Stereoselective synthesis of \(N,O,O,O\)-tetraacetyl-\(D\)-ribo-phytosphingosine, \(N,O,O\)-triacyl-\(D\)-erythro-sphingosine and \(N,O,O\)-triacyl sphinganine from a common chiral intermediate derived from \(D\)-mannitol

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Abstract
An efficient protocol for the stereoselective synthesis of tetraacetyl-\(D\)-ribo-phytosphingosine, triacyl-\(D\)-erythro-sphingosine and triacyl sphinganine has been devised from a common chiral intermediate derived from commercially available \(D\)-mannitol. The key steps involved are Sharpless epoxidation, Miyashita C(2) selective endo mode azide opening of 2,3-epoxy alcohol, and selective \(E\)-Wittig olefination.

Keywords: Sphingolipids, \(D\)-mannitol, epoxidation, regioselective, Wittig olefination

Introduction

Sphingoid bases are long-chain aliphatic compounds typically possessing 2-amino-1,3-diol and 2-amino-1,3,4-triol functionality (Figure 1). They are the structural backbone of sphingolipids (cerebrosides, sphingomyelins, gangliosides and ceramides), which are an important membrane constituents of eukaryotic cells, plasma membranes and intracellular organelles, and responsible for many physiological processes including cell growth, adhesion, differentiation and also play a prominent role in cell signaling. Apart from this, many sphingolipids from marine organisms display pronounced antifungal, antitumor, antiviral, immunostimulatory, neuritogenic, antidiabetic, cytotoxic, and protein kinase inhibitor activities.

Among the naturally occurring sphingoid bases \(D\)-erythro-sphingosine 2 is the first isolated compound from human brain by Thudichum in 1884. In addition to 2, several sphingoid bases were isolated, among them \(C_{18}\)-phytosphingosines are the more biologically active sphingoid bases have that been isolated from plants, yeast, fungi, marine organisms and mammalian tissues such as brain, hair, intestine, uterus, liver, skin and blood plasma. \(D\)-ribo-
Phytosphingosine (1) is the most frequently occurring phytosphingosine in nature and has been shown to play an important role as a potential heat stress signal in yeast cells\(^7\) and as a cytotoxic agent against human leukemic cell lines\(^8\). Furthermore, \(D\)-erythro-sphingosine\(^9\) and \(D\)-ribo-phytosphingosines\(^10\) are essential part of more complex bioactive molecules such as GalCer (4) and KRN7000 (5) respectively.

**Figure 1.** Sphingolipids and glycolipids.

Sphingolipids are available only in limited amounts from natural sources, and their isolation and purification from natural sources is expensive and difficult task. Because of the interesting biological properties of sphingolipids, there is growing interest in developing efficient methods for their synthesis.\(^1,11\) Although, many methods for the synthesis of sphingolipids have been reported, a simple and straight forward synthesis from inexpensive starting material with high level of stereocontrol is always in demand. In continuation of our efforts on natural product synthesis\(^12\) and development of new methodologies\(^13\) we herein, disclose a simple and convenient new approach for the asymmetric synthesis of title compounds 1, 2, and 3 from \(D\)-mannitol (6), involving the steps with high stereocontrol approach.

**Results and Discussion**

Our retrosynthetic analysis for target compounds 1, 2, and 3 is depicted in Scheme 1. Retrosynthetically, it was envisioned that, all the three titled sphingolipids 1, 2 and 3 to be obtained from a common intermediate 11, could be acquired by regioselective epoxide opening with azide nucleophile followed by protection of 1,3-diol as their benzylethers and consequent deprotection of cyclohexylidene group of epoxy alcohol 8 derived from the \(D\)-mannitol (6).
Scheme 1. Retrosynthetic plan for the synthesis of targeted sphingolipids.

According to the above retrosynthetic analysis, the first target is to be synthesis of diastereomerically pure azido diol intermediate 11 (Scheme 2). Towards that object, the allylic alcohol 7, which was easily prepared from D-mannitol according to known procedures,\textsuperscript{14} was subjected to Sharpless catalytic asymmetric epoxidation\textsuperscript{15} using diethyl D-tartrate, Ti(OiPr)\textsubscript{4} and cumene hydroperoxide to afford epoxy alcohol 8 in 95% yield with high diastereoselectivity, (98% de, determined by NMR and GC-MS analysis).

Scheme 2. Synthesis of key intermediate 11.

The next crucial step in our strategy is C(2) regioselective opening of the epoxy alcohol 8 by azide nucleophile. This was accomplished by using NaN\textsubscript{3}-(CH\textsubscript{3}O)\textsubscript{3}B system developed by Miyashita and co-workers.\textsuperscript{16} This reaction proceeds \textit{via} an intramolecular boron chelate through
a novel endo-mode epoxide opening with extremely high C(2) selectivity. The same thing was observed in our case. Under Miyashita conditions, the azide nucleophile selectively opening epoxy alcohol 8 at C(2) rather than C(3) position, which is sterically hindered by neighbouring protected vicinal diol moiety (>95% de, based on crude 1H-NMR analysis). Furthermore, we have treated the crude product with NaIO4 to remove the C(3) opened compound easily in the form of aldehyde by the column chromatography. Gratifyingly, the desired azido diol 9 was isolated in 96% yield, and as a single diastereomer after purification. Benzyl ether protection of azido diol 9 followed by selective deprotection of cyclohexylidene group by treating with 10% HCl in CH3CN gave the desired intermediate 4-azido 1,2-diol 11 in 93% yield.

![Figure 2](image_url)

**Figure 2.** C(2) Selective azide nucleophile opening of epoxy alcohol borate complex.

After successful synthesis of the key intermediate 11, next we turned our attention towards the synthesis of D-ribo-phytosphingosine (1) from intermediate 11 (Scheme 3). Toward this objective, the primary alcohol of 11 was selectively protected as TBS ether and the secondary alcohol group was protected as benzyl ether to afford compound 13. At this juncture, it becomes necessary to free the compound 13 from the TBS group and it was removed by stirring a solution of 13 in dry THF in the presence of n-Bu4NF (TBAF), gave the compound 14 in 96% yield. The primary alcohol functionality of 14 was converted to the corresponding aldehyde moiety using 2-Iodoxybenzoic acid (IBX), and the obtained aldehyde was rather labile to column purification and therefore it was quickly subjected to Wittig olefination using n-C13H27P=Ph3Br+ ylide in presence of n-BuLi to afford alkene 15 as a mixture of trans and cis isomers (E/Z, 19:1; determined by 1H-NMR spectra) in 81% yield. The geometrical isomer ratio is no relevance to the planned synthetic sequence as the double bond will be reduced in the next step.

