Synthesis of New Trans Double-Bond Sphingolipid Analogues: $\Delta^{4,6}$ and Δ^{6} Ceramides

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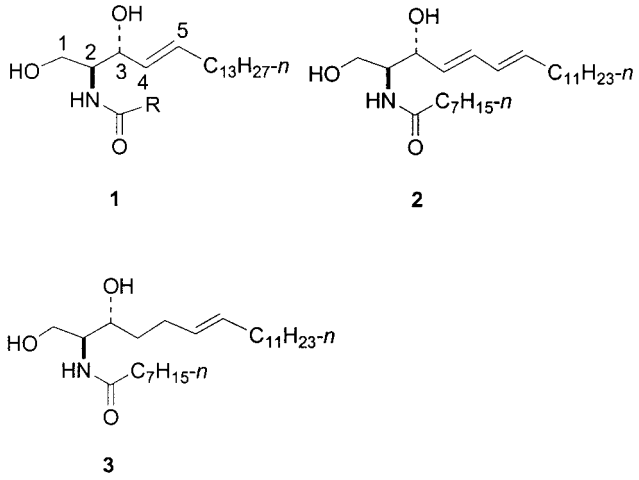
Unsaturation was introduced at $\Delta^{4,6}$ and Δ^{6} of the sphingoid chain of naturally occurring ceramide **1** via a β-keto sulfoxide (**12**) and sulfone (**18**) derived from *N*-Boc-L-serine methyl ester acetonide (**9**), affording two novel ceramide analogues, (2*S*,3*R*)-2-octanoylamidooctadeca-(4*E*,6*E*)-diene-1,3-diol (**2**) and (2*S*,3*R*)-2-octanoylamidooctadec-(6*E*)-ene-1,3-diol (**3**). After C-alkylation of **12** with (*E*)-1-bromo-2-tetradecene (**8**), a trans double bond was installed by elimination of PhS(O)H, providing conjugated dienone oxazolidine **13**. Reaction of **18** with **8**, followed by desulfonation (Al(Hg)), afforded keto-oxazolidine **20**, which bears a (*E*)-Δ^{6} double bond. The syntheses of analogues **2** and **3** from ketones **13** and **20**, respectively, were completed by the following sequence of reactions: diastereoselective reduction ($NaBH_4/CeCl_3$ or DIBAL-H), hydrolysis of the oxazolidine ring, liberation of the amino group, and installation of the *N*-amide group.

Introduction

Ceramide (*N*-acylsphingosine, **1**) is a long-chain aliphatic 2-amido-1,3-diol with a C(4),C(5)-trans double bond (Chart 1).[1] Considerable interest has been focused on its biochemistry and cell biology. Ceramide is a key intermediate in the biosynthesis of many other biologically important sphingolipids;[2] it has been implicated in many physiological events, including the regulation of cell growth and differentiation, inflammation, and programmed cell death (apoptosis).[3] Ceramide is also a regulator of many biochemical and cellular responses to stress, such as exposure to heat, radiation, oxidative conditions, and chemotherapeutic agents.[4]

Ceramide as well as more complex sphingolipids are required for activation of membrane fusion of Semliki Forest virus (SFV) and other alphaviruses.[5] The C(4)−C(5) trans double bond in the sphingoid base of naturally occurring **1** may be crucial for ceramide's capacity to modulate various fundamental biological functions.[6] For example, it was shown that unnatural ceramide analogues having a cis C(4)−C(5) or a trans C(5)−C(6) double bond do not support fusion of SFV.[6] Similarly, the presence of the (*E*)-C(4)−C(5) double bond in the long-chain base of ceramide is important for the apoptotic response of **1**, since analogues that lack this double bond have reduced activity.[7,8] An understanding of the structural biology of ceramide may be aided by the synthesis of its homologues, which can be tested as both substrates for ceramide-utilizing enzymes and as substitutes for naturally occurring ceramide in cell signaling events and other biological activities.

Chart 1



Synthetic routes to sphingosine and ceramide have been reviewed recently.[9] In an extension of our previous studies on the (*E*)-C(5)−C(6),[6] (*E*)-C(7)−C(8),[10] and (*E*)-C(15)−C(16)[10] double-bond analogues of ceramide, we report here a novel approach that allows the efficient

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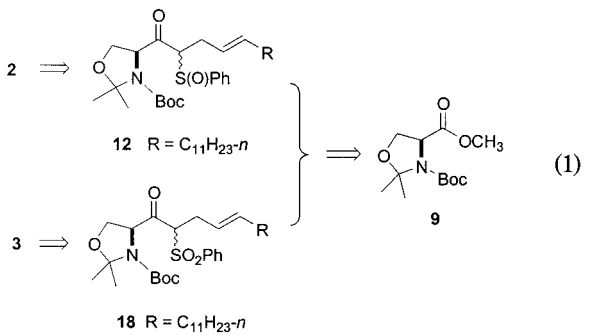
syntheses of two new unnatural analogues of ceramide in which a diene [(*E*)-C(4)−C(5) and (*E*)-C(6)−C(7), compound **2**; see Chart 1] and an (*E*)-C(6)−C(7) double bond are incorporated (compound **3**). Diene **2** may have a higher reactivity than ceramide **1** in mitochondria[11] by undergoing facilitated oxidation at C(3) to give an α,*β*-unsaturated ketone and hydrogen peroxide as the initial oxidation products, which eventually lead to apoptotic cell death. Although 4,6-sphingadienes have not been reported to occur naturally, it is of interest to note that 4,8-sphingadiene is the principal sphingoid backbone of the glucosylceramides found in soybean and wheat.[12] Its function is not known.

