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Syntheses of oligomannosides in solution and on a soluble polymer support: a comparison

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Abstract—The α-(1→6)-linked and the α-(1→2)-linked linear mannotetraose glycosides **3** and **4**, respectively, and the branched mannopentaoside **2** [R = $CH_2(CH_2)_2CH_2Cl$] were synthesised by conventional methods in solution, using trichloroacetimidate donors, and the products were obtained in 39%, 42% and 40% overall yield, respectively. For comparative purposes, the same two linear tetrasaccharides were prepared by use of MPEG as a soluble polymer support, the yields being 34% and 14%, respectively. An attempted MPEG-supported synthesis of the branched pentasaccharide was unsuccessful. The merits and shortcomings of oligosaccharide syntheses on MPEG are discussed.

Keywords: Mannose oligosaccharides; Solution synthesis; MPEG-supported synthesis

1. Introduction

Oligosaccharides of D-mannose are found in nature as essential substructures of many bioactive glycoconjugates, such as *N*-glycans, fungal cell wall mannans[1] and GPI anchors,[2] and as high affinity ligands for mannose 6-phosphate receptors.[3] There is a great demand, therefore, for efficient and practical synthetic access to these compounds. The present investigation was motivated by our requirements for sizeable quantities of selectively phosphorylated manno-oligosaccharides, such as the diphosphorylated branched pentasaccharide **1**. As essential substructures of the transport system, which is responsible for the delivery of newly synthesised acid hydrolases from the Golgi apparatus to the lysosomes in higher eukaryotic cells,[3] such compounds have great potential for targeted drug delivery.

Very effective strategies for the conventional step-wise solution synthesis of manno-oligosaccharides have been developed by, for example, Ogawa's,[4–6] Kong's,[7–9] Ley's,[10–12] Schmidt's[2,13] and Seeberger's[14] groups. High yields were achieved in relatively short reaction sequences from monosaccharide building blocks by prudent choice of protecting groups and/or fine-tuning of mannosyl donors. Such syntheses, involving selective protection, followed by glycosylation and deprotection sequences, are nevertheless time consuming as they require chromatographic purification of all intermediates. Over the last two decades efforts have been made to develop protocols for the construction of oligosaccharides on polymer supports,[15] which simplify the isolation and purification of intermediates, since non-supported reagents can be removed merely by washing the insoluble products with suitable solvents. Chromatography is required only once, after cleavage of the end-product from the support. The automation of oligosaccharide synthesis has lagged decades behind those of the oligopeptides and oligonucleotides, but significant developments have now been reported.[15d,16]

In spite of its considerable merits, progress in polymer supported oligosaccharide synthesis has, on the whole, been slow because glycosidic bond formation does not, in general, proceed with the high yield and stereoselectivity necessary for successful application of this

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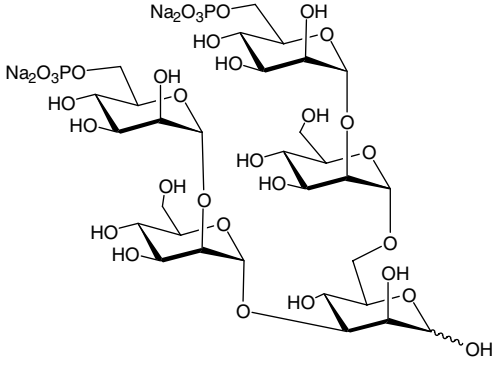
technique. α-Mannosylation is one of the few exceptions, and the polymer-supported synthesis of α-linked oligomannosides has therefore seemed a feasible alternative to classical solution synthesis. Thus, α-(1→2)-linked manno-tri-,[17] tetra-,[17] hexa-[18] and hepta[19] -ose derivatives have been prepared on Merrifield's resin with isolated yields of cleaved products of 54%, 34%, 19% and 9%, respectively, and when syntheses of such compounds were carried out under automation, higher yields were achieved.[16]

A distinct disadvantage of synthesis on the most common, insoluble supports, for example, polystyrene resins or controlled-pore glass, is the difficulty of monitoring the progress of reactions. This problem can be overcome by the use of soluble polymer supports, in particular polyethylene glycol ω-monomethyl ether [$HOCH_2$-$CH_2(OCH_2CH_2)_nOMe$ (MPEG)], introduced by Krepinski and co-workers.[20,21] While insoluble in simple dialkyl ethers, MPEG dissolves readily in halogenated solvents allowing it to react under homogeneous conditions and its reactions to be monitored by NMR spectroscopy. A successful synthesis of an α-(1→2)-linked mannotetraoside derivative on MPEG has been reported, although no yield was given.[20]

