogram of **1** and **19**, respectively. An authentic sample of the natural avenolide was also analyzed by HPLC, and its retention time was identical to that of **1** as shown in Figure 1d. This result establishes the absolute configuration of avenolide as 4*S*,10*R*.

We also synthesized (4*R*,10*R*)-avenolide (**22**) and 10-deoxy avenolide (**28**) for structure–activity relationship studies as shown in Scheme 4. (The spectrum data of compounds in Schemes 3 and 4 are provided as Supplementary information.) The synthesis of **22** was achieved



Scheme 3 Reagents and conditions: the same reaction conditions as those described in Scheme 2 were used, unless otherwise noted. (b) $K_3Fe(CN)_6$, K_2CO_3, $(DHQD)_2PHAL$, $K_2OsO_4(OH)_4$, *t*-BuOH:H_2O=1:1, 0 °C, quant., 93% ee[11–13]; (q) *t*-BuLi, pentane, Et_2O, −78 °C, then **14**, −78 °C to 0 °C, 88%.

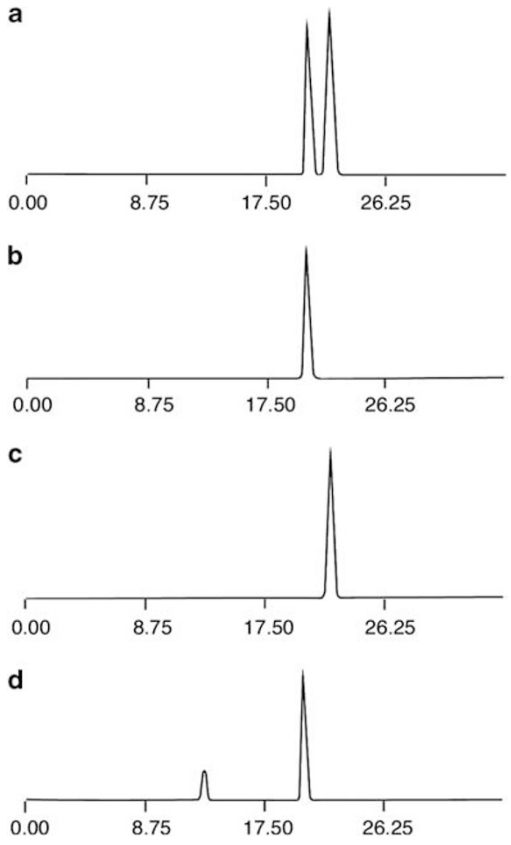


Figure 1 HPLC chromatograms of natural and synthetic samples of avenolide (**1** and **19**). The abscissa axis indicates retention time (min). (**a**) 1:1 mixture of **1** and **19**; (**b**) **1**; (**c**) **19**; (**d**) natural avenolide.

according to our synthetic procedure for **1** as shown in Scheme 2, but using (−)-DET as the chiral ligand during Sharpless epoxidation. In contrast, the synthesis of **28** required a slightly altered synthetic route. A commercially available (*S*)-2-methyl-1-butanol was subjected to TEMPO oxidation and dithioacetalization to afford **23**,[23] which was coupled with iodide **4** to give **24**. THP deprotection followed by treatment with [bis(trifluoroacetoxy)iodo]benzene furnished ketone **25**. The conversion of **25** into 10-deoxy avenolide (**28**) was carried out following the same synthetic procedure as that developed for the synthesis of **1** shown in Scheme 2.

Synthetic **1** demonstrated identical activity to that of natural **1**. In contrast, the rAvaR1-ligand activities of the C10-epimer (**19**), the C4-epimer (**22**) and the 10-deoxy avenolide (**28**) demonstrated only one-half, one twenty-fifth and one hundredth of the activity of the natural product, respectively.[6] It can therefore be concluded that the stereochemistry at C4 and C10, and the presence of the hydroxyl group at C10, are important to the activity of avenolide as a rAvaR1 ligand.

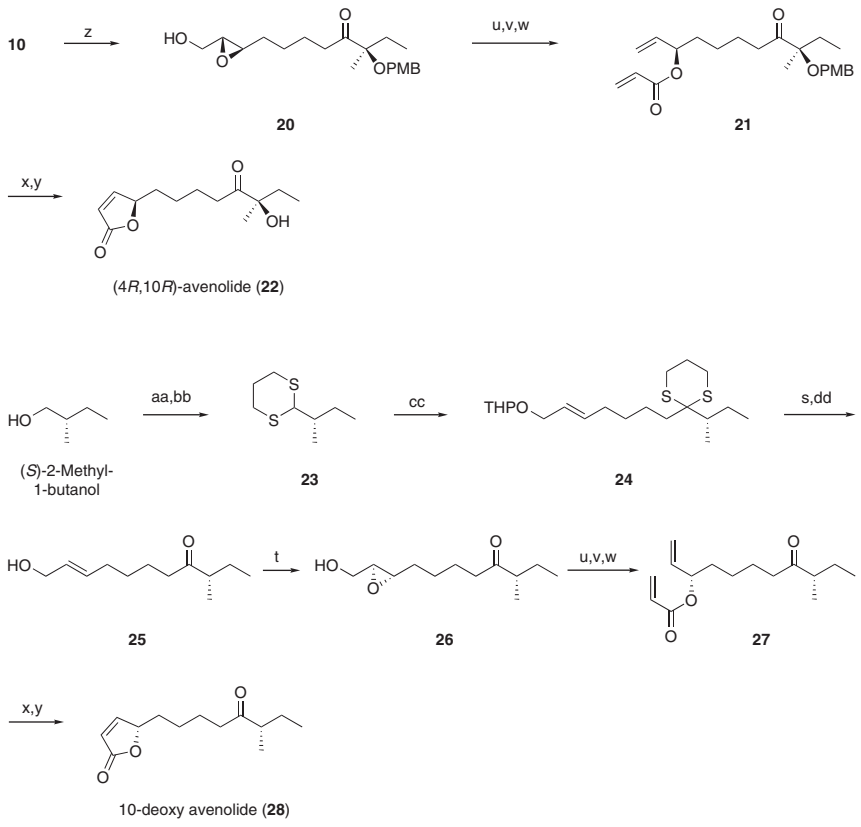
In conclusion, we have achieved the total synthesis of avenolide and determined its absolute configuration. By extending the synthetic methodology, we have generated a number of structural analogs, which provide information on the structure–activity relationship of avenolide. Further biological studies on the synthetic avenolide are currently underway and will be reported in due course.

EXPERIMENTAL PROCEDURE

General

IR spectra were obtained using a Horiba FT-710 spectrophotometer (Horiba, Kyoto, Japan). ^1H and ^{13}C-NMR spectra were obtained on Mercury-300 and UNITY-400 spectrometers (Agilent Technologies, Santa Clara, CA, USA), and chemical shifts are reported on the δ scale, using TMS as an internal reference. MS spectra were measured on JEOL JMS-700, JEOL JMS-T-100LP and JEOL JMS-AX505HA spectrometers. Optical rotations were recorded on a JASCO DIP-1000 polarimeter (Jasco, Hachioji, Japan). Commercial reagents were used without further purification unless otherwise indicated. Organic solvents were distilled and dried over molecular sieves (3 or 4 Å). Reactions were performed in flame-dried glassware under positive Ar pressure while stirring with a magnetic stirrer bar unless otherwise indicated. Flash chromatography was performed on silica gel 60N (spherical, neutral, particle size 40–50 mm). TLC was performed on 0.25 mm Merck silica gel 60 F254 plates (Merck, Darmstadt, Germany) and visualized by UV (254 nm), and using phosphomolybdic acid and *p*-anisaldehyde as TLC stains.

