Pergamon

Tetrahedron Letters 39 (1998) 7189–7192

TETRAHEDRON
LETTERS

Synthesis of Fully and Partially Benzylated Glycosyl Azides *via* Thioalkyl Glycosides as Precursors for the Preparation of N-Glycopeptides

János Kerékgyártó*[a,b], Károly Ágoston[a], Gyula Batta[c],
Johannis P. Kamerling[b], Johannes F. G. Vliegenthart[b]

[a]*Institute of Biochemistry, Lajos Kossuth University, P.O. Box 55, H-4010 Debrecen, Hungary*
[b]*Bijvoet Center, Department of Bio-Organic Chemistry, Utrecht University, P.O. Box 80.075, NL-3508 TB Utrecht, Netherlands*
[c]*Research Group for Antibiotics of the Hungarian Academy of Sciences, P.O. Box 70, H-4010 Debrecen, Hungary*

Received 29 June 1998; accepted 20 July 1998

Abstract: Fully *O*-benzylated mono-, di- and trisaccharide glycosyl azides representing the reducing terminal of the core structure of N-glycans were synthesized. Totally and partially benzylated thioalkyl glucosamine glycosides were converted into the corresponding glycosyl azides with trimethylsilyl azide in the presence of methyl triflate. The β-mannosidic linkage was created by C-2 epimerization of the initially introduced β-D-*gluco*-unit *via* oxidation followed by stereoselective reduction with tetrabutylammonium borohydride. © 1998 Elsevier Science Ltd. All rights reserved.

Keywords: Glycosidation; Carbohydrates; Thioglycosides; Azides

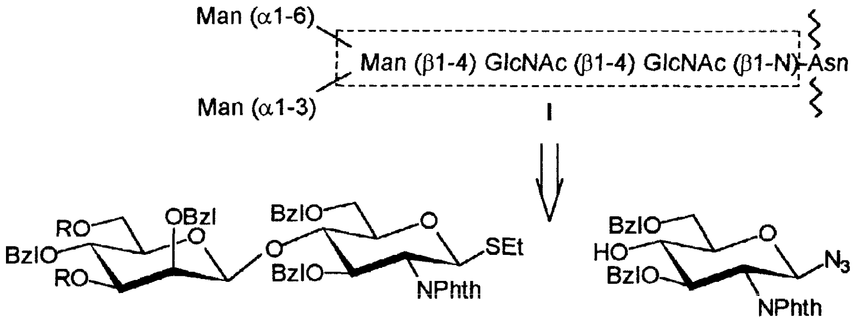
Most N-linked glycoprotein glycans share a common pentasaccharide core structure (I). The chemical synthesis of glycopeptides as glycoprotein modelling tools represents an area of significant interest [1-3]. In the framework of our research project on the synthesis of N-glycopeptides, we have studied the optimization of coupling reactions between free glycosylamines derived from GlcNAc and GlcNAc(β1-4)GlcNAc and selectively blocked oligopeptides, providing a unified and generally applicable protocol, irrespective of peptide composition and intramolecular succinimide forming tendency [4]. Using this method, two glycosylated decapeptide fragments of a lectin, containing these carbohydrate units, were prepared in a semipreparative scale [4]. To further investigate the coupling reaction between glycosylamines and selectively protected peptides, with respect to the size of each component and choice of protection for the carbohydrate hydroxyl functions, here we describe the preparation of the fully *O*-benzylated glycosyl azides GlcNAc(β1-N$_3$) (**1**), GlcNAc(β1-4)GlcNAc(β1-N$_3$) (**2**) and Man(β1-4)GlcNAc(β1-4)GlcNAc(β1-N$_3$) (**3**) representing the reducing terminal of the core structure of N-glycans.

The synthesis of *O*-acetylated glycosyl azides from *O*-acetylated glycosyl halides and from peracetates of mono- and disaccharides is well reviewed [5]. In general, glycosyl azides can be subjected to the conventional

PII: S0040-4039(98)01540-8

protection-deprotection protocols used in carbohydrate chemistry [5,6]. However, a strong base can effect the loss of the anomeric azido function [5]. Despite successful alkylations of glycosyl azides [6], in some attempts, *O*-benzylations in the presence of various bases failed [7]. Partially benzylated glycosyl azides can also be prepared from the corresponding fluorides [8] and trichloroacetimidates [9].

