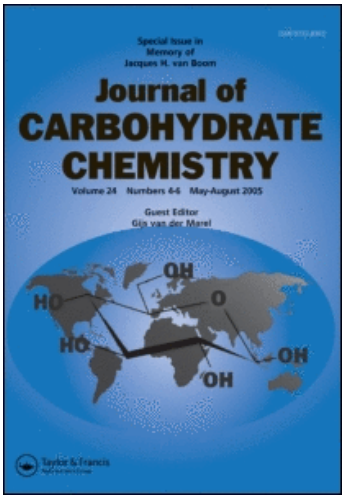


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# Synthetic Studies on Sialoglycoconjugates 50: Total Synthesis of Ganglioside GD2

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*COMMUNICATION*

# SYNTHETIC STUDIES ON SIALOGLYCOCONJUGATES 50: TOTAL SYNTHESIS OF GANGLIOSIDE GD2

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As more and more biological functions[1-10] of gangliosides are being revealed, their facile, stereocontrolled synthesis is strongly required. We have developed[11-14] an α-stereoselective glycosylation of sialic acids, α-sialyl-(2→8)-sialic acid and α-sialyl-(2→8)-α-sialyl-(2→8)-sialic acid, by using their 2-thioglycosides as the glycosyl donor and suitably protected acceptors, and dimethyl(methylthio)sulfonium triflate (DMTST) or *N*-iodosuccinimide (NIS)-trifluoromethanesufonic acid (or TMS triflate) as the glycosyl promoter in acetonitrile. In this way, we have synthesized a variety of gangliosides[15] and their analogs.[16] Previously,[13] we synthesized Ganglioside GD3 containing α-sialyl-(2-8)-sialic acid residue in the molecule, in connection with a novel approach for systematic synthesis of polysialo-glycoconjugates. As a part of our continuing studies on the synthesis and elucidation of the functions of gangliosides, we describe here a facile, stereocontrolled, total synthesis of ganglioside GD2. Ganglioside GD2, which was first isolated from human brain by R. Kuhn et al.,[17] is well known as a human melanoma associated antigen.[18]

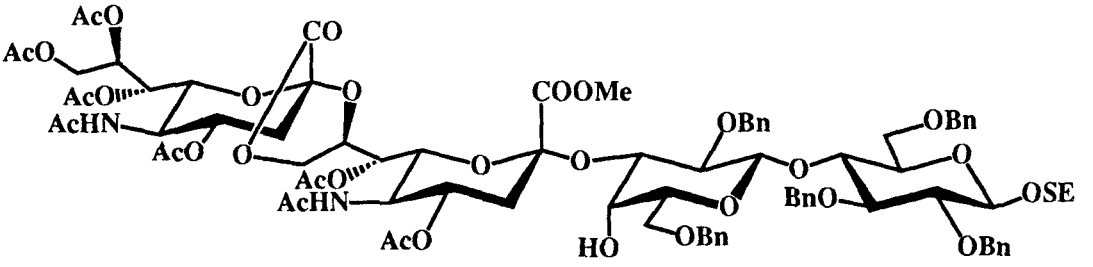
Glycosylation of 2-(trimethylsilyl)ethyl [methyl 5-acetamido-4,7-di-*O*-acetyl-3,5-dideoxy-8-*O*-(5-acetamido-4,7,8,9-tetra-*O*-acetyl-3,5-dideoxy-D-*glycero*-α-D-*galacto*-2-nonulopyrano-1',9-lactone]-D-*glycero*-α-D-*galacto*-2-nonulopyranosylonate]-(2→3)-*O*-(2,6-di-*O*-benzyl-β-D-galactopyranosyl)-(1→4)-2,3,4-tri-*O*-benzyl-β-D-glucopyranoside [13] (**1**) with methyl 6-*O*-benzoyl-2-deoxy-3,4-*O*-isopropylidene-2-phthalimido-1-thio-β-D-galactopyranoside[19] (**2**) in dichloromethane for 12 h at room temperature in the presence of *N*-iodosuccinimide (NIS)-trifluoromethanesulfonic acid (TfOH), gave the desired β-glycoside **3** {$[\alpha]_D$ +10.3° ($CHCl_3$)} in 84% yield. The structure of **3** was