(R)-2,3-Dihydroxy-2-methylpropyl-4-methoxybenzoate (**5**). To a solution of β-methallyl alcohol (3.50 ml, 41.3 mmol) in CH_2Cl_2 (83 ml) were added DMAP (504 mg, 4.13 mmol), *N*-(3-dimethylaminopropyl)-*N*′-ethylcarbodiimide hydrochloride (WSC, 8.68 g, 45.4 mmol) and *p*-anisic acid (6.91 g, 45.5 mmol) at rt under N_2. The reaction mixture was stirred for 2.5 h at rt. The reaction was quenched with H_2O and the aqueous phase was extracted with CH_2Cl_2. The combined organic extracts were dried over anhydrous Na_2SO_4 and concentrated *in vacuo*. The residue was purified by flash column chromatography (30:1 hexanes/EtOAc) to furnish the corresponding *p*-methoxybenzoate (8.38 g, 99%) as a colorless oil.



Scheme 4 Reagents and conditions: the same reaction conditions as those described in Scheme 2 were used unless otherwise noted. (z) (−)-DET, $Ti(O\textit{i}\text{-}Pr)_4$, 4 Å molecular sieves, *t*-BuOOH, CH_2Cl_2, −20 °C, 70%; (aa) $PhI(OAc)_2$, TEMPO, CH_2Cl_2, rt; (bb) 1,3-pentanedithiol, $BF_3{\cdot}Et_2O$, −5 °C, 70% (two steps); (cc) *t*-BuLi, THF:HMPA=10:1, −78 °C, then **4**, 82%; (dd) [bis(trifluoroacetoxy)iodo]benzene, CH_3CN:H_2O=9:1, quant.

IR (KBr) 3082, 2939, 2842, 1715, 1607, 1511, 1457 cm^{-1}; ^{1}H-NMR (400 MHz, $CDCl_3$) δ 8.03 (d, *J*=9.0 Hz, 2H), 6.92 (d, *J*=9.0 Hz, 2H), 5.06 (brs, 1H), 4.97 (brs, 1H), 4.72 (brs, 2H), 3.85 (s, 3H), 1.83 (brs, 3H); ^{13}C-NMR (100 MHz, $CDCl_3$) δ 166.0, 163.4, 140.2, 131.7, 122.6, 113.7, 112.7, 67.8, 55.4, 19.6; HRMS (ESI^+, TFA-Na) calcd for $C_{12}H_{14}NaO_3$ 229.0841 $[M+Na]^+$, found *m/z* 229.0841.

To a solution of the *p*-methoxybenzoate (4.00 g, 19.4 mmol) in *t*-BuOH:H_2O (1:1, 194 ml) were added $K_3Fe(CN)_6$ (19.2 g, 58.2 mmol), K_2CO_3 (8.05 g, 58.2 mmol), $K_2OsO_4(OH)_4$ (71.5 mg, 0.19 mmol) and $(DHQ)_2PHAL$ (151 mg, 0.19 mmol) at 0 °C under N_2. The reaction mixture was stirred for 4.5 h at 0 °C. The reaction was quenched with a saturated aqueous $Na_2S_2O_3$ and the aqueous phase was extracted with CH_2Cl_2. The combined organic extracts were dried over anhydrous Na_2SO_4 and concentrated *in vacuo*. The residue was purified by flash column chromatography (5:1 hexanes/EtOAc) to afford **5** (4.66 g, quant.) as a colorless oil. The enantiomeric excess (93% ee) of **5** was determined by chiral HPLC analysis (conditions: DAICEL CHIRALPAK AS-3 (0.46 cm ϕ×25 cm), mobile phase of 3:1 hexanes/2-propanol (flow rate of 1.0 ml per min) and detection at 254 nm (rt.)).

$[\alpha]_D^{26}$ −2.51 (*c* 1.0 MeOH); IR (KBr) 3478, 2976, 2842, 1706, 1606, 1512, 1462 cm^{-1}; ^{1}H-NMR (300 MHz, $CDCl_3$) δ 7.98 (d, *J*=9.1 Hz, 2H), 6.90 (d, *J*=9.1 Hz, 2H), 4.36 (d, *J*=11.3 Hz, 1H), 4.19 (d, *J*=11.3 Hz, 1H), 3.85 (s, 3H), 3.57 (d, *J*=11.6 Hz, 1H), 3.46 (d, *J*=11.4 Hz, 1H), 2.95 (brs, 1H), 1.26 (s, 3H); ^{13}C-NMR (75.0 MHz, $CDCl_3$) δ 166.9, 163.7, 131.8, 121.8, 113.8, 72.2, 68.0, 66.9, 55.5, 21.3; HRMS (ESI^+, TFA-Na) calcd for $C_{12}H_{16}NaO_5$ 263.0895 $[M+Na]^+$, found *m/z* 263.0902.

(R)-2-Methylbutane-1,2-diol (**6**). To a solution of **5** (1.58 g, 6.59 mmol) in CH_2Cl_2 (66 ml) were added Et_3N (1.80 ml, 13.2 mmol), $Me_3N{\cdot}HCl$ (63.0 mg, 0.66 mmol) and TsCl (1.90 g, 9.89 mmol) at rt under N_2. The reaction mixture was stirred for 3 h at rt. The reaction was quenched with H_2O and the aqueous phase was extracted with CH_2Cl_2. The combined organic extracts were dried over anhydrous Na_2SO_4 and concentrated *in vacuo*. The residue was purified by flash column chromatography (1:1 hexanes/EtOAc) to afford the corresponding tosylate (2.57 g, 99%) as a colorless oil.

$[\alpha]_D^{26}$ +1.94 (*c* 1.0 $CHCl_3$); IR (KBr) 3519, 2982, 2842, 1712, 1604, 1511, 1462 cm^{-1}; ^{1}H-NMR (400 MHz, $CDCl_3$) δ 7.87 (d, *J*=9.0 Hz, 2H), 7.76 (d, *J*=9.0 Hz, 2H), 7.26 (d, *J*=7.8 Hz, 2H), 6.90 (d, *J*=8.8 Hz, 2H), 4.36 (d, *J*=11.3 Hz, 2H), 4.00 (d, *J*=8.0 Hz, 2H), 3.88 (s, 3H), 2.35 (s, 3H), 1.28 (s, 3H); ^{13}C-NMR (100 MHz, $CDCl_3$) δ 166.1, 163.7, 145.2, 132.3, 131.8, 130.0, 128.0, 121.7, 113.7, 72.8, 70.8, 67.7, 55.5, 21.6, 21.6; HRMS (ESI^+, TFA-Na) calcd for $C_{19}H_{22}NaO_7S$ 417.0984 $[M+Na]^+$, found *m/z* 417.0971.

To a solution of CuCN (1.79 g, 19.9 mmol) in THF (20 ml) was added MeLi (1.07 M in diethyl ether, 37.3 ml, 39.9 mmol) at −78 °C under Ar. After stirring for 20 min at 0 °C, a solution of the tosylate (1.57 g, 3.99 mmol) in THF (20 ml) was added to the reaction mixture at −78 °C. The reaction was stirred for 30 min at −78 °C, then allowed to warm to 0 °C and stirred for 20 min. The reaction was quenched with a saturated aqueous NH_4Cl and the aqueous phase was extracted with EtOAc. The combined organic extracts were dried over anhydrous Na_2SO_4 and concentrated *in vacuo*. The resulting residue was dissolved in MeOH (40 ml) and K_2CO_3 (276 mg, 1.99 mmol) was added. After stirring for 14 h at rt, the reaction mixture was concentrated *in vacuo*. The residue was purified by flash column chromatography (5:1 hexanes/EtOAc) to give **6** (373 mg, 90% over 2 steps) as a colorless oil.

$[\alpha]_D^{25}$ +4.90 (*c* 1.0 $CHCl_3$); IR (KBr) 3424, 2926, 2856, 1654, 1462 cm^{-1}; ^{1}H-NMR (300 MHz, $CDCl_3$) δ 3.34 (d, *J*=11.1 Hz, 1H), 3.37 (d, *J*=11.1 Hz, 1H), 3.10 (brs, 1H), 2.71 (brs, 1H), 1.50 (q, *J*=7.3 Hz, 2H), 1.11 (s, 3H), 0.89 (t, *J*=7.6 Hz, 3H); ^{13}C-NMR (75.0 MHz, $CDCl_3$) δ 73.3, 69.3, 31.0, 22.4, 8.03.