Our current synthetic strategy is based on the following retrosynthetic pathway:

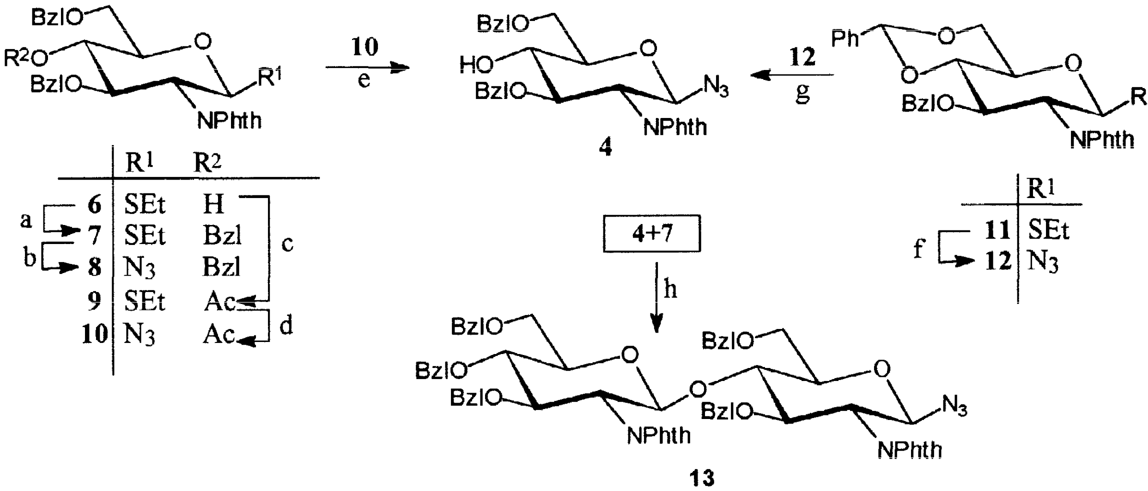


5 R = Bzl
R = Temporary protection

4

Accordingly, the approach includes the synthesis of glycosyl azides *via* thioalkyl glycosides and the preparation of suitably protected disaccharide-thioglycosides containing a β-mannosidic linkage [10,11]. The proposed protocol is suitable to prepare further extensions of the compounds described now.

Compound **6** [10] (Scheme 1) was benzylated to afford **7** or acetylated to yield **9**. The fully/selectively benzylated thioethyl glycosides 7, **9** and **11** [10] [mp 104°C] were treated with trimethylsilyl azide in the presence of methyl triflate and molecular sieves in dichloromethane, furnishing the crystalline glycosyl azides **8** (precursor of 1), **10**[1,2] and **12**[1] [mp 133°C], respectively, in high yields (83-92%). Deacetylation of **10** and reductive ring cleavage of **12** gave glycosyl acceptor **4**[1]. Condensation of thioglycoside **7** and acceptor **4** promoted by NIS-AgOTf [12] gave the chitobiose derivative **13** (precursor of **2**) in a yield of 90%.



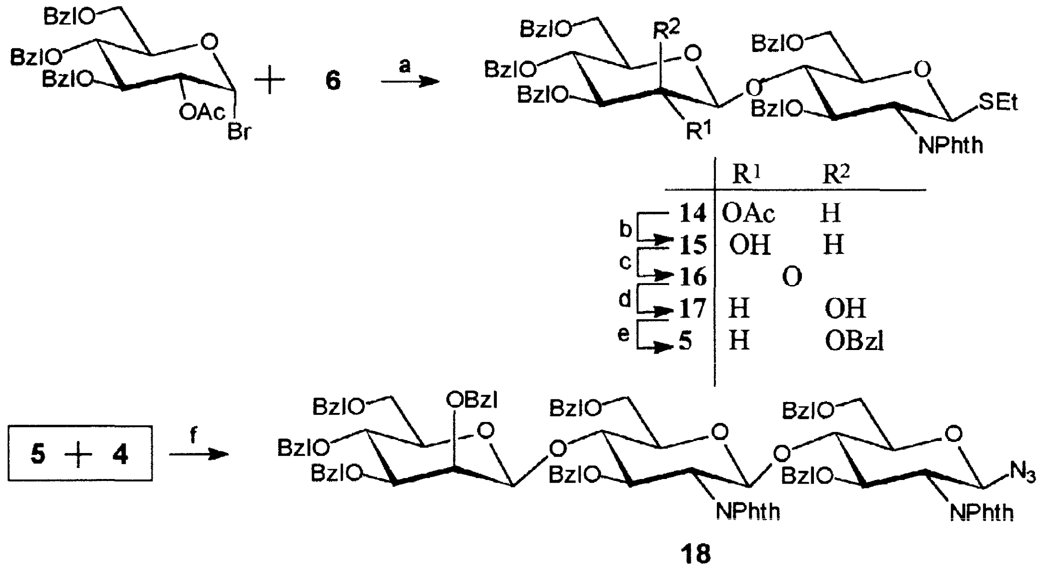
13

Scheme 1 : a) BzlBr, Ag_2O, DMF, 89%; b) TMS-N_3, MeOTf, 91%; c) Pyr, Ac_2O, quant.; d) TMS-N_3, MeOTf, 92%; e) K_2CO_3, THF/MeOH, 90%; f) TMS-N_3, MeOTf, 83%; g) Me_3NBH_3, $AlCl_3$, THF, 86%; h) NIS, AgOTf, -40°C, 90%

---

[1] Compounds **4** [6,9], **10** [9,13] and **12** [6] have been prepared earlier along other routes.