Having the desired compound at the penultimate stage, attention was focused on the final deprotection and reduction step. One-pot reduction of azide group, saturation of double bond, and deprotection of the benzyl ethers was carried out by hydrogenation using Pd(OH)2/C on 15 affording the crude residue of target molecule 1 which was difficult to purify by column chromatography and was therefore converted it into its acetyl derivative using Ac2O and pyridine to afford tetraacetyl D-ribo-phytosphingosine (1a) in good yield. The analytical and spectroscopic data of this compound is in good agreement with the reported data.
After successful synthesis of compound 1, then we turned our attention towards the synthesis of compounds 2 and 3. As per the Scheme 4, oxidative cleavage of the diol 11 with NaIO₄ yielded corresponding aldehyde and without column purification it was subjected to the Wittig olefination₁⁷ by using n-C₁₄H₂₉P⁺Ph₃Br⁻ ylide in the presence n-BuLi to afford alkene 16 as an inseparable mixture of trans and cis isomers (E/Z, 20:1, determined by ¹H-NMR spectra). The selective reduction of azide group of the compound 16 with Lindlar’s catalyst¹⁹ under hydrogen atmosphere provided the amine 17 in 83% yield. Deprotection of the benzyl groups in 17 by means of Birch reduction²⁰a gave the title compound D-erythro-sphingosine (2). For analytical purpose, compound 2 was acetylated with acetic anhydride in presence of pyridine and catalytic amount of DMAP to obtain triacetyl D-erythro-sphingosine (2a) in good yield. The analytical and spectroscopic data of both 2 and 2a were in good agreement with the reported data.⁹a-c,1₈b, 2₀

The third desired sphingoid base, sphinganine (3) was synthesized (Scheme 5) from compound 16 in a single step through the one-pot reduction of azide, hydrogenation of double bond, and deprotection of the benzyl ethers by catalytic hydrogenation using Pearlman’s catalyst. For analytical reasons, compound 3 was acetylated with acetic anhydride in presence of pyridine and DMAP (catalytic) to obtain N,O,O-triacetyl sphinganine (3a) in good yield. The analytical and spectroscopic data of 3 and 3a were in good agreement with the reported data of the respective natural product.\textsuperscript{21}

Scheme 5. Synthesis of triacetyl sphinganine 3a.

Conclusions

In conclusion, a stereoselective total synthesis of $N,O,O,O$-tetraacetyl D-ribophytopsphingosine, $N,O,O$-triacetyl D-erythro-sphingosine and $N,O,O$-triacetyl sphinganine were accomplished by versatile strategy from a common intermediate derived from D-mannitol. A combination of
Sharpless epoxidation, Miyashita C(2) selective endo mode azide opening of epoxy alcohol, and preferential E-Wittig olefination were effectively utilized in accomplishing the synthesis. We believed that the key intermediate reported in this paper serves as a good synthon for making of other natural products.

**Experimental Section**

**General.** The solvents were dried over standard drying agents and freshly distilled prior to use. The reagents were purchased from Aldrich and Lancaster, and were used without further purification unless otherwise stated. All moisture-sensitive reactions were carried out under N₂ atmosphere. Column chromatography: silica gel (SiO₂; Acme’s 60-120 mesh). Optical rotations: Perkin-Elmer P241 polarimeter and JASCO DIP-360 digital polarimeter at 25 °C, IR spectra: Perkin-Elmer IR-683 spectrophotometer. NMR: Recorded on Varian Gemini 200 or Bruker Avance 300 or Varian Unity 400 MHz spectrometer depends on their availability, using TMS as an internal standard for ¹H NMR, and CDCl₃ for ¹³C NMR (chemical shift values in δ, J in Hz). MS: Recorded either on Thermo-Finnigan MAT1020B or Micromass 7070H spectrometer operating at 70 eV using direct inlet system. All high resolution mass spectra (HRMS) were recorded on QSTAR XL hybrid MS/MS system equipped with an ESI source. GC-MS were recorded on Agilent 6890 series GC-MS system, GC (Agilent Technologies, Palo Alto, CA) equipped with a model 5973N mass selective detector and a HP-5MS capillary column (5% phenyl, 95% PDMS, 30 m x 0.25 mm i.d. x 0.25 µm film thickness).