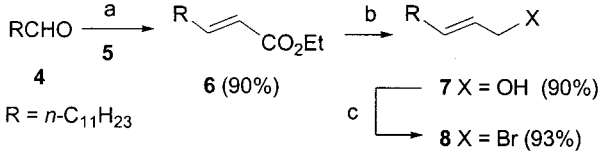
Results and Discussion

Since organosulfur compounds play important roles in the formation of carbon−carbon bonds,[13] we decided to synthesize the novel analogues of ceramide **2** and **3** by use of the α-keto-sulfoxide and sulfone intermediates shown in the retrosynthetic plan (eq 1). Both of these intermediates are derived from the commercially available protected L-serine building block **9**.



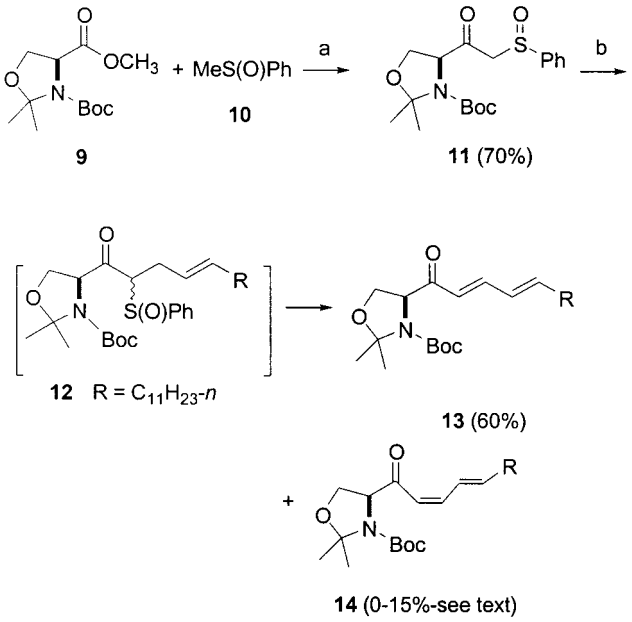
Synthesis of Allylic Bromide 8. As shown in Scheme 1, allylic bromide **8** was synthesized via α,*β*-unsaturated ester (*E*)-**6**. The latter was obtained in 90% yield (*E*/*Z* ratio, ∼18:1) by Horner−Wadsworth−Emmons (HWE) reaction of dodecanal (**4**) with the phosphonoacetate **5**. Reduction of ester **6** with DIBAL-H afforded allylic alcohol **7** in 90% yield. Treatment of allylic alcohol **7** with NBS via Mitsunobu reaction[14] gave crude allylic bromide **8** in 93% yield.

Scheme 1. Synthesis of Allylic Bromide 8[a]



[a] Reagents and conditions: (a) $(EtO)_2P(O)CH_2CO_2Et$ (**5**), LiBr, Et_3N, rt; (b) 3 equiv of DIBAL-H, THF, −78 to 0 °C; (c) NBS, Ph_3P, CH_2Cl_2, rt.

Scheme 2. Synthesis of Ketone 13[a]



[a] Reagents and conditions: (a) LDA, THF, −78 °C to rt; (b) **8**, K_2CO_3, DMF, rt, 3 d.

Synthesis of Dienone 13. The synthesis of dienone **13** started with the protected serine-derived methyl ester **9**, which is readily prepared from L-serine in three high-yielding steps.[15] Condensation of ester **9** with 2 equiv of the carbanion of methyl phenyl sulfoxide at −78 °C gave *β*-ketosulfoxide **11** in 70% yield (Scheme 2). Several methods were screened for alkylation of **11** with allylic bromide **8**. Cesium carbonate in DMF provided elimination product **13**, presumably via intermediate **12**. However, DBU in benzene was more convenient than Cs_2CO_3 in DMF, since no workup was required to obtain dienone **13**; the reaction mixture was simply passed through a pad of silica gel to remove the precipitate (DBU·HBr). Heating the filtrate in benzene for several hours afforded a mixture of two $\Delta^{4,6}$-dienones, (4*E*,6*E*)-**13** and (4*Z*,6*E*)-**14**, in a ratio of ∼4:1. We were able to avoid the formation of **14** by carrying out the reaction at room temperature, even though the reaction was slow.[16] However, we noted that the optical rotation of **13** formed by using any of the above conditions was low, suggesting that racemization had occurred. The synthesis of chiral **13** was finally realized by using K_2CO_3[17] in DMF. This two-step reaction was very efficient at room temperature, providing diene

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(14) For a review of the Mitsunobu reaction, see: Hughes, D. L. *Org. React.* **1992**, *42*, 335−656.

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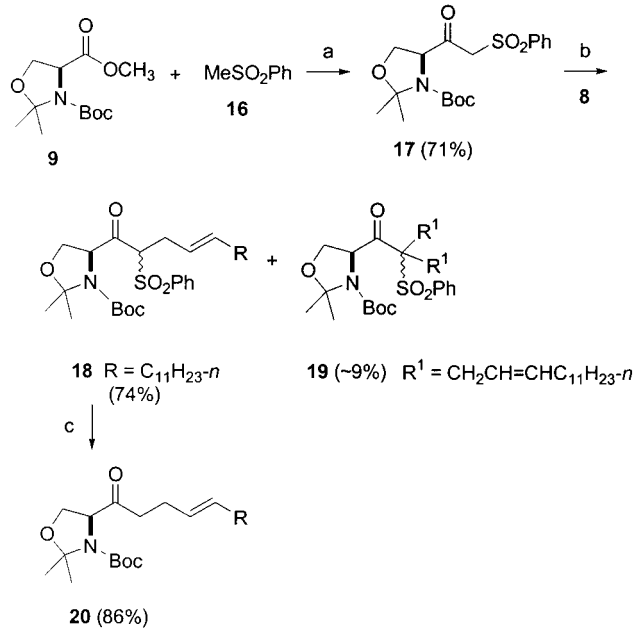
Scheme 3. Synthesis of Ketone 20[a]