[2] The conversion of the thiophenyl analogue of compound **9** into glycosyl azide **10** has been reported [13].



Scheme 2 : a) AgOTf, -40°C, 84%; b) K_2CO_3, THF/MeOH, 90%; c) DMSO, Ac_2O; d) Bu_4NBH_4, THF, 0°C, 81% (two steps); e) BzlBr, Ag_2O, DMF, 88%; f) NIS, AgOTf, -40°C, 81%

For the synthesis of the β-(1→4)-linked D-mannosyl chitobiosyl azide **18**, perbenzylated disaccharide thioethyl glycoside **5** was prepared first (Scheme 2). Condensation of 2-*O*-acetyl-3,4,6-tri-*O*-benzyl-α-D-glucopyranosyl bromide [14] and acceptor **6** in dichloromethane-toluene in the presence of AgOTf and molecular sieves gave disaccharide **14** in a yield of 84%. Removal of the 2'-*O*-acetyl group of compound **14** followed by oxidation of the HO-2' function afforded ulose derivative **16**. Reduction of ulose **16** with Bu_4NBH_4 [15] in tetrahydrofuran furnished disaccharide thioglycoside **17** (β-*manno*-epimer) in a stereo-selective manner (81% yield from **15**). Only a faint spot of the β-*gluco*-epimer **15** was detectable by TLC. Then, compound **17** was benzylated to give glycosyl donor **5**. Condensation of thioglycoside **5** and acceptor **4** in dichloromethane-acetonitrile in the presence of NIS-AgOTf and molecular sieves gave the mannosyl chitobiose derivative **18** (precursor of **3**).

The phthalimido functions of **8**, **13** and **18** were removed by treatment with ethylenediamine [16] and the resulting products were *N*-acetylated to furnish the fully *O*-benzylated glycosyl azides[3] **1**, **2** and **3** in yields of 84%, 83% and 79%, respectively.

---

[3] All compounds gave satisfactory microanalytical and/or spectroscopic data. Selected spectroscopic and physical data are the following: compound **7**: $[\alpha]_D$ +51.4° (c 0.5, $CHCl_3$); δ_H ($CDCl_3$) 5.25 (d, 1H, H-1, $J_{1,2}$ 10.3 Hz). Compound **8**: mp 103°C (ethanol); $[\alpha]_D$ +36.7° (c 0.4, $CHCl_3$); δ_H ($CDCl_3$) 5.37 (d, 1H, H-1, $J_{1,2}$ 9.4 Hz); IR ν 2105 cm^{-1} (N_3). Compound **13**: $[\alpha]_D$ +17.3° (c 0.2, $CHCl_3$); δ_H ($CDCl_3$) 5.32 (d, 1H, H-1, $J_{1,2}$ 8.3 Hz), 5.15 (d, 1H, H-1′, $J_{1',2'}$ 9.3 Hz); δ_C ($CDCl_3$) 96.92 (C-1′), 85.51 (C-1). Compound **14**: $[\alpha]_D$ +23.3° (c 0.3, $CHCl_3$); δ_H ($CDCl_3$) 5.18 (d, 1H, H-1, $J_{1,2}$ 10.6 Hz), 4.59 (d, 1H, H-1′, $J_{1',2'}$ 9.8 Hz); δ_C ($CDCl_3$) 100.31 (C-1′), 83.02 (C-1). Compound **5**: $[\alpha]_D$ +18.2° (c 0.1, $CHCl_3$); δ_H ($CDCl_3$) 5.22 (d, 1H, H-1, $J_{1,2}$ 10.0 Hz), 4.90 (bs, 1H, H-1′); δ_C ($CDCl_3$) 101.54 (C-1′), 82.69 (C-1). Compound **18**: $[\alpha]_D$ +9.5° (c 0.5, $CHCl_3$); δ_H ($CDCl_3$) 5.27 (d, 1H, H-1, $J_{1,2}$ 8.0 Hz), 5.16 (d, 1H, H-1′, $J_{1',2'}$ 9.0 Hz), 4.89 (bs, 1H, H-1″); δ_C ($CDCl_3$) 101.54 (C-1″), 97.06 (C-1′), 85.54 (C-1). Compound **1**: mp 171°C (ethanol); $[\alpha]_D$ -34° (c 0.4, MeOH), δ_H ($CDCl_3$) 4.99 (d, 1H, H-1, $J_{1,2}$ 10.4 Hz), 1.89 (s, 3H, NAc), δ_C ($CDCl_3$) 87.81 (C-1); IR ν 2105 cm^{-1} (N_3). Compound **2**: $[\alpha]_D$ -34.7° (c 0.4, $CHCl_3$); δ_H ($CDCl_3$) 4.80 (d, 1H, H-1, $J_{1,2}$ 8.1 Hz), 4.38 (d, 1H, H-1′, $J_{1',2'}$ 8.0 Hz), 1.92 and 1.72 (each 3H, 2NAc); δ_C ($CDCl_3$) 100.41 (C-1′), 88.77 (C-1); IR ν 2100 cm^{-1} (N_3). Compound **3**: $[\alpha]_D$ +48.7° (c 1.2, $CHCl_3$); δ_H ($CDCl_3$) 4.76 (d, 1H, H-1, $J_{1,2}$ 7.6 Hz), 4.49 (d, 1H, H-1″, $J_{1'',2''}$ < 2 Hz), 4.47 (d, 1H, H-1′, $J_{1',2'}$ 7.6 Hz); δ_C ($CDCl_3$) 101.24 (C-1″, $J_{C-1'',H-1''}$ 156 Hz), 99.83 (C-1′, $J_{C-1',H-1'}$ 160 Hz), 88.38 (C-1, $J_{C-1,H-1}$ 160 Hz); IR ν 2100 cm^{-1} (N_3).

