Experiments directed towards synthesis of complex glycosphingolipids: Ganglioganglioside GQ1b

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Abstract: Synthesis of a properly protected octasaccharide which could be regarded as a reasonable precursor for the synthesis of GQ1b was carried out in a stereo- and regio-controlled manner.

Tetrasialo ganglio-ganglioside GQ1b was first isolated in 1967 from human brain and then in 1972 from fish brain. The structure of 1 was characterized around 1980 through chemical and enzymic degradation as well as by Mass spectral data. The trophic effect of GQ1b on the nervous system has been shown by employing human neuroblastoma cell line. Since structurally most simple ganglioside GM3 was synthesized for the first time in 1985, more efficient strategies for the synthesis of gangliogangliosides have been developed. A total synthesis of GQ1b, however, still remains to be accomplished. In this paper, we highlight successful synthesis of a properly protected octasaccharide derivative which may be regarded as a key intermediate for further conversion into GQ1b.
Aiming at a convergent-type synthesis of GQ1b 1, strategic bond disconnection was planned as shown in scheme 1 which led to design completely protected building blocks 2, 3, 4, and 5. It is to be noted that 4-methylbenzyl and pivaloyl groups at O-2 of monosaccharide residue 4 and 7 in compounds 2 and 5, respectively, are required to serve as a stereocontrolling auxiliary.

In order to study the reactivity of O-4 at residue 2 of the building block 5, first we prepared 5 ([R=Bn, RF 0.37 in 4:1 PhMe-Me2CO; δH 5.107 (dd, 8.0 and 9.5 Hz, H-2), 3.748 and 3.510 (2s, 2 x OMe), 2.816 and 2.398 (2dd, 4.4 and 12.8 Hz, H-3eq,5eq), 1.863 and 1.708 (2s, 2 x Ac), 1.104 (s, tBu)] from known compound 6 in 3 steps (i Ph3SnH, AIBN in PhH, ii NaOMe, MeOH-PhMe, iii CH2N2 in 39% overall yield). All of the attempted glycosylations of 5 (R=Bn) as well as 6 with 3 (X=Me, Br, F, and OCNHCCl3) in our hands occurred at NHAc groups linked to either residue 5 or 6 but not at OH group of residue 2 of 5 and 6, resulting in the formation of only acid labile imidates.

In order to lower the nucleophilicity of NHAc groups in compound 5 (R=Bn) toward glycosyl donors, we had to change the protective group of sialic acid residues from benzyl to electron withdrawing acetyl groups. According to this scenario, we studied compound 5 (R=Ac) as a glycosyl acceptor. To our delight, the glycosylation of 5 (R=Ac, ref 11) with 11 went smoothly in the presence of PhSeOTf in CH3CN to give an 89% of 12: [α]D +6.7° (c 0.6, CHCl3); RF 0.43 in 20:1 CHCl3-MeOH; δH 6.235 (s, CHPh), 5.306 (d, 8.8 Hz, H-13), 2.699 and 2.671 (2dd, 4.4 and 12.8 Hz, H-3eq,5eq), 1.744 (t, 12.8 Hz, H-3ax,5ax), 1.197 (s, tBu). Having successfully carried out a chain elongation at O-4 of residue 2 of
compoun 5, N-phthaloyl function of the protected pentasaccharide 12 was then converted into N-acetyl group in 4 steps (i Li-Py, ii NH₂NH₂-H₂O, iii Ac₂O-MeOH, iv CH₂N₂, 95% overall) to give 13: [α]D -5.5° (c 0.9, CHCl₃); Rᶠ 0.33 in 5:3 Me₂CO-hexane; δH 6.276 (s, CHPh), 5.117 (dd, 8.4 and 9.2 Hz, H-2'), 3.996 (s, OMe), 2.242 (dd, 4.7 and 13.5 Hz, H-3eq₅), 1.789 (t, 12.8 Hz, H-3ax₅), 1.169 (s, tBu). Treatment of 13 with 4:1 AcOH-H₂O afforded 87% of 14 which is ready for further chain elongation at O-3 of residue 3. Compound 14 had [α]D -5.5° (c 0.3, CHCl₃); Rᶠ 0.33 in 19:1 CHCl₃-MeOH; δH 5.200 (ddd, 5.5, 11.3 and 16.5 Hz, H-4₆), 5.135 (dd, 8.1 and 9.2 Hz, H-2'), 5.024 (ddd, 5.1, 12.1 and 16.8 Hz, H-4₅), 2.346 (dd, 5.5 and 14.0 Hz, H-3eq₅), 2.240 (t, 14.0 Hz, H-3ax₅), 2.196 (dd, 5.1 and 14.3 Hz, H-3eq₅), 1.716 (t, 14.3 Hz, H-3ax₅), 1.170 (s, tBu).

The building block 2 was prepared¹⁰ starting from 15 (ref.10). Glycosylation of 16 with 0.66 equivalent of 15 in the presence of Hg(CN)₂-HgBr₂-MS₄A in CCl₄ and subsequent acetylation gave 33% of 17: δH 5.311 (d, 3.4 Hz, H-4₄), 3.421 and 2.963 (2d, 10.3 and 10.6 Hz, H-3₇₈). Further conversion of 17 into 2 was achieved in 4 steps (i Ph₃SnH, AIBN in PhH, ii H₂, 10% Pd-C in MeOH, iii Ac₂O-Py-DMAP, iv MeSSnBu₃-SnCl₄, 17% overall). Compound 2 had Rᶠ 0.38 in 20:1 CHCl₃-MeOH; [α]D +29.3° (c 0.4
in CHCl₃; δH 4.864 (d, 9.6 Hz, H-1⁴), 3.828 and 3.796 (2s, 2 x OMe), 2.365 (s, C₆H₅Me), 2.221, 2.183, 2.166, 2.165, 2.137, 2.047, 2.046, 2.045, 1.956, 1.915 and 1.829 (11s, 10 x Ac and SMe), 1.202 (s, tBu). Crucial coupling of 14 with 1 equivalent of 2 in the presence of PhSeOTf in (CH₂Cl₂)₄ afforded 16% of the designed octasaccharide 18 along with 67% of recovered pentasaccharide 14. It is to be noted that reaction of trichloroacetimidate corresponding to 2 with 14 in the presence of either TMSOTf or BF₃·OEt₂ in the same solvent failed completely. Stereochemistry of the glycosylation was confirmed by the following ¹H NMR of the compound 18: RF 0.28 in 1:2 CHCl₃-THF; δH 5.395 (dd, 7.7 and 9.5 Hz, H-2⁴), 4.801 (d, 7.7 Hz, H-1⁴), 3.800, 3.785, and 3.785 (3s, 3 x OMe), 2.368 (s, PhMe), 1.177 and 1.162 (2s, 2 x tBu). The regiochemistry of the coupling was confirmed by ¹H NMR of the acetate of 18 which contained a deshielded signal for H-4 at δ 5.420 (d, 3.7 Hz). The Compound 18 could be regarded as a plausible precursor for the imidate 19.

In summary, a first synthesis of properly protected octasaccharide 18 which was regarded a key intermediate for the synthesis of GQ1b was achieved by controlling the reactivity of OH-4 of residue 2 relative to NHAc of residues 5 and 6 in compound 5 by changing the protective groups of hydroxyl functions from benzyl to acetyl. Crucial coupling between pentasaccharide block 14 and trisaccharide block 2 was finally executed in the presence of thiophilic promoter.

REFERENCES