(R)-2-((4-Methoxybenzyl)oxy)-2-methylbutanal (**3**). To a solution of **6** (776 mg, 7.46 mmol) in CH_2Cl_2 (30 ml) were added PPTS (93.7 mg, 0.37 mmol) and *p*-anisaldehyde dimethylacetal (1.27 ml, 7.46 mmol) at rt under N_2. The reaction mixture was stirred for 6 h at rt. The reaction was quenched with H_2O and the aqueous phase was extracted with EtOAc. The combined organic extracts were

dried over anhydrous Na_2SO_4 and concentrated *in vacuo*. The residue was purified by flash column chromatography (50:1 hexanes/EtOAc) to afford the corresponding *p*-methoxybenzylidene acetal (1.66 g, quant.) as a colorless oil.

$[\alpha]_D^{26}$ +1.40 (*c* 1.0 $CHCl_3$); IR (KBr) 2969, 2929, 2874, 1614, 1515, 1461 cm^{-1}; ^{1}H-NMR (300 MHz, $CDCl_3$) δ 7.42 (d, *J*=8.8 Hz, 2H), 6.90 (d, *J*=8.8 Hz, 2H), 5.82 (s, 1H), 3.83 (s, 2H), 3.81 (s, 3H), 1.77–1.68 (m, 2H), 1.38 (s, 3H), 1.00 (t, *J*=7.5 Hz, 3H); ^{13}C-NMR (75.0 MHz, $CDCl_3$) δ 160.4, 130.5, 128.1, 113.7, 103.6, 81.7, 75.4, 55.3, 31.2, 24.3, 8.54; HRMS (ESI$^+$, TFA-Na) calcd for $C_{13}H_{19}O_3$: 223.1334 $[M+H]^+$, found *m/z* 223.1342.

To a solution of the *p*-methoxybenzylidene acetal (697 mg, 3.14 mmol) in CH_2Cl_2 (31 ml) was added DIBAL (1.02 M in hexanes, 9.00 ml, 9.41 mmol) at 0 °C under N_2. After stirring for 1 h at 0 °C, the reaction was cautiously quenched with MeOH, diluted with CH_2Cl_2 and treated with celite (2.80 g) and $Na_2SO_4{\cdot}10H_2O$ (2.80 g). The mixture was allowed to warm to rt and stirred for 2 h. It was then filtered through a pad of celite and the filtrate was concentrated. The residue was purified by flash column chromatography (10:1 hexanes/EtOAc) to give the corresponding alcohol (615 mg, 87%) as a colorless oil.

$[\alpha]_D^{26}$ +1.40 (*c* 1.0 MeOH); IR (KBr) 3444, 2970, 2936, 2879, 2839, 1613, 1514 cm^{-1}; ^{1}H-NMR (300 MHz, $CDCl_3$) δ 7.27 (d, *J*=8.7 Hz, 2H), 6.89 (d, *J*=8.7 Hz, 2H), 4.37 (s, 2H), 3.81 (s, 3H), 3.59 (d, *J*=4.8 Hz, 1H), 3.55 (d, *J*=5.0 Hz, 1H), 2.06 (brs, 1H), 1.66 (dq, *J*=7.6, 2.3 Hz, 1H), 1.23 (s, 3H), 0.94 (t, *J*=7.5 Hz, 3H); ^{13}C-NMR (75.0 MHz, $CDCl_3$) δ 159.0, 131.2, 129.0, 129.0, 113.9, 113.9, 77.8, 66.9, 63.3, 55.3, 27.8, 19.7, 8.09; HRMS (FAB, *m*-NBA) calcd for $C_{13}H_{19}O_3$ 223.1334 $[M+H]^+$, found *m/z* 223.1338.

To a solution of the alcohol (239 mg, 0.94 mmol) in CH_2Cl_2 (9.4 ml) were added DMSO (666 µl, 9.37 mmol), Et_3N (1.31 ml, 9.37 mmol) and $SO_3{\cdot}$pyr (895 mg, 5.62 mmol) at rt under N_2. The reaction mixture was stirred for 3.5 h at rt. The reaction was quenched with H_2O and the aqueous phase was extracted with CH_2Cl_2. The combined organic extracts were dried over anhydrous Na_2SO_4 and concentrated *in vacuo*. The residue was purified by flash column chromatography (50:1 hexanes/EtOAc) to furnish **3** (188 mg, 89%) as a colorless oil.

$[\alpha]_D^{22}$ +6.30 (*c* 1.0 $CHCl_3$); IR (KBr) 2974, 2937, 2838, 1734, 1613, 1514 cm^{-1}; ^{1}H-NMR (300 MHz, $CDCl_3$) δ 9.62 (s, 1H), 7.27 (d, *J*=8.8 Hz, 2H), 6.87 (d, *J*=8.8 Hz, 2H), 4.39 (d, *J*=17.6 Hz, 1H), 4.35 (d, *J*=17.6 Hz, 1H), 3.78 (s, 3H), 1.81–1.63 (m, 2H) 1.29 (s, 3H), 0.91 (t, *J*=7.6 Hz, 3H); ^{13}C-NMR (75.0 MHz, $CDCl_3$) δ 205.3, 159.3, 130.4, 129.1, 113.9, 82.8, 65.9, 55.3, 27.7, 17.7, 7.30; HRMS (FAB, *m*-NBA) calcd for $C_{13}H_{18}NaO_3$ 245.1154 $[M+Na]^+$, found *m/z* 245.1162.

(E)-7-((tert-Butyldimethylsilyl)oxy)hept-2-en-1-ol (**7**). To a solution of NaH (1.94 g, 47.5 mmol) in THF (158 ml) were added 1,5-pentanediol (5.00 ml, 47.5 mmol) and TBSCl (7.31 g, 47.5 mmol) at 0 °C under N_2. The reaction mixture was stirred for 5 min at 0 °C, then allowed to warm to rt and stirred for 2.5 h. The reaction was quenched with H_2O at 0 °C and the aqueous phase was extracted with CH_2Cl_2. The combined organic extracts were dried over anhydrous Na_2SO_4 and concentrated *in vacuo*. The residue was purified by flash column chromatography (5:1 hexanes/EtOAc) to afford the corresponding TBS ether (8.30 g, 80%) as a colorless oil.

IR (KBr) 3359, 2933, 2860, 1468, 1254, 1100 cm^{-1}; ^{1}H-NMR (400 MHz, $CDCl_3$) δ 3.64–3.59 (m, 4H), 1.58–1.52 (m, 4H), 1.40–1.39 (m, 2H), 0.88 (s, 9H), 0.04 (s, 6H); ^{13}C-NMR (100 MHz, $CDCl_3$) δ 63.2, 62.9, 32.5, 32.5, 26.0, 22.0, 18.4, –2.28; HRMS (FAB, *m*-NBA) calcd for $C_{11}H_{27}O_2Si$ 219.1780 $[M+H]^+$, found *m/z* 219.1776.

To a solution of the TBS ether (4.00 g, 18.3 mmol) in CH_2Cl_2 (183 ml) were added iodobenzene diacetate (8.86 g, 27.5 mmol) and TEMPO (573 mg, 3.67 mmol) at rt under N_2. The reaction mixture was stirred for 5 h at rt. The reaction was quenched with an aqueous $Na_2S_2O_3$ solution, and the aqueous phase was extracted with CH_2Cl_2. The combined organic extracts were dried over anhydrous Na_2SO_4 and concentrated *in vacuo*. The residue was purified by flash column chromatography (50:1 hexanes/EtOAc) to afford the corresponding aldehyde (3.96 g, quant.) as a colorless oil.

