Synthesis of *Neolacto* Ganglioside LM1

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The synthesis of the ganglioside LM1 (**1**) has been performed by glycosidation of **2** with azidosphingosine **3**, followed by reduction of the azido group, acylation with stearine anhydride (**4**), and solvolysis. **2** was obtained from sialyl xanthate **5** and tetrasaccharide **6** in the presence of phenylsulfenyl triflate. The tetrasaccharide acceptor **6** was prepared from the lactose derivatives **7** and **8**.

Introduction

Gangliosides are neuraminic acid containing cell surface oligosaccharides associated with a variety of biological recognition processes. Their involvement in the pathogenesis of cancer is a widely accepted fact[1] and many research groups are currently engaged in the synthesis of complex carbohydrate conjugates.[2] Recently, the ganglioside LM1 (**1**), belonging to the *neolacto* series, has gained special interest. It consists of a pentasaccharide moiety [Neu-Acα2→3Galβ1→4GlcNAcβ1→3Galβ1→4Glc] glycosidically bound to ceramide. It is found to be widespread in nerve cells, seems to be involved in diseases such as Guillain-Barre syndrome (GBS)[3] and caturastic lenses,[4] and its potential application as a prognostic indicator for brain tumors has been discussed.[5] LM1 has also been identified as one of the two cytostatically active compounds of mouse macrophages,[6] which are effectors of the natural immune system by recognizing tumor cells and inhibiting their growth. Their cytostatic effect is accompanied by differentiation of the target mouse mastocytoma cells.[7] A synthesis of LM1 (**1**) has already been described.[8] However, it is rather long and is not ideally suited for the preparation of larger quantities. Here we describe a more efficient synthesis of LM1 (**1**), with a view to providing the material needed for further biological testing.

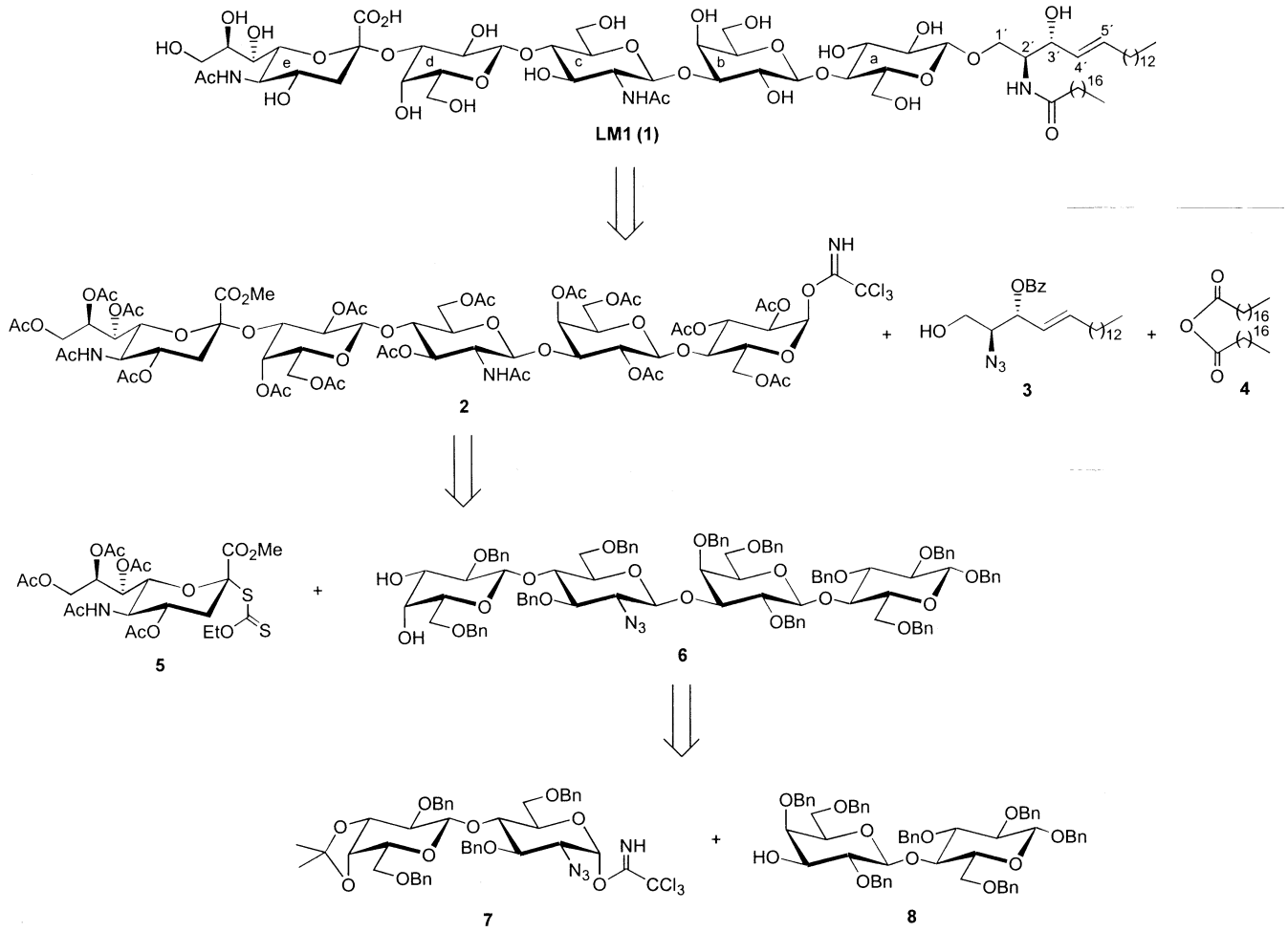
Results and Discussion

The retrosynthetic analysis of **1** leads first to the pentasaccharide **2**, which is broken down into the tetrasaccharide **6** and the neuraminic acid moiety **5**, and further to two disaccharides (Scheme 1). Thus, the tetrasaccharide **6** was prepared by connection of the azidolactose imidate **7** as donor and the lactose derivative **8** with a free hydroxy group at C-3b as acceptor. The final step in the synthesis of **2** was the glycosidation of **6** with the neuraminic acid xanthate **5**[9a] in the presence of the promoter phenylsulfenyl triflate. This protocol was established by Whitesides et al.,[9b] and has recently been successfully employed by ourselves in the synthesis of a GM3-lactone analogue.[10] The described approach to LM1 (**1**) allows the introduction of the rather valuable neuraminic acid at a late state of the synthesis, thereby keeping the costs to a minimum.

Compound **9** could be obtained as a mixture of the two anomers by azidonitration of hexa-*O*-acetyllactal (Scheme 2).[11] Saponification of the nitro group with calcium carbonate in aqueous acetone led quantitatively to **10**, which was protected at the anomeric position with *tert*-butyldiphenylsilyl chloride (TBDPSCl) and imidazole to afford 92% of **11** as a single diastereomer. Solvolysis of the acetyl groups with sodium methoxide in methanol gave **12** in 98% yield, which was transformed into the 3b,4b-*O*-isopropylidene derivative **13** in 66% yield using dry acetone in the presence of a catalytic amount of *p*-toluenesulfonic acid (PTS). As a side product, the corresponding 4b,6b-acetal was obtained (7%), which could be separated by column chromatography. In addition, we also prepared **12a**, with a *tert*-butyldimethylsilyloxy group at C-1.[12] However, due to the lower stability of this protecting group under acidic conditions, the yield of the corresponding 3b,4b-*O*-isopropylidene derivative was not satisfactory. The benzylation of **13** with benzyl bromide in the presence of sodium hydride is a very critical step in the synthesis. This has previously been observed by Schmidt et al.[12] in relation to similar systems. Exclusion of moisture and low reaction temperatures are absolutely essential. The work-up had to be performed by rapid filtration of the reaction mixture through a pad of Celite. In this way, 78% of **14** was obtained after chromatographic purification. The TBDPS group in **14** was cleaved with tetrabutylammonium fluoride (TBAF) to give **15** in quantitative yield. Formation of trichloroacetimidate **7** was accomplished in 99% yield using trichloroacetonitrile (CCl_3CN) and 1,8-diazabicyclo[5.4.0]undec-7-ene (DBU).[13]