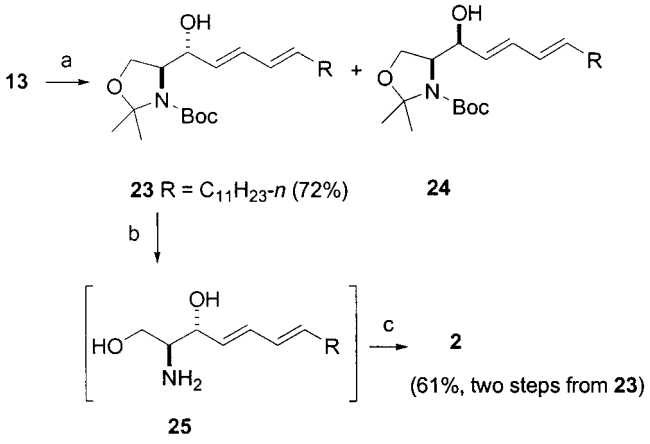
[a] Reagents and conditions: (a) *n*-BuLi, THF, −78 °C; (b) DBU, benzene, rt; (c) Al(Hg), THF/H_2O (20/1), rt.

13 exclusively in 60% yield.[18] These conditions not only avoided the difficult task of separating dienones **13** and **14**, which are formed together at elevated temperature, but also afforded **13** without racemization.

Synthesis of Ketone 20. Since α-ketosulfoxide **12** undergoes elimination even at room temperature it is not a suitable synthon for ketone **20**. Scheme 3 outlines the use of sulfone chemistry for the synthesis of **20**. Reaction of **9** with 2 equiv of the carbanion of methyl phenyl sulfone (**16**) at −78 °C gave *β*-ketosulfone **17** in 71% yield. Alkylation of **17** with allylic bromide **8** in the presence of a variety of bases was investigated. In Cs_2CO_3/DMF and DBU/DMF, dialkylation was the predominant reaction, giving **19** as the principal product. In DBU/benzene, monoalkylation of **17** was very efficient, giving **18**/**19** in a ratio of ~8:1. Sulfone intermediates **18** and **19** were readily separated by silica gel column chromatography.[19] Desulfonylation of *β*-ketosulfone **18** with aluminum amalgam[20] afforded ketone **20** in 86% yield.

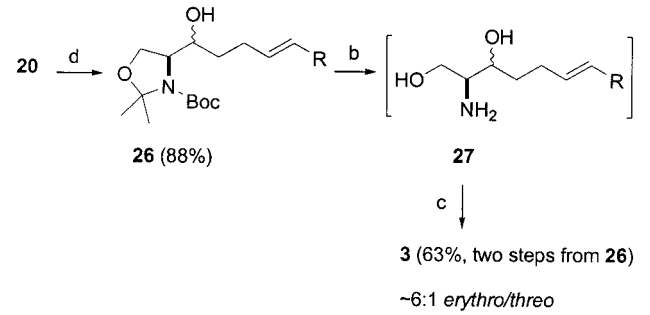
Synthesis of Ceramide Analogues 2 and 3. Several reducing agents were screened for the attempted diastereoselective reduction of ketone **13** (Scheme 4).[21] A higher erythro selectivity was observed with $NaBH_4$/$CeCl_3$ in MeOH or DIBAL-H in THF, which gave *erythro*-**23**/*threo*-**24** in a ratio of 4−5:1. However, the undesired threo diastereomer was formed as the major product with the other reducing agents we used; L-Selectride gave a ~1.3:1 ratio of threo/erythro isomers, and Red-Al gave **24**:**23** in 2:1 ratio). After diastereomers **23** and **24** were separated by column chromatography, acid hydrolysis of **23** (1 M HCl in THF, 70 °C) provided D-*erythro*-sphingosine **25**. Ceramide analogue **2** was obtained by *N*-acylation of sphingosine **25** with *p*-nitrophenyl octanoate.

Scheme 4. Synthesis of Ceramide Analogue 2[a]



[a] Reagents and conditions: (a) $NaBH_4$, $CeCl_3$, MeOH or DIBAL-H, THF, −15 to 0 °C; (b) 1 M HCl, THF, 70 °C; (c) *p*-$O_2NC_6H_4CO_2C_7H_{15}$-*n*, THF, rt.

Scheme 5. Synthesis of Ceramide Analogue 3[a]



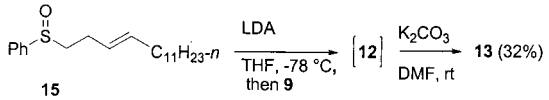
[a] Reagents and conditions: (a) (d) $NaBH_4$, MeOH, or DIBAL-H, THF, −15 °C; (b) 1 M HCl, THF, 70 °C; (c) *p*-$O_2NC_6H_4CO_2C_7H_{15}$-*n*, THF, rt.