IR (KBr) 2952, 2933, 2859, 1727, 1468, 1254, 1102 cm^{-1}; ^{1}H-NMR (400 MHz, $CDCl_3$) δ 9.75 (s, 1H), 3.61 (t, *J*=6.2 Hz, 2H), 2.46–2.42 (m, 2H), 1.70–1.65 (m, 2H), 1.57–1.51 (m, 2H), 0.87 (s, 9H), 0.03 (s, 6H); ^{13}C-NMR (100 MHz, $CDCl_3$) δ 203.7, 62.6, 43.6, 32.1, 25.9, 18.6, 18.3, –5.34; HRMS (FAB, *m*-NBA) calcd for $C_{11}H_{25}O_2Si$ 217.1624 $[M+H]^+$, found *m/z* 217.1618.

To a solution of the aldehyde (3.19 g, 14.8 mmol) in benzene (148 ml) was added ethyl(triphenylphosphoranylidene)acetate (7.71 g, 22.1 mmol) under N_2. The reaction mixture was stirred for 3 h at 80 °C. The reaction was concentrated *in vacuo*. The residue was purified by flash column chromatography (80:1 hexanes/EtOAc) to give the corresponding α,β-unsaturated ester (4.19 g, quant.) as a colorless oil.

IR (KBr) 2933, 2859, 1724, 1655, 1468, 1259, 1101 cm^{-1}; ^{1}H-NMR (400 MHz, $CDCl_3$) δ 6.94 (dt, *J*=15.7, 7.0 Hz, 1H), 5.79 (dt, *J*=15.7, 1.6 Hz, 1H), 4.16 (q, *J*=7.2 Hz, 2H), 3.59 (t, *J*=6.1 Hz, 2H), 2.21–2.19 (m, 2H), 1.52–1.49 (m, 4H), 1.26 (t, *J*=7.1 Hz, 3H), 0.87 (s, 9H), 0.02 (s, 6H); ^{13}C-NMR (100 MHz, $CDCl_3$) δ 166.7, 149.1, 121.4, 62.7, 60.1, 32.2, 31.9, 25.9, 24.4, 18.3, 14.3, –5.32; HRMS (ESI$^+$, TFA-Na) calcd for $C_{15}H_{30}NaO_3Si$ 309.1862 $[M+Na]^+$, found *m/z* 309.1877.

To a solution of the α,β-unsaturated ester (1.82 g, 8.02 mmol) in CH_2Cl_2 (80 ml) was added DIBAL (1.02 M in hexanes, 15.7 ml, 16.0 mmol) at 0 °C under N_2. After stirring for 1 h, the reaction was cautiously quenched with MeOH, diluted with CH_2Cl_2 and treated with celite (5.90 g) and $Na_2SO_4{\cdot}10H_2O$ (5.90 g). The mixture was allowed to warm to rt and stirred for 2 h, then filtered through a pad of celite and the filtrate was concentrated. The residue was purified by flash column chromatography (20:1 hexanes/EtOAc) to furnish **7** (1.96 g, quant.) as a colorless oil.

IR (KBr) 3339, 2932, 2859, 1467, 1254, 1101 cm^{-1}; ^{1}H-NMR (300 MHz, $CDCl_3$) δ 5.68–5.63 (m, 2H), 4.07 (d, *J*=4.7 Hz, 2H), 3.60 (t, *J*=6.3 Hz, 2H), 2.09–2.02 (m, 2H), 1.55–1.39 (m, 4H), 0.88 (s, 9H), 0.04 (s, 6H); ^{13}C-NMR (75.0 MHz, $CDCl_3$) δ 133.2, 129.1, 63.8, 63.0, 32.3, 31.9, 26.0, 26.0, 25.4, 18.4, −5.29; HRMS (FAB, *m*-NBA) calcd for $C_{13}H_{29}O_2Si$ 245.1937 $[M+H]^+$, found *m/z* 245.1934.

(E)-7-((Tetrahydro-2H-pyran-2-yl)oxy)hept-5-en-1-ol (**8**). To a solution of **7** (1.96 g, 8.02 mmol) in CH_2Cl_2 (40 ml) were added PPTS (202 mg, 0.80 mmol) and DHP (7.25 ml, 80.2 mmol) at 0 °C under N_2. The reaction mixture was stirred for 6 h at 0 °C. The reaction was quenched with H_2O and the aqueous phase was extracted with EtOAc. The combined organic extracts were dried over anhydrous Na_2SO_4 and concentrated *in vacuo*. The residue was purified by flash column chromatography (50:1 hexanes/EtOAc) to afford the corresponding THP ether (2.63 g, quant.) as a colorless oil.

IR (KBr) 2935, 2859, 1467, 1254, 1102 cm^{-1}; ^{1}H-NMR (400 MHz, $CDCl_3$) δ 5.72–5.67 (m, 1H), 5.60–5.54 (m, 1H), 4.64–4.62 (m, 1H), 4.17 (ddd, *J*=11.9, 5.7, 1.2 Hz, 1H), 3.92 (ddd, *J*=11.9, 5.7, 1.2 Hz, 1H), 3.90–3.84 (m, 1H), 3.59 (t, *J*=6.5 Hz, 2H), 3.51–3.48 (m, 1H), 2.08–2.03 (m, 2H), 1.82–1.56 (m, 6H) 1.55–1.39 (m, 4H), 0.88 (s, 9H), 0.03 (s, 6H); ^{13}C-NMR (100 MHz, $CDCl_3$) δ 134.5, 126.3, 97.8, 67.9, 63.1, 62.3, 32.4, 32.1, 30.7, 26.0, 25.5, 25.3, 19.6, 18.4, −5.26; HRMS (ESI$^+$, TFA-Na) calcd for $C_{18}H_{36}NaO_3Si$ 351.2325 $[M+Na]^+$, found *m/z* 351.2331.

To a solution of the THP ether (2.11 g, 6.42 mmol) in THF (64 ml) was added TBAF (7.71 ml, 7.71 mmol) at rt under N_2. The reaction mixture was stirred for 4.5 h at rt. The reaction was quenched with H_2O, and the aqueous phase was extracted with CH_2Cl_2. The combined organic extracts were dried over anhydrous Na_2SO_4 and concentrated *in vacuo*. The residue was purified by flash column chromatography (1:1 hexanes/EtOAc) to give **8** (1.34 g, quant.) as a colorless oil.

IR (KBr) 3393, 2939, 2866, 1499, 1351, 1120 cm^{-1}; ^{1}H-NMR (300 MHz, $CDCl_3$) δ 5.72–5.65 (m, 1H), 5.61–5.54 (m, 1H), 4.63–4.60 (m, 1H), 4.16 (ddd, *J*=11.9, 5.5, 1.1 Hz, 1H), 3.90 (ddd, *J*=11.9, 5.5, 1.1 Hz, 1H), 3.89–3.82 (m, 1H), 3.61 (t, *J*=6.5 Hz, 2H), 3.50–3.46 (m, 1H), 2.10–2.03 (m, 2H), 1.83–1.41 (m, 10H); ^{13}C-NMR (75.0 MHz, $CDCl_3$) δ 133.0, 127.0, 97.8, 67.8, 62.7, 62.2, 32.2, 32.0, 30.6, 25.4, 25.2, 19.5; HRMS (FAB, *m*-NBA) calcd for $C_{12}H_{22}NaO_3$ 273.1467 $[M+Na]^+$, found *m/z* 273.1467.

(E)-2-((7-Iodohept-2-en-1-yl)oxy)tetrahydro-2H-pyran (**4**). To a solution of **8** (657 mg, 3.07 mmol) in CH_2Cl_2 (31 ml) were added Et_3N (855 µl, 6.14 mmol), $Me_3N{\cdot}$HCl (29.3 mg, 0.31 mmol) and MsCl (356 µl, 4.61 mmol) at 0 °C under N_2. The reaction mixture was allowed to warm to rt and stirred for 3 h. The reaction was quenched with H_2O and the aqueous phase was extracted with CH_2Cl_2. The combined organic extracts were dried over anhydrous Na_2SO_4 and concentrated *in vacuo*. The residue was purified by flash column chromatography (10:1 hexanes/EtOAc) to afford the corresponding mesylate (837 mg, 93%) as a colorless oil.