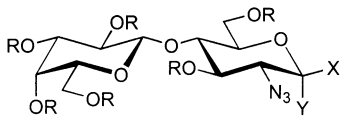
  1434−193X/98/■−■ $ 17.50+.50/0

Scheme 1



The glycosidation of the donor **7** with acceptor **8**, which had been synthesized from lactose in 8 steps as described in the literature,[12] was performed using trimethylsilyl trifluoromethanesulfonate (TMSOTf) as mediator at −40°C in acetonitrile (Scheme 3). The desired tetrasaccharide **16** was obtained in 80% yield in a highly stereoselective reaction. In spite of the absence of a neighbouring group at C-2a in **7** − position 2a carries an azido group − only a β-glycosidic bond was formed. The stereocontrol can be explained by the nitrile effect of acetonitrile as a coordinating solvent.[14] Unreacted acceptor **8** could be recovered almost quantitatively; however, in the preparations of larger amounts of **16** it is more appropriate to separate unreacted **8** after the next step. The 3b,4b-*O*-isopropylidene group in **16** was then cleaved with aqueous acetic acid (80%) at 100°C, leading to the diol **6** in 79% yield. Diol **6** served as the acceptor in the subsequent sialylation using the very reactive xanthate **5**, which was synthesized according to the literature.[9] The promoter phenylsulfenyl triflate was prepared in situ from silver triflate and phenylsulfenyl chloride in the presence of 2,6-di-*tert*-butyl-4-methylpyridine (DTBMP), which serves as a proton scavenger. The reaction was carried out in a 2:1 mixture of acetonitrile and dichloromethane at −70°C, again making use of the nitrile effect,[14] to give the α-glycosidic sialoside **17** highly selectively in 58% yield. A regioisomer of **17**, which might have been formed by reaction of the free hydroxy group at C-4d in **6**, was not found, demonstrating the low reactivity of the axial hydroxy group in galactose-containing oligosaccharides. It should be noted that exclusion of light and moisture is essential, and that the reaction temperature should be carefully monitored, since the mixture tends to freeze just below −70°C. The described procedure can also be used for the preparation of **17** on a gram scale. The structures of the oligosaccharides **16** and **17** were confirmed principally by ^{1}H-NMR spectroscopy using total correlation spectroscopy (TOCSY) experiments. This procedure allows the selective identification of all hydrogen atoms of the different monosaccharide moieties. Thus, 1c-H of **16** resonates at δ = 4.46 with J = 8.0 Hz, clearly demonstrating the existence of a β-glycosidic bond; for the 3e-H_{ax} and 3e-H_{eq} in **17**, signals are observed at δ = 2.11 and δ = 2.69 with J = 13.0 Hz and J = 4.5, 13.0 Hz, respectively. The coupling constant $J_{7e,8e}$ was 8.0 Hz and the Δδ = δ(9e-H_b) − δ(9e-

Scheme 2

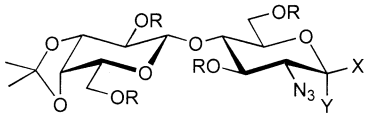


9: R = Ac, X,Y = H,ONO_2
10: R = Ac, X,Y = H,OH
11: R = Ac, X = OTBDPS, Y = H
12: R = H, X = OTBDPS, Y = H

12a: R = H, X = OTBDMS, Y = H

$CaCO_3$, acetone
TBDPSCl, imidazole, DMF
NaOMe, methanol

PTS, acetone



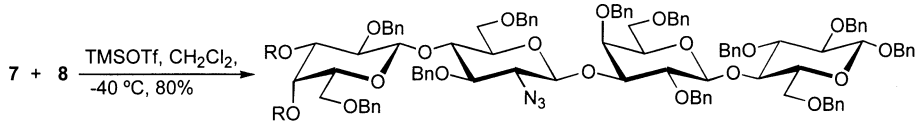
13: R = H, X = OTBDPS, Y = H
14: R = Bn, X = OTBDPS, Y = H
15: R = Bn, X,Y = H,OH
7: R = Bn, X = H, Y = OC(NH)CCl_3

BnBr, NaH, DMF
TBAF, THF
CCl_3CN, DBU, CH_2Cl_2

H_a) value was 0.47, indicating an α-glycosidic bond at the neuraminic acid.[15] In case of a β-glycosidic bond a coupling constant $J_{7e,8e}$ of approximately 2 Hz and a $\Delta\delta = \delta(9e\text{-}H_b) - \delta(9e\text{-}H_a)$ value > 0.9 should be expected.[16]

To complete the synthesis of **1**, the benzyl groups in **17** were removed by hydrogenation under a pressure of 5 bar using freshly prepared Pearlman's catalyst[17] [palladium(II) hydroxide]. Under these conditions, the azido group in **17** was transformed into the desired amino group, which was directly acetylated by treating the crude product with acetic anhydride and pyridine to give **18** in 80% overall yield as a 1:1 mixture of the α and β anomers. Selective removal of the acetate group at C-1a with hydrazinium acetate[18] in DMF led to **19** in 92% yield. Reaction of **19** with CCl_3CN and DBU in dichloromethane gave exclusively the trichloroacetimidate **2** with α configuration at C-1a, which was treated with azidosphingosine[19] in the presence of catalytic amounts of boron trifluoride in dichloromethane at 0°C to afford **20** in 43% yield (Scheme 4). The azido group in **20** was reduced in situ with Lindlar's catalyst under hydrogen, and the resulting amine was directly transformed into the stearine amide with stearine anhydride (**4**) to yield 92% of **21**.[20] Saponification of all the ester groups under Zemplén conditions[21] gave 95% of the desired LM1 (**1**). Spectroscopic data were in complete agreement with those reported in the literature,[8] thus confirming the assigned structure.

Scheme 3

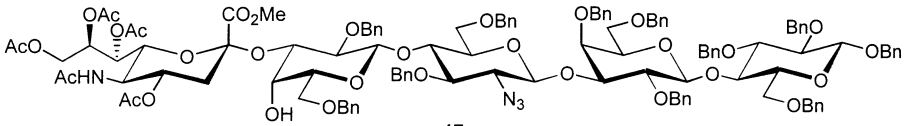


7 + **8** — TMSOTf, CH_2Cl_2, -40 °C, 80%

16: R = isopropylidene
6: R = H

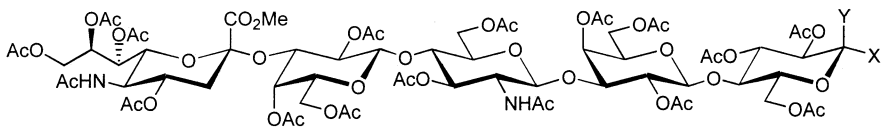
HOAc / H_2O, 100 °C, 79%

5, AgOTf, PhSCl, DTBMP, CH_3CN, CH_2Cl_2, -70 °C, 58%



17

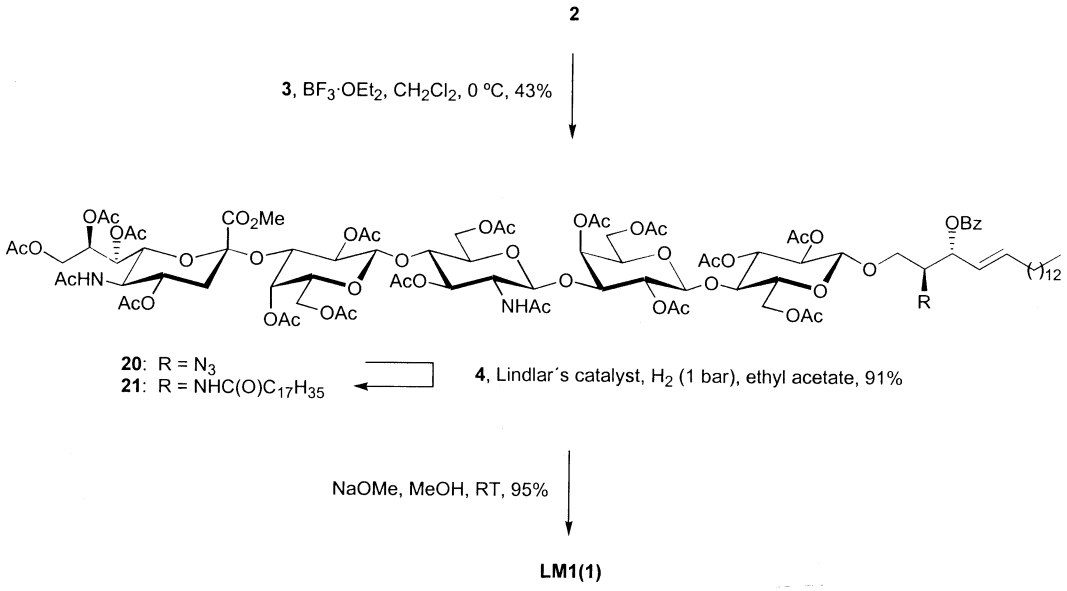
1. H_2 (5 bar), $Pd(OH)_2$, MeOH, RT
2. Ac_2O, pyridine, RT, 80% (overall)



18: X, Y = H, OAc
19: X, Y = H, OH
2: X = H, Y = OC(NH)CCl_3

N_2H_4·HOAc, DMF, 40 °C, 92%
CCl_3CN, DBU, CH_2Cl_2, 0 °C, 75%

Scheme 4



Conclusions

The ganglioside LM1 (**1**) is of great importance in the pathogenesis and treatment of cancer. Of particular interest is the differentiation of mouse mastocytoma cells.[6][7] We have described herein a selective and efficient synthesis of LM1 (**1**) that allows the preparation of this compound in 19 steps starting from lactose, the lactose acceptor **8**, sialyl xanthate **5**, and azidosphingosine **3**. The synthesis is 7 steps shorter than a previously described procedure.[8]

The synthesized LM1 (**1**) is currently being tested in cooperation with the Department of Medicinal Microbiology at the University of Göttingen.

We thank the *Deutsche Forschungsgemeinschaft* (SFB 500) and the *Fonds der Chemischen Industrie* for generous support.

Experimental Section

General: Solvents were purified and dried by standard procedures; the petroleum ether (PE) used had a boiling range of 35−65°C. − Organic phases were dried with sodium sulfate and were concentrated in vacuo with a bath temperature < 40°C. − NMR: Varian XL-200, UNITY 300 or INOVA 500; internal standard tetramethylsilane or residual protons in the solvent. − Optical rotations were measured with a Perkin-Elmer 241 digital polarimeter in a 1-dm cell. − IR: Bruker IFS. − Column chromatography: Silica gel 60 (Macherey-Nagel & Co, particle size 63−200 μm). − Flash chromatography: Silica gel 40 (Macherey-Nagel & Co, particle size 40−63 μm). − Thin-layer chromatography (TLC): Macherey-Nagel & Co, SIL G/UV_{254}. − Elemental analyses: Microanalytical Laboratory of the Institute of Organic Chemistry, University of Göttingen.