In summary, fully and partially benzylated glycosyl azides have been prepared from the corresponding thioalkyl glycosides by direct activation of the anomeric center. Glycosyl azides **1**, **2** and **3** are stable compounds. Chemoselective reduction (PtO_2/H_2) of their azido functions resulted in fully *O*-benzylated glycosylamines. The investigation of their coupling reactions by *in situ* trapping their amines with selectively protected oligopeptides as well as the preparation of larger oligoglycosyl azides are in progress.

Acknowledgements

We are grateful for the grants from the Netherlands Organization for Scientific Research (NWO), the Hungarian National Science Foundation (OTKA-T-019404) and the Ministry of Education Foundation (FKFP 0329/1997), and for drx 500 purchase grants (OMFB MEC 93 0098, Phare Accord H-9112-0198, OTKA A084).

References

[1] Garg HG, von dem Bruch K, Kunz H. Adv. Carbohydr. Chem. Biochem. 1994;50:277-310.
[2] Rio-Anneheim S, Paulsen H, Meldal M, Bock K. J. Chem. Soc. Perkin Trans 1 1995:1071-1080.
[3] Guo Z-W, Nakahara Yu, Nakahara Yo, Ogawa T. Bioorg. Med. Chem. 1997;5:1917-1924.
[4] Varga J, Kerékgyártó J, Váradi Gy, Székely Z, Tóth G, Penke B. J. Peptide Sci. in press.
[5] Györgydeák Z, Szilágyi L, Paulsen H. J. Carbohydr. Chem. 1993;12:139-163.
[6] Bröder W, Kunz H. Carbohydr. Res. 1993;249:221-241.
[7] Bröder W, Kunz H. Bioorg. Med. Chem. 1997;5:1-19.
[8] Unverzagt C. Angew. Chem. Int. Ed. Engl. 1994;33:1102-1104.
[9] Matsuo I, Nakahara Yu, Ito Y, Nukada T, Nakahara Yo, Ogawa T. Bioorg. Med. Chem. 1995;3:1455-1463.
[10] Kerékgyártó J, van der Ven JGM, Kamerling JP, Vliegenthart JFG. Carbohydr. Res. 1993;238:135-145.
[11] Kerékgyártó J, Ágoston K, Kamerling JP, Vliegenthart JFG. Abstr. 9th Eur. Carbohydr. Symp., Utrecht, Netherlands 1997:A91.
[12] Kaine O, Ito Y, Ogawa T. J. Am. Chem. Soc. 1994;116:12073-12074.
[13] Matsuo I, Isomura M, Walton R, Ajisaka K. Tetrahedron Lett. 1996;37:8795-8798.
[14] Shaban MAE, Jeanloz RW. Carbohydr. Res. 1976;52:103-114.
[15] van Seeventer PB, Kerékgyártó J, van Dorst JALM, Halkes KM, Kamerling JP, Vliegenthart JFG. Carbohydr. Res. 1997;300:127-138.
[16] Kanie O, Crawley SC, Palcic MM, Hindsgaul O. Carbohydr. Res. 1993;243:139-164.