Total Synthesis of a Natural Cerebroside from Euphorbiaceae

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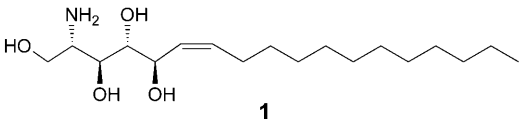
The regioselective total synthesis of the natural cerebroside (2*R*,15*Z*)-*N*-{(1*S*,2*S*,3*R*,4*R*,5*Z*)-1-[(β-D-glucopyranosyloxy)methyl]-2,3,4-trihydroxyheptadec-5-en-1-yl}-2-hydroxytetracos-15-enamide (**2**), originally isolated from Euphorbiaceae, is reported in full detail.

Introduction. – Lipids and sphingoglycolipids play significant roles in numerous biological processes. They have been considered to be present at the outer layer of biological cell membranes and to take part in antigen–antibody reactions and in the communication between cells [1].

In our previous work [2], we described the isolation, identification, and biological evaluation of several cerebrosides from Euphorbiaceae. Complex sphingoglycolipids, in which a tetrahydroxy 'long-chain base' (LCB) occurs as part of the cerebroside moiety, were isolated from *Euphorbia characias* L. [3] and *E. wulfenii* HOPPE ex KOCH [4], and characterized by physical and chemical methods. Unfortunately, sphingoglycolipids are usually available from natural sources in only limited quantities as often hardly separable mixtures, especially in terms of their fatty acid compositions. Hence, a versatile synthesis of these important natural products is of importance, especially for biochemical investigations.

The C_{18} sphingosine (2*S*,3*S*,4*R*,5*R*,6*Z*)-2-aminooctadec-6-ene-1,3,4,5-tetrol (C_{18}-sphingoside; **1**) present in the natural cerebroside **2**, has been isolated from the latex of Euphorbiaceae. Various syntheses of **1** have been described, either starting from D-mannose [5], D-glutamic acid [6], or, more recently, from D-allosamines [7]. Herein, we report the total synthesis of the more-complex cerebroside **2** from Euphorbiaceae to confirm the structure of the natural isolate and to obtain higher quantities for biological studies.

Results and Discussion. – A retrosynthetic analysis is outlined in *Scheme 1*. Bond disconnection of **2** leads to D-glucose (D-Glc), C_{18}-sphingosine (**1**), and (15*Z*)-2-hydroxytetracos-15-enoic acid (**3**) as building blocks. Whereas D-Glc is a readily available natural product, a suitably protected derivative of **1** had to be prepared from L-serine (L-Ser) *via* the oxazolidine **4** [8], to which 2-(trimethylsilyl)-1,3-thiazole (**5**) was added according to *Dondoni et al.* [9]. Since the way to connect the three building blocks, Glc, **1**, and **3**, is somewhat problematic, this was performed in a convergent manner, *i.e.*, (**1**+**3**)+Glc → **2**.



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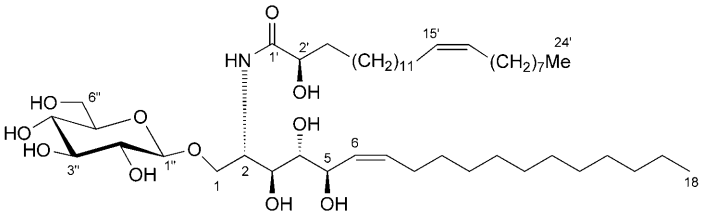

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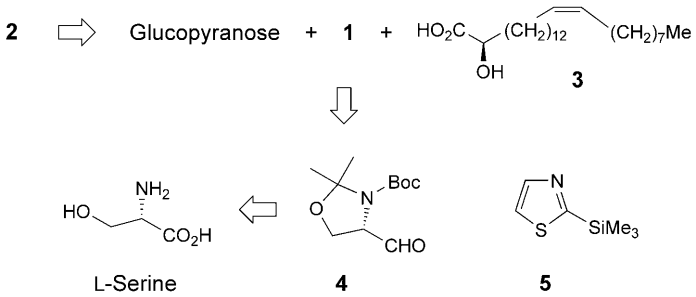
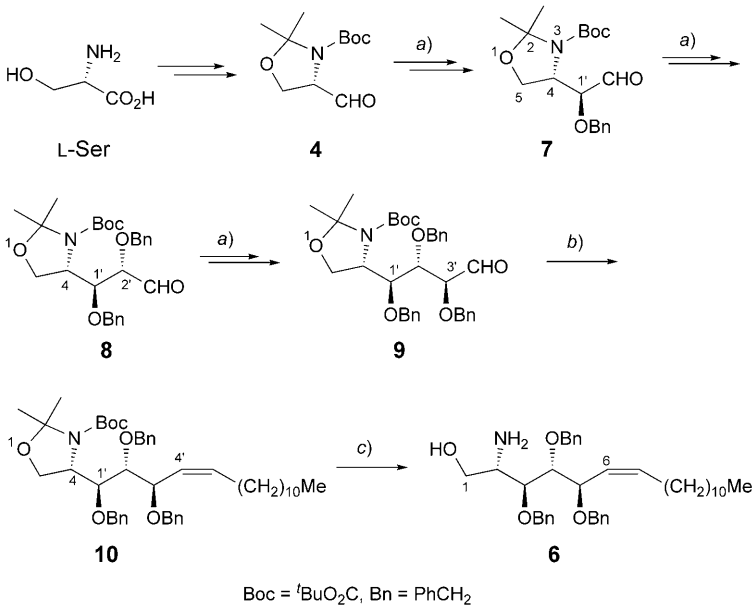
Scheme 1



First, the C_{18}-sphingosine derivative **6** was prepared according to [9]. Thus, the oxazolidinecarbaldehyde **4** was treated with **5**, which afforded, after desilylation of the initial adduct, the corresponding α-hydroxyalkylthiazole with a high level of diastereoselectivity. The *anti*-configured alcohol was separated from the *syn* adduct, and then subjected to the following aldehyde-releasing four-step sequence: *1*) OH protection as benzyl (Bn) ether, which gave essentially the *anti*-isomer; *2*) *N*-methylation of the thiazole ring with MeI affording the *N*-methylthiazolium salt; *3*) reduction with $NaBH_4$ to the corresponding thiazolidine; and 4) Hg-mediated hydrolysis to the aldehyde **7** in high yield [9]. Hence, **5** is a formyl-anion synthon adding to the aldehyde in a stereoselective manner, creating a new stereogenic center. Thus, repetition of the above processes, *i.e.*, stereoselective addition of **5** followed by aldehyde release, over three consecutive cycles, each proceeding with high stereo-induction and chemical yield, finally provided compounds **8** [9] and **9**.