3,6-Di-O-acetyl-2-azido-2-deoxy-4-O-(2,3,4,6-tetra-O-acetyl-β-D-galactopyranosyl)-β-D-glucopyranose (**10**): A solution of **9** (18.0 g, 27.1 mmol) in a 1:1 mixture of acetone and water (300 ml) was vigorously stirred with calcium carbonate (4.5 g, 45.0 mmol) for 48 h at room temperature. Then, 2 N HCl (200 ml) was added and the aqueous phase was extracted with dichloromethane (3 × 150 ml). The combined organic phases were washed with brine, dried, and concentrated to give 16.8 g (quantitative) of **10**, which was used without further purification. R_f = 0.24 (PE/ethyl acetate, 1:1). The physical data were in agreement with those reported in the literature.[22]

tert-Butyldiphenylsilyl 3,6-Di-O-acetyl-2-azido-2-deoxy-4-O-(2,3,4,6-tetra-O-acetyl-β-D-galactopyranosyl)-β-D-glucopyranoside (**11**): To a stirred solution of **10** (2.50 g, 4.04 mmol) in DMF (25 ml), *tert*-butyldiphenylsilyl chloride (2.21 g, 8.04 mmol) and imidazole (1.10 g, 16.2 mmol) were added at 0°C. The solution was stirred at room temperature for 12 h, and then diluted with ice-cold water (200 ml). The resulting mixture was extracted with dichloromethane (2 × 50 ml), and the combined extracts were washed with 1 N HCl, water, and brine. After drying and concentration, the residue was purified by column chromatography (PE/ethyl acetate, 2:1) yielding 3.20 g (92%) of **11**. R_f = 0.66 (PE/ethyl acetate, 1:1). − $[\alpha]_D$ = 0.5 (c = 1.0, $CHCl_3$). − IR (KBr): $\tilde{\nu}$ = 2960 cm^{-1}, 2938, 2896, 2112, 1754, 1370, 1224, 1064. − ^{1}H NMR (200 MHz, $CDCl_3$): δ = 1.09 [s, 9 H, $C(CH_3)_3$], 1.96, 1.97, 2.05, 2.06, 2.09, 2.13 (6 s, 18 H, 6 × Ac), 3.18 (ddd, J = 2.0, 5.5, 10.5 Hz, 1 H, 5a-H), 3.46 (dd, J = 8.0, 10.0 Hz, 1 H, 2a-H), 3.65 (t, J = 9.5 Hz, 1 H, 4a-H), 3.68−4.25 (m, 6 H, 5a-H, 5b-H, 6a-H_2, 6b-H_2), 4.40 (d, J = 8.0 Hz, 1 H, 1-H), 4.45 (d, J = 8.0 Hz, 1 H, 1b-H), 4.80−4.90 (m, 2 H, 3a-H, 3b-H), 5.05 (dd, J = 8.0, 10.0 Hz, 1 H, 2b-H), 5.34 (d, J = 3.0 Hz, 1 H, 4b-H), 7.25−7.80 (m, 10 H, Ph-H). − ^{13}C NMR (50 MHz, $CDCl_3$): δ = 19.03 [$C(CH_3)_3$], 20.46, 20.52, 20.83, 26.48, 26.64 (6 × Ac), 20.59 [$C(CH_3)_3$], 60.80, 61.69 (C-6a, C-6b), 66.53, 66.59, 68.97, 70.60, 70.88, 72.06, 72.36, 76.22 (C-2a, C-2b, C-3a, C-3b, C-4a, C-4b, C-5a, C-5b), 96.41, 100.9 (C-1a, C-1b), 127.3, 127.4, 127.6, 127.8, 129.8, 130.1, 132.0, 132.7 (aromatic), 168.8, 169.4, 170.0, 170.1, 170.2 (Ac). − MS (DCI, ammonia): m/z (%) = 875 (85) $[M + 18]^+$, 364 (90), 302 (100). − $C_{38}H_{55}N_3O_{16}Si$ (837.9): calcd. C 56.00, H 5.99; found C 56.35, H 6.08.

tert-Butyldiphenylsilyl 2-Azido-2-deoxy-4-O-(β-D-galactopyranosyl)-β-D-glucopyranoside (**12**): A solution of **11** (4.50 g, 5.25 mmol) in methanol (90 ml) was stirred with sodium methoxide (0.23 ml of a 5.4 N solution in methanol, 4.73 mmol) at room temperature for 3 h, and then neutralized with Duolite resin (H^+ form). After

filtration, the solvent was removed from the filtrate and the residue was purified by column filtration (dichloromethane/methanol, 6:1) to give 2.92 g (98%) of **12** as a white resin. R_f = 0.38 (dichloromethane/methanol, 6:1). − $[\alpha]_D$ = −12.5 (*c* = 0.6, $CHCl_3$). − IR (KBr): $\tilde{\nu}$ = 3420 cm^{-1}, 2932, 2860, 2112, 1114, 1071. − ^1H NMR (300 MHz, [D$_6$]DMSO/D$_2$O): δ = 1.03 [s, 9 H, C(CH$_3$)$_3$], 3.05 (m, 1 H, 5a-H), 3.32 (dd, *J* = 7.5, 9.5 Hz, 1 H, 2a-H), 3.25−3.55 (m, 9 H, 2b-H, 3a-H, 3b-H, 4a-H, 5b-H, 6a-H$_2$, 6b-H$_2$), 3.57−3.59 (m, 1 H, 4b-H), 4.14 (d, *J* = 7.5 Hz, 1 H, 1b-H), 4.50 (d, *J* = 7.5 Hz, 1 H, 1a-H), 7.15−7.70 (m, 10 H, Ph-H). − ^{13}C NMR (50 MHz, [D$_6$]DMSO): δ = 18.70 [C(*C*H$_3$)$_3$], 26.53 [*C*(CH$_3$)$_3$], 60.12, 60.39 (C-6a, C-6b), 68.08, 68.57, 70.50, 72.84, 73.13, 74.18, 75.54, 80.19 (C-2a, C-2b, C-3a, C-3b, C-4a, C-4b, C-5a, C-5b), 95.39, 103.7 (C-1a, C-1b), 127.5, 129.8, 132.5, 135.2 (aromatic). − MS (DCI, ammonia): *m/z* (%) = 623 (100) [M + 18]$^+$. − $C_{28}H_{39}N_3O_{10}Si$ (605.7): calcd. C 55.52, H 6.49; found C 55.40, H 6.66.

tert-Butyldiphenylsilyl 2-Azido-2-deoxy-4-O-(3,4-O-isopropylidene-β-D-galactopyranosyl)-β-D-glucopyranoside (**13**): Compound **12** (0.34 g, 0.60 mmol) was dissolved in dry acetone (15 ml) and stirred with *p*-toluenesulfonic acid (30 mg, 0.16 mmol) at room temperature for 18 h. Then, $NaHCO_3$ (15 mg, 0.18 mmol) and Na_2SO_4 (5 g) were added and the mixture was filtered. Evaporation of the solvent from the filtrate and chromatographic purification (ethyl acetate/acetone, 2:1 → 1:1) of the residue gave 0.26 g (66%) of **13**. 26 mg (7%) of the corresponding 4b,6b-*O*-isopropylidene derivative and 55 mg (15%) of **12** were additionally isolated. R_f = 0.70 (ethyl acetate/acetone, 1:1). − $[\alpha]_D$ = +13.0 (*c* = 0.5, $CHCl_3$). − IR (KBr): $\tilde{\nu}$ = 3440 cm^{-1}, 3052, 2934, 2890, 2112, 1162. − ^1H NMR (300 MHz, $CDCl_3$): δ = 1.10 [s, 9 H, C(CH$_3$)$_3$], 1.32, 1.49 [2 s, 6 H, C(CH$_3$)$_2$], 2.96 (m$_c$, 1 H, 5a-H), 3.35−3.45 (m, 2 H, 2a-H, 3a-H), 3.46−3.60 (m, 4 H, 2b-H, 5b-H, 6a-H$_2$), 3.78−3.88 (m, 1 H, 6b-H$_a$), 3.90−4.00 (m, 2 H, 4a-H, 6b-H$_b$), 4.05−4.10 (m, 1 H, 3b-H), 4.12 (dd, *J* = 1.5, 7.0 Hz, 1 H, 4b-H), 4.27 (d, *J* = 8.0 Hz, 1 H, 1b-H), 4.53 (d, *J* = 7.0 Hz, 1 H, 1a-H), 7.28−7.84 (m, 10 H, Ph-H). − ^{13}C NMR (75 MHz, $CDCl_3$): δ = 19.04 [*C*(CH$_3$)$_3$], 26.19, 26.54 [C(*C*H$_3$)$_2$], 26.74 [C(*C*H$_3$)$_3$], 61.61, 62.03 (C-6a, C-6b), 68.03 (C-2a), 73.17, 73.25, 73.70, 73.92, 74.88, 79.13, 80.66 (C-2b, C-3a, C-3b, C-4a, C-4b, C-5a, C-5b), 96.80 (C-1a), 102.7 (C-1b), 110.6 [*C*(CH$_3$)$_2$], 127.7, 129.9, 130.0, 132.4, 133.6, 2 × 135.7 (aromatic). − MS (DCI, ammonia): *m/z* (%) = 663 (40) [M + 18]$^+$, 274 (100). − $C_{31}H_{43}N_3O_{10}Si$ (645.7): calcd. C 57.66, H 6.71; found C 57.45, H 6.84.