(25,3S,4R)-2,3-Epoxy-4,5-(cyclohexylidenedioxy)-pentan-1-ol (8). To activated 4Å molecular sieves powder (2.8 g, 35% (wt/wt)) in dry CH₂Cl₂ (175 mL) under N₂ were sequentially added Ti(OiPr)₄ (0.95 mL, 3.2 mmol) and diethyl d-tartrate (0.67 mL, 4 mmol) at -20 °C, and the mixture was stirred for 30 min. A solution of 7 (8 g, 40.4 mmol) in CH₂Cl₂ (50 mL) was added, and the resulting mixture was stirred at -20 °C for 30 min. Cumene hydroperoxide (11.8 mL, 80.8 mmol) was added dropwise to the reaction mixture, and the resulting solution was stored at -20 °C in freezer for 72 h. Aq. tartaric acid (10%, 40 mL) was added slowly at -20 °C, and the whole mass was allowed to warm to r.t. After being stirred for 1 h, the reaction mixture was filtered, and the filtrate was extracted with CH₂Cl₂ (50 mL). The combined org. layers were treated with a pre-cooled (0°C) soln. of 25 mL of 30% NaOH (w/v) in brine at 0 °C [25 mL of 30% NaOH soln. in brine are prepared by adding 1.25 g of NaCl to a soln. of 7.5 g of NaOH in 22.5 mL of H₂O] and stirred for 20 min. The two layers were separated and the aq. layer was extracted with CH₂Cl₂ (2x20 mL). The combined organic layers were washed with brine, dried (anh. Na₂SO₄), filtered and concentrated in vacuo. The residue was purified by column chromatography (SiO₂; EtOAc/hexane, 1:4) to give 8 as colorless oil (8.2 g, 95%, 98% de). [α]D²⁵ +27.5 (c 1.9, CHCl₃). ¹H NMR (300 MHz, CDCl₃): δH 4.09 (dt, 1H, J 3.2, 8.6 Hz), 3.93-3.80 (m, 3H), 3.64 (brd, 1H, J 12.4 Hz), 3.04 (dd, 1H, J 2.2, 3.9 Hz), 3.01 (dd, 1H, J 2.0, 5.8 Hz),
2.34 (brs, 1H), 1.62-1.51 (m, 8H), 1.41 (brs, 2H). 13C NMR (50 MHz, CDCl3): δC 110.5, 74.8, 66.4, 61.0, 57.2, 55.4, 36.0, 34.6, 24.9, 23.8, 23.7. IR (neat) (νmax, cm^-1): 3444, 2935, 2860, 1449, 1367, 1162, 1098. ESI-MS: m/z 237 [M+Na]^+. HRMS calculated for C11H18O4Na: 237.1102 [M+Na]^+, found: 237.1100. GC-MS data: The inlet and GC-MS interface temperatures were kept at 280 °C. Helium was used as the carrier gas at flow rate of 1 mL/min., and the sample was injected in split mode 1:10 ratio. Oven program was 80 °C for 2 min; raised temperature 10 °C/min to 280 °C; hold 5 min; a single peak was found at the retention time of 13.77 min with mass m/z 214 [M+H]^+.

(2S,3S,4R)-2-Azido-4,5-(cyclohexyldenedioxy)-pentane-1,3-diol (9). A mixture of epoxy alcohol 8 (7.92 g, 37 mmol), B(OMe)_3 (6.3 mL, 55.5 mmol), and NaN_3 (4.81 g, 74 mmol) in DMF (60 mL) under N_2 atmosphere were stirred at 50 °C for 8 h. After cooling to 0 °C, saturated NaHCO_3 (50 mL) was added, and the mixture was stirred for 30 min at the same temp. The mixture was separated, and the aq. layer was extracted with Et_2O (3x75 mL). The combined organic layers were successively washed with H_2O (25 mL), brine (45 mL), and dried (Na_2SO_4). Concentration under reduced pressure gave oily residue, was dissolved in CH_2CN (60 mL) and treated with NaIO_4 (3.96 g, 18.5 mmol) dissolved in 40 mL water at r.t. for 30 min. and filtered. The filtrate was mixed with H_2O (10 mL) and extracted with EtOAc (3x70 mL). The combined org. layers were washed with H_2O (20 mL), brine (30 mL), dried (Na_2SO_4), filtered and concentrated in vacuo. The residue was purified by column chromatography (SiO_2; EtOAc/hexane, 1:4) to afford the azido diol 9 as colorless oil (9.12 g, 96%). [α]D^25 +26.7 (c 1.1, CHCl3). 1H NMR (300 MHz, CDCl3): δH 4.18 (dd, 1H, J 6.0, 12.0 Hz), 4.09 (dd, 1H, J 6.0, 8.3 Hz), 3.96-3.91 (m, 3H), 3.83 (dd, 1H, J 5.2, 9.8 Hz) 3.61 (dd, 1H, J 4.5, 9.8 Hz), 2.77 (brd, 1H, J 4.5 Hz), 2.54 (brs, 1H), 1.60-1.55 (m, 8H), 1.41 (brs, 2H). 13C NMR (75 MHz, CDCl3): δC 110.2, 74.9, 72.5, 65.7, 63.9, 62.3, 36.2, 34.5, 24.9, 23.9, 23.6. IR (neat) (νmax, cm^-1): 3405, 2937, 2857, 2105, 1447, 1275, 1101. ESI-MS: m/z 280 [M+Na]^+. HRMS calculated for C_{11}H_{19}N_{3}O_{4}Na: 280.1273 [M+Na]^+, found: 280.1271.