IR (KBr) 2940, 2866, 1354, 1174, 1121 cm^{-1}, ^{1}H-NMR (400 MHz, $CDCl_3$) δ 5.69–5.64 (m, 1H), 5.62–5.55 (m, 1H), 4.61–4.60 (m, 1H), 4.21 (t, *J*=6.5 Hz, 2H), 4.17 (ddd, *J*=12.0, 5.3, 1.2 Hz, 1H), 3.91 (ddd, *J*=12.0, 5.3, 1.2 Hz, 1H),

3.88–3.47 (m, 1H), 3.51–3.47 (m, 1H), 2.99 (s, 3H), 2.12–2.06 (m, 2H), 1.84–1.46 (m, 10H); ^{13}C-NMR (100 MHz, $CDCl_3$) δ 133.0, 127.0, 97.8, 69.8, 67.6, 62.2, 37.2, 31.5, 30.5, 28.5, 25.3, 24.7, 19.4; HRMS (ESI$^+$, TFA-Na) calcd for $C_{13}H_{24}NaO_5S$ 315.1242 $[M+Na]^+$, found *m/z* 315.1206.

To a solution of NaI (2.42 g, 16.2 mmol) in acetone (216 ml) was added the mesylate (3.15 g, 10.8 mmol) under N_2. The reaction mixture was stirred for 8.5 h at reflux. After cooling to rt, the reaction was quenched with H_2O and the aqueous phase was extracted with EtOAc. The combined organic extracts were dried over anhydrous Na_2SO_4 and concentrated *in vacuo*. The residue was purified by flash column chromatography (80:1 hexanes/EtOAc) to give **4** (3.49 g, quant.) as a colorless oil.

IR (KBr) 2941, 2869, 1451, 1350, 1121 cm^{-1}; ^{1}H-NMR (300 MHz, $CDCl_3$) δ 5.71–5.54 (m, 2H), 4.61 (dd, *J*=4.3, 2.9 Hz, 1H), 4.17 (ddd, *J*=12.0, 5.4, 1.0 Hz, 1H), 3.91 (ddd, *J*=12.0, 5.3, 1.2 Hz, 1H), 3.88–3.82 (m, 1H), 3.53–3.47 (m, 1H), 3.17 (t, *J*=7.0 Hz, 2H), 2.10–2.03 (m, 2H), 1.87–1.44 (m, 10H); ^{13}C-NMR (75.0 MHz, $CDCl_3$) δ 133.5, 126.8, 97.8, 67.7, 62.3, 33.0, 31.2, 30.6, 29.8, 25.5, 19.6, 6.80; HRMS (FAB, *m*-NBA): calcd for $C_{12}H_{21}NaO_2I$ 347.0484 $[M+Na]^+$, found *m/z* 347.0484.

(3R,E)-3-((4-Methoxybenzyl)oxy)-3-methyl-11-((tetrahydro-2H-pyran-2-yl)oxy)undec-9-en-4-ol (**9**). To a solution of iodide **4** (844 mg, 3.8 mmol) in pentane (22 ml) were added Et_2O (8.2 ml) and *t*-BuLi (1.55 M in pentane, 5.64 ml, 8.74 mmol) at −78 °C under Ar. After stirring for 15 min at −78 °C, a solution of aldehyde **3** (1.35 g, 4.18 mmol) in Et_2O (7.0 ml) was added. The reaction mixture was allowed to warm to 0 °C and stirred for 3 h. The mixture was quenched with an aqueous NH_4Cl solution and the aqueous phase was extracted with EtOAc. The combined organic extracts were dried over anhydrous Na_2SO_4 and concentrated *in vacuo*. The residue was purified by flash column chromatography (15:1 hexanes/EtOAc) to afford **9** (1.54 g, 96%) as a colorless oil.

$[\alpha]_D^{22}$ +3.88 (*c* 1.0 $CHCl_3$); IR (KBr) 3561, 2837, 1612, 1512, 1458 cm^{-1}; ^{1}H-NMR (300 MHz, $CDCl_3$) δ 7.26 (d, *J*=8.8 Hz, 2H), 6.88 (d, *J*=8.8 Hz, 2H), 5.75–5.68 (m, 1H), 5.62–5.55 (m, 1H), 4.64 (dd, *J*=3.5, 3.0 Hz, 1H), 4.34 (s, 2H), 4.19 (ddd, *J*=11.9, 5.6, 1.1 Hz, 1H), 3.92 (ddd, *J*=11.9, 6.7, 0.8 Hz, 1H), 3.88–3.82 (m, 1H), 3.80 (s, 3H), 3.68–3.64 (m, 1H), 3.54–3.47 (m, 1H), 2.17–2.05 (m, 2H), 1.86–1.15 (m, 14H), 1.14 (s, 3H), 0.92 (t, *J*=7.5 Hz, 3H); ^{13}C-NMR (75.0 MHz, $CDCl_3$) δ 159.0, 134.9, 131.0, 128.9, 126.1, 113.8, 97.7, 80.1, 73.9, 67.8, 62.8, 62.2, 55.2, 32.3, 30.8, 30.6, 29.6, 29.1, 26.5, 26.4, 25.4, 19.5, 17.6, 14.1, 7.34; HRMS (ESI$^+$, TFA-Na) calcd for $C_{25}H_{40}NaO_5$ 443.2773 $[M+Na]^+$, found *m/z* 443.2753.

(R,E)-11-Hydroxy-3-((4-methoxybenzyl)oxy)-3-methylundec-9-en-4-one (**10**). To a solution of **9** (674 mg, 1.60 mmol) in CH_2Cl_2 (16 ml) were added NMO (380 mg, 3.21 mmol), TPAP (28.2 mg, 0.08 mmol) and 4 Å molecular sieves (674 mg) at rt under N_2. The reaction mixture was stirred for 1.5 h at rt. The mixture was filtered through a pad of celite. The filtrate was diluted with H_2O and extracted with CH_2Cl_2 followed by EtOAc. The combined organic extracts were dried over anhydrous Na_2SO_4 and concentrated *in vacuo*. The residue was purified by flash column chromatography (40:1 hexanes/EtOAc) to afford the corresponding ketone (652 mg, 97%) as a colorless oil.

$[\alpha]_D^{27}$ +7.80 (*c* 1.0 $CHCl_3$); IR (KBr) 2940, 2868, 1712, 1613, 1514, 1459 cm^{-1}; ^{1}H-NMR (300 MHz, $CDCl_3$) δ 7.27 (d, *J*=8.8 Hz, 2H), 6.89 (d, *J*=8.8 Hz, 2H), 5.71–5.51 (m, 1H), 5.60–5.51(m, 1H), 4.63–4.25 (m, 1H), 4.32 (d, *J*=10.7 Hz, 1H), 4.28 (d, *J*=10.7 Hz, 1H), 4.17 (ddd, *J*=12.0, 5.5, 1.1 Hz, 1H), 3.91 (ddd, *J*=12.0, 5.5, 1.1 Hz, 1H), 3.88–3.83 (m, 1H) 3.80 (s, 3H), 3.80–3.46 (m, 1H), 2.64 (dt, *J*=7.3, 3.5 Hz, 2H), 2.09–2.01 (m, 2H), 1.87–1.35 (m, 10H), 1.42–1.35 (m, 2H), 1.32 (s, 3H), 0.84 (t, *J*=7.5 Hz, 3H); ^{13}C-NMR (75.0 MHz, $CDCl_3$) δ 214.9, 159.0, 134.2, 130.7, 128.6, 126.4, 113.8, 97.8, 84.9, 67.8, 65.1, 62.6, 55.3, 36.8, 32.2, 30.7, 29.3, 28.8, 25.5, 23.0, 19.9, 19.6, 7.89; HRMS (ESI$^+$, TFA-Na) calcd for $C_{25}H_{38}NaO_5$ 441.2617 $[M+Na]^+$, found *m/z* 441.2605.