tert-Butyldiphenylsilyl 2-Azido-3,6-di-O-benzyl-2-deoxy-4-O-(2,6-di-O-benzyl-3,4-O-isopropylidene-β-D-galactopyranosyl)-β-D-glucopyranoside (**14**): To a stirred solution of **13** (10.20 g, 15.79 mmol) and benzyl bromide (18.76 ml, 157.95 mmol) in DMF (100 ml) at −10°C, sodium hydride (2.46 g, 102.64 mmol) was added portionwise over a period of 30 min. After stirring for 16 h at −10°C, the reaction mixture was directly filtered at this temperature through a pad of Celite into ice-cooled water (50 ml). The aqueous phase was quickly extracted with dichloromethane (3 × 10 ml) and the combined organic phases were washed with brine, dried, and concentrated. Chromatographic purification (PE/ethyl acetate, 6:1) yielded 12.31 g (78%) of **14**. R_f = 0.44 (PE/ethyl acetate, 1:1). − $[\alpha]_D$ = +3.8 (*c* = 1.0, $CHCl_3$). − IR (KBr): $\tilde{\nu}$ = 3068 cm^{-1}, 3032, 2956, 2934, 2112, 1458, 1372, 1112. − ^1H NMR (300 MHz, $CDCl_3$): δ = 1.08 [s, 9 H, C(CH$_3$)$_3$], 1.32, 1.36 [2 s, 6 H, C(CH$_3$)$_2$], 2.85 (m$_c$, 1 H, 5a-H), 3.15−3.22 (m, 2 H, 3a-H, 6a-H$_a$), 3.27 (dd, *J* = 8.0, 8.1 Hz, 1 H, 2b-H), 3.34 (dd, *J* = 8.0, 10.0 Hz, 1 H, 2a-H), 3.50 (dd, *J* = 5.0, 9.0 Hz, 1 H, 6b-H$_a$), 3.58−3.73 (m, 3 H, 5b-H, 6a-H$_b$, 6b-H$_b$), 3.92−4.15 (m, 3 H, 3b-H, 4a-H, 4b-H), 4.20−4.74 (m, 8 H, 4 × C*H*$_2$Ph), 4.29 (d, *J* = 8.0 Hz, 1 H, 1a-H), 4.39 (d, *J* = 8.1 Hz, 1 H, 1b-H), 7.10−7.75 (m, 30 H, Ph-H). − ^{13}C NMR (75 MHz, $CDCl_3$): δ = 19.13 [*C*(CH$_3$)$_3$], 26.38, 26.80 [2 × C(*C*H$_3$)$_2$], 26.74 [C(*C*H$_3$)$_3$], 67.30 (C-6a), 68.38 (C-2a), 68.91 (C-6b), 72.23, 73.67, 75.07, 79.30, 80.62, 81.24 (C-2b, C-3a, C-3b, C-4a, C-4b, C-5a, C-5b), 73.27, 73.35, 73.37, 75.16 (4 × *C*H$_2$Ph), 96.74, 101.7 (C-1a, C-1b), 109.8 [*C*(CH$_3$)$_2$], 127.8, 2 × 128.1, 128.2, 128.3, 128.8, 129.6, 129.8, 129.8, 132.6, 133.3, 138.2, 138.3, 138.6 (aromatic). − MS (DCI, ammonia): *m/z* (%) = 1023 (100) [M + 18]$^+$. − $C_{59}H_{67}N_3O_{10}Si$ (1006.2): calcd. C 70.42, H 6.71; found C 70.39, H 6.75.

2-Azido-3,6-di-O-benzyl-2-deoxy-4-O-(2,6-di-O-benzyl-3,4-O-isopropylidene-β-D-galactopyranosyl)-β-D-glucopyranose (**15**): A stirred solution of **14** (12.31 g, 12.23 mmol) in dichloromethane (250 ml) was treated with a solution of tetrabutylammonium fluoride (5.79 g, 18.35 mmol) in THF (40 ml) at −20°C. After stirring for 3 h at this temperature, the reaction mixture was poured onto crushed ice. The resulting mixture was extracted with dichloromethane (2 × 150 ml) and the combined organic phases were washed with brine, dried, and concentrated. The residue was purified by column filtration (pentane/ethyl acetate, 3:1 → 1:1) and yielded 9.36 g (quantitative) of **15** as a white foam. R_f = 0.45 (pentane/ethyl acetate, 2:1). − $[\alpha]_D$ = +24.3 (*c* = 1.0, $CHCl_3$). − IR (KBr): $\tilde{\nu}$ = 3348 cm^{-1}, 2968, 2930, 2110, 1106, 1052. − ^1H NMR (300 MHz, C_6D_6): δ = 1.21, 1.38 [2 s, 6 H, C(CH$_3$)$_2$], 2.20 (dd, *J* = 8.0, 10.0 Hz, 1 H, 2b-H), 2.64 (dd, *J* = 7.5, 10.0 Hz, 1 H, 2a-H), 3.08−3.30 (m, 2 H, 5a-H, 5b-H), 3.55−4.94 (m, 16 H, 1a-H, 1b-H, 3a-H, 3b-H, 4a-H, 4b-H, 6a-H$_2$, 6b-H$_2$, 3 × C*H*$_2$Ph), 5.14 (d, *J* = 10.5 Hz, 1 H, C*H*Ph), 5.27 (d, *J* = 11.0 Hz, 1 H, C*H*Ph), 7.00−7.68 (m, 5 H, Ph-H). − ^{13}C NMR (75 MHz, $CDCl_3$): δ = 26.40, 27.95 [2 × C(*C*H$_3$)$_2$], 66.97 (C-2b), 67.83, 68.87 (C-6a, C-6b), 70.65, 72.15, 72.30, 73.24, 73.62, 75.04, 75.28, 75.36, 75.99, 76.54, 78.17, 79.30, 80.49, 80.60, 81.25 (C-2a, C-3a, C-3b, C-4a, C-4b, C-5a, C-5b, 4 × *C*H$_2$Ph), 91.78 (C-1a), 102.0 (C-1b), 109.8 [*C*(CH$_3$)$_2$], 127.3, 127.4, 127.5, 127.6, 127.7, 127.8, 127.9, 128.1, 128.2, 128.4, 128.7, 128.9, 2 × 137.7, 138.0, 138.1, 138.3, 138.4, 138.5 (aromatic). − MS (DCI, ammonia): *m/z* (%) = 741 (35) [M + 18]$^+$, 418 (100).

O-[2-Azido-3,6-di-O-benzyl-2-deoxy-4-O-(2,6-di-O-benzyl-3,4-O-isopropylidene-β-D-galactopyranosyl)]trichloroacetimidate (**7**): To a solution of compound **15** (0.88 g, 1.15 mmol) in dichloromethane (50 ml) at −15°C were added trichloroacetonitrile (3.59 ml, 35.60 mmol) and 1,8-diazabicyclo[5.4.0]undec-7-ene (25 µl, 0.17 mmol). After stirring for 2 h at this temperature, the volatiles were removed and the residue was directly subjected to chromatographic purification (pentane/ethyl acetate, 3:1) to give 1.04 g (99%) of **7** as a white foam. R_f = 0.34 (pentane/ethyl acetate, 5:1). − $[\alpha]_D$ = +3.8 (*c* = 1.0, $CHCl_3$). − ^1H NMR (300 MHz, $CDCl_3$): δ = 1.21, 1.40 [2 s, 6 H, C(CH$_3$)$_2$], 3.11 (dd, *J* = 3.5, 10.0 Hz, 1 H, 2a-H), 3.53−3.81 (m, 6 H, 2b-H, 5a-H, 5b-H, 6a-H$_2$, 6b-H$_a$), 3.92 (t, *J* = 6.5 Hz, 1 H, 4a-H), 4.05−4.30 (m, 5 H, 3a-H, 3b-H, 4b-H, 6b-H$_b$, C*H*Ph), 4.36−4.52 (m, 3 H, 3 × C*H*Ph), 4.61 (d, *J* = 8.0 Hz, 1 H, 1b-H), 4.59−5.27 (m, 4 H, 4 × C*H*Ph), 6.40 (d, *J* = 3.5 Hz, 1 H, 1a-H), 7.00−7.71 (m, 5 H, Ph-H), 8.54 (s, 1 H, NH). − ^{13}C NMR (75 MHz, $CDCl_3$): δ = 26.45, 28.14 [2 × C(*C*H$_3$)$_2$], 62.71 (C-2b), 67.75, 69.40 (C-6a, C-6b), 73.12, 74.15, 74.43, 76.30, 78.78, 80.07, 81.23 (C-2b, C-3a, C-3b, C-4a, C-4b, C-5a, C-5b), 73.37, 73.48, 73.54, 75.50 (4 × *C*H$_2$Ph), 91.63 (CCl_3), 95.08, 102.3 (C-1a, C-1b), 109.9 [*C*(CH$_3$)$_2$], 127.5, 127.6, 127.8, 127.9, 128.1, 128.5, 128.6, 128.8, 129.2, 138.8, 138.9, 139.0, 139.3 (aromatic), 160.7 [*C*(NH)CCl_3]. − MS (FAB, 3-NBA): *m/z* (%) = 912 (100) [M]$^+$.