Next, the aldehyde **9** was subjected to *Wittig* reaction using dodecylidenetriphenylphosphorane generated *in situ* from the appropriate phosphonium salt and BuLi in the presence of 'hexamethylphosphoramide' (HMPA) so as to ensure *cis* selectivity [10]. This reaction led exclusively to the (*Z*)-alkene **10**, which was isolated in relatively modest overall yield (35%) after flash chromatography. The (*Z*)-configuration of the newly

Scheme 2



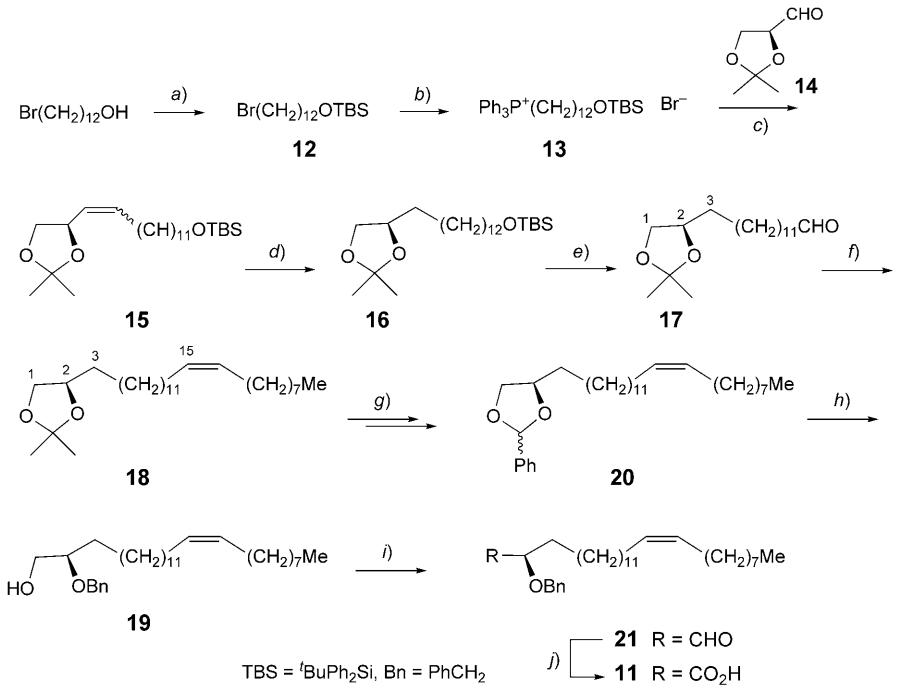
a) 1. **5**, CH_2Cl_2; 2. BnBr, NaH; 3. MeI, $NaBH_4$, Hg^{2+}, H_2O. *b*) $Me(CH_2)_{11}(Ph_3)P^+Br^-$, BuLi, HMPA, $-78°$. *c*) TFA, H_2O.

generated C=C bond of **10** was verified by NMR spectroscopy. Thus, two distinct downfield signals were observed in the ^{1}H-NMR spectrum at δ(H) 5.70 (*dd*, $J=10.3$, 8.4 Hz, H–C(5′)) and 5.59 (*dd*, $J=10.4$, 9.0 Hz, H–C(4′)); and in the ^{13}C-NMR spectrum, a typical chemical shift of δ(C) 27.7 was found for the CH_2 group adjacent to the olefinic C(6′), which is expected to resonate at δ(C) *ca.* 33.0 in the (*E*)-configured long-chain isomer. Finally, one-pot deprotection of **10** with TFA/H_2O 9:1 afforded the target tris(*O*-benzyl)-protected C_{18}-sphingosine derivative **6**.

The next step was the synthesis of the Bn-protected 2-hydroxytetracos-15-enoic acid **11** (*Scheme 3*). Treatment of 12-bromododecan-1-ol with (*tert*-butyl)diphenylsilyl chloride (TBSCl) gave **12**. The latter was heated with Ph_3P at 120° [11] to yield the corresponding phosphonium salt **13**. After deprotonation with BuLi, the resulting phosphorane was subjected to *Wittig* reaction with the aldehyde **14**, an oxidized, acetal-protected derivative of D-mannitol [12], which afforded **15** in 60% yield as a mixture of geometric isomers. Hydrogenation with H_2/PtO_2 gave **16** in 85% yield, and silyl deprotection of the latter with Bu_4NF (TBAF), followed by oxidation of the resulting OH group with pyridinium chlorochromate (PCC), provided the aldehyde **17** in 87% yield. Next, *Wittig* reaction between **17** and nonyl(triphenyl)phosphonium bromide in the presence of BuLi at $-70°$ gave a mixture of geometric isomers, which were easily separated by flash column chromatography on silica gel. The major (*Z*)-isomer **18** (40% yield) was then converted to the benzyl ether **19** in satisfactory overall yield through the following sequence: *1*) acetal deprotection followed by re-protection with $PhCH(OMe)_2$ to **20**, and *2*) selective reduction of **20** with $(i\text{-}Bu)_2AlH$ (DIBALH), gen-

erating the mono-bezylated target compound **19**. Finally, the primary OH group of **19** was oxidized with PCC to afford the corresponding aldehyde **21**, and then further oxidized to the target synthon **11**, which was obtained in an overall yield of 59%.