(2S,3S,4R)-2-Azido-4,5-(cyclohexyldenedioxy)-1,3-(dibenzyl)pentane (10). To a well stirred soln. of NaH (60% dispersion in mineral oil, 2.4 g, 60 mmol) in dry THF (75 mL) under nitrogen was added azido diol 9 (5.14 g, 20 mmol) dissolved in THF (30 mL) via syringe very slowly at 0 °C and allowed to stir at same temp. for 20 min. TBAI (50 mg, cat.) followed by benzyl bromide (6 mL, 50 mmol) were added at 0 °C and allowed to stir at r.t. for overnight. The reaction mixture was quenched by addition of H_2O until clear soln. results. THF was removed under reduced pressure, the residue was treated with NaIO_4 (20 mL), brine (45 mL), and dried (Na_2SO_4). Concentration under reduced pressure gave oily residue, was dissolved in CH_2CN (60 mL) and treated with NaIO_4 (3.96 g, 18.5 mmol) dissolved in 40 mL water at r.t. for 30 min. and filtered. The filtrate was mixed with H_2O (10 mL) and extracted with EtOAc (3x70 mL). The combined org. layers were washed with H_2O (20 mL), brine (30 mL), dried (Na_2SO_4), filtered and concentrated in vacuo. The residue was purified by column chromatography (SiO_2; EtOAc/hexane, 1:4) to afford the azido diol 9 as colorless oil (8.04 g, 92%). [α]D^25 +26.4 (c 1.0, CHCl3). 1H NMR (300 MHz, CDCl3): δH 7.30-7.23 (m, 10H), 4.69 (dd, 2H, J 11.3, 22.3 Hz), 4.57 (dd, 2H, J 12.0, 13.9 Hz), 4.15 (dd, 1H, J 6.2, 12.6 Hz), 4.00 (dd, 1H, J 6.4, 8.3 Hz), 3.86-3.75 (m, 2H), 3.72-3.59 (m, 3H), 1.56-1.53 (m, 8H), 1.38 (brs, 2H). 13C NMR (50 MHz, CDCl3): δC 137.7, 137.6, 128.4, 127.9, 127.7, 127.6, 109.8, 79.3, 74.6, 73.8, 73.3, 69.6, 66.2, 62.5, 36.2, 34.7, 25.1, 23.6, 23.5, 20.9, 20.8, 14.1, 13.6.
To a cooled (0 °C) solution of compound 10 (7 g, 16 mmol) in CH$_3$CN (85 mL) was added 10% HCl (85 mL) and allowed to stir at r.t. for 4 h. The reaction was quenched with solid NaHCO$_3$ until neutralized at r.t. The CH$_3$CN was removed under vacuum, then it was diluted with EtOAc (70 mL) and after separation of the layers, the aq. layer was further extracted with EtOAc (2x30 mL). The combined organic layers were washed with brine (20 mL), dried (Na$_2$SO$_4$), filtered and concentrated. The residue was purified by column chromatography (SiO$_2$; EtOAc/hexane, 1:4) to give 11 as colorless oil (5.31 g, 93%). [α]$_D$$^{25}$ +38.8 (c 1.1, CHCl$_3$). $^1$H NMR (400 MHz, CDCl$_3$): δH 7.34–7.25 (m, 10H), 4.68 (d, 1H, J 10.9 Hz), 4.59 (d, 1H, J 10.9 Hz), 4.55 (s, 2H), 3.90 (dt, 1H, J 4.4, 6.6 Hz), 3.80–3.72 (m, 2H), 3.70–3.66 (m, 3H), 3.64 (m, 1H) 2.82 (brs, 1H), 2.00 (brs, 1H). $^{13}$C NMR (50 MHz, CDCl$_3$): δC 137.4, 137.3, 128.4, 128.5, 128.4, 128.2, 128.1, 127.9, 127.7, 79.2, 73.9, 73.5, 71.2, 69.3, 63.3, 61.9. IR (neat) (ν$_{max}$, cm$^{-1}$): 3405, 2872, 2100, 1455, 1267, 1090. ESI-MS: m/z 380 [M+Na]$^+$. HRMS calculated for C$_{19}$H$_{33}$N$_3$O$_4$Na: 380.1586 [M+Na]$^+$, found: 380.1597.

(2R,3S,4S)-1-(tert-Butyldimethylsilyloxy)-4-azido-3,5-(dibenzyloxy)-pentane-2-ol (12). To a well stirred soln. of diol 11 (3.57 g, 10 mmol) in dry CH$_2$Cl$_2$ (75 mL) was added imidazole (1.36g, 20 mmol) TBSCI (1.73 g, 11.5 mmol) and DMAP (122 mg, 10 mol%) at 0 °C. After 30 min, the reaction mixture was left to warm to r.t., and stirred for overnight. The reaction mixture was treated with saturated aq. NH$_4$Cl (20 mL) and extracted with CH$_2$Cl$_2$ (2x40 mL). The combined organic layers were washed with H$_2$O (20 mL) and brine (20 mL), dried (Na$_2$SO$_4$), filtered and concentrated in vacuo. The residue was purified by column chromatography (SiO$_2$; hexane 100%, followed by EtOAc/hexane, 1:99) to give 12 as a colorless oil (4.47 g, 95%). [α]$_D$$^{25}$ +22.9 (c 1, CHCl$_3$). $^1$H NMR (300 MHz, CDCl$_3$): δH 7.32–7.23 (m, 10H), 4.72 (d, 1H, J 11.3 Hz), 4.55 (s, 2H), 4.54 (d, 1H, J 11.3 Hz), 4.01–3.96 (m, 1H), 3.81 (dd, 1H, J 3.7, 9.8 Hz), 3.72–3.63 (m, 3H), 3.59–3.54 (m, 2H), 2.43 (brd, 1H, J 4.7 Hz), 0.89 (s, 9H), 0.06 (s, 3H), 0.05 (s, 3H). $^{13}$C NMR (50 MHz, CDCl$_3$): δC 137.8, 137.7, 128.4, 127.9, 127.8, 127.7, 127.6, 79.1, 73.8, 73.3, 70.9, 69.6, 63.6, 62.3, 25.8, 18.2, -5.4. IR (neat) (ν$_{max}$, cm$^{-1}$): 3456, 2928, 2857, 2097, 1461, 1256, 1096. ESI-MS: m/z 494 [M+Na]$^+$. HRMS calculated for C$_{25}$H$_{37}$N$_3$O$_4$SiNa: 494.2451 [M+Na]$^+$, found: 494.2465.

(2R,3S,4S)-1-(tert-Butyldimethylsilyloxy)-4-azido-2,3,5-(tribenzyloxy)-pentane (13). To a well stirred soln. of NaH (60% dispersion in mineral oil, 1.04 g, 26.1 mmol) in dry THF (75 mL) under nitrogen was added compound 12 (4.1 g, 8.7 mmol) dissolved in THF (30 mL) via syringe very slowly at 0 °C and allowed to stir for 20 min. Then it was added n-Bu$_4$NI (50 mg cat.) followed by benzyl bromide (3.1 mL, 26.1 mmol) drop wise with help of syringe and allowed to stir for overnight. The reaction mixture was quenched by addition of water until clear soln. results. THF was removed under reduced pressure, the residue was diluted with H$_2$O (40 mL) and extracted with Et$_2$O (3x35 mL). The combined organic layers was dried (Na$_2$SO$_4$), filtered and concentrated. The residue was purified by column chromatography (SiO$_2$; hexane 100%,
followed by EtOAc/hexane 1:19) to give 13 as a pale yellow oil (4.53 g, 93%). $[\alpha]_D^{25} +2.6$ (c 1.1, CHCl$_3$). $^1$H NMR (300 MHz, CDC$_3$): $\delta_H$ 7.26-7.22 (m, 15H), 4.68 (d, 1H, J 12.0 Hz), 4.59 (s, 2H), 4.53 (d, 1H, J 12.0 Hz), 4.45 (s, 2H), 3.92-3.82 (m, 2H), 3.74-3.66 (m, 2H), 3.64-3.56 (m, 3H), 0.89 (s, 9H), 0.03 (s, 6H). $^{13}$C NMR (50 MHz, CDC$_3$): $\delta_C$ 138.2, 137.9, 137.8, 128.3, 127.9, 127.8, 127.7, 127.5, 79.3, 78.2, 73.8, 73.2, 72.4, 69.9, 62.2, 60.5. IR (neat) ($v_{\text{max}}$, cm$^{-1}$): 2931, 2859, 2097, 1458, 1256, 1098. ESI-MS: $m/z$ 584 [M+Na]$^+$; HRMS calculated for C$_{23}$H$_{32}$N$_3$O$_3$SiNa: 584.2920 [M+Na]$^+$, found: 584.2912.