Only a few direct comparisons between solution and polymer-supported approaches to oligosaccharide synthesis have been published. Hewitt and Seeberger[22] achieved nearly equal yields (17% and 18%, respectively) in the syntheses of an α-glycoside of the branched tetrasaccharide α-D-Man-(1→2)-α-D-Man-(1→2)-[β-D-Gal-(1→4)]-Man in solution and on Merrifield's resin. The polymer-supported approach was however considerably faster. Whitfield and coworkers[23] found MPEG-supported methodology advantageous in the preparation of trisaccharide derivative β-D-GlcNAc-(1→3)-β-D-Gal-(1→4)-β-D-GlcSPh and tetrasaccharide derivative β-D-Gal-(1→4)-β-D-GlcNAc-(1→3)-β-D-Gal-(1→4)-β-D-GlcSPh, as the classical solution syntheses were laborious and low yielding (32% and 28% for the tri- and tetra-saccharide, respectively) because of the extensive chromatographic purification required following the formation of the β-D-GlcNAc-(1→3)-D-Gal glycosidic bonds. On MPEG, both the tri- and tetra-saccharide were obtained with less effort and in 45% yield. Lam and Gervay-Hague,[24] on the other hand, gave preference to conventional synthesis in solution over synthesis on Tentagel (polystyrene resin with PEG-chains grafted onto the backbone) for the step-wise assembly of α-(1→6)-linked glucosyl oligomers using monosaccharide glycosyl iodides as donors. Not only were the yields slightly higher, but the glycosylations needed far smaller excesses of glycosyl donors and proceeded more rapidly in solution. Chromatography, although necessary at each step, was in this case uncomplicated and fast.



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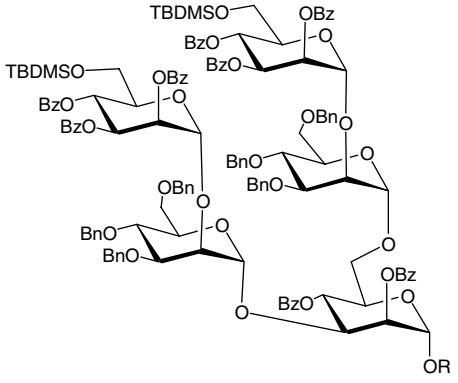

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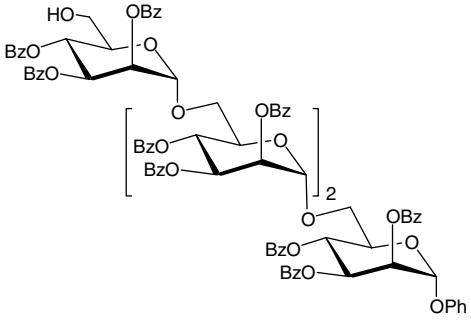

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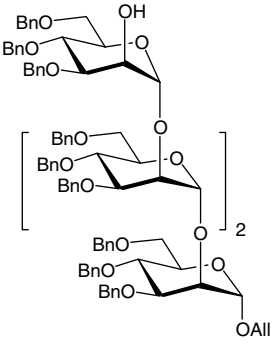

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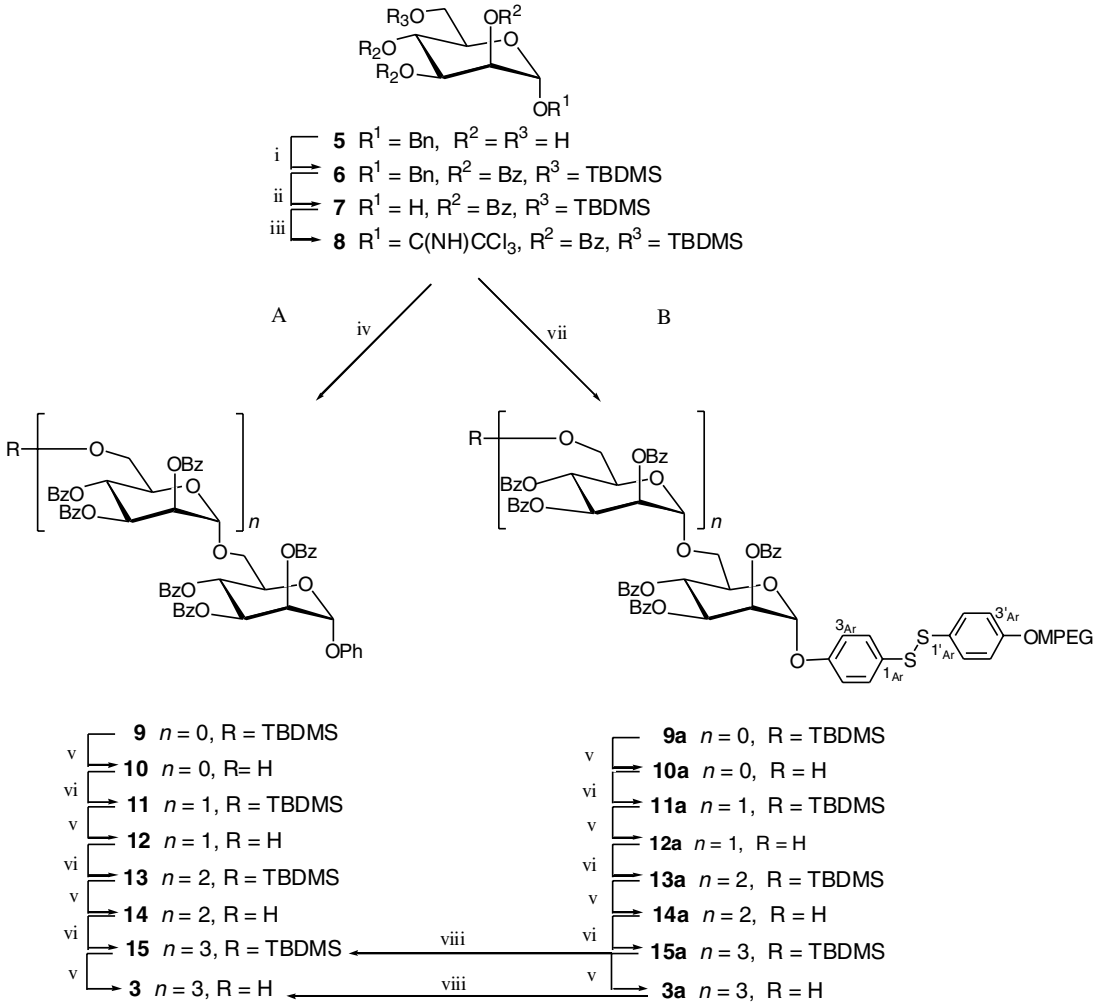
2. Results and discussion