Scheme 3

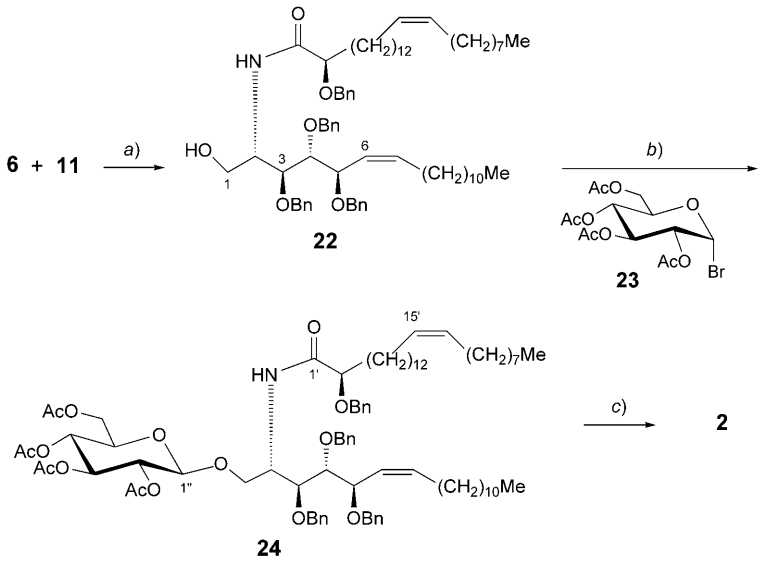


a) ^{t}Bu(Ph)$_2$SiCl (TBSCl), CH_2Cl_2. *b*) Ph_3P, 120°. *c*) BuLi, −30°. *d*) H_2/PtO_2, MeOH. *e*) 1. Bu_4NF (TBAF), THF; 2. pyridinium chlorochromate (PCC), CH_2Cl_2. *f*) $Me(CH_2)_7CH_2(Ph_3)P^+Br^-$, BuLi, THF, −70°. *g*) AcOH, H_2O, 2N H_2SO_4, $PhCH(OMe)_2$, TFA, CH_2Cl_2. *h*) DIBALH, toluene. *i*) PCC, CH_2Cl_2. *j*) ^{t}BuOH, 2-methylbut-2-ene; 2. H_2O, $NaClO_2$, NaH_2PO_4.

The next stage of the synthesis was the coupling of the building blocks **6** and **11** (*Scheme 4*). The reaction furnished the protected ceramide **22** in 50% yield upon coupling with DCC/HOBt (DCC = dicyclohexylcarbodiimide, HOBt = 1-hydroxy-1*H*-benzotriazole) in THF. β-Glucosidation of **22** with the protected Glc derivative **23** under *Koenigs–Knorr* conditions in the presence of $Hg(CN)_2$ in nitromethane afforded **24** in 50% yield [13]. Finally, removal of the Ac protecting groups upon alkaline hydrolysis, followed by debenzylation with lithium naphthalenide, afforded the target cerebroside **2** in 85% yield [14].

The structure of compound **2** was secured by EI-MS, FAB-MS, ^{1}H-NMR, ^{13}C-NMR, and ^{1}H,^{13}C-COSY experiments (see *Exper. Part*). Also, the high-field NMR data of the synthetic material were found to be identical with those reported for the compound isolated from natural sources [3][4].

Scheme 4



a) HOBT, DCC, THF. *b*) Benzene, $Hg(CN)_2$. *c*) 1. 5% KOH in MeOH; 2. Li naphthalenide, THF, $-25°$.

Experimental Part

General. All solvents were distilled before use, and all reactions were carried out under N_2 atmosphere. Column chromatography (CC): *Kieselgel 60* (70–230 mesh; *Merck*). Thin-layer chromatography (TLC): pre-coated *Kieselgel 60* F_{254} plates (*Merck*). Melting points (m.p.): *Büchi-510* micro-melting-point apparatus; uncorrected. FT-IR Spectra: *Jasco IR-700* spectrometer, in cm^{-1}. ^1H- and ^{13}C-NMR Spectra: *Varian Gemini-200* and *Unity-400* spectrometers, in $CDCl_3$ or C_5D_5N soln.; δ in ppm rel. to Me_4Si, *J* in Hz. EI-MS (4 kV, 70 eV) and FAB-MS (8 kV, Xe): *Kratos MS-80-RFA* apparatus; FAB-MS in MeOH soln. with glycerol/NaCl matrix; all values in *m*/*z* (rel. %).

tert-*Butyl (4*S*)-4-[(1*S*)-1-(Benzyloxy)-2-oxoethyl]-2,2-dimethyl-1,3-oxazolidine-3-carboxylate* (**7**). Prepared according to [15], and purified by CC (SiO_2; hexane/AcOEt 7:3). Yield: 1.6 g (85%). Colorless powder. M.p. 170–171°. IR (KBr): 1735, 1700, 1170. ^1H-NMR ($CDCl_3$)[1]): 1.46 (*s*, Me); 1.51 (br. *s*, ^{t}Bu); 1.60 (*s*, Me); 3.40 (*m*, H–C(1′)); 3.80–4.20 (*m*, CH_2(5), H–C(4)); 4.40–4.80 (*m*, PhCH_2); 7.30 (*m*, 5 arom. H); 9.60 (*d*, CHO). ^{13}C-NMR ($CDCl_3$): 22.2–28.1 (Me); 58.1 (C(4)); 65.0 (C(5)); 65.5 (*C*Me_3); 73.7 (Ph*C*H_2); 82.3 (BnO*C*H); 123.0–128.0 (arom. C); 137.0; 173.0 (O=C–O); 200.2 (CHO). EI-MS: 349 (15, M^+), 320 (100, ([$M-29$]$^+$), 247 (71).

tert-*Butyl (4*S*)-4-[(1*S*,2*S*)-1,2-Bis(benzyloxy)-3-oxopropyl]-2,2-dimethyl-1,3-oxazolidine-3-carboxylate* (**8**). Prepared according to [15] from **7** (0.9 g, 2.57 mmol) and **5** (0.2 g, 1.5 mmol). Repetition of the usual functionalization–unmasking sequence afforded **8** (*ribo* configuration) in 85% yield, together with 15% of the *arabino*-configured isomer. IR (film): 1739, 1700, 1665, 1170. ^1H-NMR ($CDCl_3$): 1.45 (*s*, Me); 1.50 (br. *s*, ^{t}Bu); 1.61 (*s*, Me); 3.42–3.60 (*m*, H–C(1′), H–C(2′)); 3.82–4.22 (*m*, CH_2(5), H–C(4)); 4.45–4.89 (*m*, 2 Ph*C*H_2); 7.30 (*m*, 10 arom. H); 9.75 (br. *s*, CHO). ^{13}C-NMR ($CDCl_3$): 26.2–29.3 (Me); 58.3 (C(4)); 64.7 (C(5)); 65.6 (Me_3*C*); 72.9, 73.9 (2 Ph*C*H_2); 79.3 (BnO*C*H); 80.7, 84.2 (BnO*C*H); 123.0–128.0 (arom. C); 137.2; 173.1 (O=C–O); 201.2 (CHO). EI-MS: 469 (20, M^+), 440 (100, [$M-29$]$^+$).