(2R,3S,4S)-4-Azido-2,3,5-(tribenzyloxy)-pentan-1-ol (14). To a stirred soln. of 13 (4.1 g, 7.3 mmol) in dry THF (80 mL) under nitrogen was added TBAF (1M in THF, 14.6 mL, 14.6 mmol) at 0$^\circ$. The reaction mixture was allowed to warm gradually to rt., and stirred for 4 h. THF was removed under reduced pressure, the residue was purified by column chromatography (SiO$_2$: hexane 100%, followed by EtOAc/hexane 1:9) to give 14 as a pale yellow oil (3.26 g, 96%). $[\alpha]_D^{25} +13.8$ (c 1.0, CHCl$_3$). $^1$H NMR (300 MHz, CDC$_3$): $\delta_H$ 7.34-7.24 (m, 15H), 4.65 (s, 2H), 4.60 (dd, 2H, J 11.5, 22.0 Hz), 4.48 (d, 2H, J 3.0 Hz), 3.90 (dd, 1H, J 4.9, 10.0 Hz), 3.80-3.69 (m, 3H), 3.63-3.58 (m, 3H), 1.91 (s, 1H). $^{13}$C NMR (50 MHz, CDC$_3$): $\delta_C$ 137.7, 137.6, 137.5, 128.5, 128.4, 128.3, 128.1, 127.9, 127.7, 127.6, 78.5, 78.1, 74.1, 73.3, 71.9, 69.6, 62.2, 60.5. IR (neat) ($v_{\text{max}}$, cm$^{-1}$): 3452, 3031, 2923, 2866, 2096, 1454, 1095. ESI-MS: $m/z$ 470 [M+Na]$^+$; HRMS calculated for C$_{26}$H$_{26}$N$_3$O$_4$Na: 470.2055 [M+Na]$^+$, found: 470.2043.

(2S,3S,4R,5E)-2-Azido-1,3,4-(tribenzyloxy)-octadeca-5-ene (15). To a stirred soln. of IBX (4.7 g, 16.8 mmol) in DMSO (8 mL) was added alcohol 14 (2.5 g, 5.6 mmol) dissolved in CH$_3$CN (32 mL) at rt., and stirred until completion of the reaction (24 h). Filtered the reaction mixture through pad of celite and repeatedly washed with Et$_2$O (50 mL). The combined org. filtrates were washed with ice cold water (2x25 mL) followed by saturated hypo soln. (50 mL), dried and concentrated under reduced pressure to give corresponding aldehyde as a colourless viscous oil (2.36 g, 95%), that was used without purification for further step.

$n$-BuLi (1.6M in hexane, 13 mL, 21.8 mmol) was added dropwise via syringe to a stirred soln. of (1-tridecyl)-triphenylphosphonium bromide (11.06 g, 21.1 mmol) in dry THF (150 mL) at -78 $^\circ$C. After 30 min, hexane (100 mL) was added, followed by the dropwise addition of a soln. of above freshly prepared aldehyde (2.36 g, 5.28 mmol) in THF (50 mL) via syringe. The reaction mixture was stirred at -78 $^\circ$C for 2 h before the addition of MeOH (50 mL). The reaction mixture was allowed to warm to rt. over further 12 h and quenched with saturated aq. NH$_4$Cl (100 mL). Brine soln. (50 mL) was added, the organic layer was separated and the aq. layer was extracted with Et$_2$O (3x75 mL). The combined organic extracts were dried (Na$_2$SO$_4$), filtered and concentrated. The residue was purified by column chromatography (SiO$_2$: hexane 100%, followed by EtOAc/hexane 1:99) to give 15 as a colourless oil (2.61 g, 81%). $[\alpha]_D^{25} -23.9$ (c 0.45, CHCl$_3$). $^1$H NMR (300 MHz, CDC$_3$): $\delta_H$ 7.32-7.25 (m, 15H), 5.79 (dt, 1H, J 6.7, 15.4 Hz), 5.51 (dd, 1H, J 8.5, 15.3 Hz), 4.78 (dd, 1H, J 3.7, 11.1 Hz), 4.62 (dd, 2H, J 5.8, 11.8 Hz), 4.51 (d, 2H, J 4.5 Hz), 3.12 (dd, 1H, J 2.4, 11.8 Hz), 3.06 (dd, 1H, J 3.7, 8.3 Hz), 2.77-3.63 (m, 4H),
2.13 (dt, 2H, J 6.9, 13.5 Hz), 1.41-1.25 (m, 20H), 0.89 (t, 3H, J 6.7 Hz). 13C NMR (75 MHz, CDCl3): δC 138.4, 138.1, 137.9, 137.8, 136.8, 128.4, 128.3, 128.2, 128.0, 127.9, 127.8, 127.6, 127.5, 127.4, 126.1, 125.7, 80.7, 74.4, 73.2, 69.9, 69.8, 69.7, 61.9, 32.5, 31.9, 29.6, 29.4, 29.3, 29.2, 29.1, 28.0, 22.7, 14.1. IR (neat) (vmax, cm⁻¹): 2924, 2854, 2098, 1456, 109. ESI-MS: m/z 634 [M+Na]⁺. HRMS calculated for C30H53N5O3Na: 634.3984 [M+Na]⁺, found: 634.3971.

