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Note

2-Azidoethyl glycosides: glycosides potentially useful for the preparation of neoglycoconjugates

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Glycosides with a terminal amino group in the aglycon moiety are often used in the synthesis of neoglycoconjugates by attachment to carrier molecules or particles *via* amide bonds or their hetero-analogues[1,2]. The acrylamido group can be introduced into this type of aglycon *via* *N*-acryloylation, and the resulting glycosides can be transformed into high-molecular-weight neoglycoconjugates by copolymerisation with acrylamide[3–6]. In syntheses of the glycosides, the terminal amino group of the aglycon is usually protected *e.g.*, with an *N*-trifluoroacetyl[4,5] or *N*-benzyloxycarbonyl[7,8] group, and amino groups can also be generated from azido groups[9,10].

We now report the synthesis of 2-azidoethyl glycosides that can be converted easily by catalytic hydrogenation into 2-aminoethyl glycosides, which can then be used for the preparation of neoglycoproteins or for *N*-acryloylation[11].

The C_2 spacer in 2-aminoethyl glycosides can be elongated by condensation with *N*-trifluoroacetylglycine or *N*-trifluoroacetylglycylglycine in the presence of ethyl 2-ethoxy-1,2-dihydroquinoline-1-carboxylate (EEDQ) to afford peptide-like spacer-arm glycosides with diminished hydrophobicity[12].

Acetylated methyl (2-azidoethyl β-D-glucopyranosid)uronate (**1**) and 2-azidoethyl β-D-galactopyranoside (**3**) were prepared by reacting 2-azidoethanol (easily accessible from commercial 2-chloroethanol and sodium azide[13,14]) with the appropriate acetobromo sugar in the presence of mercury(II) salts. By using the boron trifluoride etherate method, the appropriate acetylated sugars were converted easily into the acetylated derivatives of **1**, 2-azidoethyl β-D-glucopyranoside (**2**), **3**, 2-azidoethyl α-D-mannopyranoside (**4**), and 2-azidoethyl α-L-rhamnopyranoside (**5**). Acetylated 2-azidoethyl glycosides prepared by the reaction promoted by boron trifluoride etherate had mobilities in

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t.l.c. (as for 2-bromoethyl glycosides[15]) similar to those of the starting acetylated sugars, but the reactions could be monitored by g.l.c.

G.l.c. of the reaction of 2-azidoethanol with 1,2,3,4,6-penta-*O*-acetyl-β-D-glucopyranose in the presence of Lewis acids (boron trifluoride etherate, trimethylsilyl triflate) revealed that formation of **2** was accompanied by anomerisation of the starting β-acetate and the degradation of **2**. In the boron trifluoride etherate-promoted reaction (see Experimental), the anomerisation was more rapid than glycosylation. However, in the trimethylsilyl triflate-promoted reaction in dichloromethane at 20° with 0.02% of the promoter, anomerisation of β-penta-acetate into the α anomer did not occur as glycosidation proceeded. Higher concentrations of the promoter accelerated the glycosylation but also increased the rate of degradation of the product **2**. When the glycosylation was performed on a preparative scale with 0.1% of trimethylsilyl triflate, 26% of a crystalline 1:1 mixture (^{13}C-n.m.r. data) of **2** and the α-penta-acetate was isolated. Thus, boron trifluoride etherate is preferred as the promoter for the preparation of 2-azidoethyl glycosides (see Table I).

When a solution of methyl (2-chloroethyl 2,3,4-tri-*O*-acetyl-β-D-glucopyranosid)uronate and tetrabutylammonium azide in benzene was boiled under reflux, the 2-azidoethyl glycoside **1** was obtained. Similarly, 2-chloroethyl 2,3,4,6-tetra-*O*-acetyl-β-D-galactopyranoside gave the 2-azidoethyl glycoside **3** in high yield.

2-Azidoethyl glycosides of mono- and di-saccharides have been used in the synthesis of fragments of bacterial polysaccharides from *Escherichia coli*[16] and *Proteus mirabilis*[17] for further transformation into neoglycoconjugates.