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[1]) Arbitrary atom numbering for all compounds (see *Schemes 2–4*).

tert-*Butyl (4*S*)-2,2-Dimethyl-4-[(1*S*,2*S*,3*S*)-1,2,3-tris(benzyloxy)-4-oxobutyl]-1,3-oxazolidine-3-carboxylate* (**9**). Repetition of the usual functionalization–unmasking sequence according to [15] with **8** afforded **9** (0.5 g, 78%) as an oil. IR (film): 1739, 1700, 1660, 1175. ^{1}H-NMR ($CDCl_3$): 1.46 (*s*, Me); 1.55 (br. *s*, ^{t}Bu); 1.65 (*s*, Me); 3.45–3.70 (*m*, H–C(1′), H–C(2′), H–C(3′)); 3.83–4.50 (*m*, H_2(5), H–C(4)); 4.60–4.80 (*m*, 3 PhCH_2); 7.32–7.40 (*m*, 15 arom. H); 9.35 (*s*, CHO). ^{13}C-NMR ($CDCl_3$): 26.0–29.5 (Me); 58.0 (C(4)); 63.7 (C(5)); 65.9 (Me_3C); 72.2, 73.3 (2 Ph*C*H_2); 74.6; 75.6 (BnO*C*H); 80.2 (Ph*C*H_2); 82.2, 82.4 (2 BnO*C*H); 127.0–128.0 (arom. C), 137.0; 176.1 (O=C–O); 201.0 (CHO). EI-MS: 589 (15, M^+), 560 (100, ([$M-29$]$^+$), 487 (70).

*(4*S*)-2,2-Dimethyl-4-[(1*S*,2*R*,3*R*,4*Z*)-1,2,3-tris(benzyloxy)hexadec-4-en-1-yl]-1,3-oxazolidine* (**10**). To a suspension of dodecyl(triphenyl)phosphonium bromide (0.43 g, 0.85 mmol) in anh. THF (20 ml) and HMPA (1.5 ml) at $-78°$ was added under N_2 a 1.6M soln. of BuLi in hexane (0.6 ml, 0.90 mmol). The mixture was stirred at this temp. for 1 h. Then, a soln. of **9** (0.5 g, 0.85 mmol) in THF (10 ml) was added dropwise under stirring at $-78°$, and the resulting mixture was stirred at this temp. for 1 h. Then, the temp. was allowed to rise to $-10°$ within 2 h, the mixture was quenched by addition of brine (10 ml), and extracted with Et_2O (3×20 ml). The combined org. extracts were dried (Na_2SO_4) and concentrated *in vacuo*. The residue was purified by CC (SiO_2; hexane/AcOEt 4:1) to yield **10** (0.22 g, 35%) as an oil. A small amount of the corresponding (*E*)-isomer was obtained by further elution. IR (film): 2970, 1700, 1630. ^{1}H-NMR ($CDCl_3$): 0.87 (*t*, *J* = 6.0, *Me*(CH_2)$_{10}$); 1.23 (br. *s*, 9 CH_2); 1.46 (*s*, Me); 1.57 (br. *s*, ^{t}Bu); 1.65 (*s*, Me); 2.30 (*m*, CH_2–CH=); 3.50–3.70 (*m*, H–C(1′), H–C(2′), H–C(3′)); 3.80–4.55 (*m*, CH_2(5), H–C(4)); 4.63–4.79 (*m*, 3 PhCH_2); 5.59 (*dd*, *J* = 10.4, 9.0, H–C(4′)); 5.70 (*dd*, *J* = 10.3, 8.4, H–C((5′)); 7.30–7.40 (*m*, 15 arom. H). ^{13}C-NMR ($CDCl_3$): 14.3 (*Me*(CH_2)$_{10}$); 22.0–26.1 (Me); 27.7 (C(6′)); 28.1–30.0 (CH_2); 58.3 (C(4)); 63.8 (C(5)); 66.0 (Me_3C); 70.4, 73.3 (2 Ph*C*H_2); 73.8; 74.8 (BnO*C*H); 80.1 (Ph*C*H_2); 81.7, 82.1 (2 BnO*C*H); 123.6 (C(5′)); 127.0–128.0 (arom. C); 136.0 (C(4′)); 137.0; 175.6 (O=C–O). EI-MS: 725 (22, M^+), 584 (60, [$M-C_{10}H_{21}$]$^+$).

*(2*S*,3*S*,4*R*,5*R*,6*Z*)-6-Octadecasphingenine* (= *(2*S*,3*S*,4*R*,5*R*,6*Z*)-2-Amino-3,4,5-tris(benzyloxy)octadec-6-en-1-ol*; **6**). A mixture of trifluoroacetic anhydride (10 ml), H_2O (1 ml), and **10** (0.28 g, 0.37 mmol) was stirred at r.t. for 1 h. Then, the mixture was washed with sat. aq. $NaHCO_3$ soln. and brine, and extracted with CH_2Cl_2 (3×5 ml). The combined org. extracts were dried (Na_2SO_4) and concentrated *in vacuo*. The residue was purified by CC (SiO_2; hexane/AcOEt 4:1) to afford **6** (0.16 g, 90%) as an oil. $[\alpha]_D^{20} = -33.6$ (*c* = 0.2, $CHCl_3$). IR (film): 3390, 3300, 3250, 1585, 1045. ^{1}H-NMR ($CDCl_3$): 0.88 (*t*, *J* = 6.0, Me(18)); 1.27 (br. *s*, 9 CH_2); 1.73 (br. *s*, OH, NH_2); 2.05 (*m*, CH_2(8)); 2.88 (*m*, H–C(2)); 3.65 (br. *d*, *J* = 5.0, CH_2(1)); 3.60–3.78 (*m*, H–C(3), H–C(4), H–C(5)); 4.65–4.80 (*m*, 3 PhCH_2); 5.56 (*dd*, *J* = 10.5, 9.0, H–C(6)); 5.75 (*dd*, *J* = 10.3, 8.4, H–C(7)); 7.10–7.50 (*m*, 15 arom. H). ^{13}C-NMR ($CDCl_3$): 14.2 (C(18)); 22.7–31.8 (CH_2); 27.8 (C(8)); 52.9 (C(2)); 59.1 (C(1)); 70.7, 72.2, 72.8 (3 Ph*C*H_2); 73.3, 75.5, 81.2 (3 BnO*C*H); 125.3 (C(7)); 128.2–129.3 (arom. C); 136.1 (C(6)); 137.6. EI-MS: 601 (23, M^+), 460 (73, [$M-C_{10}H_{21}$]$^+$).