N,O,O,O-Tetraacetyl-D-ribo-phytosphingosine ((2S,3S,4R)-2-acetamino-1,3,4-(triacetoxy)-octadecane) (1a). A mixture of olefin 15 (1.22 g, 2 mmol) and Pd(OH)2/C (20% content, 30% wt/wt), 0.366 g) in EtOAc (40 mL) containing trace amount of HCl (5 µl) was stirred for 48 h at r.t. under H2 atmosphere (balloons). The catalyst was filtered through pad of celite, repeatedly washed with EtOAc (20 mL) and filtrate was concentrated in vacuo to give crude phytosphingosine. This crude product was dissolved in pyridine (15 mL), DMAP (0.1 g, cat.) and acetic anhydride (3 mL, excess) were added under N2 atmosphere (balloons). The solvent was removed under reduced pressure, saturated aq. NaHCO3 (10 mL) was added to the residue and the mixture was extracted with Et2O (3×30 mL). The combined organic layers were sequentially washed with saturated aq. CuSO4 (2×15 mL), H2O (10 mL) and brine (10 mL), dried (Na2SO4), filtered and concentrated in vacuo and the residue was purified by column chromatography (SiO2; EtOAc/hexane 1:7) to afford compound 1a as a semi solid (0.8 g, 80%). [α]D +19.1 (c 1.0, CHCl3). 1H NMR (400 MHz, CDCl3): δH 5.98 (d, 1H, J 9.3 Hz), 5.05 (dd, 1H, J 3.1, 8.5 Hz), 4.88 (td, 1H, J 3.8, 7.0 Hz), 4.33-4.37 (m, 1H), 4.28 (dd, 1H, J 5.4, 11.6 Hz), 3.95 (dd, 1H, J 3.1, 11.6 Hz), 2.01 (s, 3H), 1.97 (s, 6H), 1.95 (s, 3H), 1.59-1.52 (m, 2H), 1.24-1.18 (m, 24H), 0.82 (t, 3H, J 7.0 Hz). 13C NMR (75 MHz, CDCl3): δC 171.1, 170.8, 170.1, 169.7, 72.9, 71.8, 62.8, 47.5, 31.8, 31.3, 30.1, 29.6, 29.4, 29.3, 29.2, 28.0, 25.4, 23.2, 22.6, 20.9, 20.6, 20.7, 14.0. IR (neat) (vmax, cm⁻¹): 2925, 2854, 1745, 1660, 1371, 1279. ESI-MS: m/z 508 [M+Na]⁺. HRMS calculated for C50H87NO7Na: 508.3250 [M+Na]⁺, found: 508.3274.

(2S,3R,4E)-2-Azido-1,3-(dibenzzyloxy)-octadec-4-ene (16). To a stirred soln. of 11 (2.5 g, 7 mmol) in 30 mL of CH2CN at r.t. was added NaIO4 (3 g, 14 mmol) dissolved in H2O 20 mL over a period of 10 min. The mixture was stirred about 30 min, filtered through pad of celite and repeatedly washed with Et2O. The filtrate was mixed with H2O (20 mL) and extracted with EtOAc (3×50 mL). The combined organic layers were washed with H2O (20 mL), brine soln. (25 mL) and dried (Na2SO4). Solvent removal under reduced pressure afforded the corresponding aldehyde in almost quantitative yield (2.2 g, 98%). This was sufficiently pure and hence used as such for the next step. [α]D -16.4 (c 1.0, CHCl3). 1H NMR (300 MHz, CDCl3): δH 9.56 (d, 1H, J 1.5 Hz), 7.32-7.21 (m, 10H), 4.72 (d, 1H, J 11.3 Hz), 4.65 (d, 1H, J 12.1 Hz), 4.49 (s, 2H), 3.91 (dd, 1H, J 1.5, 3.7 Hz), 3.87 (dt, 1H, J 3.7, 6.0 Hz), 3.71 (dd, 1H, J 6.7, 9.8 Hz), 3.63 (dd, 1H, J 6.0, 9.8 Hz). 13C NMR (75 MHz, CDCl3): δC 200.7, 137.2, 128.5, 128.4, 128.3, 128.1, 127.8, 127.7, 127.6, 82.5, 73.4, 67.6, 61.4. IR (neat) (vmax, cm⁻¹): 2923, 2867, 2101, 1731, 1454, 1267, 1100. ESI-MS: m/z 380 [M+Na]⁺.

n-BuLi (1.6M in hexanes, 16.9 mL, 27 mmol) was added dropwise via syringe to a stirred soln. of (1-tetradecyl)triphenylphosphonium bromide (14.7 g, 27 mmol) in dry THF (150 mL) at -78 °C. After 30 min, hexane (150 mL) was added, followed by the dropwise addition of a soln. of
above freshly prepared aldehyde (2.2 g, 6.76 mmol) in THF (30 mL) via syringe. The reaction mixture was stirred at -78 °C for 2 h before the addition of MeOH (50 mL). The reaction mixture was allowed to warm to r.t. over further 12 h and quenched with saturated aq. NH₄Cl (75 mL).

Brine (50 mL) was added, the organic layer was separated and the aq. layer was extracted with Et₂O (3x50 mL). The combined org. extracts were dried (Na₂SO₄), filtered and concentrated. The residue was purified by column chromatography (SiO₂; hexane 100%, followed by EtOAc/hexane 1:9) to give 16 as a pale yellow oil (2.9 g, 85%, mixture of E and Z isomers).

[α]D²⁵ -33.7 (c 1.3, CHCl₃). ¹H NMR (300 MHz, CDCl₃): δH 7.30-7.23 (m, 10H), 5.72 (dt, 1H, J 7.2, 15.6 Hz), 5.41 (dd, 1H, J 8.3, 15.6 Hz), 4.58 (d, 1H, J 12.2 Hz), 4.54 (d, 1H, J 12.2 Hz), 4.50 (d, 1H, J 11.3 Hz) 4.32 (d, 1H, J 11.3 Hz), 3.88 (dd, 1H, J 5.2, 8.3 Hz), 3.61-3.53 (m, 3H), 2.10 (dd, 2H, J 7.2, 14.5 Hz), 1.41-1.25 (m, 22H), 0.89 (t, 3H, J 6.2 Hz). ¹³C NMR (75 MHz, CDCl₃): δc 138.1, 137.9, 137.8, 128.3, 128.2, 127.7, 127.6, 127.5, 125.9, 79.4, 73.3, 69.9, 69.4, 64.4, 32.3, 31.9, 29.6, 29.4, 29.3, 29.1, 29.0, 27.9, 22.6, 14.1. IR (neat) (νmax cm⁻¹): 2924, 2854, 2852, 1509, 1457, 1098. ESI-MS: m/z 528 [M+Na]+. HRMS calculated for C₃₂H₄₇N₃O₂Na: 528.3565 [M+Na]+, found: 528.3548.