unambiguously proved by 270 MHz $^1$H NMR spectroscopy. Significant signals were a one-proton doublet of doublets at δ 3.37 ($J_{1,2}$ = 7.7 Hz, $J_{2,3}$ = 9.0 Hz) due to H-2e and a one-proton doublet at δ 4.34 due to H-1e, indicating the newly formed glycosidic linkage to be β. Other $^1$H NMR data are consistent with the structure assigned. *O*-Deisopropylidenation of **3** with aqueous 80% acetic acid at 50 °C gave **4** {$[\alpha]_D$ +6.8°($CHCl_3$)} in 80% yield. Catalytic hydrogenolysis (10% Pd-C) in 1: 1 EtOH-$CH_3COOH$ of the benzyl group in **4** for 3 days at 45 °C and subsequent *O*-acetylation gave compound **5** {$[\alpha]_D$ -1.2°($CHCl_3$)} in 85% yield. The $^1$H NMR spectrum of **5** showed the presence of fifteen, three-proton signals at δ 1.85-2.23 (2AcN and 13 AcO), a three-proton singlet at δ 3.85 (MeO), and multiplets at δ 7.43-8.12 due to nine aromatic protons, indicating the structure assigned. Treatment[20] of **5** with trifluoroacetic acid in dichloromethane for 2 h at room temperature gave the 1-hydroxy compound **6** in 94% yield. When treated with trichloroacetonitrile in dichloromethane in the presence of 1,8-diazabicyclo[5.4.0]undec-7-ene for 2 h at 0 °C, compound **6** gave the α-trichloroacetimidate **7** {$[\alpha]_D$ +14.0° ($CHCl_3$)} in 76% yield, after column chromatography. Significant signals in the $^1$H NMR spectrum of **7** were at δ 6.49 ($J_{1,2}$ = 3.8 Hz, H-1a) and at δ 8.66 (s, C=NH), indicating the configuration of the imidate to be α. Glycosylation[21,22] of (2*S*,3*R*,4*E*)-2-azido-3-*O*-benzoyl-4-octadecene-1,3-diol[22,23] (**8**) in dichloromethane by **7** in the presence of TMS triflate and molecular sieves 4Å (AW 300) at 0 °C, yielded the expected β-glycoside **9** {$[\alpha]_D$ -5.8° ($CHCl_3$)} in 81% yield. Selective reduction[22,24] of the azide group in **9** with $H_2S$ gas in 5:1 pyridine-water gave the amine, which on condensation with octadecanoic acid by use of 1-(3-dimethylaminopropyl)-3-ethylcarbodiimide hydrochloride (WSC) in dichloromethane gave the protected ganglioside GD2 **10** {$[\alpha]_D$ +1.2° ($CHCl_3$)} in 85% yield.

Finally, *O*-deacylation of **10** with sodium methoxide in methanol, subsequent hydrolysis of the methyl ester and lactone, and treatment with ethylenediamine in *n*-butyl alcohol for 1 h at 70 °C, followed by *N*-acetylation with acetic anhydride in methanol, yielded ganglioside GD2 (**11**) {$[\alpha]_D$ -3.5° (5:5:1 $CHCl_3$-MeOH-$H_2O$)} in 72% yield. The $^1$H NMR data of **11** in 98:2 $CD_3SOCD_3$-$D_2O$ included δ 0.89 (t, 6H, 2*Me*$CH_2$), 1.27 (s, 52H, 26$CH_2$), 1.98, 2.03 (2) (3s, 3AcN), 5.45 (dd, 1H, $J_{3,4}$ = 7.5 Hz, $J_{4,5}$ = 15.4 Hz, H-4 of sphingosine) and 5.70 (m, 1H, H-5 of sphingosine).

In summary, a facile, stereocontrolled first total synthesis of GD2 was accomplished. This work shows that the glycosyl acceptor **1** and the disialyl pentasaccaride **4** described herein could be used as the intermediates suitable for polysialogangliosides syntheses.

Elemental analyses, as well as IR and $^1$H NMR data of all the new compounds reported here were satisfactory with the assigned structures.