EXPERIMENTAL

Solvents were evaporated at <40° (bath). Optical rotations were determined at 24–32°, using a Jasco DIP-360 polarimeter, and i.r. spectra were recorded with a Perkin–Elmer 577 instrument. N.m.r. spectra were recorded at 25° with Bruker WM 250 (^{1}H) and AM 300 (^{13}C) spectrometers. T.l.c. was performed on Silica Gel 60F_{254} (Merck) with u.v. detection, charring with sulfuric acid, reaction with 1% of potassium permanganate in aqueous sodium carbonate (for unsaturated compounds), or with 0.3% of ninhydrin in acetone (for amines). Column chromatography was performed on silica gel L (40–100 μm, 100–160 μm), Silpearl (20–40 μm) (Czechoslovakia), or LiChroprep Si 60 (40–63 μm) (Merck). H.p.l.c. was performed on columns (6 × 150 mm, 5 μm analytical; 16 × 250 mm, 10 μm semi-preparative) of Silasorb 600, with a Knauer differential refractometer or an ISCO model UA-5 u.v. detector (254 or 280 nm). For g.l.c., a Hewlett–Packard 5890 instrument equipped with a flame-ionisation detector and an HP 3393A integrator was used. Separations were performed on a glass capillary column (0.2 mm × 25 m) coated with Ultra-1 (0.33-μm layer) at 200° with nitrogen as the carrier gas at 140 kPa. Syrupy or amorphous compounds were purified by column chromatography and characterised by n.m.r. spectroscopy. Organic solvents were distilled over appropriate drying agents.

TABLE I

Data for the acetylated 2-azidoethyl glycosides **1–5**

Compound	*Procedure*[a] *(reaction time)*	*Yield* (%)	*M.p.* (°)	$[\alpha]_D$ *(chloroform)*	*Molecular formula*	*Analysis* (%)					
						Calc.			*Found*		
						C	*H*	*N*	*C*	*H*	*N*
1	*a* (22 h)	39	96–98	−58° (*c* 1)	$C_{15}H_{21}N_3O_{10}$	44.67	5.25	10.42	44.43	5.32	10.56
	b	69									
	d	33									
2	*a* (26 h)	38	114–115	−40° (*c* 1.5)	$C_{16}H_{23}N_3O_{10}$	46.04	5.55	10.07	45.79	5.63	10.12
3	*a* (24 h)	75	syrup	−12° (*c* 1.7)							
	c	90									
	e	71									
4	*a* (26 h)	42	75–77	+39° (*c* 1.5)	$C_{16}H_{23}N_3O_{10}$	46.04	5.55	10.07	46.02	5.56	9.89
5	*a* (3 h)	90	syrup	−47° (*c* 1.5)							

[a] The procedures *a–e* are detailed in the Experimental section.

2-Azidoethanol. — 2-Chloroethanol (25.2 mL, 375 mmol) was added to a solution of sodium azide (30 g, 461 mmol) and sodium hydroxide (1.5 g, 37.5 mmol) in water (115 mL). The mixture was stirred at 20° for 2 days, sodium sulfate (35 g) was added, and, after 10 min, the mixture was extracted with dichloromethane (3 × 70 mL). The combined extracts were dried (Na_2SO_4) and concentrated, and the residue (~30 mL) was distilled to give 2-azidoethanol (28.7 g, 88%), b.p. 76–78°/25 mmHg, n_D^{21} 1.4615, ν_{max} 2120 cm^{-1} (N_3); lit.[13,14] b.p. 73°/20 mmHg, n_D^{25} 1.4578.

