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Note

A practical synthesis of 2-azidoethyl α-glycosides: useful spacer-arm glycosides for the synthesis of neoglycoconjugates †

Gangavaram V.M. Sharma * and Palakodety Radha Krishna

Bio-Organic Laboratory, Indian Institute of Chemical Technology, Hyderabad 500 007 (India)

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Progress in the chemistry of glycoconjugates during the last decade reflects the availability of a wide array of methods for stereoselective O-glycosylation[1–3]. Carbohydrate segments of biological interest can be coupled through a spacer[4] to a soluble protein to yield synthetic antigens, and attachment to solid phases leads to immunoadsorbents. Spacer-arm glycosides have been made[5] and used[6] for several purposes. Our studies of the synthesis of the lipopolysaccharide of *Proteus* sps. culminated in the development of a method for the synthesis of 2-azidoethyl β-glycosides[7] and was utilised for the synthesis of determinants as neoglycoconjugates[8,9] which gave positive serodiagnostic results.

The stereoselective synthesis of α-linked saccharides is important since they are constituents of many biologically active glycoconjugates[8]. We now report an extension of the methodology for glycosylation with 2-pyridyl 1-thioglycosides[10,11] for the synthesis of 2-azidoethyl α-glycosides.

The glycosyl donor 2-pyridyl 2,3,4-tri-*O*-benzyl-6-*O*-trityl-1-thio-β-D-glucopyranoside (**1**) was prepared from 1,2,3,4,6-penta-*O*-acetyl-β-D-glucopyranose; 2-pyridyl 2,3,4,6-tetra-*O*-benzyl-1-thio-β-D-galactopyranoside (**2**), 2-pyridyl 2,3,4-tri-*O*-benzyl-1-thio-α,β-L-rhamnopyranoside (**3**), and 2-pyridyl 2,3,5-tri-*O*-benzyl-1-thio-β-D-ribofuranoside (**4**) were prepared by known procedures[10,11]; and 2-pyridyl 2,3,6-tri-*O*-benzyl-4-*O*-(2,3,4,6-tetra-*O*-benzyl-β-D-galactopyranosyl)-1-thio-β-D-glucopyranoside (**5**) was obtained from α-acetobromolactose[12] by adapting a reported[10] procedure.

When the 2-pyridyl 1-thioglycosides **1–5** (1.0 mmol) were each heated with 2-azidoethanol (3 mmol) in anhydrous dichloromethane (10 mL, containing 3% of methyl iodide) in the presence of 4A molecular sieves at 50°C (12–72 h), the

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* Author for correspondence.

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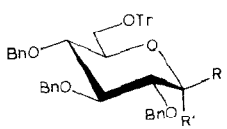
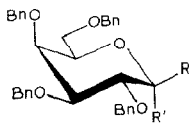


1 R = SPy, R' = H
7 R = H, R' = O⌒⌒N3



2 R = SPy, R' = H
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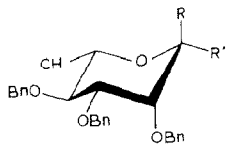
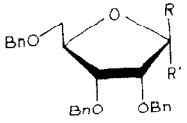
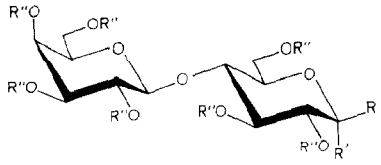


3 R, R' = H, SPy
9 R = O⌒⌒N3, R' = H



4 R = SPy, R' = H
10 R = H, R' = O⌒⌒N3



5 R = SPy, R' = H, R'' = Bn
6 R = SPy, R' = H, R'' = Ac
11 R = H, R' = O⌒⌒N3, R'' = Bn

corresponding α-glycosides, 2-azidoethyl 2,3,4-tri-*O*-benzyl-6-*O*-trityl-α-D-glucopyranoside (**7**), 2-azidoethyl 2,3,4,6-tetra-*O*-benzyl-α-D-galactopyranoside (**8**), 2-azidoethyl 2,3,4-tri-*O*-benzyl-α-L-rhamnopyranoside (**9**), 2-azidoethyl 2,3,5-tri-*O*-benzyl-α-D-ribofuranoside (**10**), and 2-azidoethyl 2,3,6-tri-*O*-benzyl-4-*O*-(2,3,4,6-tetra-*O*-benzyl-β-D-galactopyranosyl)-α-D-glucopyranoside (**11**) were obtained after chromatography. That these compounds were α-glycosides was indicated by the $[\alpha]_D$ values and the NMR data (δ 97.1–101.7 for C-1; $J_{C\text{-}1,H\text{-}1}$ for **8** and **9** = 165 and 169 Hz, respectively). β-Glycosides were not detected during the glycosylation reactions.