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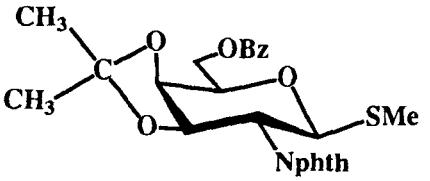
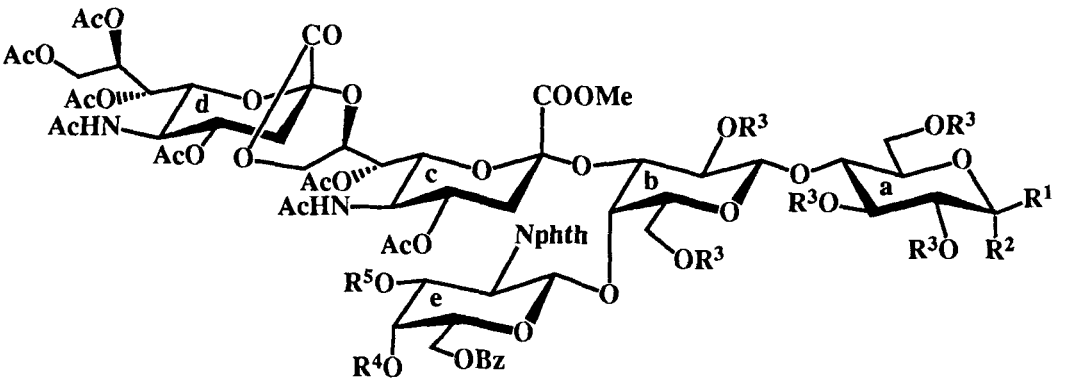
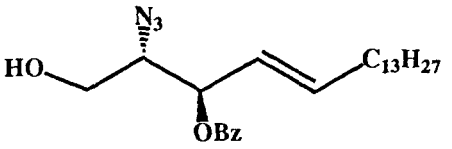


2

SE = 2-(trimethylsilyl)ethyl
Bn = benzyl
Bz = benzoyl
phth = phthaloyl



| | $R^1$ | $R^2$ | $R^3$ | $R^4$ | $R^5$ |
|---|---|---|---|---|---|
| 3 | OSE | H | Bn | - ipd - | |
| 4 | OSE | H | Bn | H | H |
| 5 | OSE | H | Ac | Ac | Ac |
| 6 | H , OH | | Ac | Ac | Ac |
| 7 | H | $OC(=NH)CCl_3$ | Ac | Ac | Ac |



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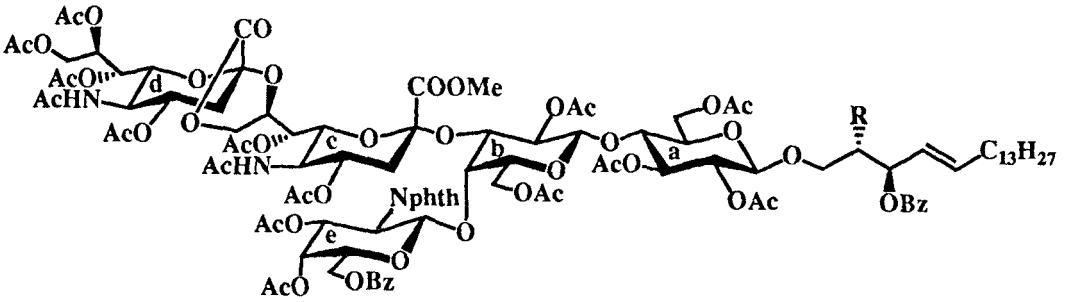
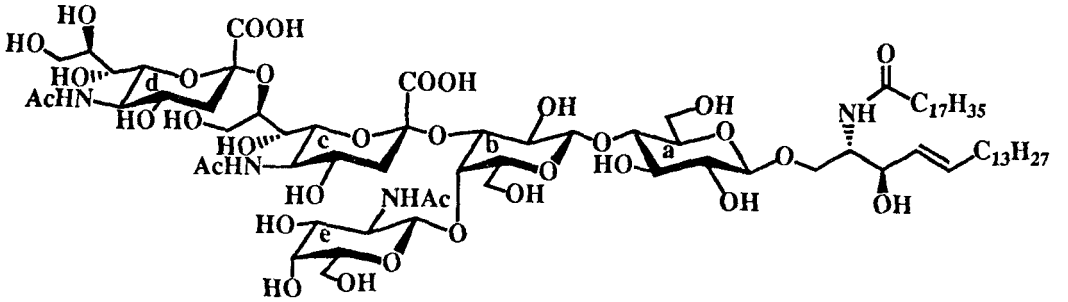


9 R = $N_3$
10 R = $NHCOC_{17}H_{35}$



11 Ganglioside GD2

## ACKNOWLEDGMENT

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