Benzyl O-(2,6-Di-O-benzyl-3,4-isopropylidene-β-D-galactopyranosyl)-(1→4)-[2-azido-3,6-di-O-benzyl-2-deoxy-β-D-glucopyranosyl-(1→3)-2,4,6-tri-O-benzyl-β-D-galactopyranosyl]-

(1→4)-2,3,6-tri-O-benzyl-β-D-glucopyranoside (**16**): A solution of acceptor **8**[12] (1.08 g, 1.11 mmol) in acetonitrile (15 ml) was stirred together with 3 Å molecular sieves (400 mg) for 90 min at room temperature and then the mixture was treated with trimethylsilyl trifluoromethanesulfonate (35 µl, 0.20 mmol) at −40°C. A cooled solution of donor **7** (1.43 g, 1.57 mmol) in acetonitrile (5 ml) was subsequently added dropwise over a period of 30 min. After stirring for a further 60 min, the reaction was quenched with triethylamine (0.1 ml) and the mixture was filtered. Chromatographic purification (PE/ethyl acetate, 4:1) yielded 1.53 g (80%) of a white foam, which was found to contain traces of acceptor **8**. 0.16 g (15%) of acceptor **8** was additionally recovered. R_f = 0.48 (PE/ethyl acetate, 4:1). − $[\alpha]_D$ = −10.0 (*c* = 1.0, $CHCl_3$). − IR (KBr): $\tilde{\nu}$ = 3062 cm^{-1}, 3032, 2974, 2112, 1118, 1072. − ^1H NMR (500 MHz, C_6D_6): δ = 1.22, 1.39 [2 s, 6 H, $C(CH_3)_2$], 3.09 (ddd, *J* = 1.0, 3.0, 10.0 Hz, 1 H, 5c-H), 3.32 (ddd, *J* = 1.5, 4.0, 10.0 Hz, 1 H, 5b-H), 3.38 (m_c, 2 H, 3c-H, 2c-H), 3.49 (dd, *J* = 5.0, 8.5 Hz, 1 H, 6d-H_a), 3.53 (dd, *J* = 1.0, 11.0 Hz, 1 H, 6c-H_a), 3.56−3.69 (m, 7 H, 2a-H, 2b-H, 3b-H, 5a-H, 5d-H, 6a-H_a, 6d-H_b), 3.71−3.80 (m, 4 H, 3a-H, 4d-H, 6b-H_a, 6a-H_b), 3.85 (dd, *J* = 3.0, 10.0 Hz, 1 H, 3d-H), 3.89 (dd, *J* = 3.5, 11.0 Hz, 1 H, 6c-H_b), 3.96 (t, *J* = 6.0 Hz, 1 H, 4a-H), 4.02 (dd, *J* = 4.0, 11.0 Hz, 6b-H_b), 4.10−4.40 (m, 10 H, 2d-H, 4b-H, 4c-H, 7 × PhC*H*), 4.49 (d, *J* = 8.0 Hz, 1 H, 1a-H), 4.59 (d, *J* = 8.0 Hz, 1 H, 1b-H), 4.51−4.68 (m, 8 H, 1c-H, 7 × PhC*H*), 4.76−5.35 (m, 9 H, 1d-H, 8 × PhC*H*), 7.00−7.70 (m, 55 H, Ph-H). − ^{13}C NMR (125 MHz, $CDCl_3$): δ = 26.44, 28.14 [2 × $C(CH_3)_2$], 67.30 (C-2b), 68.10 (C-6c), 68.61 (C-6d), 68.73 (C-6b), 69.38 (C-6a), 70.84 (CH_2Ph), 73.02 (C-5a), 73.35, 73.37, 73.56, 73.60 (4 × CH_2Ph), 73.94 (C-5d), 74.14 (C-4d), 75.14 (CH_2Ph), 75.45 (C-5c), 75.58 (CH_2Ph), 75.67 (C-5b), 75.69, 75.99, 75.72, 75.80 (4 × CH_2Ph), 76.51 (C-4c), 76.83 (C-4b), 77.37 (C-4a), 79.95 (C-3d), 80.47 (C-2d), 81.39 (C-3b), 81.56 (C-3c), 82.09 (C-3a), 82.36 (C-2b), 83.18 (C-2a), 102.4, 103.0, 103.1, 103.3 (C-1a, C-1b, C-1c, C-1d), 109.9 [$C(CH_3)_2$], 127.3−140.1 (aromatic). − MS (positive FAB, 3-NBA): *m/z* (%) = 1746 (100) $[M + 23]^+$; MS (negative FAB, 3-NBA): *m/z* (%) = 1875 (100) $[M + 153]^-$.

Benzyl O-(2,6-Di-O-benzyl-β-D-galactopyranosyl)-(1→4)-[2-azido-3,6-di-O-benzyl-2-deoxy-β-D-glucopyranosyl-(1→3)-2,4,6-tri-O-benzyl-β-D-galactopyranosyl]-(1→4)-2,3,6-tri-O-benzyl-β-D-glucopyranoside (**6**): A solution of **16** (0.27 g, 0.15 mmol) in aqueous acetic acid (5 ml, 80%) was stirred for 30 min at 100°C. Water (15 ml) was then added and the mixture was extracted with dichloromethane (3 × 15 ml). The combined organic phases were washed with satd. $NaHCO_3$ solution, water, and brine, dried and concentrated. Chromatographic purification yielded 0.21 g (79%) of **6** as a white foam. R_f = 0.25 (PE/ethyl acetate, 2:1). − $[\alpha]_D$ = −2.4 (*c* = 0.5, $CHCl_3$). − IR (KBr): $\tilde{\nu}$ = 3438 cm^{-1}, 3032, 2922, 2112, 1092. − ^1H NMR (500 MHz, C_6D_6): δ = 3.05 (m_c, 1 H, 5c-H), 3.26 (br. t, *J* = 6.0 Hz, 1 H, 5b-H), 3.28−3.70 (m, 15 H, 2a-H, 2b-H, 2c-H, 3a-H, 3b-H, 3c-H, 4a-H, 4d-H, 5a-H, 5d-H, 6a-H_2, 6b-H_2, 6c-H_a), 3.78 (dd, *J* = 1.5, 11.5 Hz, 1 H, 6d-H_a), 3.81 (dd, *J* = 4.0, 10.5 Hz, 1 H, 6c-H_b), 3.85 (dd, *J* = 3.0, 9.5 Hz, 1 H, 3d-H), 4.02 (dd, *J* = 4.0, 10.5 Hz, 1 H, 6d-H_b), 4.10−4.40 (m, 9 H, 2d-H, 4b-H, 4c-H, 7 × PhC*H*), 4.46 (d, *J* = 12.5 Hz, 1 H, PhC*H*), 4.49 (d, *J* = 7.5 Hz, 1 H, 1a-H), 4.56 (d, *J* = 7.0 Hz, 1 H, 1b-H), 4.59−4.68 (m, 5 H, 1c-H, 4 × PhC*H*), 4.77 (d, *J* = 8.0 Hz, 1 H, 1d-H), 4.80−5.35 (m, 10 H, 10 PhC*H*), 7.00−7.70 (m, 55 H, Ph-H). − ^{13}C NMR (125 MHz, C_6D_6): δ = 67.38 (C-2c), 68.10 (C-6c), 68.61 (C-6d), 68.72 (C-6b), 69.27 (C-6a, C-3d), 70.84 (CH_2Ph), 73.35 (C-5a, CH_2Ph), 73.56, 73.60, 73.82, 73.92, 73.97, 75.13 (6 × CH_2Ph), 75.40 (C-5c), 75.46, 75.68 (CH_2Ph), 75.74 (C-5b), 75.79 (CH_2Ph), 76.50 (C-4c), 76.82 (C-4b), 77.40 (C-4b), 80.44 (C-2d), 80.58 (C-2a, C-3a), 81.44 (C-3c), 82.36 (C-2b), 83.16 (C-3b), 103.0, 103.1, 103.2, 103.3 (C-1a, C-1b, C-1c, C-1d), 127.3−140.1 (aromatic). − MS (negative FAB, 3-NBA): *m/z* (%) = 1682 (100) $[M]^-$. − $C_{101}H_{107}N_3O_{20}$ (1682.9): calcd. C 72.08, H 6.41; found C 72.02, H 6.66.