Synthesis of 2-azidoethyl glycosides. — (*a*) To a cooled (0°) solution of acetylated sugar (1 mmol, β anomer or an α,β-mixture) and 2-azidoethanol (1.2 mmol) in anhydrous dichloromethane (2 mL) was added BF_3-etherate (5 mmol) dropwise. The mixture was stirred for 1 h at 0°, then at room temperature (2–26 h), and the reaction was monitored by g.l.c. The mixture was diluted with chloroform (5 mL), washed with cold water (5 mL) and aqueous sodium hydrogencarbonate (5 mL), and filtered through cotton wool, and the solvent was evaporated. The 2-azidoethyl glycoside was isolated from the residue by column chromatography on silica gel (gradient of ether in toluene) and/or by crystallisation from ethyl acetate–hexane.

Methyl (2-azidoethyl 2,3,4-tri-*O*-acetyl-β-D-glucopyranosid)uronate (**1**), 2-azidoethyl 2,3,4,6-tetra-*O*-acetyl-β-D-glucopyranoside (**2**), 2-azidoethyl 2,3,4,6-tetra-*O*-acetyl-β-D-galactopyranoside (**3**), 2-azidoethyl 2,3,4,6-tetra-*O*-acetyl-α-D-mannopyranoside (**4**), and 2-azidoethyl 2,3,4-tri-*O*-acetyl-α-L-rhamnopyranoside (**5**) were synthesised by this procedure. See Tables I and II.

(*b*) Crystalline methyl (2,3,4-tri-*O*-acetyl-α-D-glucopyranosyl bromide)uronate[18] (1.19 g, 3 mmol) was added in one portion to a hot (105°) solution of mercury(II) cyanide (780 mg, 3.1 mmol) in 2-azidoethanol (3.27 mL, 43.2 mmol). The mixture was stirred at 105–110° for 10 min, then overnight at room temperature. T.l.c. (hexane–ethyl acetate, 6:4) revealed no starting material (R_F 0.59), but a product (R_F 0.30) appeared. An excess of 2-azidoethanol was removed *in vacuo* (< 1 mm). A solution of the residue in

TABLE II

^{13}C-N.m.r. data (δ in p.p.m.) for **1–5**[a]

Compound	*C-1* ($J_{C-1,H-1}$ *in Hz*)	*C-2,3,4,5*				*C-6*	*OCH_2*	*CH_2N_3*
1	100.6[b]	72.6	72.0	71.0	69.4	167.1	68.8	50.5
2	100.7							
	(160)	72.9	72.1	71.2	68.6	62.0	68.3	50.5
3	101.0							
	(159)	70.8	70.7	68.4	66.9	61.2	68.3	50.4
4	97.7							
	(168)	69.3	68.8(2C)		66.0	62.4	66.9	50.3
5	97.5							
	(159)	70.8	69.5	68.8	66.6	17.2	66.6	50.3

[a] Other resonances: $COCH_3$ 20.3–20.7, $COOCH_3$ 52.9, $COCH_3$ 169.3–170.3 p.p.m. [b] The β configuration of **1** was confirmed by the signal for H-1 at δ 4.64 (d, $J_{1,2}$ 7.8 Hz).

chloroform (50 mL) was washed with water (50 mL). The aqueous layer was extracted with chloroform (3 × 50 mL). The combined organic phases were then washed with M sodium iodide (4 × 200 mL), saturated sodium hydrogencarbonate (200 mL), and water (200 mL), dried (Na_2SO_4), and concentrated. Chromatography of the residue (1.25 g) on a column (25 × 250 mm) of Silpearl with light petroleum–ethyl acetate (65:35) gave crystalline **1** (840 mg, 69%). G.l.c. of the crude product revealed 3% of the α anomer. The analytical sample of **1** was obtained by recrystallisation from ether (see Tables I and II) and had ν_{max} 2120 cm^{-1} (N_3).