(2S,3R,4E)-2-Amino-1,3-(dibenzyloxy)-octadeca-4-ene (17). A mixture of olefin 16 (1.01 g, 2 mmol) and Lindlar’s catalyst (Pd/CaCO₃, 20% content, 35% (wt/wt), 0.353 g) in EtOAc (30 mL) was stirred for 8 h at r.t. under H₂ atmosphere (balloons). The catalyst was filtered through pad of celite, repeatedly washed with EtOAc (20 mL) and filtrate was concentrated in vacuo to give crude amino compound which was purified by column chromatography (SiO₂; EtOAc/hexane 1:9) to give 17 as colourless oil (0.795 g, 83%). [α]D²⁵ -28.4 (c 0.68, CHCl₃). ¹H NMR (400 MHz, CDCl₃): δH 7.28-7.21 (m, 10H), 5.70 (dt, 1H, J 6.8, 15.6 Hz), 5.38 (dd, 1H, J 7.8, 15.6 Hz), 4.54 (d, 1H, J 11.7 Hz), 4.47 (s, 2H), 4.28 (d, 1H, J 11.7 Hz), 3.69 (t, 1H, J 7.8 Hz), 3.56 (dd, 1H, J 3.9, 8.7 Hz), 3.46 (dd, 1H, J 7.8, 15.6 Hz), 3.03 (q-like, 1H, J 3.9 Hz), 2.11 (dd, 2H, J 6.8, 13.6 Hz), 1.39-1.25 (m, 22H), 0.89 (t, 3H, J 6.8 Hz). ¹³C NMR (75 MHz, CDCl₃): δc 138.4, 138.1, 137.9, 128.3, 128.2, 127.7, 127.6, 127.4, 126.5, 80.6, 73.2, 70.5, 70.0, 54.4, 32.4, 31.9, 29.7, 29.5, 29.3, 29.2, 29.1, 28.0, 22.7, 14.1. IR (neat) (νmax cm⁻¹): 3386, 3062, 3030, 2924, 2855, 1457, 1093. ESI-MS: m/z 480 [M+H]+. HRMS calculated for C₃₂H₅₀NO₂: 480.3841 [M+H]+, found: 480.3865.

D-erythro-Sphingosine ((2S,3R,4E)-2-amino-octadec-4-ene-1,3-diol (2). A soln. of compound 17 (410 mg, 0.860 mmol) in dry THF (10 mL) was added dropwise to a soln. of Na (198 mg, 8.60 mmol) in dry liq. NH₃ (ca. 40 mL) at -78 °C. After 45 min, solid NH₄Cl was added, and the mixture was allowed to warm to r.t. (ammonia evaporated). The resulting solid residue was stirred with EtOAc (30 mL), filtered and washed with EtOAc (25 mL). The combined organic phases were dried (Na₂SO₄), and concentrated. The residue was purified by column chromatography (SiO₂; MeOH/ EtOAc, 1:9) gave compound 2 as a white waxy compound (205 mg, 80%). m.p. 72-74 °C, [α]D²⁵ -3.1 (c 1.0, CHCl₃), [lit.]²⁰ mp 73-75 °C, [α]D²⁵ -2.86 (c 1.1, CHCl₃)] and [lit.]²⁰ mp 76-77 °C, [α]D²⁴ -3.0 (c 1.0, CHCl₃)]. ¹H NMR (300 MHz, CDCl₃): δH 6.54 (brd, 1H, J 7.5 Hz), 6.51 (brd, 1H, J 7.5 Hz), 5.83 (dt, 1H, J 6.6, 14.3 Hz), 5.56 (dd, 1H, J 6.2, 15.1 Hz), 4.31 (brs, 1H), 3.97-3.68 (m, 4H), 3.00 (brs, 1H), 2.06-1.98 (m, 2H), 1.36-1.10 (m,
22H), 0.88 (t, 3H, J 6.9 Hz). $^{13}$C NMR (50 MHz, CDCl$_3$): $\delta$C 134.1, 128.6, 74.3, 62.2, 54.5, 32.2, 31.8, 30.68, 29.6, 29.4, 29.3, 29.2, 27.8, 23.3, 22.6, 14.1. IR (KBr) ($v_{\text{max}}$, cm$^{-1}$): 3292, 2919, 2850, 1649, 1552, 1464, 1050, 722. ESI-MS: $m/z$ 300 [M+H]$^+$