Benzyl O-[Methyl-(5-acetamido-4,7,8,9-tetra-O-acetyl-3,5-dideoxy-α-D-glycero-D-galacto-2-nonulopyranosyl)onate]-(2→3)-O-(2,6-di-O-benzyl-β-D-galactopyranosyl)-(1→4)-[2-azido-3,6-di-O-benzyl-2-deoxy-β-D-glucopyranosyl-(1→3)-2,4,6-tri-O-benzyl-β-D-galactopyranosyl]-(1→4)-2,3,6-tri-O-benzyl-β-D-glucopyranoside (**17**): A solution of acceptor **6** (0.67 g, 0.40 mmol) and donor **5**[9] (0.47 g, 0.80 mmol) in a 2:1 mixture of acetonitrile and dichloromethane (13.5 ml) was stirred together with 3 Å and 4 Å molecular sieves (0.5 g) for 1 h at room temperature. Under exclusion of light, the mixture was cooled to −50°C and 2,6-di-*tert*-butyl-4-methylpyridine (0.17 g, 0.83 mmol) and silver triflate (0.20 g, 0.78 mmol) were added. Finally, at −70°C, a solution of phenylsulfuryl chloride (92 µl, 0.2 mmol) in 0.5 ml of dichloromethane was added in 50 µl portions at 5-min intervals by running the solution down the cold wall of the reaction flask. After stirring for a further 30 min at −70°C, the reaction mixture was diluted with ethyl acetate (50 ml) containing triethylamine (0.1 ml), filtered, and the filtrate was washed with water and brine. Drying, evaporation of the solvent, and chromatographic purification (pentane/ethyl acetate, 1:1 → pure ethyl acetate) yielded 0.50 g (58%) of **17** as a white foam. In addition, 0.24 g (36%) of acceptor **6** was recovered. R_f = 0.48 (pure ethyl acetate). − $[\alpha]_D$ = −10.0 (*c* = 0.5, $CHCl_3$). − IR (KBr): $\tilde{\nu}$ = 3400 cm^{-1}, 3032, 2922, 2112, 1368, 1224, 1060. − ^1H NMR (500 MHz, C_6D_6): δ = 1.55, 1.61, 1.72, 1.80, 2.06 (5 s, 15 H, 5 × Ac), 2.11 (t, *J* = 13.0 Hz, 1 H, 3e-H_{ax}), 2.69 (dd, *J* = 4.5, 13.0 Hz, 1 H, 3e-H_{eq}), 2.81 (br. s, 1 H, OH), 3.08 (ddd, *J* = 1.5, 3.5, 9.5 Hz, 1 H, 5c-H), 3.31 (m_c, 1 H, 5b-H), 3.28−3.35 (m, 2 H, 2c-H, 3c-H), 3.47 (s, 3 H, CO_2Me), 3.49 (dd, *J* = 5.5, 9.0 Hz, 1 H, 6d-H_a), 3.55−3.65 (m, 7 H, 2a-H, 2b-H, 3b-H, 5a-H, 5d-H, 6a-H_a, 6d-H_b), 3.74−4.02 (m, 6 H, 3d-H, 4d-H, 5e-H, 6a-H_b, 6b-H_b, 6c-H_b), 3.94 (dd, *J* = 4.0, 11.0 Hz, 1 H, 4a-H), 4.12 (dd, *J* = 7.5, 9.5 Hz, 1 H, 2d-H), 4.20 (dd, *J* = 6.0, 12.0 Hz, 1 H, 9e-H_a), 4.23−4.40 (m, 8 H, 4b-H, 4c-H, 6 × PhC*H*), 4.45−4.65 (m, 5 H, 1b-H, 1c-H, 3 × PhC*H*), 4.67 (dd, *J* = 2.5, 12.5 Hz, 1 H, 9e-H_b), 4.74 (d, *J* = 8.0 Hz, 1 H, 1a-H), 4.80 (d, *J* = 12.0 Hz, 1 H, PhC*H*), 4.76−5.33 (m, 14 H, 1d-H, 4e-H, 12 × PhC*H*), 5.45 (dd, *J* = 2.0, 8.0 Hz, 1 H, 7e-H), 5.79 (m_c, 1 H, 8e-H), 7.10−7.89 (m, 55 H, Ph-H). − ^{13}C NMR (125 MHz, $CDCl_3$): δ = 20.30, 20.38, 20.60, 21.08, 22.90 (5 × Ac), 37.29 (C-3e), 49.10 (C-5e), 52.56 (CO_2CH_3), 62.86 (C-9e), 67.42 (C-7e), 67.44 (C-2c), 68.08 (C-6), 68.33 (C-6d), 68.65 (C-4e), 68.85 (C-6b), 69.25 (C-6c), 69.73 ($PhCH_2$), 70.84 (C-8e), 73.33 (C-5a), 73.38, 73.51, 73.58, 73.97 (4 × $PhCH_2$), 75.12 (C-6e), 75.36 (C-5c), 75.45, 75.60, 75.65 (3 × $PhCH_2$), 75.74 (C-5b), 75.82 ($PhCH_2$), 76.23 (C-4c), 76.83 (C-4b), 76.92 ($PhCH_2$), 77.50 (C-4a), 79.17, 80.42 (C-2d, C-3b), 81.52 (C-3c), 82.23 (C-3a), 82.38 (C-2a), 83.20 (C-2b), 98.77 (C-2e), 102.9, 103.0, 103.2, 103.3 (C-1a, C-1b, C-1c, C-1d), 127.2−170.2 (aromatic, Ac). − MS (negative FAB, 3-NBA): *m/z* (%) = 2309 (100) $[M + 153]^-$. − $C_{121}H_{134}N_4O_{32}\cdot 2H_2O$ (2156.4 + 2 × 18.0): calcd. C 66.29, H 6.34; found C 66.16, H 6.29.

Acetyl O-[Methyl-(5-acetamido-4,7,8,9-tetra-O-acetyl-3,5-dideoxy-α-D-glycero-D-galacto-2-nonulopyranosyl)onate]-(2→3)-O-(2,6-di-O-acetyl-β-D-galactopyranosyl)-(1→4)-[2-acetamido-3,6-di-O-acetyl-2-deoxy-β-D-glucopyranosyl-(1→3)-2,4,6-tri-O-acetyl-β-D-galactopyranosyl]-(1→4)-2,3,6-tri-O-acetyl-α,β-D-glucopyranoside (**18**): A mixture of compound **17** (40 mg, 0.019 mmol) and palladium hydroxide on carbon (80 mg, 20% Pd) in methanol (20 ml) was shaken under hydrogen (5 bar) for 24 h at room temperature. After filtration through Celite, the organic solvent was removed. The residue was taken up in a 2:1 mixture of pyridine

and acetic anhydride (7 ml), the resulting solution was stirred for 18 h at room temperature, and then poured onto crushed ice (50 ml). The mixture was extracted with dichloromethane (3 × 20 ml). The combined organic phases were washed with 1 N HCl, water, and brine, dried and concentrated. Chromatographic purification (ethyl acetate/acetone, 2:1) of the residue gave 25 mg (80%) of **18** as a 1:1 mixture of the anomers. R_f = 0.41 (ethyl acetate/acetone, 2:1). − $[\alpha]_D$ = −4.7 (*c* = 0.5, $CHCl_3$). − IR (KBr): $\tilde{\nu}$ = 1748 cm^{-1}, 1372, 1230, 1130. − ^1H NMR (500 MHz, $CDCl_3$): δ = 1.68 (t, *J* = 12.4 Hz, 1 H, 3e-H_{ax}), 1.82−2.25 (m, 54 H, 18 × Ac), 2.58 (dd, *J* = 4.5, 12.8 Hz, 1 H, 3e-H_{eq}), 3.85 (s, 3 H, CO_2H), 5.55 (ddd, *J* = 2.8, 5.5, 8.9 Hz, 1 H, 8e-H), 5.66 (d, *J* = 8.2 Hz, 1 H, 1a-Hα), 6.25 (d, *J* = 3.9 Hz, 1 H, 1a-Hβ). − ^{13}C NMR (125 MHz, $CDCl_3$): δ = 89.41, 91.98, 100.8, 101.42, 101.70 (C-1aα, C-1aβ, C-1b, C-1c, C-1d). − MS (DCI, ammonia): *m/z* (%) = 1703 (100) $[M + 18]^+$. − $C_{70}H_{96}N_2O_{45}\cdot H_2O$ (1685.5 + 18.0): calcd. C 49.35, H 5.80; found C 49.36, H 5.66.

O-[Methyl-(5-acetamido-4,7,8,9-tetra-O-acetyl-3,5-dideoxy-α-D-glycero-D-galacto-2-nonulopyranosyl)onate]-(2→3)-O-(2,6-di-O-acetyl-β-D-galactopyranosyl)-(1→4)-[2-acetamido-3,6-di-O-acetyl-2-deoxy-β-D-glucopyranosyl-(1→3)-2,4,6-tri-O-acetyl-β-D-galactopyranosyl]-(1→4)-2,3,6-tri-O-acetyl-α.β-D-glucopyranose (**19**): A solution of compound **18** (195 mg, 0.116 mmol) in DMF (2 ml) was treated with hydrazinium acetate (13 mg, 0.14 mmol) and the mixture was stirred at 45°C for 30 min. The solution was then diluted with dichloromethane (40 ml), washed with brine, and dried. Evaporation of the solvents and chromatographic purification (acetone/toluene, 3:2) of the residue yielded 176 mg (92%) of a white foam. R_f = 0.22 (acetone/toluene, 2:1). − ^1H NMR (500 MHz, $CDCl_3$): δ = 1.59−2.39 (m, 55 H, 18 × Ac, 3e-H_{ax}), 2.81 (dd, *J* = 4.2, 12.5 Hz, 1 H, 3e-H_{eq}), 3.76 (s, 3 H, CO_2Me), 5.89−6.96 (m, 1 H, 8e-H). − MS (DCI, ammonia): *m/z* (%) = 1661 (32) $[M + 18]^+$, 1643 (8) $[M]^+$, 1373 (100). − $C_{68}H_{94}N_2O_{44}\cdot H_2O$ (1643.5 + 18.0): calcd. C 49.16, H 5.82; found C 49.33, H 5.58.