*[(12-Bromododecyl)oxy](*tert*-butyl)diphenylsilane* (**12**). A soln. of 12-bromododecanol (4.0 g, 15 mmol) in anh. CH_2Cl_2 (20 ml) containing ^{t}BuPh_2SiCl (TBSCl; 3.2 ml, 12.4 mmol) and 1*H*-imidazole (1.7 g, 2.2 mmol) was stirred for 12 h at r.t. The mixture was then poured into ice-cooled aq. $NaHCO_3$ soln. and extracted with CH_2Cl_2 (3×50 ml). The combined org. extracts were dried (Na_2SO_4) and concentrated *in vacuo*. The residue was purified by CC (SiO_2; hexane) to afford **12** (7 g, 95%) as colorless needles. M.p. 131–132°. IR: 1585, 1440, 1110, 1065. ^{1}H-NMR ($CDCl_3$): 1.08 (*s*, ^{t}Bu); 1.20–1.70 (br. *s*, 10 CH_2); 3.42 (*t*, *J* = 6.2, CH_2(1)); 3.65 (*m*, CH_2(12)); 7.30–7.50 (*m*, 6 arom. H); 7.60–7.80 (*m*, 4 arom. H). EI-MS: 503 (30, M^+).

*(12-{[(*tert*-Butyl)(diphenyl)silyl]oxy}dodecyl)(triphenyl)phosphonium Bromide* (**13**). A mixture of Ph_3P (0.16 g, 0.6 mmol) and **12** (0.3 g, 0.6 mmol) was stirred under N_2 at 120° for 12 h. The resulting *Wittig* salt **13** was used in the next step without further purification.

*(*tert*-Butyl)({(12*Z*)-13-[(4*R*)-2,2-dimethyl-1,3-dioxolan-4-yl]tridec-12-en-1-yl}oxy)diphenylsilane* (**15**). A 1.6M BuLi soln. in hexane (0.4 ml, 0.6 mmol) was added to a stirred suspension of crude **13** (0.46 g, 0.6 mmol) in anh. THF (30 ml) under N_2 at $-30°$, and the mixture was stirred for 1 h at this temp. Then, a soln. of 2,3-*O*-isopropylidene-D-glyceraldehyde (**14**; 0.05 g, 0.43 mmol) in anh. THF (10 ml) was added, and stirring was continued for 1 h at $-30°$, and for 2 h at 0°. The mixture was poured into ice-cooled sat. NH_4Cl soln. (15 ml) and extracted with Et_2O (3×25 ml). The combined org. extracts were washed with

brine, dried (Na_2SO_4), filtered, and concentrated *in vacuo*. The residue was purified by CC (SiO_2; hexane/AcOEt 10:1) to afford a mixture of (*E*)- and (*Z*)-**15** (4.8 g, 60%). IR (film): 1589, 1466, 1430, 1380, 1110. ^{1}H-NMR ($CDCl_3$): 1.10 (*s*, ^{t}Bu); 1.25 (br. *s*, 8 CH_2); 1.32, 1.37 (2*s*, Me_2C); 1.65 (*m*, CH_2(6)); 2.22 (*m*, CH_2(5)); 3.49–4.70 (*m*, CH_2(1), H–C(2), CH_2(15)); 5.30–5.60 (*m*, CH=CH); 7.30–7.50 (*m*, 6 arom. H); 7.60–7.80 (*m*, 4 arom. H). ^{13}C-NMR ($CDCl_3$): 19.2 (SiC); 22.0–26.0 (Me); 28.2–30.1 (CH_2), 32.3 (CH_2–CH=); 64.0 (CH_2OSi); 69.5 (C(1)); 72.1 (C(2)); 109.0 (Me_3C); 127.1 (C(3)); 128.0–136.1 (arom. C); 135.3 (C(4)); 134.0. EI-MS: 536 (20, M^+), 479 (79, $[M-C_4H_9]^+$).

*(*tert*-Butyl)({13-[(4*R*)-2,2-dimethyl-1,3-dioxolan-4-yl]tridecyl}oxy)diphenylsilane* (**16**). A soln. of **15** (2.5 g, 4.65 mmol) in anh. MeOH (50 ml) was hydrogenated at 100 atm H_2 over PtO_2 (1.2 g) at r.t. When the reaction was complete (GC control), the pressure was released, and the catalyst was removed by filtration. The filtrate was concentrated *in vacuo* to afford **16** (2.2 g, 85%) as an oil, which was used in the next step without purification. IR (film): 1600, 1110. ^{1}H-NMR ($CDCl_3$): 1.10 (*s*, ^{t}Bu); 1.26 (br. *s*, 12 CH_2); 1.31, 1.36 (2*s*, Me_2C); 3.50–4.65 (*m*, CH_2(1), H–C(2), CH_2(15)); 7.30–7.50 (*m*, 6 arom. H); 7.60–7.80 (*m*, 4 arom. H). EI-MS: 538 (12, M^+).