The above mild procedure has potential for the synthesis of neoglycoconjugates.

EXPERIMENTAL

General methods.—Melting points were determined in open capillaries and are uncorrected. NMR spectra were recorded for solutions in $CDCl_3$ (internal Me_4Si) with a Varian 200-Gemini spectrometer (^1H, 200 MHz; ^{13}C, 50 MHz). Optical rotations were measured with a JASCO DIP 360 or 370 polarimeter. Silica gel (60–120 mesh, Acme) was used for column chromatography. TLC was performed on Silica Gel 60 F_{254} (Merck) with detection using a solution of 2% of phosphomolybdic acid and 1% of $Ce_2SO_4 \cdot 4H_2O$ in aq 20% H_2SO_4 at 130°C. All the reactions were carried out in dry solvents under anhydrous conditions unless otherwise stated.

2-Pyridyl 2,3,4-tri-O-benzyl-6-O-trityl-1-thio-β-D-glucopyranoside (**1**).—2-Pyridyl 2,3,4,6-tetra-*O*-acetyl-1-thio-β-D-glucopyranoside[11] (2.2 g, 5.0 mmol) was *O*-deacetylated conventionally with methanolic NaOMe. A mixture of the resulting 2-pyridyl 1-thio-β-D-glucopyranoside and trityl chloride (1.53 g, 5.4 mmol) in pyridine (15 mL) was heated at 50°C for 7 h. Methanol was added and the mixture was distilled azeotropically with 4:1 toluene–heptane. Column chromatography (1:1 light petroleum–EtOAc, then EtOAc) of the residue gave 2-pyridyl 6-*O*-trityl-1-thio-β-D-glucopyranoside (2.2 g, 88%); $[\alpha]_D$ −19° (*c* 1.0, MeOH). ^{1}H NMR data: δ 3.2–4.0 (m, 9 H), 5.1 (d, 1 H, $J_{1,2}$ 9.5 Hz, H-1), 7.0–7.6 (m, 18 H, ArH), and 8.4 (m, 1 H, SPy).

A solution of the foregoing product in DMF (5 mL) was benzylated[11] to give **1** (2.2 g, 65%); $[\alpha]_D$ −6.8° (*c* 1.0, $CHCl_3$). NMR data: ^{1}H, δ 3.2–3.7 (m, 5 H), 4.3 (d, 1 H, $J_{1,2}$ 9 Hz, H-2), 4.6–5.0 (m, 6 H), 5.35 (d, 1 H, H-1), 6.7–8.5 (m, 34 H, ArH); ^{13}C, δ 66.0, 70.5, 73.0, 75.0, 76.0, 78.0, 80.0, and 82.0 (C-2/6 and 3 O*C*H$_2$Ph), 84.0 (C-1), and 124.0–130.0 (aromatic). Anal. Calcd for $C_{51}H_{47}NO_5S$: C, 77.93; H, 6.02. Found: C, 78.00; H, 6.16.

2-Pyridyl 2,3,6-tri-O-acetyl-4-O-(2,3,4,6-tetra-O-acetyl-β-D-galactopyranosyl)-1-thio-β-D-glucopyranoside (**6**).—A solution of 2-mercaptopyridine (0.586 g, 5.28 mmol) in dry acetone (10 mL) was stirred with K_2CO_3 (1.38 g, 10.0 mmol) for 1 h. A solution of α-acetobromolactose (3.5 g, 5 mmol) in dry benzene (10 mL) was added and stirring was continued for 4 h. The mixture was washed with water, aq 1% KOH, and water, dried (Na_2SO_4), and concentrated. Column chromatography (2:1 light petroleum–EtOAc) of the residue afforded **6** (2.1 g, 58%); mp 147–150°C (from hexane–EtOAc); $[\alpha]_D$ +27° (*c* 1.0, $CHCl_3$). NMR data: ^{1}H, δ 1.9–2.2 (7 s, 21 H, 7 OAc), 3.7–3.9 (m, 3 H), 4.0–4.15 (m, 3 H), 4.3–4.6 (m, 2 H), 4.9–5.4 (m, 5 H), 5.8 (d, 1 H, $J_{1,2}$ 10 Hz, H-1), and 7.0–8.4 (m, 4 H, SPy); ^{13}C, δ 20.2 (7 *C*H$_3$CO), 60.7, 62.0, 66.5, 68.8, 69.6, 70.4, 70.7, 73.6, 75.9, 76.3, and 81.2 (C-1/6 and C-2′/6′), 100.6 (C-1), 120.5, 122.9, 136.4, 149.2, 155.2 (5 SPy), and 168.0–172.0 (7 O*C*OCH$_3$). Anal. Calcd for $C_{31}H_{39}NO_{17}S$: C, 51.02; H, 5.38. Found: C, 51.10; H, 5.50.