O-[Methyl-(5-acetamido-4,7,8,9-tetra-O-acetyl-3,5-dideoxy-α-D-glycero-D-galacto-2-nonulopyranosyl)onate]-(2→3)-O-(2,6-di-O-acetyl-β-D-galactopyranosyl)-(1→4)-[2-acetamido-3,6-di-O-acetyl-2-deoxy-β-D-glucopyranosyl-(1→3)-2,4,6-tri-O-acetyl-β-D-galactopyranosyl]-(1→4)-2,3,6-tri-O-acetyl-α-D-glucopyranosyl Trichloroacetimidate (**2**): To a solution of compound **19** (137 mg, 0.077 mmol) in dichloromethane (5 ml), trichloroacetonitrile (0.32 ml, 3.09 mmol) and 1,8-diazabicyclo[5.4.0]undec-7-ene (18 µl, 0.113 mmol) were added at 0°C. After stirring for 2 h at room temperature, the volatiles were removed. Chromatographic purification (acetone/pentane, 3:2) of the residue gave 137 mg (75%) of **2** as a yellowish foam. R_f = 0.33 (acetone/pentane, 3:2). − $[\alpha]_D$ = −10.3 (*c* = 1.0, $CHCl_3$). − IR (KBr): $\tilde{\nu}$ = 1750 cm^{-1}, 1372, 1228, 1050. − ^1H NMR (500 MHz, C_6D_6): δ = 1.58−2.37 (m, 52 H, 17 × Ac, 3e-H_{ax}), 2.81 (dd, *J* = 4.5, 12.2 Hz, 1 H, 3e-H_{eq}), 3.38−3.46 (m, 1 H, 5c-H), 3.52 (dd, *J* = 3.0, 12.0 Hz, 1 H, 6e-H), 3.61 (dt, *J* = 8.2, 9.5 Hz, 1 H, 2c-H), 3.75−3.82 (m, 5 H, 5b-H, 5e-H, CH_2OMe), 3.86−3.91 (m, 2 H, 4a-H, 4b-H), 4.03 (t, *J* = 9.0 Hz, 1 H, 4c-H), 4.14−4.38 (m, 10 H, 5a-H, 5d-H, 6b-H_2, 6c-H_2, 6d-H_2, 9e-H_a), 4.45 (m, 1 H, 6a-H_a), 4.82 (d, *J* = 8.0 Hz, 1 H, 5e-NH), 4.65 (m, 1 H, 6a-H_b), 4.72 (dd, *J* = 2.8, 12.5 Hz, 1 H, 9e-H_b), 4.83 (d, *J* = 8.0 Hz, 1c-H), 4.85 (dt, *J* = 4.0, 10.0 Hz, 1 H, 4e-H), 4.91 (m, 1 H, 3b-H), 4.96 (dd, *J* = 3.1, 10.5 Hz, 1 H, 3d-H), 5.05 (d, *J* = 8.2 Hz, 1 H, 2c-NH), 5.15 (d, *J* = 8.0 Hz, 1d-H), 5.27 (br. d, *J* ≈ 3 Hz, 1 H, 4d-H), 5.33 (dd, *J* = 3.5, 10.0 Hz, 1 H, 2a-H), 5.42 (dd, 2.8, 9.0 Hz, 1 H, 7e-H), 5.49 (dd, *J* = 8.0, 10.0 Hz, 1 H, 2b-H), 5.55−5.63 (m, 3 H, 1b-H, 2d-H, 3c-H), 5.92 (m_c, 1 H, 8e-H), 6.00 (t, *J* = 10.0 Hz, 3a-H), 6.82 (d, *J* = 3.5 Hz, 1 H, 1a-H), 8.47 (br. s, 1 H, NH). − ^{13}C NMR (125 MHz, $CDCl_3$): δ = 20.01, 20.07, 3 × 20.31, 20.48, 20.64, 3 × 20.80, 20.92, 3 × 21.01, 21.53, 22.98, 23.20 (17 × Ac), 37.67 (C-3e), 48.82 (C-5e), 52.93 (CO_2CH_3), 61.68, 61.70, 62.72, 62.86, 62.93 (C-6a, C-6b, C-6c, C-6d, C-9e), 67.01, 67.68, 68.66, 69.27, 69.37, 70.27, 70.38, 70.57, 70.91, 71.29, 71.31, 71.51, 71.80, 72.02, 72.40, 73.01, 73.15, 76.20, 76.71 (C-2a−d, C-3a−d, C-4a−e, C-5a−d, C-6e, C-7e, C-8e), 91.42 (CCl_3), 93.55, 100.7, 101.4, 101.8 (C-1a−d), 97.29 (CO_2CH_3), 161.5 [C(NH)CCl_3], 168.9−170.1 (17 × Ac). − MS (DCI, ammonia): *m/z* (%) = 1805 (10) $[M + 18]^+$, 1661 (12) $[M - OC(NH)CCl_3 + 18]^+$, 1642.9 (100) $[M - OC(NH)CCl_3]^+$. − Compound **2** was used for the next step without further purification.

O-[Methyl-(5-acetamido-4,7,8,9-tetra-O-acetyl-3,5-dideoxy-α-D-glycero-D-galacto-2-nonulopyranosyl)onate]-(2→3)-O-(2,6-di-O-acetyl-β-D-galactopyranosyl)-(1→4)-[2-acetamido-3,6-di-O-acetyl-2-deoxy-β-D-glucopyranosyl-(1→3)-2,4,6-tri-O-acetyl-β-D-galactopyranosyl]-(1→4)-2,3,6-tri-O-acetyl-β-D-glucopyranosyl-(1→1)-(2S,3R,4E)-2-azido-3-O-benzoyl-4-octadecene-1,3-diol (**20**): A solution of imidate **2** (100 mg, 0.056 mmol) and the azidosphingosine derivative **3**[19] (60 mg, 0.140 mmol) in dichloromethane (3 ml) was stirred together with 4 Å molecular sieves (100 mg) for 2 h at room temperature. After cooling to 0°C, $BF_3\cdot OEt_2$ (24 µl, 0.200 mmol) was added, and stirring was continued for a further 2 h at room temperature. The mixture was then filtered and the filtrate was washed with satd. $NaHCO_3$ solution, water and brine, and then dried. Evaporation of the solvent gave a residue, which was purified by flash chromatography (toluene/acetone, 3:2 → 1:2) yielding 50 mg (43%) of **20** as a white foam. R_f = 0.45 (acetone/toluene, 3:2). − ^1H NMR (500 MHz, C_6D_6): δ = 0.92 (br. t, *J* = 7.0 Hz, 3 H, CH_3), 1.30 (br. s, 22 H, CH_2), 1.59−2.35 (m, 54 H, 17 × Ac, 6′-H_2, 3e-H_{eq}), 2.59 (dd, *J* = 4.5, 12.5 Hz, 1 H, 3e-H_{ax}), 3.20 (m, 1 H, 1′-H_a), 3.39 (m, 1 H, 5c-H), 3.48−3.84 (m, 11 H, 1′-H_b, 2′-H, 2c-H, 4a-H, 4b-H, 5b-H, 5e-H, 6e-H, CO_2Me), 4.01 (t, *J* = 9.0 Hz, 1 H, 4c-H), 4.19−4.36 (m, 11 H, 5a-H, 5d-H, 6a-H_2, 6b-H_2, 6c-H_2, 6d-H_2, 9e-H_a), 4.70 (dd, *J* = 2.5, 12.5 Hz, 1 H, 9e-H_b), 4.78 (d, *J* = 8.0 Hz, 1 H, 1c-H), 4.82 (dt, *J* = 4.5, 11.0 Hz, 1 H, 4e-H), 4.89 (m, 1 H, 3b-H), 4.94 (dd, *J* = 3.0, 10.5 Hz, 1 H, 3d-H), 5.14 (d, *J* = 8.0 Hz, 1 H, 1d-H), 5.26 (d, *J* = 3.0 Hz, 1 H, 4d-H), 5.30 (dd, *J* = 8.0, 9.0 Hz, 1 H, 2a-H), 5.40 (m, 2 H, 3a-H, 7e-H), 5.48 (dd, *J* = 8.0, 10.0 Hz, 1 H, 2b-H), 5.52−5.60 (m, 3 H, 1b-H, 2d-H, 3c-H), 5.66 (br. dd, *J* = 7.0, 15.0 Hz, 1 H, 4′-H), 5.90 (m_c, 1 H, 8e-H), 5.93 (dd, *J* = 3.0, 8.0 Hz, 1 H, 3′-H), 6.00 (dt, *J* = 15.0, 7.0 Hz, 1 H, 5′-H), 7.10, 8.22 (2 m, 5 H, Ph-H). − MS (positive FAB, 3-NBA): *m/z* (%) = 2055 (81) $[M]^+$, 2077 (100) $[M + Na]^+$. − Compound **20** was used for the next step without further purification.