Reduction of **20** with $NaBH_4$ or DIBAL-H gave alcohol **26**, but unfortunately the two diastereoisomers could not be isolated at this step (Scheme 5). Acid hydrolysis of **26** (1 M HCl in THF, 70 °C) provided crude sphingosine **27**. *N*-Acylation of sphingosine **27** with *p*-nitrophenyl oc-

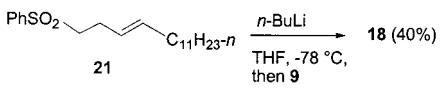
(16) It is well known that elimination of a sulfoxide moiety from *β*-ketosulfoxides at high temperature affords (*E*)-enones; see: Trost, B. M.; Salzmann, T. N.; Hiroi, K. *J. Am. Chem. Soc.* **1978**, *98*, 4887−4902. Pyrolytic elimination of the sulfinyl moiety from an analogue of **12** that lacked the Δ^6 double bond gave the conjugated (*E*)-Δ^4-3-keto product exclusively. When allylic *β*′-protons are present in a *β*-ketosulfoxide (e.g., **12**), we observed that the elimination reaction proceeds even at room temperature, affording a conjugated dienone (e.g., **13**).

(17) Anhydrous K_2CO_3 in CH_3CN did not cause racemization of an oxazolidine in an enone derived from a chiral *β*-ketophosphonate. See: Koskinen, A. M. P.; Koskinen, P. M. *Synlett* **1993**, 501−502.

(18) A less desirable route to dienone **13** is as follows. Treatment of sulfoxide **15** with LDA, followed by reaction of the α-sulfinyl carbanion with ester **9** at −78 °C, gave the presumed *β*-ketosulfoxide intermediate **12**. After workup, the reaction mixture was dissolved in DMF and a small amount of K_2CO_3 was added, affording dienone **13** in ~32% overall yield.



(19) A less efficient approach to sulfone **18** is as follows. Reaction of methyl phenyl sulfone (**16**) with (*n*-BuLi) then with allylic bromide **8**, gave sulfone **21** in 50% yield, together with dialkylation byproduct **22**. Treatment of **21** with *n*-BuLi and reaction with ester **9** gave sulfone **18** in 40% yield.



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tanoate gave two diastereoisomers, which were separated by column chromatography. The erythro/threo ratio was about 6:1.

In summary, a simple and convenient method has been established for the regiospecific introduction of unsaturation (both at a single site and as a conjugated diene) into the sphingoid backbone of ceramide by employing organosulfur chemistry.

Experimental Section

General Information. Melting points were measured on a Hoover capillary melting point apparatus and are uncorrected. ^{1}H NMR and ^{13}C NMR spectra were recorded at 400 and 100 MHz on a Bruker spectrometer, respectively, and were referenced to the residual $CHCl_3$ at 7.24 (^{1}H) and 77.00 ppm (^{13}C). $CDCl_3$ was the only solvent used for the NMR analyses unless otherwise indicated. IR spectra were recorded on a Perkin-Elmer 1600-series FT-IR spectrophotometer, and $CHCl_3$ was used as the sole solvent. Optical rotations were measured in a 1.0-dm cell on a JASCO model DIP-140 digital polarimeter. High-resolution mass spectra were recorded the University of California at Riverside. THF was distilled from sodium and benzophenone immediately before use. CH_2Cl_2 and DMF were dried over CaH_2. Spectral-grade benzene was distilled from sodium wire. Flash chromatography and TLC were carried out with Merck silica gel 60 (230−400 ASTM mesh) and Merck 60F_{254} (0.25-mm thick) sheets, respectively. $NaBH_4$, L-Selectride, and *p*-nitrophenyl octanoate were purchased from Acros. Red-Al was purchased from Fluka. Ester **9** and diisopropylamine were purchased from Aldrich. Methyl phenyl sulfoxide (**10**) and methyl phenyl sulfone (**16**) were purchased from Alfa-Aesar.

Ethyl (*E*)-2-Tetradecenoate (6). To a nitrogen-flushed solution of 21.0 g (245 mmol) of LiBr in 200 mL of dry THF was injected 11.7 mL (13.2 g, 59 mmol) of $(EtO)_2P(O)CH_2CO_2$-Et (**5**) at rt. After the solution was stirred at rt for 10 min, 13.6 mL (98 mmol) of Et_3N was injected, and stirring was continued for 10 min. A solution of dodecanal (**4**, 9.0 g, 49 mmol) in 20 mL of dry THF was injected. The reaction mixture was stirred vigorously at rt until the full consumption of the aldehyde **4** was observed (TLC). The precipitate was removed by passing the reaction mixture through a pad of silica gel in a sintered glass funnel. The pad was washed with hexane/EtOAc 10:1. Concentration gave a pale yellow oil that was purified by column chromatography (hexane/EtOAc 20:1), providing 11.2 g (90%) of ester **6** as a colorless oil. The NMR data are in full accord with the literature data.[22]

(*E*)-2-Tetradecen-1-ol (7). To a solution of 5.0 g (20 mmol) of ester **6** in 150 mL of dry THF was injected 40 mL of DIBAL-H (a 1.5 M solution in toluene, 60 mmol) at −78 °C under argon. After 1 h, the solution was allowed to warm to 0 °C. The reaction was quenched by slow addition of 5 mL of MeOH followed by 20 mL of cold 5% aqueous potassium sodium tartrate solution. The product was extracted with EtOAc (3 × 20 mL), and the combined organic layers were washed with brine (15 mL), dried (Na_2SO_4), and concentrated. Purification of the residue by column chromatography (hexane/EtOAc 4:1) gave 4.0 g (90%) of alcohol **7** as a colorless oil. The NMR data are in full accord with the literature data.[23]