To a solution of the ketone (597 mg, 1.43 mmol) in MeOH (14 ml) was added PPTS (359 mg, 1.43 mmol) at rt under N_2. The reaction mixture was stirred for 5 h at rt. The reaction was quenched with H_2O and the aqueous phase was extracted with CH_2Cl_2. The combined organic extracts were dried over anhydrous Na_2SO_4 and concentrated *in vacuo*. The residue was purified by flash column chromatography (10:1 hexanes/EtOAc) to afford **10** (475 mg, 99%) as a colorless oil.

$[\alpha]_D^{22}$ +4.35 (*c* 1.0 $CHCl_3$); IR (KBr) 3561, 3298, 2866, 1712, 1612, 1513, 1458 cm^{-1}; ^{1}H-NMR (300 MHz, $CDCl_3$) δ 7.27 (d, *J*=8.8 Hz, 2H), 6.89 (d, *J*=8.8 Hz, 2H), 5.69–5.58 (m, 2H), 4.32 (d, *J*=10.7 Hz, 1H), 4.29 (d, *J*=10.7 Hz, 1H), 4.07–4.05 (m, 2H), 3.80 (s, 3H), 2.64 (dt, *J*=7.3, 3.8 Hz, 2H), 2.08–2.01 (m, 2H), 1.87–1.68 (m, 2H), 1.62–1.52 (m, 2H), 1.41–1.34 (m, 2H), 1.32 (s, 3H), 0.84 (t, *J*=7.5 Hz, 3H); ^{13}C-NMR (75.0 MHz, $CDCl_3$) δ 215.0, 159.0, 132.8, 130.7, 129.3, 128.6, 113.8, 113.8, 65.1, 63.7, 55.3, 36.8, 32.1, 29.2, 28.8, 23.0, 19.9, 7.90; HRMS (ESI$^+$, TFA-Na) calcd for $C_{20}H_{30}NaO_4$ 357.2042 $[M+Na]^+$, found *m/z* 357.2031.

(R)-8-((2S,3S)-3-(Hydroxymethyl)oxiran-2-yl)-3-((4-methoxybenzyl)oxy)-3-methyloctan-4-one (**2**). To a suspension of 4 Å molecular sieves (134 mg) and (+)-DET (177 µl, 1.03 mmol) in CH_2Cl_2 (5.0 ml) was added $Ti(OiPr)_4$ (305 µl, 1.03 mmol) at −20 °C under Ar. After stirring for 0.5 h, *t*-BuOOH (413 µl, 2.06 mmol) was slowly added to the suspension at −20 °C and the resulting mixture was stirred for 0.5 h. A solution of **10** (335 mg, 1.03 mmol) in CH_2Cl_2 (5.3 ml) was then added dropwise to the reaction mixture and the mixture was stirred for 18 h at −20 °C. The reaction was quenched with Me_2S (105 µl, 1.43 mmol), diluted with CH_2Cl_2 and treated with celite (1.10 g) and $Na_2SO_4{\cdot}10H_2O$ (1.10 g). The suspension was allowed to warm to rt and then stirred for 2 h. The resulting mixture was filtered through a pad of celite and the filtrate was concentrated *in vacuo*. The residue was purified by flash column chromatography (5:1 hexanes/EtOAc) to afford **2** (311 mg, 86%) as a colorless oil.

$[\alpha]_D^{28}$ −2.89 (*c* 1.0 $CHCl_3$); IR (KBr) 3460, 2935, 1711, 1613, 1514, 1461 cm^{-1}; ^{1}H-NMR (400 MHz, $CDCl_3$) δ 7.27 (d, *J*=8.8 Hz, 2H), 6.89 (d, *J*=8.8 Hz, 2H), 4.33 (d, *J*=10.7 Hz, 1H), 4.29 (d, *J*=10.7 Hz, 1H), 3.90–3.82 (m, 1H), 3.80 (s, 3H), 3.64–3.62 (m, 1H), 2.96–2.88 (m, 2H), 2.66 (dt, *J*=7.2, 2.6 Hz, 2H), 1.88–1.75 (m, 2H), 1.73–1.54 (m, 4H), 1.48–1.39 (m, 2H), 1.33 (s, 3H), 0.84 (t, *J*=7.5 Hz, 3H); ^{13}C-NMR (75.0 MHz, $CDCl_3$) δ 214.9, 159.0, 130.7, 128.6, 113.8, 84.9, 65.1, 61.7, 58.4, 55.8, 55.3, 36.8, 31.4, 29.2, 25.7, 23.1, 20.0, 7.91; HRMS (ESI$^+$, TFA-Na) calcd for $C_{20}H_{30}NaO_5$ 373.1991 $[M+Na]^+$, found *m/z* 373.1975.

(3S,9R)-9-((4-Methoxybenzyl)oxy)-9-methyl-8-oxoundec-1-en-3-yl acrylate (**11**). To a solution of **2** (195 mg, 0.56 mmol) in THF:MeCN (4:1, 5.6 ml) were added imidazole (227 mg, 3.33 mmol), PPh_3 (437 mg, 1.67 mmol) and I_2 (423 mg, 1.67 mmol) at 0 °C under N_2. After warming to rt, the reaction mixture was stirred for 1 h. The reaction was quenched with H_2O and the aqueous phase was extracted with CH_2Cl_2. The combined organic extracts were dried over anhydrous Na_2SO_4 and concentrated *in vacuo*. The residue was purified by flash column chromatography (30:1 hexanes/EtOAc) to give the corresponding iodide (204 mg, 80%) as a colorless oil.

$[\alpha]_D^{25}$ +6.68 (*c* 1.0 $CHCl_3$); IR (KBr) 2936, 2864, 1712, 1613, 1514, 1460 cm^{-1}; ^{1}H-NMR (300 MHz, $CDCl_3$) δ 7.28 (d, *J*=8.8 Hz, 2H), 6.90 (d, *J*=8.8 Hz, 2H), 4.33 (d, *J*=10.7 Hz, 1H), 4.30 (d, *J*=10.7 Hz, 1H), 3.80 (s, 3H), 3.26–3.21 (m, 1H), 3.05–3.00 (m, 1H), 2.97 (ddd, *J*=4.7, 2.3 Hz, 1H), 2.79 (ddd, *J*=5.6, 5.5, 1.9 Hz, 1H), 2.66 (dt, *J*=7.0, 3.8 Hz, 2H), 1.88–1.66 (m, 2H), 1.64–1.52 (m, 4H), 1.48–1.40 (m, 2H), 1.33 (s, 3H), 0.84 (t, *J*=7.5 Hz, 3H); ^{13}C-NMR (75.0 MHz, $CDCl_3$) δ 214.7, 159.1, 130.7, 128.6, 113.9, 84.9, 65.1, 62.4, 58.3, 36.8, 31.6, 29.2, 25.6, 23.2, 20.0, 7.93, 5.02; HRMS (ESI$^+$, TFA-Na) calcd for $C_{20}H_{29}INaO_4$ 483.1008 $[M+Na]^+$, found *m/z* 483.1012.

To a solution of the iodide (163 mg, 0.35 mmol) in MeOH (707 µl) were added NaI (133 mg, 0.88 mmol) and Zn (69.4 mg, 1.06 mmol) under N_2. After stirring for 2.5 h at 90 °C, the reaction was quenched with H_2O and the aqueous phase was extracted with CH_2Cl_2. The combined organic extracts were dried over anhydrous Na_2SO_4 and concentrated *in vacuo*. The residue was purified by flash column chromatography (10:1 hexanes/EtOAc) to afford the corresponding allyl alcohol (118 mg, quant.) as a colorless oil.