In an initial model study, we compared the efficiency and convenience of conventional solution synthesis with synthesis on MPEG as polymer-support by preparing the linear tetrasaccharide derivatives **3** and **4** under conditions, which were otherwise as similar as possible. Analogous parallel approaches to the synthesis of precursor **2** (R = H) of the diphosphorylated branched pentasaccharide **1** were then undertaken.

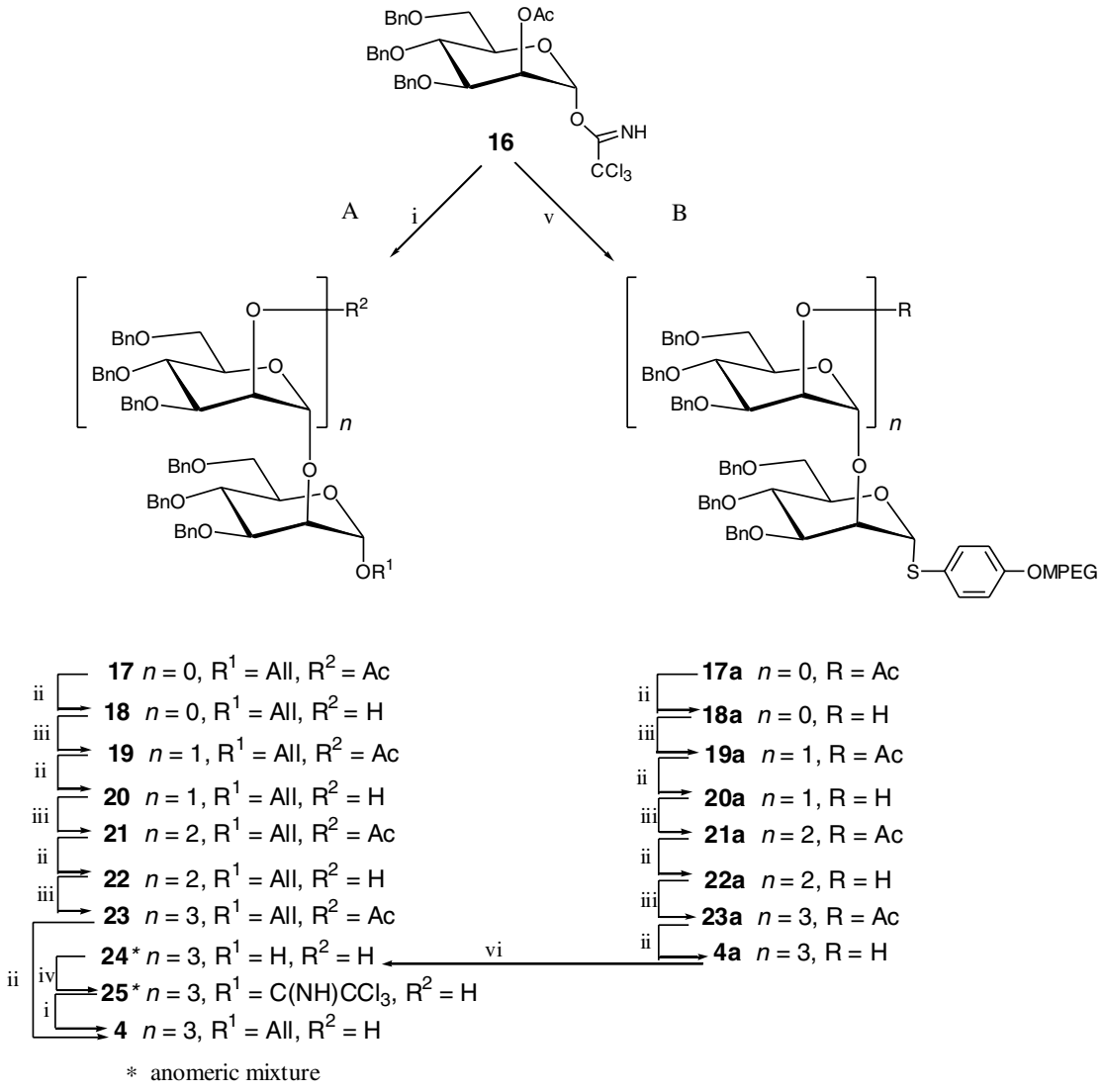
2.1. Linear tetrasaccharide glycosides 3 and 4