(*E*)-1-Bromo-2-tetradecene (8). To a solution of 2.12 g (10 mmol) of alcohol **7** and 2.75 g (1.05 mmol) of Ph_3P in 30 mL of dry CH_2Cl_2 was added 1.96 g (11 mmol) of NBS at 0 °C under argon. The reaction mixture was stirred at 0 °C for 1 h, allowed to warm to rt, and then stirred for 1 h. The mixture was diluted with 60 mL of hexane and passed through a pad of silica gel with suction to remove the precipitate of Ph_3PO. The filtrate was concentrated, and the resulting residue was dissolved in 60 mL of hexane and passed through a pad of silica gel to remove the precipitate of Ph_3PO again. Concentration gave 2.56 g (93%) of bromide **8** as a colorless oil: ^{1}H NMR δ 0.86 (t, 3H, J = 6.5 Hz), 1.10−1.50 (m, 18H), 2.04 (m, 2H), 3.92 (d, 2H, J = 7.3 Hz), 5.69 (m, 2H); ^{13}C NMR δ 14.1, 22.7, 28.8, 29.1, 29.3, 29.4, 29.57, 29.62, 29.64, 31.9, 32.1, 33.7, 126.2, 136.8.

***N-tert*-Butoxycarbonyl (4*S*)-4-[(Phenylsulfinyl)acetyl]-2,2-dimethyl-1,3-oxazolidine (11).** To a solution of diisopropylamine (930 μL, 6.6 mmol) in 8 mL of THF was added 2.64 mL of *n*-butyllithium (a 2.5 M solution in hexane, 6.6 mmol) at −15 °C under nitrogen. After the mixture was stirred at this temperature for 30 min, a solution of methyl phenyl sulfoxide (**10**, 841 mg, 6.0 mmol) in 5 mL of THF was added dropwise. The mixture was stirred at −15 °C for 30 min and then chilled to −78 °C. A solution of ester **9** (778 mg, 3.0 mmol) in 5 mL of THF was added dropwise. The solution was stirred at −78 °C for 2 h and allowed to warm to rt overnight. After saturated aqueous NH_4Cl (10 mL) solution was added, the product was extracted with EtOAc, washed with brine, and dried ($MgSO_4$). Purification by column chromatography (hexane/EtOAc 1:4, R_f 0.72) gave 771 mg (70%) of **11** as a yellow solid: mp 82.5−85.0 °C; IR 1710, 1385, 1170, 1090, 1055 cm^{-1}; ^{1}H NMR (C_6D_6, 70 °C) δ 1.35 (s, 9H), 1.40 (s, 3H), 1.67 (s, 3H), 3.75 (m, 4H), 4.26 and 4.40 (two sets of s, 1H), 7.10 (m, 3H), 7.49 (m, 2H); the ^{13}C NMR spectrum was very complex because several diastereoisomers are present (see the Supporting Information); HR-MS (FAB, MH^+) calcd for m/z $C_{18}H_{26}NO_5S$ 368.1532, found 368.1516.

(3*E*)-Pentadecenyl Phenyl Sulfoxide (15). To a solution of diisopropylamine (930 μL, 6.6 mmol) in 8 mL of THF was added 2.64 mL of *n*-butyllithium (a 2.5 M solution in hexane, 6.6 mmol) at −15 °C under nitrogen. After the mixture was stirred at this temperature for 30 min, a solution of PhS(O)-Me (**10**, 841 mg, 6.0 mmol) in 5 mL of THF was added dropwise. The mixture was stirred at −15 °C for 30 min. A solution of bromide **8** (1.65 g, 6.0 mmol) in 5 mL of THF was added dropwise. The mixture was stirred at −15 °C for 2 h and allowed to warm to rt overnight. Saturated aqueous NH_4-Cl solution (10 mL) was added, and the product was extracted with EtOAc. The organic layer was washed with brine, dried ($MgSO_4$), and concentrated. Purification of the residue by column chromatography (hexane/EtOAc 3:2, R_f 0.72) gave 1.32 g (66%) of **15** as a colorless liquid: ^{1}H NMR δ 0.83 (t, 3H, J = 6.6 Hz), 1.10−1.40 (m, 18H), 1.92 (q, 2H, J = 6.5 Hz), 2.23 (dt, 1H, J = 21.5, 7.1 Hz), 2.41 (dt, 1H, J = 22.3, 7.5 Hz), 2.76 (q, 2H, J = 8.0 Hz), 5.30 (m, 1H), 5.49 (m, 1H), 7.45 (m, 3H), 7.58 (m, 2H); ^{13}C NMR δ 14.0, 22.6, 25.2, 29.0, 29.17, 29.24, 29.4, 29.5, 29.53, 29.56, 29.6, 31.8, 32.4, 56.9, 57.0, 123.9, 125.9, 129.1, 130.8, 133.5, 143.8.