**N,O,O-Triacetyl D-erythro-Sphingosine ((2S,3R,4E)-1,3-diacetoxy-2-acetamido-octadec-4-ene (2a).** The compound 2 (100 mg, 0.33 mmol) was dissolved in pyridine (6 mL) and Ac$_2$O (0.5 mL) and DMAP (3 mg, cat.) were added sequentially. The reaction mixture was stirred overnight and the solvent removed under reduced pressure. Et$_2$O (30 mL) was added to the residue and washed sequentially with saturated aq. CuSO$_4$ (2x5 mL), H$_2$O (5 mL) and brine (5 mL), dried (Na$_2$SO$_4$) and concentrated. The residue was purified by column chromatography (SiO$_2$; EtOAc/hexane, 1:6) to afford pure compound 2a as a white solid (120 mg, 85%). m.p. 97-99 °C; $[\alpha]_D$ $^{25}$ +11.8 (c 1.0, CHCl$_3$); [{lit.$^{9a}$} mp 99-101 °C, $[\alpha]_D^{20}$ -12.0 (c 1.0, CHCl$_3$)] and [lit.$^{9c}$ mp 99-101 °C, $[\alpha]_D^{20}$ -12.1 (c 1.0, CHCl$_3$)]. $^1$H NMR (300 MHz, CDCl$_3$): $\delta$H 5.84 (dt, 1H, J 6.6, 15.3 Hz), 5.66 (d, 1H, J 8.8 Hz), 5.41 (dd, 1H, J 7.3, 15.3 Hz), 5.29 (t, 1H, J 6.6 Hz), 4.46-4.40 (m, 1H), 4.32 (dd, 1H, J 11.7, 5.9 Hz), 4.06 (dd, 1H, J 3.6, 11.7 Hz), 2.07 (s, 3H), 2.06 (s, 3H), 2.04-1.99 (m, 2H), 1.98 (s, 3H), 1.34-1.25 (m, 22H), 0.89 (t, 3H, J 5.8 Hz). $^{13}$C NMR (50 MHz, CDCl$_3$): $\delta$C 171.0, 170.0, 169.6, 137.5, 124.1, 73.8, 62.5, 50.6, 32.2, 31.9, 29.7, 29.6, 29.4, 29.3, 29.1, 28.9, 23.3, 22.6, 21.1, 20.7, 14.1. IR (KBr) ($v_{\text{max}}$, cm$^{-1}$): 3289, 2919, 2852, 1736, 1654, 1549, 1230. ESI-MS: $m/z$ 448 [M+Na]$^+$; HRMS calculated for C$_{24}$H$_{43}$NO$_5$Na: 448.3038 [M+Na]$^+$, found: 448.3034.

**Sphinganine ((2S,3R)-2-amino-octadecane-1,3-diol (3).** A mixture of olefin 16 (252 mg, 0.5 mmol) and Pd(OH)$_2$/C (20% content, 30 mg) in EtOAc:MeOH (1:1, 30 mL) containing trace amount of HCl (5 µl) was stirred for 48 h at r.t. under H$_2$ atmosphere (balloons). The catalyst was filtered through pad of celite, repeatedly washed with aq. MeOH and filtrate was concentrated in vacuo to give the solid compound which on recrystallization from CH$_2$CN afforded compound 3 as a white solid (130 mg, 87%). m.p. 70-73 °C; $[\alpha]_D$ $^{25}$ +4.5 (c 0.5, MeOH), [{lit.$^{21a}$} $[\alpha]_D^{25}$ +8.1 (c 1.0, MeOH) and [lit.$^{21b}$] mp 71-73 °C, $[\alpha]_D^{25}$ +1.83 (c 1.0, pyridine). $^1$H NMR (400 MHz, CDCl$_3$+DMSO-d$_6$): $\delta$H 8.10 (brs, 2H), 4.86 (brs, 1H), 3.87 (brs, 1H), 3.78 (brs, 2H), 3.12 (brs, 1H), 1.48-1.25 (m, 28H), 0.89 (t, 3H, J 7.3 Hz). $^{13}$C NMR (75 MHz, CDCl$_3$+CD$_3$OD): $\delta$C 70.2, 58.6, 57.7, 34.0, 32.8, 30.5, 30.4, 30.2, 26.8, 23.5, 14.7. IR (KBr) ($v_{\text{max}}$, cm$^{-1}$): 3387, 2918, 2849, 1599, 1507, 1466, 1058. ESI-MS: $m/z$ 302 [M+H]$^+$; HRMS calculated for C$_{18}$H$_{34}$NO$_5$: 302.3059 [M+H]$^+$, found: 302.3060.

**N,O,O-Triacetyl sphinganine ((2S,3R)-1,3-diacetoxy-2-acetamido-octadecane (3a).** The compound 3 (100 mg, 33.2 mmol) was dissolved in pyridine (6 mL) and Ac$_2$O (0.5 mL) and DMAP (3 mg, cat.) were added sequentially. The reaction mixture was stirred overnight and removed the solvent under vacuo. Et$_2$O (30 mL) was added to the residue and washed sequentially with saturated aq. CuSO$_4$ (2x5 mL), water (5 mL) and brine (5 mL), dried (Na$_2$SO$_4$) and concentrated. The residue was purified by column chromatography (SiO$_2$; EtOAc/hexane, 1:6) to afford pure compound 3a as a white solid (114 mg, 81%). m.p. 92-95 °C, $[\alpha]_D$ $^{25}$ +14.6 °C (c 0.5, CHCl$_3$) [{lit.$^{21c}$} mp 90-93 °C, $[\alpha]_D^{19}$ +16.0 (c 0.5, CHCl$_3$) and lit.$^{21d}$ $[\alpha]_D^{27.5}$ +13.8 (c 1.0, CHCl$_3$). $^1$H NMR (300 MHz, CDCl$_3$): $\delta$H 5.90 (brs, 1H), 4.85 (dd, 1H, J 5.8, 12.6 Hz), 4.34-4.31
General procedure for the synthesis of tridecyl and tetradecyl triphenyl phosphonium bromides, 1-Bromotridecane or 1-bromotetradecane (30 mmol) and triphenyl phosphine (30 mmol) were refluxed in 125 mL dry CH$_3$CN for overnight. The solvent was removed under reduced pressure and the residue was repeatedly washed with hexane or diethyl ether until the compound precipitate out. Further, it was recrystallized with benzene, ether (1:1) mixture to give white solid.

Data of n-tridecyl triphenyl phosphonium bromide$^{22a}$: (13.05 g, 83%); mp 83-87 ºC. $^1$H NMR (300 MHz, CDCl$_3$): $\delta$H 7.96-7.64 (m, 15H), 4.02-3.93 (m, 2H), 1.60 (brs, 4H), 1.22 (brd, 18H), 0.89 (t, 3H, $J$ 6.7 Hz); ESI-MS: $m/z$ 445 [M-Br]$^+$.

Data of n-tetradecyl triphenyl phosphonium bromide$^{22b}$: (14.2 g, 88%). mp 82-85 ºC. $^1$H NMR (300 MHz, CDCl$_3$): $\delta$H 7.96-7.67 (m, 15H), 4.01-3.96 (m, 2H), 1.60 (brs, 4H), 1.28-1.17 (m, 20H), 0.89 (t, 3H, $J$ 6.7 Hz); ESI-MS: $m/z$ 459 [M-Br]$^+$.

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References