*13-[(4*R*)-2,2-Dimethyl-1,3-dioxolan-4-yl]tridecanal* (**17**). *a*) A 1M soln. of Bu_4NF (TBAF) in THF (6 ml, 6.0 mmol) was added to a soln. of **16** (1.6 g, 3.0 mmol) in THF (50 ml), and the mixture was stirred for 3 h at r.t. Then, the mixture was diluted with $CHCl_3$ and washed with sat. aq. NH_4Cl soln. The aq. layer was re-extracted with $CHCl_3$, the combined org. layers were washed with brine, dried (Na_2SO_4), and concentrated *in vacuo*. The residue was purified by CC (SiO_2; hexane/AcOEt 7:3) to afford *13-[(4*R*)-2,2-dimethyl-1,3-dioxolan-4-yl]tridecan-1-ol* (0.8 g, 89%).

b) A soln. of the above intermediary alcohol (0.8 g, 2.6 mmol) in CH_2Cl_2 (15 ml) was added to a stirred suspension of pyridinium chlorochromate (PCC; 0.6 g, 2.7 mmol) in anh. CH_2Cl_2 (20 ml), and the mixture was stirred at r.t. for 2 h. The mixture was treated with *Celite*, extracted with Et_2O (3×20 ml), and the combined org. extracts were filtered through a short plug of SiO_2, and concentrated *in vacuo*. The resulting residue was subjected to CC (SiO_2; hexane/AcOEt 5:1) to afford **17** (0.7 g, 87%) as an oil. IR (film): 2750, 1730, 1430, 1115. ^{1}H-NMR ($CDCl_3$): 1.28 (br. *s*, 12 CH_2); 1.30, 1.36 (2*s*, Me_2C); 2.40 (*m*, CH_2(14)); 3.50–4.05 (*m*, CH_2(1), H–C(2)); 9.70 (*s*, CHO). ^{13}C-NMR ($CDCl_3$): 24.1, 24.3 (Me_2C); 27.0–31.9 (CH_2); 62.1 (C(14)); 69.5 (C(1)); 72.3 (C(2)); 108.7 (Me_2C); 204.0 (CHO). EI-MS: 298 (25, M^+), 269 (100, $[M-29]^+$).

*(4*R*)-4-[(13*Z*)-Docos-13-en-1-yl]-2,2-dimethyl-1,3-dioxolane* (**18**). A 1.6M BuLi soln. in hexane (1.5 ml, 2.4 mmol) was added to a stirred suspension of nonyl(triphenyl)phosphonium bromide (1.12 g, 2.4 mmol) in anh. THF (30 ml) and HMPA (3 ml) under N_2 at −78°, and the mixture was stirred for 40 min at this temp. Then, a soln. of **17** (0.65 g, 2.2 mmol) in anh. THF (20 ml) was added, and stirring was continued for 1 h at −78°, and for 2 h at 0°. The mixture was poured into ice-cooled brine (20 ml), and extracted with Et_2O (3×20 ml). The combined org. extracts were dried (Na_2SO_4), filtered, and concentrated *in vacuo*. The residue was purified by CC (SiO_2; hexane/AcOEt 9:1) to afford pure **18** (0.35 g, 40%) as an oil, together with a small amount (0.09 g, 10%) of the (*E*)-isomer (oil). IR (film): 1650, 1120. ^{1}H-NMR ($CDCl_3$): 0.90 (*t*, Me); 1.26 (br. *s*, 17 CH_2); 1.31, 1.36 (2*s*, Me_2C); 1.90–2.15 (*m*, CH_2(14), CH_2(17)); 3.50–4.05 (*m*, CH_2(1), H–C(2)); 5.35 (*dd*, *J* = 9.0, 9.2, CH=CH). ^{13}C-NMR ($CDCl_3$): 14.0 (Me); 24.1, 24.3 (Me_2C); 26.2–31.3 (CH_2); 27.8 (C(14)); 27.9 (C(17)); 69.4 (C(1)); 72.5 (C(2)); 108.9 (Me_2C); 130.0 (CH=CH). EI-MS: 408 (12, M^+), 295 (100, $[M-C_8H_{17}]^+$).

*(4*R*)-4-[(13*Z*)-Docos-13-en-1-yl]-2-phenyl-1,3-dioxolane* (**20**). *a*) A soln. of **18** (0.35 g, 0.80 mmol) in THF (3.6 ml), AcOH (20 ml), H_2O (12 ml), and 2N aq. H_2SO_4 (10 ml) was stirred at r.t. for 24 h. The mixture was washed with sat. aq. $NaHCO_3$ soln. (20 ml), and extracted with CH_2Cl_2 (3×25 ml). The combined org. extracts were dried (Na_2SO_4) and concentrated *in vacuo*, and the resulting crude diol, *i.e.*, *(2*R*,15*Z*)-tetracos-15-ene-1,2-diol*, was used in the next step without further purification.

b) A mixture of the above crude diol (0.2 g, 0.5 mmol) in CH_2Cl_2, benzaldehyde dimethyl acetal ($PhCH(OMe)_2$; 0.12 ml, 0.80 mmol), and CF_3CO_2H (TFA; 0.1 ml) was stirred under N_2 at 50° for 2 h. The mixture was then washed with sat. aq. $NaHCO_3$ soln. (10 ml), and extracted with CH_2Cl_2 (3×20 ml). The combined org. extracts were washed with brine, and concentrated *in vacuo*. The resulting residue was purified by CC (SiO_2; hexane/Et_2O 10:1) to afford **20** (0.12 g, 50%) as an oil. A small amount (20 mg) of the (*E*)-isomer was also obtained by further elution. IR (film): 1650, 1590, 1110. ^{1}H-NMR ($CDCl_3$): 0.90 (*t*, Me); 1.27 (br. *s*, 17 CH_2); 1.90–2.10 (*m*, CH_2(14), CH_2(17)); 3.40–4.20 (*m*, CH_2(1),

H–C(2)); 5.35 (*m*, CH=CH); 5.80 (*s*, PhC*H*); 7.35 (*m*, 3 arom. H); 7.50 (*m*, 2 arom. H). ^{13}C-NMR ($CDCl_3$): 14.0 (Me); 26.0–30.9 (CH_2); 27.8 (C(14)); 27.9 (C(17)); 69.5 (C(1)); 72.6 (C(2)); 107.1 (Ph*C*H); 130.1 (CH=CH); 128.0–135.0 (arom. C). EI-MS: 456 (30, M^+), 343 (100, $[M-C_8H_{17}]^+$).