2-Pyridyl 2,3,6-tri-O-benzyl-4-O-(2,3,4,6-tetra-O-benzyl-β-D-galactopyranosyl)-1-thio-β-D-glucopyranoside (**5**).—A solution of **6** (0.48 g, 0.65 mmol) in dry MeOH (5 mL) was treated with a catalytic amount of NaOMe. After the usual work up, a solution of the product in DMF (3 mL) was benzylated[11] to furnish **5** (0.45 g, 66%); mp 103–106°C (from hexane–EtOAc); $[\alpha]_D$ +3.2° (*c* 1.0, $CHCl_3$). NMR data: ^{1}H, δ 3.1–3.8 (m, 10 H), 4.0–4.9 (m, 17 H), 5.2 (d, 1 H, $J_{1,2}$ 10 Hz, H-1), 6.7–7.2 (m, 38 H, ArH), and 8.15 (s, 1 H, SPy); ^{13}C, δ 68.0 (2 C), 72.4, 72.8, 72.9, 73.2, 73.6, and 74.6 (2 C), 75.1 (4 C), 76.2 (2 C), 82.4, 83.6, and 84.8 (C-1/6, C-2′/6′, and 7 O*C*H$_2$Ph), 102.7 (C-1), 120.2 (C-3″), 123.1 (C-4″), 127.1–128.3 (aromatic), 136.4 (C-5″), and 149.5 (C-6″). Anal. Calcd for $C_{66}H_{67}NO_{10}S$: C, 74.33; H, 6.33. Found: C, 74.20; H, 6.52.

Preparation of benzylated 2-azidoethyl α-glycosides.—A solution of the benzylated 2-pyridyl 1-thioglycoside (1 mmol) in CH_2Cl_2 (10 mL, containing 3% of MeI)

was treated with 4A molecular sieves (0.200 g) and 2-azidoethanol (3 mmol), and then heated at 50°C until the reaction was complete (TLC, usually 12–72 h). A 1 : 1 mixture of Celite and Na_2SO_4 was added, the mixture was filtered, the insoluble material was washed with CH_2Cl_2, and the combined filtrate and washings were concentrated. Column chromatography (light petroleum–EtOAc) of the residue afforded the α-glyoside in good yield.

*2-Azidoethyl 2,3,4-tri-*O*-benzyl-6-*O*-trityl-α-*D*-glucopyranoside* (**7**).—Reaction of **1** (0.50 g, 0.63 mmol) with azidoethanol for 16 h afforded **7** (0.329 g, 68%) isolated as a syrup; $[\alpha]_D$ +7.4° (*c* 1.0, $CHCl_3$). NMR data: ^{1}H, δ 3.5 (t, 2 H, CH_2N_3), 3.6–3.75 (m, 2 H), 3.85–4.4 (m, 5 H), 4.7–5.1 (m, 8 H), and 7.15–7.6 (m, 30 H, ArH); ^{13}C, δ 50.5 (CH_2N_3), 62.0, 66.2, 70.7, 73.1, 74.8, 75.8, 77.9, 80.2, and 81.9 (C-2/6, OCH_2, and 3 OCH_2Ph), 97.1 (C-1), and 126.1–129.9 (aromatic). Anal. Calcd for $C_{48}H_{47}N_3O_6$: C, 75.66; H, 6.21. Found: C, 75.81; H, 6.32.