$[\alpha]_D^{26}$ +8.93 (*c* 1.0 $CHCl_3$); IR (KBr) 3518, 3075, 2941, 1710, 1613, 1513 cm^{-1}; ^{1}H-NMR (300 MHz, $CDCl_3$) δ 7.28 (d, *J*=8.8 Hz, 2H), 6.89 (d, *J*=8.8 Hz, 2H), 5.90–5.79 (m, 1H), 5.20 (ddd, *J*=17.2, 1.4 Hz, 1H), 5.09 (ddd, *J*=10.4, 1.4 Hz, 1H), 4.32 (d, *J*=10.8 Hz, 1H), 4.29 (d, *J*=10.8 Hz, 1H), 4.12–4.05 (m, 1H) 3.80 (s, 3H), 2.65 (dt, *J*=7.3, 4.3 Hz, 2H), 1.87–1.68 (m, 2H), 1.61–1.34 (m, 6H), 1.32 (s, 3H), 0.84 (t, *J*=7.5 Hz, 3H); ^{13}C-NMR (75.0 MHz, $CDCl_3$) δ 215.0, 159.0, 141.1, 130.7, 128.6, 128.6, 114.5, 113.8, 113.8, 84.8, 72.9, 65.1, 55.2, 36.8, 36.8, 29.2, 25.0, 23.2, 20.0, 7.84; HRMS (ESI$^+$, TFA-Na) calcd for $C_{20}H_{30}NaO_4$ 357.2042 $[M+Na]^+$, found *m/z* 357.2025.

To a solution of the allyl alcohol (179 mg, 0.53 mmol) in CH_2Cl_2 (5.3 ml) were added acryloyl chloride (65 µl, 0.80 mmol), Et_3N (223 µl, 1.60 mmol) and DMAP (cat.) at 0 °C under N_2. After stirring for 1 h at rt, the reaction was

quenched with H_2O and the aqueous phase was extracted with CH_2Cl_2. The combined organic extracts were dried over anhydrous Na_2SO_4 and concentrated *in vacuo*. The residue was purified by flash column chromatography (30:1 hexanes/EtOAc) to afford **11** (187 mg, 90%) as a colorless oil.

$[\alpha]_D^{24}$ +6.32 (*c* 1.0 $CHCl_3$); IR (KBr) 2940, 1718, 1615, 1514, 1460 cm^{-1}; ^{1}H-NMR (300 MHz, $CDCl_3$) δ 7.27 (d, *J*=8.8 Hz, 2H), 6.89 (d, *J*=8.8 Hz, 2H), 6.40 (dd, *J*=17.3, 1.5 Hz, 1H), 6.12 (dd, *J*=17.3, 10.4 Hz, 1H), 5.84–5.73 (m, 2H), 5.31–5.27 (m, 1H), 5.24 (ddd, *J*=17.3, 9.3, 1.3 Hz, 1H), 5.16 (ddd, *J*=10.5, 5.9, 1.2 Hz, 1H), 4.32 (d, *J*=10.7 Hz, 1H), 4.29 (d, *J*=10.7 Hz, 1H), 3.81 (s, 3H), 2.64 (dt, *J*=7.3, 3.9 Hz, 2H), 1.87–1.53 (m, 8H), 1.32 (s, 3H), 0.84 (t, *J*=7.5 Hz, 3H); ^{13}C-NMR (75.0 MHz, $CDCl_3$) δ 214.8, 165.5, 159.0, 136.3, 130.7, 130.6, 128.7, 128.6, 116.8, 113.8, 113.8, 84.5, 74.8, 65.1, 55.3, 36.8, 34.1, 29.2, 24.8, 23.3, 20.0, 7.90; HRMS (ESI$^+$, TFA-Na) calcd for $C_{23}H_{32}NaO_5$ 411.2147 $[M+Na]^+$, found *m/z* 411.2136.

*(4S,10R)-avenolide (***1***).* To a solution of **11** (187 mg, 0.48 mmol) in CH_2Cl_2:H_2O (20:1, 4.6 ml) was added DDQ (146 mg, 0.53 mmol) at rt under N_2. After stirring for 1 h at rt, the reaction was quenched with H_2O and the aqueous phase was extracted with CH_2Cl_2. The combined organic extracts were dried over anhydrous Na_2SO_4 and concentrated *in vacuo*. The residue was purified by flash column chromatography (40:1 hexanes/EtOAc) to give the corresponding alcohol (159 mg, quant.) as a colorless oil.

$[\alpha]_D^{25}$ −8.51 (*c* 1.0 $CHCl_3$); IR (KBr) 3486, 2936, 2863, 1721, 1637, 1459 cm^{-1}; ^{1}H-NMR (300 MHz, $CDCl_3$) δ 6.41 (dd, *J*=17.3, 1.5 Hz, 1H), 6.12 (dd, *J*=17.3, 10.4 Hz, 1H), 5.85–5.73 (m, 2H), 5.34–5.16 (m, 2H), 3.82 (brs, 1H), 2.54–2.45 (m, 2H), 1.75–1.59 (m, 8H), 1.33 (s, 3H), 0.79 (t, *J*=7.4 Hz, 3H); ^{13}C-NMR (75.0 MHz, $CDCl_3$) δ 214.3, 165.5, 136.2, 130.8, 128.6, 116.9, 78.9, 74.6, 35.5, 34.0, 32.3, 25.1, 24.7, 23.3, 7.65; HRMS (ESI$^+$, TFA-Na) calcd for $C_{15}H_{24}NaO_4$ 291.1572 $[M+Na]^+$, found *m/z* 291.1576.

To a solution of the alcohol (38.1 mg, 0.14 mmol) in CH_2Cl_2 (5.0 ml) was added a solution of Grubbs second-generation catalyst (6.0 mg, 0.01 mmol) in CH_2Cl_2 (9.2 ml) at rt under N_2. The reaction mixture was stirred for 2 h at 40 °C. After the reaction was complete, Quadrasil AP (500 mg) was added to the reaction mixture. The suspension was stirred for 5 min at rt and then allowed to stand for 10 min. The mixture was filtered through a pad of celite and the filtrate was washed with H_2O. The organic layers were dried over anhydrous Na_2SO_4 and concentrated *in vacuo*. The residue was purified by flash column chromatography (5:1 hexanes/EtOAc) to afford **1** (43.2 mg, quant.) as a colorless oil.

$[\alpha]_D^{26}$ +2.27 (*c* 1.0 $CHCl_3$); IR (KBr) 3480, 2929, 2858, 1746, 1710, 1459 cm^{-1}; ^{1}H-NMR (300 MHz, $CDCl_3$) δ 7.44 (dd, *J*=5.8, 1.5 Hz, 1H), 6.11 (dd, *J*=5.7, 2.0 Hz, 1H), 5.05–5.02 (m, 1H), 3.76 (brs, 1H), 2.56–2.47 (m, 2H), 1.83–1.61 (m, 6H), 1.61–1.42 (m, 2H), 1.33 (s, 3H), 0.79 (t, *J*=7.4 Hz, 3H); ^{13}C-NMR (75.0 MHz, $CDCl_3$) δ 214.2, 173.0, 156.1, 121.7, 83.0, 78.9, 35.4, 33.0, 32.4, 25.2, 24.6, 23.2, 7.66; HRMS (FAB, *m*-NBA) calcd for $C_{13}H_{21}O_4$ 241.1440 $[M+H]^+$, found *m/z* 241.1447.

*(4S,10S)-avenolide (***19***).* $[\alpha]_D^{24}$+48.91 (*c* 1.0, $CHCl_3$); IR (KBr) 3483, 2940, 2874, 1748, 1711, 1459 cm^{-1}; ^{1}H-NMR (300 MHz, $CDCl_3$) δ 7.45 (dd, *J*=5.7, 1.5 Hz, 1H), 6.12 (dd, *J*=5.7, 2.1 Hz, 1H), 5.08–5.03 (m, 1H), 3.77 (brs, 1H), 2.60–2.45 (m, 2H), 1.87–1.41 (m, 8H), 1.34 (s, 3H), 0.81 (t, *J*=7.5 Hz, 3H); ^{13}C-NMR (75.0 MHz, $CDCl_3$) δ 214.1, 173.0, 156.0, 121.7, 83.0, 78.9, 35.4, 33.0, 32.4, 25.2, 24.6, 23.1, 7.66; HRMS (FAB, *m*-NBA) calcd for $C_{13}H_{21}O_4$ 241.1440 $[M+H]^+$, found *m/z* 241.1438.