O-[Methyl-(5-acetamido-4,7,8,9-tetra-O-acetyl-3,5-dideoxy-α-D-glycero-D-galacto-2-nonulopyranosyl)onate]-(2→3)-O-(2,6-di-O-acetyl-β-D-galactopyranosyl)-(1→4)-[2-acetamido-3,6-di-O-acetyl-2-deoxy-β-D-glucopyranosyl-(1→3)-2,4,6-tri-O-acetyl-β-D-galactopyranosyl]-(1→4)-2,3,6-tri-O-acetyl-β-D-glucopyranosyl-(1→1)-(2S,3R,4E)-3-O-benzoyl-2-octadecanamido-4-octadecene-1,3-diol (**21**): A mixture of **20** (50 mg, 0.024 mmol), stearine anhydride (40 mg, 0.072 mmol) and Lindlar's catalyst (100 mg) in ethyl acetate (3 ml) was stirred under hydrogen (1 bar) for 8 h. Subsequent filtration through a pad of Celite and evaporation of the solvent yielded a residue, which was purified by flash chromatography (acetone/toluene, 1:1) to give 50 mg (91%) of **21** as a white solid. R_f = 0.47 (acetone/toluene, 3:2). − ^1H NMR (500 MHz, C_6D_6): δ = 0.91 (m, 6 H, 2 × CH_3), 1.30, 1.34 (2 br. s, 54 H, CH_2), 1.58−2.32 (m, 54 H, 17 × Ac, 6′-H_2, 3e-H_{eq}), 2.78 (dd, *J* = 4.5, 12.5 Hz, 1 H, 3e-H_{ax}), 3.12 (m, 1 H, 1′-H_a), 3.40 (m, 1 H, 5c-H), 3.49−3.77 (m, 9 H, 2c-H, 4a-H, 4b-H, 5b-H, 5e-H, 6e-H, CO_2Me), 3.86−4.02 (m, 3 H, 1′-H_b, 2′-H, 4c-H), 4.11−4.35 (m, 11 H, 5a-H,

5d-H, 6a-H_2, 6b-H_2, 6c-H_2, 6d-H_2, 9e-H_a), 4.70 (dd, J = 2.5, 12.5 Hz, 1 H, 9e-H_b), 4.74−4.91 (m, 3 H, 1c-H, 3b-H, 4e-H), 4.93 (dd, J = 3.0, 10.5 Hz, 1 H, 3d-H), 5.10 (d, J = 8.0 Hz, 1 H, 1d-H), 5.24 (d, J = 3.0 Hz, 1 H, 4d-H), 5.34−5.58 (m, 7 H, 1b-H, 2a-H, 2b-H, 2d-H, 3′-H, 3a-H, 3c-H), 5.66 (br. dd, J = 7.0, 15.0 Hz, 1 H, 4′-H), 5.87−5.95 (m, 2 H, 5′-H, 8e-H), 7.15, 8.30 (2 m, 5 H, Ph-H). − MS (negative FAB, 3-NBA): *m/z* (%) = 2449 (100) [M + matrix]$^-$, 2330 (85) [M + matrix − OC(O)Ph]$^+$, 2407 (20) [M + matrix − OC(O)CH_3]$^+$. − Compound **21** was used for the next step without further purification.

O-[(5-Acetamido-3,5-dideoxy-α-D-glycero-D-galacto-2-nonulopyranosyl)onic acid]-(2→3)-O-(β-D-galactopyranosyl)-(1→4)-[2-acetamido-2-deoxy-β-D-glucopyranosyl-(1→3)-β-D-galactopyranosyl]-β-D-glucopyranosyl-(1→1)-(2S,3R,4E)-2-octadecanamido-4-octadecene-1,3-diol (LM1, **1**): A solution of **21** (50 mg, 0.022 mmol) in methanol (6 ml) was stirred with sodium methoxide (45 µl of a 5.4 N solution in methanol, 0.024 mmol) for 17 h at room temperature. Water (0.4 ml) was then added, and stirring was continued for a further 4 h. The mixture was then neutralized with Amberlyte IR-120 resin (H^+ form) and filtered. Chromatographic purification (chloroform/methanol/water, 60:60:5) and lyophilization yielded 32 mg (95%) of **1** as a white solid. R_f = 0.33 (chloroform/methanol/water, 60:60:5). − ^{1}H NMR (500 MHz, [D_6]DMSO/D_2O, 98:2): δ = 0.85 (br. t, J = 7.0 Hz, 6 H, 2 × CH_3), 1.23 (br. s, 50 H, CH_2), 1.46 [m, 2 H, C(O)CH_2CH_2], 1.81, 1.88 (2 s, 6 H, 2 × Ac), 2.03 [m, 2 H, C(O)CH_2], 2.75 (dd, J = 5.0, 12.0 Hz, 1 H, 3e-H_{eq}), 3.04 (m, 1 H, 2a-H), 3.86 (d, J = 2.8 Hz, 1 H, 4d-H), 3.92 (m, 1 H, 3′-H), 4.16 (d, J = 8.0 Hz, 1 H, 1a-H), 4.21 (d, J = 7.9 Hz, 1 H, 1d-H), 4.27 (d, J = 7.1 Hz, 1 H, 1b-H), 4.67 (d, J = 8.5 Hz, 1 H, 1c-H), 5.37 (br. dd, J = 7.0, 15.5 Hz, 1 H, 4′-H), 5.54 (br. dt, J = 7.0, 15.5 Hz, 1 H, 5′-H). − The spectral data were in complete agreement with those reported in the literature.[8]

[1] [1a] Y. C. Lee, R. T. Lee, *Acc. Chem. Rev.* **1995**, *28*, 321−327. − [1b] H. F. Oettgen, *Gangliosides and Cancer*, 1st ed., VCH, Weinheim, **1989**.

[2] [2a] A. Hasegawa, M. Kiso in *Preparative Carbohydrate Chemistry* (Ed.: S. Hanessian), Dekker, New York, **1997**, 357−379. − [2b] G.-J. Boons, *Tetrahedron* **1996**, *4*, 1095−1121. − [2c] R. R. Schmidt, *A.C.S. Symp. Ser.* **1994**, *560 (Synthetic Oligosaccharides)*, 276−296.

[3] [3a] S. Kusunoki, A. Chiba, K. Kon, S. Ando, K. Arisawa, A. Tate, I. Kanazawa, *Ann. Neurol.* **1994**, *35*, 570−576. − [3b] P. Fredman, C. A. Vedeler, H. Nyland, J. A. Aarli, L. Svennerholm, *J. Neurol.* **1991**, *238*, 75−79.

[4] T. Ariga, R. V. Tao, B. C. Lee, M. Yamawaki, H. Yoshino, N. J. Scarsdale, T. Kasama, Y. Kushi, R. K. Yu, *J. Biol. Chem.* **1994**, *269*, 2667−2675.

[5] C. C. Sung, D. K. Pearl, S. W. Coons, B. W. Scheithauer, P. C. Johnson, M. Zheng, A. J. Yates, *Cancer* **1995**, *75*, 851−859.

[6] [6a] L. Schaade, K. Ritter, *J. Planar Chromatogr.* **1995**, *8*, 362−365. − [6b] K. Ritter, L. Schaade, R. Thomssen, *J. Planar Chromatogr.* **1991**, *4*, 150−153.

[7] K. Ritter, R. Thomssen, personal communication.

[8] A. Kameyama, H. Ishida, M. Kiso, A. Hasegawa, *Carbohydr. Res.* **1990**, *200*, 269−285.

[9] [9a] Sinaÿ, Pierre, *Phosphorus, Sulphur, Silicon Relat. Elem.* **1994**, *96*, 89−102. − [9b] V. Martichonok, G. M. Whitesides, *J. Org. Chem.* **1996**, *61*, 1702−1706.

[10] L. F. Tietze, H. Keim, *Angew. Chem.* **1997**, *109*, 1704−1706; *Angew. Chem. Int. Ed. Engl.* **1997**, *36*, 1615−1617.

[11] R. U. Lemieux, R. M. Ratcliffe, *Can. J. Chem.* **1979**, *57*, 1244−1251.

[12] K.-H. Jung, M. Hoch, R. R. Schmidt, *Liebigs Ann. Chem.* **1989**, 1099−1106.

[13] R. R. Schmidt, *Angew. Chem.* **1986**, *98*, 213−236; *Angew. Chem. Int. Ed. Engl.* **1986**, *25*, 212−235.

[14] [14a] A. J. Ratcliffe, B. Fraser-Reid, *J. Chem. Soc., Perkin Trans. 1* **1990**, 746−751. − [14b] R. R. Schmidt, M. Behrendt, A. Toepfer, *Synlett* **1990**, 694−696. − [14c] Y. D. Vankar, P. S. Vankar, M. Behrendt, R. R. Schmidt, *Tetrahedron* **1991**, *48*, 9985−9992.

[15] Y. Nagao, T. Nekado, K. Ikeda, K. Achiwa, *Chem. Pharm. Bull.* **1995**, *43*, 1536−1542.

[16] K. Okamoto, T. Kondo, T. Goto, *Bull. Chem. Soc. Jpn.* **1987**, *60*, 637−643.

[17] W. M. Pearlman, *Tetrahedron Lett.* **1967**, *17*, 1663−1664.

[18] G. Excoffier, D. Gagnaire, J.-P. Utille, *Carbohydr. Res.* **1975**, *39*, 368−373.

[19] R. R. Schmidt in *Comprehensive Organic Chemistry* (Eds.: B. M. Trost, I. Fleming), 1st ed., Pergamon Press, Oxford, **1991**, 33−63.

[20] T. K. Park, I. J. Kim, S. Hu, M. T. Bilodeau, J. T. Randolph, O. Kwon, S. J. Danishefsky, *J. Am. Chem. Soc.* **1996**, *118*, 11488−11500.

[21] G. Zemplén, *Ber. Dtsch. Chem. Ges.* **1927**, *60*, 1555−1564.

[22] G. Grundler, R. R. Schmidt, *Liebigs Ann. Chem.* **1984**, 1826−1847.

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