*(2*R*,15*Z*)-2-(Benzyloxy)tetracos-15-en-1-ol* (**19**). A soln. of **20** (0.1 g, 0.2 mmol) was added to a 1M suspension of diisobutylaluminum hydride (DIBALH) in toluene (2 ml, 2 mmol), and the mixture was stirred at r.t. for 24 h. The reaction was quenched with MeOH, the mixture was washed with sat. aq. $NaHCO_3$ soln. (5 ml), and extracted with CH_2Cl_2 (3×10 ml). The org. extracts were combined, dried (Na_2SO_4), and concentrated under reduced pressure. The residue was purified by CC (SiO_2; hexane/Et_2O 1:1) to afford **19** (73 mg, 80%) as colorless needles. M.p. 95–96°. IR (KBr): 3400, 1650, 1585, 1110. ^{1}H-NMR ($CDCl_3$): 0.90 (*t*, Me); 1.26 (br. *s*, 17 CH_2); 1.90–2.10 (*m*, CH_2(14), CH_2(17)); 3.41 (*m*, H–C(2)); 4.20–4.72 (*m*, CH_2(1), PhCH_2); 5.30–5.50 (*m*, CH=CH); 7.30–7.50 (*m*, 5 arom. H). EI-MS: 458 (20, M^+), 345 (80, $[M-C_8H_{17}]^+$).

*(2*R*,15*Z*)-2-(Benzyloxy)tetracos-15-enal* (**21**). Prepared from **19** (0.7 g, 1.5 mmol) in analogy to the procedure described for the preparation of **17** (see above). Yield: 0.62 g (90%). IR (film): 2745, 1730, 1650, 1585, 1110. ^{1}H-NMR ($CDCl_3$): 0.90 (*t*, Me); 1.27 (br. *s*, 17 CH_2); 1.90–2.15 (*m*, 2 CH_2); 3.75 (*m*, 1 H); 4.50–4.75 (*m*, PhCH_2); 5.35 (*m*, CH=CH); 7.30–7.45 (*m*, 5 arom. H); 9.60 (*s*, CHO). ^{13}C-NMR ($CDCl_3$): 14.3 (Me); 26.0–30.6 (CH_2); 73.3 (Ph*C*H$_2$); 83.8; 128.1, 130.0 (CH=CH); 130.3–135.1 (arom. C); 204.1 (CHO). EI-MS: 456 (10, M^+), 427 (100, ($[M-29]^+$), 314 (65, $[M-29-C_8H_{17}]^+$).

*(2*R*,15*E*)-2-(Benzyloxy)tetracos-15-enoic Acid* (**11**). Aldehyde **21** (4.3 g, 9.6 mmol) was dissolved in ^{t}BuOH (20 ml) and 2-methylbut-2-ene (4.8 ml). A soln. of $NaClO_2$ (0.8 g, 8.8 ml) and NaH_2PO_4 (1.0 g, 6.6 mmol) in H_2O (8 ml) was added over 10 min. The pale-yellow mixture was stirred at r.t. overnight. The volatile components were removed under reduced pressure. The residue was dissolved in H_2O (20 ml), and the soln. was extracted with hexane (3×20 ml). The aq. layer was acidified to pH 2 with 2N HCl, and extracted with Et_2O (3×20 ml). The combined org. layers were washed with H_2O, dried (Na_2SO_4), and concentrated to afford **11** (2.6 g, 59%) as an oil. IR (film): 3060, 1730, 1650, 1580, 1110. ^{1}H-NMR ($CDCl_3$): 0.90 (*t*, Me(23)); 1.27 (br. *s*, 17 CH_2); 1.90–2.10 (*m*, CH_2(14), CH_2(17)); 3.75 (*m*, H–C(2)); 4.50–4.75 (*m*, PhCH_2); 5.35 (*m*, CH=CH); 7.30–7.45 (*m*, 5 arom. H); 12.1 (br. *s*, COOH). ^{13}C-NMR ($CDCl_3$): 14.2 (C(24)); 26.0–30.6 (CH_2); 27.8 (C(14)); 27.9 (C(17)); 73.2 (Ph*C*H$_2$); 83.7 (C(1)), 128.0 (CH=CH); 130.0, 130.2–135.0 (arom. C); 220.1 (COOH). EI-MS: 472 (15, M^+), 359 (50, $[M-C_8H_{17}]^+$).

*(2*R*,15*Z*)-2-(Benzyloxy)-*N*-[(1*S*,2*S*,3*R*,4*R*,5*Z*)-2,3,4-tris(benzyloxy)-1-(hydroxymethyl)heptadec-5-en-1-yl]tetracos-15-enamide* (**22**). To a soln. of **6** (1 g, 1.6 mmol) and **11** (0.7 g, 1.6 mmol) in anh. THF (20 ml) was added a soln. of 1,3-dicyclohexylcarbodiimide (DCC; 0.4 g, 2 mmol) and 1-hydroxy-1*H*-benzotriazole (HOBt; 0.3 g, 2 ml) in anh. THF (15 ml), and the mixture was stirred for 48 h at r.t. The solvent was evaporated at reduced pressure, and the residue was purified by CC (SiO_2; hexane/AcOEt 8:2). Yield: 0.76 g (50%). Colorless oil. IR (film): 3442, 1739, 1670. ^{1}H-NMR ($CDCl_3$): 0.88 (*m*, Me(18), Me(24′)); 1.28 (br. *s*, 21 CH_2); 1.90–2.20 (*m*, CH_2(8), CH_2(14′), CH_2(17′)); 3.50–4.27 (*m*, CH_2(1), H–C(2), H–C(3), H–C(4), H–C(5), H–C(2′)); 4.52–4.82 (*m*, 4 PhCH_2); 5.24–5.90 (*m*, H–C(6), H–C(7), H–C(15′), H–C(16′)); 7.10–7.41 (*m*, 20 arom. H, NH). ^{13}C-NMR ($CDCl_3$): 14.0, 14.2 (C(18), C(24′)); 23.9–30.7 (CH_2); 27.8, 27.9, 28.1 (C(8), C(14′), C(17′)); 51.7 (C(2)); 62.3 (C(1)); 70.2 (Ph*C*H$_2$); 72.2 (BnO*C*H); 74.3 (Ph*C*H$_2$); 74.4 (Ph*C*H$_2$); 74.6 (Ph*C*H$_2$); 79.9 (BnO*C*H); 80.3 (BnO*C*H); 80.5 (BnO*C*H); 126.1–130.2 (arom. C); 137.5, 131.0–138.2 (C(6), C(7), C(15′), C(16′)); 179.2 (C(1′)). EI-MS: 1056 (10, M^+).