*(4R,10R)-avenolide (***22***).* $[\alpha]_D^{26}$ −51.07 (*c* 1.0. $CHCl_3$); IR (KBr) 3482, 3093, 2930, 1748, 1458 cm^{-1}; ^{1}H-NMR (300 MHz, $CDCl_3$) δ 7.45 (dd, *J*=5.7, 1.5 Hz, 1H), 6.12 (dd, *J*=5.7, 2.1 Hz, 1H), 5.07–5.02 (m, 1H), 3.76 (brs, 1H), 2.59–2.45 (m, 2H), 1.86–1.34 (m, 8H), 1.34 (s, 3H), 0.80 (t, *J*=7.4 Hz, 3H); ^{13}C-NMR (75.0 MHz, $CDCl_3$) δ 214.4, 173.2, 156.2, 121.9, 83.2, 79.1, 35.6, 33.2, 32.6, 35.4, 24.9, 23.3, 7.86; HRMS (ESI$^+$, TFA-Na) calcd for $C_{13}H_{20}NaO_4$ 263.1259 $[M+Na]^+$, found *m/z* 263.1271.

*10-Deoxy avenolide (***28***).* $[\alpha]_D^{25}$ +47.75 (*c* 1.0. $CHCl_3$); IR (KBr) 3503, 3091, 2930, 1753, 1708 cm^{-1}; ^{1}H-NMR (300 MHz, $CDCl_3$) δ 7.45 (dd, *J*=5.7, 1.5 Hz, 1H), 6.11 (dd, *J*=5.7, 2.1 Hz, 1H), 5.07–5.02 (m, 1H), 2.47–2.40 (m, 3H), 1.81–1.28 (m, 8H), 1.05 (d, *J*=7.0 Hz, 3H), 0.87 (t, *J*=7.5 Hz, 3H); ^{13}C-NMR (75.0 MHz, $CDCl_3$) δ 214.4, 173.1, 156.2, 121.6, 83.1, 47.9, 40.6, 33.1, 25.9, 24.7, 23.1, 15.9, 11.7; HRMS (ESI$^+$, TFA-Na) calcd for $C_{13}H_{20}NaO_3$ 247.1310 $[M+Na]^+$, found *m/z* 247.1322.

Evaluation of biological activity

Biological activity of the compounds were measured either as the DNA dissociation activity from the rAvaR1–DNA complex in gel-shift experiment or by avermectin-inducing activity of *S. avermitilis aco* mutant.[6]

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1 Horinouchi, S. & Beppu, T. Autoregulators. in *Genetics and Biochemistry of Antibiotic Production* (ed. Vining, L.) 103–119 (Butterworth-Heinemann, Newton, MA, 1994).
2 Yamada, Y. Autoregulatory factors and regulation of antibiotic production in Streptomyces. In: *Microbial Signalling and Communication* (eds. England, R., Hobbs, G., Bainton, N. & Roberts, D.McL.) 177–196 (Society for General Microbiology, Cambridge, 1999).
3 Kondo, K., Higuchi, Y., Sakuda, S., Nihira, T. & Yamada, Y. New viriginiae butanolides from *Streptomyces virginiae*. *J. Antibiot.* **42,** 769–778 (1989).
4 Sato, K., Nihira, T., Sakuda, S., Yanagimoto, M. & Yamada, Y. Isolation and structure of a new butyrolactone autoregulator from *Streptomyces* sp. FRI-5. *J. Ferment. Bioeng.* **68,** 170–173 (1989).
5 Takano, E. *et al.* Purification and structural determination of SCB1, a γ-butyrolactone that elicits antibiotic production in *Streptomyces coelicolor* A3 (2). *J. Biol. Chem.* **275,** 11010–11016 (2000).
6 Kitani, S. *et al.* Avenolide, a novel *Streptomyces* hormone controlling antibiotic production in *Streptomyces avermitilis*. *Proc. Natl. Acad. Sci. USA* **108,** 16410–16415 (2011).
7 Mukku, V. J. R., Speitling, M., Laatsch, H. & Helmke, E. New butenolides from two marine streptomycetes. *J. Nat. Prod.* **63,** 1570–1572 (2000).
8 Gawronski, J. K., van Oeveren, A., van der Deen, H., Leung, C. W. J. & Feringa, B. L. Simple circular dichroic method for the determination of absolute configuration of 5-substituted 2(5*H*)-furanones. *J. Org. Chem.* **61,** 1513–1515 (1996).
9 Jacobsen, E. N., Marko, I., Mungall, W. S., Schroder, G. & Sharpless, K. B. Asymmetric dihydroxylation via ligand-accelerated catalysis. *J. Am. Chem. Soc.* **110,** 1968–1970 (1988).
10 Kolb, H., VanNiewenhze, M. S. & Sharpless, K. B. Catalytic asymmetric dihydroxylation. *Chem. Rev.* **94,** 2483–2547 (1994).
11 Corey, E. J., Guzman-Perez, A. & Noe, M. C. The application of a mechanistic model leads to the extension of the Sharpless asymmetric dihydroxylation to allylic 4-methoxybenzoates and conformationally related amine and homoallylic alcohol derivatives. *J. Am. Chem. Soc.* **117,** 10805–10816 (1995).
12 Nagamitsu, T. *et al.* Total synthesis of nafuredin-γ, a γ-lactone related to nafuredin with selective inhibitory activity against NADH-fumarate reductase. *Tetrahedron Lett.* **44,** 6441–6444 (2003).
13 Nagamitsu, T. *et al.* The total synthesis and biological evaluation of nafuredin-γ and its analogues. *Tetrahedron* **64,** 8117–8127 (2008).
14 Parikh, J. R. & Doering, W.v.E. Sulfur trioxide in the oxidation of alcohols by dimethyl sulfoxide. *J. Am. Chem. Soc.* **89,** 5505–5507 (1967).
15 Liu, L. & Floreancig, P. E. Cyclization reactions through DDQ-mediated vinyl oxazolidinone oxidation. *Org. Lett.* **11,** 3152–3155 (2009).
16 Liu, J.- H., Song, L.- D. & Long, Y.- Q. Studies toward the total synthesis of cyclodidemniserinol trisulfate. Part I: 3,5,7-Trisubstituted 6,8-dioxabicyclo [3.2.1] octane core structure construction via a convergent and a linear stereoselective synthesis. *Tetrahedron Lett.* **50,** 4587–4591 (2009).
17 Katsuki, T. & Sharpless, K. B. The first practical method for asymmetric epoxidation. *J. Am. Chem. Soc.* **102,** 5974–5976 (1980).
18 Trnka, T. M. & Grubbs, R. H. The development of L_2X_2Ru=CHR olefin metathesis catalysts: an organometallic success story. *Acc. Chem. Res.* **34,** 18–29 (2001).
19 Fürstner, A. Olefin metathesis and beyond. *Angew. Chem. Int. Ed. Engl.* **39,** 3012–3043 (2000).
20 Grubbs, R. H. & Chang, S. Recent advances in olefin metathesis and its application in organic synthesis. *Tetrahedron* **54,** 4413–4450 (1998).
21 Schuster, M. & Blechert, S. Olefin metathesis in organic chemistry. *Angew. Chem. Int. Ed. Engl.* **36,** 2036–2056 (1997).
22 Fürstner, A. Recent advancements in ring closing olefin metathesis. *Top. Catal.* **4,** 285–299 (1997).
23 Seebach, D. & Steinmueller, D. Preparation of (*S*)-2-methylbutyl and (*S*)-*sec*-butyl ketones from optically active 2-methyl-1-butanol by the dithiane method. *Angew. Chem. Int. Ed. Engl.* **7,** 619–620 (1968).

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