The α-(1→6)-linked tetrasaccharide derivative **3** and the α-(1→2)-linked tetrasaccharide derivative **4** were assembled step-wise by use of mannosyl trichloroacetimidates **8** and **16**,[25,26] respectively, the solution phase (A) and the polymer-support (B) routes being outlined in Schemes 1 and 2, respectively. Donor **8**, used in making tetramer **3**, was prepared from the known benzyl α-D-mannopyranoside **5**[27] in three standard steps: (i) selective silylation followed by benzoylation to give the fully protected **6**; (ii) debenzylation to give free sugar **7**; (iii) introduction of trichloroacetimidate functionality (Scheme 1). The overall yield was 74%. Donor **16** has been reported previously.[25,26]

2.1.1. Solution synthesis of the α-(1→6)-linked tetrasaccharide derivative 3. Phenyl glycoside **10**, obtained by reaction of trichloroacetimidate **8** with phenol to give **9** and subsequent desilylation with aqueous acetic acid, served as the first mannosyl-based acceptor. Iterative glycosylations with donor **8**, followed by desilylations, furnished target **3** via disaccharide derivatives **11** and **12**, trisaccharide compounds **13** and **14** and the tetrasaccharide derivative **15** in 39% overall yield from the substituted phenyl mannoside **9** (Scheme 1A).



Scheme 1. Reagents: (i) a, TBDMSCl, py, b, BzCl; (ii) Pd/C, HCO_2NH_4, MeOH; (iii) a, K_2CO_3, CH_2Cl_2, b, Cl_3CCN; (iv) a, PhOH, CH_2Cl_2, sieves, b, TMSOTf; (v) AcOH, H_2O; (vi) **8**, CH_2Cl_2, TMSOTf; (vii) a, $HOC_6H_4(p)SSC_6H_4(p)OMPEG$, CH_2Cl_2, sieves, b, $BF_3{\cdot}OEt_2$; (viii) $NiCl_2$, H_3BO_3, $NaBH_4$, EtOH, H_2O.

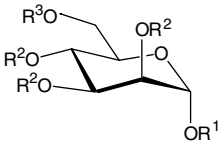


Scheme 2. Reagents: (i) a, AllOH, CH_2Cl_2, sieves, b, TMSOTf; (ii) DBU, MeOH; (iii) **16**, TMSOTf, CH_2Cl_2, sieves; (iv) a, K_2CO_3, CH_2Cl_2, b, CCl_3CN; (v) a, $HSC_6H_4OMPEG(p)$, sieves, b, CH_2Cl_2, $BF_3{\cdot}OEt_2$; (vi) NBS, 2,6-di-*tert*-butylpyridine, Me_2CO, H_2O.

2.1.2. MPEG-supported synthesis of the α-(1→6)-linked tetrasaccharide derivative 3. The strategy chosen employed the polymer support 4-hydroxy-4′-MPEG-oxydiphenyl disulfide,[28] which was glycosylated with trichloroacetimidate **8** in CH_2Cl_2 in the presence of $BF_3{\cdot}OEt_2$. Threefold excesses of donor and of promoter were required to achieve complete glycosylation of the phenolic hydroxyl group. The resulting mannosylated polymer **9a**, after precipitation with ether and recrystallisation from ethanol, was desilylated by heating in aqueous acetic acid to give **10a**, and the product was again precipitated with ether and recrystallised from ethanol. Highly selective and near quantitative glycosylation of the monohydroxy polymer to **10a** was clearly indicated by NMR spectroscopy.