***N-tert*-Butoxycarbonyl (4*S*)-4-[1′-Oxo-(2′*E*,4′*E*)-hexadecadienyl]-2,2-dimethyl-1,3-oxazolidine [(−)-13].** To a solution of β-ketosulfoxide **11** (370 mg, 1.0 mmol) in 4 mL of DMF was added K_2CO_3 (170 mg, 1.2 mmol) at rt. After the mixture was stirred at rt for 1 h under nitrogen, a solution of bromide **8** (275 mg, 1 mmol) in 2 mL of DMF was added. The mixture was stirred at rt for 3 days. Water (5 mL) was added, and the product was extracted with Et_2O (3 × 15 mL), washed with brine, and dried ($MgSO_4$). Concentration and purification by flash column chromatography (hexane/EtOAc 4:1, R_f 0.75) gave 261 mg (60%) of **13** as a white solid: mp 48.5−50.0 °C; $[\alpha]^{25}_D$ −36.8° (*c* 2.5, $CHCl_3$); IR 1700, 1385 cm^{-1}; ^{1}H NMR (C_6D_6, 70 °C) δ 0.88 (t, 3H, J = 7.0 Hz), 1.10−1.30 (m, 18H), 1.39 (s, 9H), 1.55 (s, 3H), 1.82 (s, 3H), 1.92 (q, 2H, J = 6.9 Hz), 3.78−3.84 (m, 2H), 4.43 (br s, 1H), 5.81−5.89 (m, 1H), 5.95−6.01 (m, 1H), 6.23 (d, 1H, J = 15.3 Hz), 7.29 (dd, 1H, J = 15.3, 10.7 Hz); ^{13}C NMR (C_6D_6)[24] δ 14.1, 20.1, 28.4, 29.0, 29.1, 29.4, 29.5, 29.7, 32.2, 33.2, 33.3, (64.7) 65.0, (66.0) 66.3, (79.9) 80.0, (94.4) 95.3, 123.3 (124.4), (128.2) 129.2, (144.0) 144.2, (145.8) 146.4, 151.7 (152.2), (195.5) 196.0; HR-MS (FAB, MNa^+) calcd for m/z $C_{26}H_{45}NO_4Na$ 458.3247, found 458.3249.

***N-tert*-Butoxycarbonyl (4*S*)-4-[1′-oxo-(2′*Z*,4′*E*)-hexadecadienyl]-2,2-dimethyl-1,3-oxazolidine (14):** ^{1}H NMR

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(24) The oxazolidine system undergoes a dynamic equilibrium at ambient temperature.

(C_6D_6, 70 °C) δ 0.89 (t, 3H, J = 6.2 Hz), 1.10−1.30 (m, 16H), 1.39 (s, 9H), 1.52 (s, 3H), 1.50−1.60 (m, 2H), 1.81 (s, 3H), 2.81 (m, 2H), 3.77 (m, 2H), 4.27 (br s, 1H), 5.13 (d, 1H, J = 10.8 Hz), 5.50 (d, 1H, J = 17.4 Hz), 6.13 (m, 2H).

***N-tert*-Butoxycarbonyl (4*S*)-4-[1′-Hydroxy-(2′*E*,4′*E*)-hexadecadienyl]-2,2-dimethyl-1,3-oxazolidine [(−)-23].** To a solution dienone **13** (325 mg, 0.75 mmol) in 15 mL of dry MeOH was added anhydrous $CeCl_3$ (61.5 mg, 0.25 mmol) at −15 °C. After the mixture was stirred for 10 min, $NaBH_4$ (32 mg, 0.85 mmol) was added. The temperature was gradually raised to 0 °C. After 2 h, water (20 mL) was added, and the product was extracted with Et_2O (3 × 25 mL), washed with brine, and dried ($MgSO_4$). Concentration and purification by column chromatography (hexane/EtOAc 4:1, R_f 0.45) gave 235 mg (72%) of **23** as a colorless oil: $[\alpha]^{25}_D$ −18.6° (*c* 1.5, $CHCl_3$); IR 1690, 1375 cm^{-1}; ^1H NMR (C_6D_6, 70 °C) δ 0.89 (t, 3H, J = 6.6 Hz), 1.28 (m, 18H), 1.38 (s, 9H), 1.43 (s, 3H), 1.61 (s, 3H), 2.02 (q, 2H, J = 6.8 Hz), 3.65 (m, 1H), 3.79 (br s, 1H), 3.94 (br s, 1H), 4.34 (m, 1H), 5.59−5.66 (m, 2H), 6.09 (m, 1H), 6.38 (m, 1H); ^{13}C NMR (C_6D_6, 70 °C) δ 12.5, 14.1, 19.0, 22.9, 24.4, 26.8, 28.1, 28.4, 29.5, 29.59, 29.68, 29.71, 29.88, 29.99, 30.04, 32.3, 32.9, 33.1, 62.8, 65.0, 73.6, 80.2, 94.6, 128.7, 130.5, 131.78, 134.82; HR-MS (FAB, MNa^+) calcd for m/z $C_{26}H_{47}NO_4Na$ 460.3403, found 460.3419.

(2*S*,3*R*)-(4*E*,6*E*)-2-Octanoylamidooctadecadiene-1,3-diol [(−)-2]. A solution of 88 mg (0.2 mmol) of **23** in 4 mL of 1 M HCl and 4 mL of THF was heated at 70 °C with stirring for 10 h under argon. The reaction mixture was cooled to rt and neutralized with 1 M NaOH (4 mL). The product was extracted with EtOAc (3 × 10 mL), and the combined organic layers were washed with brine and dried (Na_2SO_4). Removal of the solvent provided crude sphingosine analogue **25** as a white solid, which was used in the next reaction without further purification. To a solution of **25** in 6 mL of dry THF was added 108 mg (0.40 mmol) of *p*-nitrophenyl octanoate at rt. The mixture was stirred for 48 h and concentrated. Purification by flash chromatography ($CHCl_3$/MeOH 9:1) afforded 51 mg (61%, two steps) of $\Delta^{4,6}$-ceramide analogue **2** as a white solid: mp 69.0−71.0 °C; $[\alpha]^{25}_D$ −4.30° (*c* 2.2, $CHCl_3$); IR 1620, 1540, 1455 cm^{-1}; ^1H NMR δ 0.86 (t, 6H, J = 6.6 Hz), 1.10−1.40 (m, 26H), 1.60 (m, 2H), 2.05 (q, 2H, J = 7.1 Hz), 2.20 (t, 2H, J = 7.4 Hz), 3.69 (m, 1H), 3.90 (m, 2H), 4.37 (br s, 1H), 5.60 (dd, 1H, J = 15.3, 6.3 Hz), 5.73 (m, 1H), 6.00 (m, 1H), 6.27 (m, 2H); ^{13}C NMR δ 14.05, 14.10, 22.6, 22.7, 25.8, 29.0, 29.15, 29.21, 29.3, 29.49, 29.59, 29.62, 29.65, 31.7, 31.9, 32.7, 36.8, 54.5, 62.5, 74.5, 128.9, 132.7, 136.7, 174.0; HR-MS (FAB, MNa^+) calcd for m/z $C_{26}H_{49}NO_3Na$ 446.3610, found 446.3598.