(*c*) A solution of mercury(II) cyanide (252 mg, 1 mmol), mercury(II) bromide (360 mg, 1 mmol), and 2-azidoethanol (0.15 mL, 2 mmol) in acetonitrile (5 mL, freshly distilled over calcium hydride) was stirred with molecular sieves type 4A for 15 min under argon. A solution of 2,3,4,6-tetra-*O*-acetyl-α-D-galactopyranosyl bromide (822 mg, 2 mmol) in acetonitrile (5 mL) was added dropwise over ~10 min and the mixture was then stirred at 20° for 12 h in the dark. T.l.c. (ethyl acetate–hexane, 3:2; R_F 0.50) showed complete reaction. The mixture was diluted with chloroform (50 mL), filtered, washed with cold water (25 mL) and M sodium iodide (25 mL), dried ($MgSO_4$), and concentrated. Column chromatography (ethyl acetate–hexane, 35:65) gave **3**, isolated as a syrup (750 mg, 90%), homogeneous in t.l.c. (see Tables I and II).

(*d*) Crystalline methyl (2,3,4-tri-*O*-acetyl-α-D-glucopyranosyl bromide)uronate[18] (500 mg, 1.26 mmol) was added in one portion to a hot (105°) solution of mercury(II) cyanide (317 mg, 1.26 mmol) in 2-chloroethanol (5 mL, 66.5 mmol). The mixture was stirred at 105–110° for 15 min, then concentrated *in vacuo*. A solution of the residue in chloroform (40 mL) was washed with M sodium iodide (2 × 20 mL), saturated aqueous sodium hydrogencarbonate (20 mL), and water (20 mL), dried (Na_2SO_4) and concentrated. The residue (460 mg) was crystallised from ethanol to give methyl (2-chloroethyl 2,3,4-tri-*O*-acetyl-β-D-glucopyranosid)uronate (336 mg, 67%), m.p. 136–138°, $[\alpha]_D$ −22° (*c* 1.3, chloroform). N.m.r. data ($CDCl_3$): ^{13}C, δ 170.1, 169.4 (2 C, 2 C=O), 167.2 ($COOCH_3$), 101.1 (C-1, $J_{C\text{-}1,H\text{-}1}$ 161 Hz), 72.8 (C-3), 71.8 (C-5), 71.0 (C-2), 70.1 (OCH_2), 69.4 (C-4), 53.0 ($COOCH_3$), 42.5 (CH_2Cl), 20.6 ($COCH_3$); ^1H, δ 5.30 (dd, 1 H, $J_{3,4}$ 9.5 Hz, H-3), 5.22 (dd, 1 H, H-4), 5.05 (dd, 1 H, $J_{2,3}$ 9.4 Hz, H-2), 4.62 (d, 1 H, $J_{1,2}$ 8.4 Hz, H-1), 4.12–4.22 (m, 1 H, OCH_{2A}), 4.07 (d, 1 H, $J_{4,5}$ 9.9 Hz, H-5), 3.88 (s, 3 H, COOMe), 3.72–3.83 (m, 1 H, OCH_{2B}), 3.61–3.67 (m, 2 H, CH_2Cl), 2.05 (s, 3 H, Ac), 2.01 (s, 6 H, 2 Ac).

Anal. Calc. for $C_{15}H_{21}ClO_{10}$: C, 45.41; H, 5.33; Cl, 8.94. Found: C, 45.56; H, 5.57; Cl, 8.08.

To a solution of tetrabutylammonium azide[19,20] (426 mg, 1.5 mmol) in dry benzene (6 mL) was added methyl (2-chloroethyl 2,3,4-tri-*O*-acetyl-β-D-glucopyranosid)uronate (300 mg, 0.757 mmol). The mixture was boiled under reflux, and the reaction was monitored by g.l.c. After 1 h, the mixture was washed with water, the organic layer was treated with activated carbon, and the solvent was evaporated. The residue (233 mg) was crystallised from ether to give **1** (100 mg, 33%).