The ^{1}H NMR resonances for H-1–H-5 of the sugar residues of **9a** and **10a** were very similar to those of the corresponding unsupported compounds **9** and **10**, respectively (see Table 1). In the ^{1}H NMR spectrum of **9a**, integration of the signals for the two pairs of aromatic protons *meta* to the disulfide bridge of the linker (H-3_{Ar}, H-$3'_{Ar}$, δ 7.15, 6.84) and of those for H-1–H-4 of the carbohydrate moiety (δ 6.09–5.75) showed a ratio of 1:1, indicating that the support was fully glycosylated. The resonances of the sugar CH_2 protons and the aromatic protons *ortho* to the disulfide bridge were concealed by the strong signals for the polyether methylene and the benzoyl groups, respectively. Both compounds **9a** and **10a** had a single ^{13}C resonance characteristic of C-1 of α-mannopyranosides (**9a**: δ 96.1, $J_{C\text{-}1,H\text{-}1}$ 175 Hz; **10a**: δ 96.1, $J_{C\text{-}1,H\text{-}1}$ 173 Hz).[29] The resonances of all other carbon atoms of the sugar rings were hidden by the massive signal of the polyether methylene groups. Complete deprotection at O-6 of **9a** was

Table 1. ^{1}H and ^{13}C NMR chemical shifts (δ) for the monosaccharide precursors of (1→6)-linked oligosaccharide derivatives[a]



Compd	R^1	R^2	R^3	H-1	H-2	H-3	H-4	H-5	H-6a	H-6b	C-1	C-2	C-3	C-4	C-5	C-6
5[b]	Bn	H	H	4.99	3.96	3.81	3.77	3.6–3.7	3.88	~3.7	99.8	70.5	71.0	67.2	73.3	61.3
6	Bn	Bz	TBDMS	5.12	5.71	5.86	5.94	4.15	3.75–3.85		97.1	71.1[c]	71.0[c]	67.4	72.2	62.7
7	H	Bz	TBDMS	5.48	5.68	5.90	5.97	4.37	3.8–3.9		92.8	71.6	70.6	67.4	71.9	62.9
8	C(NH)-CCl_3	Bz	TBDMS	6.54	5.85–5.9		6.14	4.32	3.8–3.9		95.4	70.7[c]	69.4[c]	66.4	74.6	62.2
9[d]	Ph	Bz	TBDMS	5.78	5.88	6.06	6.10	4.26	3.84	3.80	96.4	70.9	70.8	66.9	72.4	62.3
9a[e]	MPEG[f]	Bz	TBDMS	5.75	5.83	6.03	6.09	~4.2	—	—	96.1, $J_{C\text{-}1,H\text{-}1}$ 175 Hz	—	—	66.8	—	61.9
10	Ph	Bz	H	5.84	5.88	6.22	5.97	4.19	3.88	3.73	96.4	70.9	69.9	67.4	72.1	61.5
10a[e]	MPEG[f]	Bz	H	5.82	5.85	6.16	5.95	~4.1	—	—	96.1, $J_{C\text{-}1,H\text{-}1}$ 173 Hz	—	—	67.0	—	61.1

[a] The resonances for benzyl, benzoyl, phenyl and TBDMS groups were present as required but are not reported. Coupling constants were recorded as follows: $J_{1,2}$ 1.6 ± 0.2, $J_{2,3}$ 3.2 ± 0.1, $J_{3,4} = J_{4,5}$ 9.95 ± 0.15, $J_{5,6a}$ 2.6 ± 0.6, $J_{5,6b}$ 3.85 ± 0.55, $J_{6a,6b}$ 12.5 ± 0.5.

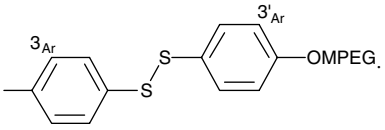
[b] Spectra recorded in D_2O.

[c] Assignments may be reversed.

[d] ^{1}H NMR spectrum recorded at 500 MHz.

[e] For polymer-supported compounds, selected assignments were made by comparison with the spectra of the corresponding solution-synthesised compounds. In their ^{1}H NMR spectra singlets for OMe at δ 3.38, triplets for $ArOCH_2CH_2$ at δ 4.11 and doublets for H-3_{Ar} and H-$3'_{Ar}$ at δ 7.15 and 6.86, respectively, were also observed but are not reported as were signals for OMe at δ 58.9 in their ^{13}C NMR spectra. The signals for H-6a, H-6b, C-2, C-3 and C-5 were hidden under the massive polyether methylene peaks.

[f] 'MPEG' =



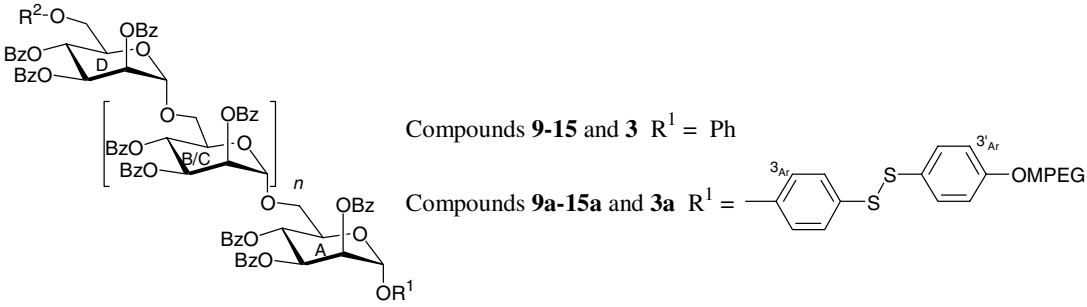
ascertained by the absence of signals for the silyl group in the ^{1}H and ^{13}C NMR spectra of compound **10a**.