***N-tert*-Butoxycarbonyl (4*S*)-4-[(Phenylsulfonyl)acetyl]-2,2-dimethyl-1,3-oxazolidine [(−)-17].** A solution of sulfone **16** (937 mg, 6.0 mmol) in 10 mL of THF was added 2.64 mL of *n*-butyllithium (a 2.5 M solution in hexane, 6.6 mmol) at −15 °C under nitrogen. The reaction mixture was stirred at −15 °C for 30 min and then chilled to −78 °C. A solution of ester **9** (778 mg, 3.0 mmol) in 5 mL of THF was added dropwise. The reaction mixture was stirred at −78 °C for 2 h and allowed to warm to rt overnight. Saturated aqueous NH_4Cl solution (10 mL) was added, and the product was extracted with EtOAc, washed with brine, and dried ($MgSO_4$). Purification by column chromatography (hexane/EtOAc 1:1, R_f 0.72) gave 817 mg (71%) of **17** as a white solid: mp 105−106 °C; $[\alpha]^{25}_D$ −93.2° (*c* 2.2, $CHCl_3$); ^1H NMR (C_6D_6, 70 °C) δ 1.33 (s, 9H), 1.45 (s, 3H), 1.58 (s, 3H), 3.90 (br s, 2H), 3.99 (d, 1H, J = 14.0 Hz), 4.15 (br s, 1H), 4.53 (br s, 1H), 6.99 (m, 3H), 7.78 (m, 2H); HR-MS (FAB, MH^+) calcd for m/z $C_{18}H_{26}NO_6S$ 384.1481, found 384.1487.

(3*E*)-Pentadecenyl Phenyl Sulfone (21). To a solution of sulfone **16** (780 mg, 5.0 mmol) in 5 mL of THF was added 2.4 mL of *n*-butyllithium (a 2.5 M solution in hexane, 6.0 mmol) at −78 °C under nitrogen. The solution was stirred at −78 °C for 30 min. A solution of bromide **8** (140 mg, 5.0 mmol) in 5 mL of THF was added dropwise. The mixture was stirred at −78 °C for 2 h and allowed to warm to rt overnight. After saturated aqueous NH_4Cl solution (10 mL) was added, the product was extracted with EtOAc, washed with brine, and dried ($MgSO_4$). Purification by column chromatography (hexane/EtOAc 3:2, R_f 0.72) gave 175 mg (50%) of **21** as a colorless liquid: ^1H NMR δ 0.83 (t, 3H, J = 7.0 Hz), 1.10−1.40 (m, 18H), 1.87 (m, 2H), 2.35 (m, 2H), 3.08 (m, 2H), 5.22 (m, 1H), 5.41 (m, 1H), 7.51 (m, 2H), 7.61 (m, 1H), 7.86 (m, 2H); ^{13}C NMR δ 14.0, 22.6, 25.8, 29.1, 29.2, 29.3, 29.47, 29.53, 31.8, 32.3, 56.0, 124.8, 128.0, 129.1, 133.5, 139.0.

***N-tert*-Butoxycarbonyl (4*S*)-4-[1′-Oxo-2′-phenylsulfonyl-(4′*E*)-hexadecenyl]-2,2-dimethyl-1,3-oxazolidine (18).** To a solution of β-ketosulfone **17** (383 mg, 1.0 mmol) in 10 mL of benzene was added DBU (153 mg, 1.0 mmol) at rt. After the mixture was stirred at rt for 1 h under nitrogen, a solution of bromide **8** (275 mg, 1 mmol) in 5.0 mL of benzene was added dropwise. The reaction mixture was stirred at rt for 3 h and passed through a pad of silica gel to remove the precipitate (DBU·HBr). The pad was washed with benzene. Purification by column chromatography (hexane/EtOAc 4:1, R_f 0.70) gave 428 mg (74%) of **18** as a colorless liquid: IR 1732, 1698, 1390, 1360, 1315, 1175, 1145 cm^{-1}; ^1H NMR (C_6D_6, 70 °C) δ 0.96 (t, 3H, J = 7.0 Hz), 1.35 (m, 18H), 1.44 (s, 9H), 1.55 (s, 3H), 1.74 (s, 3H), 1.88 (m, 1H), 1.97 (m, 1H), 2.60−3.00 (m, 2H), 3.80−4.19 (m, 1H), 4.01−4.30 (m, 1.5H), 4.78−4.90 (m, 1H), 5.39 (br s, 1H), 5.46−5.49 (m, 1.5H), 7.05−7.17 (m, 3H), 7.23 (m, 1H), 8.04 (br s, 1H); HR-MS (FAB, MH^+) calcd for m/z $C_{32}H_{52}NO_6S$ 578.3515, found 578.3515.