(*e*) To a cooled (ice–water), stirred solution of 1,2,3,4,6-penta-*O*-acetyl-β-D-galactopyranose (780 mg, 2 mmol) and 2-chloroethanol (0.175 mL, 2.3 mmol) in

anhydrous dichloromethane (4 mL) was added BF_3-etherate (1.23 mL, 10 mmol) dropwise. Stirring was continued for 1 h at 0°, then at room temperature (6 h), and the reaction was monitored by g.l.c. The mixture was worked-up as in (*a*) to give 2-chloroethyl 2,3,4,6-tetra-*O*-acetyl-β-D-galactopyranoside (758 mg, 92%), isolated as a syrup, which was used in the next step without further purification. ^{13}C-N.m.r. data ($CDCl_3$): δ 169.5–170.3 (C=O), 101.6 (C-1, $J_{C\text{-}1,H\text{-}1}$ 161 Hz), 70.8, 70.7, 68.5, and 66.9 (C-2,3,4,5), 69.9 (OCH_2), 61.2 (C-6), 42.5 (CH_2Cl), 20.6–20.7 ($COCH_3$).

A solution of the above glycoside (205 mg, 0.5 mmol) and tetrabutylammonium azide (284 mg, 1 mmol) in anhydrous benzene (3 mL) was boiled under reflux and the reaction was monitored by g.l.c. After 2.5 h, the mixture was diluted with chloroform (20 mL), washed with water (10 mL), filtered through cotton wool, and concentrated. Column chromatography (ethyl acetate–hexane, 35:65) of the residue gave **3** (147 mg, 71%).

*2-(*N*-Trifluoroacetylglycylamino)ethyl and 2-(*N*-trifluoracetylglycylglycylamino)ethyl* α-D-*mannopyranosides.* — A mixture of **4** (121 mg, 0.29 mmol) and Dowex 1-X8 (HO^-) resin (2mL) in methanol–water (4:1, 4 mL) was stirred for 2 h at 20°, then filtered and concentrated. A solution of the residue in methanol (2 mL) was hydrogenated over 10% Pd/C for 2 h at 20°, and the reaction was monitored by t.l.c. (ethanol–1-butanol–pyridine–water–acetic acid, 100:10:10:10:3). The catalyst was removed and the filtrate was concentrated, to leave 2-aminoethyl α-D-mannopyranoside (65 mg, 0.29 mmol), to a solution of which in dry *N,N*-dimethylformamide (3 mL) were added *N*-trifluoroacetylglycine[22] (50 mg, 0.29 mmol) and EEDQ[23] (80 mg, 0.32 mmol). The mixture was kept at 20°. After 24 h, t.l.c. revealed a product with R_F 0.55. The mixture was co-concentrated with toluene, and a solution of the residue in methanol (5 mL) was treated with KU-2 (H^+) resin, then filtered, and concentrated. Column chromatography on silica gel (hexane–ethyl acetate, 3:2) of the residue gave 2-(*N*-trifluoroacetylglycylamino)ethyl α-D-mannopyranoside (90 mg, 82%), isolated as a syrup homogeneous according to t.l.c., $[\alpha]_D$ + 66.5° (*c* 1, methanol). ^{13}C-N.m.r. data (CD_3OD): δ 170.0 (CONH), 101.6 (C-1), 74.8 (C-5), 72.5 (C-3), 72.0 (C-2), 68.7 (C-4), 67.1 (OCH_2CH_2NH), 62.9 (C-6), 43.2 ($COCH_2NH$), 40.4 (OCH_2CH_2NH).

Similarly, 2-(*N*-trifluoroacetylglycylglycylamino)ethyl α-D-mannopyranoside was obtained (82%), R_F 0.55 (hexane–ethyl acetate, 3:2), $[\alpha]_D$ + 58° (*c* 1, methanol). ^{13}C-N.m.r. data (CD_3OD): δ 171.6, 170.7 (CONH), 101.6 (C-1), 74.7 (C-5), 72.5 (C-3), 72.0 (C-2), 68.7 (C-4), 67.1 (OCH_2CH_2NH), 62.9 (C-6), 43.4 (2C) ($COCH_2NH$), 40.3 (OCH_2CH_2NH).

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