Three iterative glycosylation/desilylations were then carried out on compound **10a** under the conditions used in the corresponding solution synthesis, that is, with trichloroacetimidate **8** as donor and TMSOTf as promoter, followed by treatment with hot aqueous acetic acid, to furnish, successively, the MPEG-supported intermediates **11a**–**15a** and **3a** (Scheme 1B). Yields of supported materials, for example, **9a** and **10a**, were >95% in the initial steps, but diminished with growing chain-length, such that compounds **15a** and **3a** were recovered in 61% and 53% yield, respectively, based on the amount of compound **9a** used. The decreasing yields were thought to be caused by the increasing ether-solubility of the sugar–polymer conjugates, as well as gradual deterioration of the MPEG polyether backbone. At the same time, the NMR spectra of successive products became less well resolved and showed the presence of increasing proportions of by-products. The anomeric carbon atom and proton signals, as well as a number of other proton resonances, were nevertheless readily assigned, and their chemical shifts and their $^{1}H,^{1}H$-coupling constants showed remarkable agreement with those parameters of the equivalent unsupported compounds (see Tables 1–3).

To remove the tetrasaccharide product from the polymer, **15a** was reductively desulfurised using sodium borohydride with nickel chloride and boric acid in aqueous ethanol[30] to give the previously made tetrasaccharide phenyl glycoside **15** (Scheme 1A) in 67% yield, which corresponds to a 41% overall yield from the supported monosaccharide glycoside **9a**. The desilylated **3a**, on similar reductive cleavage of the sulfur–sulfur bond, released 65% of the theoretically expected amount of tetrasaccharide phenyl glycoside derivative **3**, corresponding to a 34% overall yield from the monosaccharide conjugate **9a**.

2.1.3. Solution synthesis of the α-(1→2)-linked tetrasaccharide derivative 4. The known allyl 3,4,6-tri-*O*-benzyl-α-D-mannopyranoside **18**,[5,31] prepared from trichloroacetimidate **16** by way of 2-*O*-acetate **17**,[5,31] was subjected to iterative glycosylation and deacetylation with the same donor and DBU in methanol, to afford successively disaccharides **19** and **20**, both

Table 2. ^{1}H NMR chemical shifts (δ) for (1→6)-linked oligosaccharide derivatives[a,b]



Compound	*n*	Sugar ring	R^2	H-1	H-2	H-3	H-4	H-5	H-6a	H-6b
11[c]	0	A	—	5.82	5.9–6.0	6.14	6.17	4.54	4.08	3.72
		D	TBDMS	5.08	5.71	5.86	5.9–6.0	4.04	3.65	3.58
11a	0	A	—	5.81	5.91	6.10	6.20	4.48	—	—
		D	TBDMS	5.09	5.72	5.87[d]	—	—	—	—
12	0	A	—	5.75	5.87	6.05	6.12	4.44	3.97	3.68
		D	H	5.04	5.66	5.93	5.72	3.86	3.47	3.40
12a	0	A	—	5.81	5.92[d]	6.09	6.22	4.47	—	—
		D	H	5.14	5.75[d]	6.03	—	—	—	—
13[c]	1	A	—	5.87	6.00	6.19	6.42	4.68	4.29	3.81
		B	—	5.17	5.89	6.06	5.96	4.22	3.87	3.31
		D	TBDMS	4.71	5.46	5.79	5.91	3.98	3.62	3.58
13a	1	A	—	5.85	—	6.14	6.46	~4.7	4.31	—
		B	—	5.19	5.41	5.78	—	4.21	—	—
		D	TBDMS	4.68	—	6.08	—	—	—	—
14[c]	1	A	—	5.87	6.00	6.18	6.38	4.67	4.23	~3.8
		B	—	5.14	5.88	6.03	~6.0	~4.2	~3.8	3.36
		D	H	4.81	5.50	5.95	5.73	3.88	3.53	3.44
14a	1	A	—	5.85	—	6.19	6.43	4.6–4.65	—	—
		B	—	5.16	—	—	—	—	—	—
		D	H	4.78	5.45	6.14	—	—	—	—
15[c,e]	2	A	—	5.88	6.00	6.19	6.42	4.70	4.29	3.84
		B	—	4.82	5.59	~5.9	6.09	~4.28	~3.93	3.41
		C	—	4.94	5.70	~5.9	6.07	4.22	3.86	3.44
		D	TBDMS	5.20	~5.9	~5.9	5.96	~3.93	3.54	3.46
15a[e]	2	A	—	5.85	—	6.15	6.46	4.68	—	—
		B	—	4.77	5.53	—	—	—	—	—
		C	—	4.94	5.71	—	—	—	—	—
		D	TBDMS	5.21	—	—	—	—	—	—
3[c,e]	2	A	—	~5.9	5.99	6.18	6.39	4.68	4.27	3.83
		B	—	4.79	5.60	~5.9	6.10	4.18	3.77	3.45
		C	—	4.98	5.71	~6.05	6.06	~4.25	3.91	~3.4
		D	H	5.19	~5.9	6.01	5.74	~3.8	~3.4	~3.4
3a[e]	2	A	—	—	—	6.14	6.44	~4.65	—	—
		B	—	4.76	~5.6	—	—	—	—	—
		C	—	5.00	—	—	—	—	—	—
		D	H	5.21	—	—	—	—	—	—