*(2*R*,15*Z*)-2-(Benzyloxy)-*N*-((1*S*,2*S*,3*R*,4*R*,5*Z*)-2,3,4-tris(benzyloxy)-1-{[(2,3,4,6-tetra-*O*-acetyl-β-D-glucopyranosyl)oxy]methyl}heptadec-5-en-1-yl)tetracos-15-enamide* (**24**). Anh. benzene was added to a soln. of **22** (150 mg, 0.14 mmol) in nitromethane (13 ml), and the resulting soln. was stirred at 110° to remove moisture azeotropically. The mixture was concentrated to a volume of *ca.* 15 ml, and cooled under N_2. Then, *2,3,4,6-tetra-*O*-acetyl-α-D-glucopyranosyl bromide* (**23**; 82.2 mg, 0.2 mmol) and $HgCN_2$ (50 mg, 0.2 mmol) were added, and stirring was continued for 2 h at 90° under N_2. After cooling, the mixture was diluted with $CHCl_3$, and washed with a sat. soln. of H_2S. The black precipitate (HgS) was filtered off (*Celite*), the filtrate was washed with sat. aq. NH_4Cl soln. and brine, dried (Na_2SO_4), and concentrated *in vacuo*. The residue was purified by CC (SiO_2; $CHCl_3$/AcOEt 15:1). Yield: 95.8 mg (50%). The compound was used in the next step without further purification.

*(2*R*,15*Z*)*-N*-{(1*S*,2*S*,3*R*,4*R*,5*Z*)-1-[(β-*D*-Glucopyranosyloxy)methyl]-2,3,4-trihydroxyheptadec-5-en-1-yl}-2-hydroxytetracos-15-enamide* (**2**). *a*) A soln. of crude **24** in MeOH (10 ml) was treated with 5% KOH in MeOH (10 ml). The mixture was stirred at r.t. for 30 min, and then neutralized with *Dowex 50 WX8* (H^+ form). The resin was removed by filtration, the filtrate was concentrated under reduced pressure, and the residue was purified by CC (SiO_2; $CHCl_3$/MeOH 10:1) to afford a colorless powder (60 mg; intermediate). Meanwhile, in a separate flask, to a soln. of naphthalene (30 mg, 0.28 mmol) in THF (1.5 ml) was added Li (1.5 mg, 0.2 mmol) in small pieces. The mixture was stirred at r.t. under N_2 atmosphere until the metal was completely dissolved (*ca.* 3 h) [11]. The resulting dark-green lithium naphthalenide soln. was cooled to −25°. Then, a soln. of the above colorless powder (60 mg, 0.05 mmol) in THF (2 ml) was added dropwise over 5 min. The resulting mixture was stirred at −25° for 2 h. Then, sat. aq. NH_4Cl soln. (0.5 ml) and H_2O (0.5 ml) were added. The mixture was extracted with Et_2O (3×5 ml). The combined org. extracts were washed with H_2O and brine, dried (Na_2SO_4), filtered, and concentrated. The crude product was purified by CC (SiO_2; $CHCl_3$/MeOH 5:1) to afford the target cerebroside **2** (0.36 g, 85%). $[\alpha]_D^{20} = +13.2$ ($c = 0.3$, MeOH). IR (film): 3500, 1740, 1670. ^{1}H-NMR (C_5D_5N)[1]): 0.86 (*m*, Me(18), Me(24′)); 1.25 (br. *s*, 21 CH_2); 1.90–2.40 (*m*, CH_2(8), CH_2(14′), CH_2(17′)); 3.68 (*m*, H−C(5″)); 3.90 (*t*, $J = 7.5$ Hz, H−C(2″)); 4.10 (obscured, H−C(4)); 4.13 (obscured, H−C(3″)); 4.15 (obscured, H−C(4″)); 4.23 (*m*, H−C(3)); 4.29 (*dd*, $J = 11.7$, 5.7, H_b−C(6″)); 4.45 (*dd*, $J = 11.7$, 2.2, H_a−C(6″)); 4.48 (*dd*, H_b−C(1)); 4.55 (*dd*, $J = 7.8$, 3.7, H−C(2′)); 4.66 (*dd*, $J = 10.8$, 6.6, H_a−C(1)); 4.78 (*m*, H−C(5)); 4.92 (*d*, $J = 8.0$, H−C(1″)); 5.22 (*m*, H−C(2)); 5.50 (*m*, CH=CH); 5.60 (*m*, H−C(7)); 5.80 (*dd*, $J = 10.0$, 9.7, H−C(6)); 8.51 (*d*, $J = 9.0$, NH). ^{13}C-NMR (C_5D_5N): 14.0, 14.2 (C(18), C(24′)); 23.0–30.5 (CH_2); 27.8, 27.9, 28.1 (C(8), C(14′), C(17′)); 51.7 (C(2)); 62.6 (C(6″)); 67.6 (C(5)); 70.2 (C(1)); 71.5 (C(4″)); 72.4 (C(2′)); 75.1 (C(2″)); 75.9 (C(3), C(4)); 78.4 (C(3″), C(5″)); 105.4 (C(1″)); 130.2–131.0 (C(6), C(7), C(15′), C(16′)); 175.6 (C(1′)). FAB-MS: 881 (15, $[M+Na]^+$), 679 (20, $[M-179]^+$), 661 (35, $[M-179-H_2O]^+$), 516 (100, $[C_{24}H_{47}NO_9+Na]^+$), 314 (50, $C_{18}H_{36}NO_3^+$), 296 (45, $C_{18}H_{34}NO_2^+$).

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