***N-tert*-Butoxycarbonyl (4*S*)-4-[1′-Oxo-(4′*E*)-hexadecenyl]-2,2-dimethyl-1,3-oxazolidine [(−)-20].** To a solution of ketosulfone **18** (290 mg, 0.50 mmol) in 25 mL of THF/H_2O 20/1 was added Al(Hg) (freshly prepared from aluminum foil; 135 mg, 5 mmol, 2% aqueous $HgCl_2$).[20] After the mixture was stirred at rt overnight, it was passed through a pad of silica gel with suction, which was washed with EtOAc. Concentration and purification by flash column chromatography (hexane/EtOAc 4:1, R_f 0.85) gave 186 mg (86%) of ketone **20** as a colorless oil: $[\alpha]^{25}_D$ −5.6° (*c* 2.8, $CHCl_3$); IR 1709, 1463, 1390, 1380, 1365, 1167 cm^{-1}; ^1H NMR (C_6D_6, 70 °C) δ 0.86 (t, 3H, J = 7.0 Hz), 1.29 (m, 18H), 1.38 (m, 9H), 1.47 (s, 3H), 1.75 (s, 3H), 1.96 (m, 2H), 2.30 (m, 2H), 2.39 (m, 2H), 3.69 (m, 2H), 4.16 (br s, 1H), 5.43 (m, 2H); ^{13}C NMR (C_6D_6, 70 °C) δ 14.1, 23.0, 26.7, 28.4, 29.6, 29.7, 29.9, 30.0, 30.1, 32.3, 32.9, 65.6, 65.7, 80.2, 129.1, 131.8, 206.2; HR-MS (FAB, MNa^+) calcd for m/z $C_{26}H_{47}NO_4Na$ 460.3403, found 460.3393.

***N-tert*-Butoxycarbonyl (4*S*)-4-[1′-Hydroxy-(4′*E*)-hexadecenyl]-2,2-dimethyl-1,3-oxazolidine (26).** To a solution of ketone **20** (65 mg, 0.15 mmol) in 4 mL of dry MeOH was added $NaBH_4$ (6.4 mg, 0.17 mmol) at −15 °C. The temperature was gradually raised to 0 °C. After 2 h, water (5 mL) was added, and the product was extracted with Et_2O (3 × 15 mL), washed with brine, dried ($MgSO_4$), and concentrated. Purification by column chromatography (hexane/EtOAc 4:1, R_f 0.45) gave 58 mg (88%) of **26** as a colorless oil: IR 1701, 1671, 1457, 1390, 1365 cm^{-1}; ^1H NMR (C_6D_6, 70 °C) δ 0.89 (t, 3H, J = 6.9 Hz), 1.22 (m, 20H), 1.30 (s, 9H), 1.47 (s, 3H), 1.63 (s, 3H), 2.00 (m, 2H), 2.20 (m, 1H), 2.34 (m, 1H), 3.61−3.69 (m, 2H), 3.84 (br s, 2H), 5.46−5.55 (m, 2H); ^{13}C NMR (C_6D_6, 70 °C) δ 14.1, 23.0, 27.2, 28.4, 29.1, 29.6, 29.7, 30.0, 30.1, 32.3, 33.0, 62.7, 65.1, 80.3, 94.4, 130.5, 131.2.

(2*S*,3*R*)-(6*E*)-2-Octanoylamidooctadecene-1,3-diol [(−)-3]. A solution of 88 mg (0.2 mmol) of **26** in 4 mL of 1 M HCl and 4 mL of THF was heated at 70 °C with stirring for 10 h under argon. The reaction mixture was cooled to rt and neutralized with 1 M NaOH (4 mL). The product was extracted with EtOAc (3 × 10 mL), and the combined organic layers were washed with brine and dried (Na_2SO_4). Removal of the solvent provided crude sphingosine analogue **27** as a white solid, which was used in the next reaction without further purification. To a solution of **27** in 6 mL of dry THF was added 108 mg (0.40 mmol) of *p*-nitrophenyl octanoate at rt. The mixture was stirred for 48 h and then concentrated under reduced pressure. Purification by column chromatography (EtOAc) afforded 53 mg (63%, two steps) of Δ^6-ceramide analogue **3** as a low-melting white solid: $[\alpha]^{25}_D$ −2.39° (*c* 2.2, $CHCl_3$); IR 1631, 1542 cm^{-1}; ^1H NMR δ 0.86 (t, 6H, J = 7.1 Hz), 1.10−1.40 (m, 26H), 1.52 (m, 2H), 1.62 (m, 2H), 1.93 (m, 2H), 2.07 (m, 2H), 2.21 (t, 2H, J = 7.7 Hz), 2.57 (br s, 2H), 3.78 (m, 2H), 3.88 (m, 1H),

3.96 (m, 1H), 5.38 (m, 2H), 6.20 (d, 1H, $J = 8.3$ Hz); ^{13}C NMR δ 14.05, 14.10, 22.5, 22.7, 25.8, 28.8, 29.0, 29.2, 29.4, 29.52, 29.53, 29.6, 29.7, 31.4, 31.7, 31.9, 32.6, 34.0, 36.9, 53.2, 65.4, 72.6, 129.1, 131.8, 174.1; HR-MS (DCI, MH^+) calcd for *m/z* $C_{26}H_{52}NO_3$ 426.3947, found 426.3952.

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Supporting Information Available: Copies of ^{1}H and ^{13}C NMR spectra for compounds **2**, **3**, **11**, **13**, **15**, **17**, **18**, **20**, **21**, and **23**. This material is available free of charge via the Internet at http://pubs.acs.org.

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