[a] The resonances for benzoyl, benzyl, phenyl and TBDMS groups were present as required but are not reported. Coupling constants were recorded as follows: $J_{1,2}$ 1.6 ± 0.2, $J_{2,3}$ 3.2 ± 0.1, $J_{3,4} = J_{4,5}$ 9.95 ± 0.15, $J_{5,6a}$ 2.6 ± 0.6, $J_{5,6b}$ 3.85 ± 0.55, $J_{6a,6b}$ 12.5 ± 0.5 Hz.

[b] For MPEG-supported compounds, selected assignments were made by comparison with the spectra of the corresponding solution-synthesised compounds; the primary sugar protons and most of the aromatic linker protons were hidden by the massive signals for the polyether methylene and the benzoyl groups, respectively. Singlets for OMe at δ 3.38, triplets for ArOCH_2CH$_2$ at δ 4.11 and doublets for H-3′$_{Ar}$ at δ 6.86 were also observed.

[c] Spectrum recorded at 500 MHz.

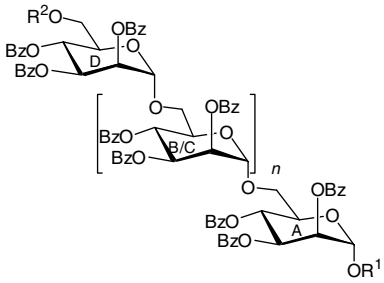
[d] Tentative assignment.

[e] The resonances in each row are derived from the same sugar moiety (^{1}H–^{1}H COSY and ^{1}H–^{13}C COSY experiments), but the assignments for rings B and C might be reversed.

reported previously,[31] trisaccharides **21** and **22**, and tetrasaccharide derivatives **23** and **4** (Scheme 2A). The final compound of this series, allyl glycoside **4**, was obtained in 42% overall yield from the initial acceptor **18**.

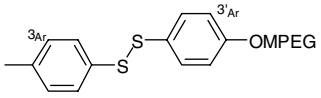
2.1.4. MPEG-supported synthesis of the α-(1→2)-linked tetrasaccharide derivative 4. For this synthesis the linker 4-MPEGoxythiophenol, readily obtained by reductive cleavage of the sulfur–sulfur bond of the phenolic

Table 3. ^{13}C NMR chemical shifts (δ) for (1→6)-linked oligosaccharide derivatives[a,b]



Compounds **9-15** and **3** R^1 = Ph

Compounds **9a-15a** and **3a** R^1 =



Compound	*n*	Sugar ring	R^2	C-1	C-2	C-3	C-4	C-5	C-6
11	0	A	—	96.8	71.0[c]	70.5	67.2	70.8[c]	66.8
		D	TBDMS	98.1	70.9[c]	70.6[c]	67.0	71.8	62.2
11a	0	A	—	96.4	—	—	—	—	66.1
		D	TBDMS	97.8	—	—	—	—	61.7
12	0	A	—	96.6	70.7[c]	70.5[c]	67.1	70.3[c]	66.9
		D	H	98.3	70.6[c]	70.1	67.4	71.3	61.3
12a	0	A	—	96.0	—	—	—	—	66.2
		D	H	97.6	—	—	—	—	60.5
13	1	A	—	96.9	70.8	70.8	66.8	70.5	66.7
		B	—	98.3	70.8	70.8	66.9	69.8	66.0
		D	TBDMS	97.6	70.8	70.8	67.3	71.8	62.1
13a	1	A	—	96.8	—	—	—	—	66.6
		B	—	98.1	—	—	—	—	65.6
		D	TBDMS	97.2	—	—	—	—	61.9
14	1	A	—	96.9	69.9[c]	69.9 or 70.7	66.9	70.5	66.8
		B	—	98.4	69.9 or 70.7	69.9 or 70.7	67.2	69.8[c]	66.3
		D	H	97.9	69.9 or 70.7	69.9 or 70.7	67.6	71.3	61.4
14a	1	A	—	96.6	—	—	—	—	—
		B	—	98.1	—	—	—	—	—
		D	H	97.4	—	—	—	—	61.0
15	2	A	—	96.9	70.6, 70.8 or 71.1		67.3 or 66.8	~71	~67
		B	—	97.9[c]	70.6, 70.8 or 71.1		67.3 or 66.8	69.8	66.0
		C	—	98.2[c]	70.6, 70.8 or 71.1		67.3 or 66.8	69.8	66.3
		D	TBDMS	98.5	70.6, 70.8 or 71.1		67.3 or 66.8	71.7	61.9
15a	2	A	—	97.2	—	—	—	—	—
		B	—	97.9[c]	—	—	—	—	—
		C	—	98.3[c]	—	—	—	—	—
		D	TBDMS	98.7	—	—	—	—	—
3	2	A	—	96.9	69.8–71.4		66.4–67.5	69.8–71.4	66.4–67.5
		B	—	98.1[c]	69.8–71.4		66.4–67.5	69.8–71.4	66.4–67.5
		C	—	98.5[c]	69.8–71.4		66.4–67.5	69.8–71.4	66.4–67.5
		D	H	98.5	69.8–71.4		66.4–67.5	71.3	61.3
3a	2	A	—	96.5	—	—	—	—	—
		B	—	98.0[c]	—	—	—	—	—
		C	—	97.9[c]	—	—	—	—	—
		D	H	97.4	—	—	—	—	—