*2-Azidoethyl 2,3,4,6-tetra-*O*-benzyl-α-*D*-galactopyranoside* (**8**).—Reaction of **2** (0.316 g, 0.5 mmol) with azidoethanol for 13 h furnished **8** (0.218 g, 72%) isolated as a syrup; $[\alpha]_D$ +15° (*c* 1.0, $CHCl_3$). NMR data: ^{1}H, δ 3.5 (t, 2 H, CH_2N_3), 4.0 (t, 2 H, $OCH_2CH_2N_3$), 4.35–4.90 (m, 15 H, H-1/6 and 4 OCH_2Ph), and 7.15–7.40 (m, 20 H, 4 Ph); ^{13}C, δ 50.5 (CH_2N_3), 66.7, 69.0, 69.6, 72.7, 73.1, 73.2, 74.6, 75.2, 76.2, and 78.6 (C-2/6, 4 CH_2Ph, and OCH_2), 98.1 (C-1), and 127.3–139.5 (aromatic); $J_{C\text{-}1,H\text{-}1}$ 169 Hz. Anal. Calcd for $C_{36}H_{39}N_3O_6$: C, 70.91; H, 6.41. Found: C, 71.01; H, 6.6.

*2-Azidoethyl 2,3,4-tri-*O*-benzyl-α-*L*-rhamnopyranoside* (**9**).—Reaction of **3** (0.36 g, 0.68 mmol) with azidoethanol for 72 h gave **9** (0.170 g, 50%) isolated as a syrup; $[\alpha]_D$ −13° (*c* 1.45, $CHCl_3$). NMR data: ^{1}H, δ 1.3 (d, 3 H, $J_{5,6}$ 5.8 Hz, H-6,6,6), 3.2–3.35 (m, 2 H, CH_2N_3), 3.5–3.9 (m, 5 H), 4.55–5.0 (m, 8 H), and 7.2–7.4 (m, 15 H, 3 Ph); ^{13}C, δ 17.9 (CH_3), 50.3 (CH_2N_3), 66.1, 66.2, 72.0, 72.8, 74.7, 75.1, 79.8, and 80.1 (C-2/5, OCH_2, and 3 OCH_2Ph), 98.1 (C-1), and 127.5–128.2 (aromatic); $J_{C\text{-}1,H\text{-}1}$ 169 Hz. Anal. Calcd for $C_{29}H_{33}N_3O_5$: C, 69.16; H, 6.61. Found: C, 69.32; H, 6.90.

*2-Azidoethyl 2,3,5-tri-*O*-benzyl-α-*D*-ribofuranoside* (**10**).—Reaction of **4** (0.5 g, 1 mmol) with azidoethanol for 70 h afforded **10** (0.34 g, 70%) isolated as a syrup; $[\alpha]_D$ +71° (*c* 1.4, $CHCl_3$). NMR data: ^{1}H, δ 3.3–3.5 (m, 2 H, CH_2N_3), 3.7–3.9 (m, 2 H, OCH_2), 4.2–4.7 (m, 11 H), 5.05 (d, 1 H, $J_{1,2}$ 3.42 Hz, H-1), and 7.2–7.4 (m, 15 H, 3 Ph); ^{13}C, δ 50.6 (CH_2N_3), 66.6, 69.7, 72.0, 72.3, 73.1, 74.9, 77.6, and 81.8 (C-2/5 and 4 OCH_2), 101.7 (C-1), and 127.4–129.6 (aromatic). Anal. Calcd for $C_{28}H_{31}N_3O_5$: C, 68.69; H, 6.38. Found: C, 68.82; H, 6.60.

*2-Azidoethyl 2,3,6-tri-*O*-benzyl-4-*O*-(2,3,4,6-tetra-*O*-benzyl-β-*D*-galactopyranosyl)-α-*D*-glucopyranoside* (**11**).—Reaction of **5** (0.310 g, 0.29 mmol) with azidoethanol for 20 h gave **11** (0.2 g, 66%) isolated as a syrup; $[\alpha]_D$ +41° (*c* 0.25, $CHCl_3$). NMR data: ^{1}H, δ 3.3–3.6 (m, 8 H), 3.7–3.9 (m, 8 H), 4.2–5.1 (m, 16 H), and 7.1–7.4 (m, 35 H, 7 Ph); ^{13}C, δ 50.4 (CH_2N_3), 66.6, 67.8, 68.0, 70.4, 72.4, 72.9, 73.2, 73.5, 73.6, 74.5, 75.0, 75.3, and 76.3 (2 C), 78.8 and 79.8 (2 C), 82.2 (C-2/6, C-2′/6′, 7 OCH_2Ph, and OCH_2), 97.5 and 102.7 (C-1,1), and 126.9–128.1 (aromatic). Anal. Calcd for $C_{63}H_{67}N_3O_{11}$: C, 72.60; H, 6.48. Found: C, 72.82; H, 6.61.

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