[a] Resonances for benzoyl, benzyl, phenyl and TBDMS groups were present as required but are not reported.

[b] For MPEG-supported compounds, C-1 and C-6 were assigned by comparison with the spectra of the corresponding solution-synthesised compounds. The signals of all other carbohydrate carbon atoms were hidden under the massive polyether methylene peaks. Signals for MeO at δ 58.9 were also observed.

[c] Assignments may be reversed.

MPEG derivative[28] used to make compound **3** (Scheme 1B), was employed. In the ^1H NMR spectrum of this thiol-containing polymer doublets appeared at δ 7.25 and 6.82, which were assigned to the protons *ortho* and *meta* to the sulfur substituent of the thiophenyl moiety, respectively. The thiol was glycosylated with trichloroacetimidate **16** (Scheme 2B) and, as in the MPEG-supported synthesis of the (1→6)-linked series, best results were achieved by use of considerable excesses of donor and $BF_3{\cdot}OEt_2$ as promoter. Near quantitative glycosylation of the support was indicated by the presence of a single doublet in the ^1H NMR spectrum of product **17a** for the two protons *meta* to the sulfur substituent ($H\text{-}3_{Ar}$) at δ 6.76, slightly up-field from its

position (δ 6.82) in the disulfide precursor. The integral of this doublet and of the signals for H-2 (δ 5.58) and H-1 (δ 5.34) of the sugar moiety showed ratios of 1.0:0.45:0.43, indicating that approximately 90% α-mannoside had been formed. The first hint that appreciable amounts (ca. 10%) of β-glycosidation had occurred was the presence of a small doublet at δ 5.77 ($J_{2,3}$ 2.4 Hz) where H-2 of the β-mannoside might be expected to resonate. This notion was confirmed by a major ^{13}C resonance characteristic of C-1 of α-mannopyranosides (δ 87.1, $J_{C\text{-}1,H\text{-}1}$ 179 Hz) and a minor one characteristic of a β-anomeric carbon atom (δ 86.2, $J_{C\text{-}1,H\text{-}1}$ 162 Hz).[29]

Since metal ion-containing reagents are not recommended for use in MPEG-supported syntheses,[20] deacetylation of **17a** was carried out with DBU in MeOH.[20,32] Complete deprotection at O-2 was ascertained by the absence of a low-field ^1H NMR signal for H-2 and of the distinctive ^1H- and ^{13}C-signals for acetate in the spectra of product **18a** (Tables 5 and 6).

Iterative glycosylation/deacetylation was carried out under the conditions used in the corresponding solution synthesis, that is, with TMSOTf as promoter in the glycosylations and with DBU as base in the deacetylations, furnishing in sequence the MPEG-supported intermediates **19a**–**23a** and **4a** (Scheme 2B).

The NMR spectra of these compounds were less well resolved than those of their analogues of the (1→6)-linked series, and no attempt was made to assign any specific resonances except those of the anomeric protons and carbon atoms, and those of the C-2 protons of the 2-O-acetylated saccharide units. For the mono-, di- and tri-saccharides these signals were consistent with those observed in the unsupported analogues. The expected α-selectivities of (1→2)-bond formations were confirmed by a value of 180 Hz for both $J_{C\text{-}1',H\text{-}1'}$ and $J_{C\text{-}1,H\text{-}1}$ of disaccharide **20a**.

The glycosylations of the disaccharide acceptor **20a** and of the trisaccharide acceptor **22a** were incomplete even after repeated treatments with donor. Similar poor coupling yields from the trisaccharide stage and beyond have also been observed in a synthesis of α-(1→2)-linked oligomannosides on Merrifield's resin.[19] In the present work, the formation of more by-products with successive reaction steps became obvious from the increasing number and relative sizes of minor peaks in the NMR spectra. In particular, several additional small doublets 0.05–0.15 ppm downfield from the main resonance for the protons *meta* to the sulfur substituent of the thiophenyl moiety were observed, as were more than the required number of peaks in the regions 4.5–5.5 (^1H spectra) and 95–105 ppm (^{13}C spectra), suggesting increasing proportions of β-linked products. For the last two compounds, the tetrasaccharide derivatives **23a** and **4a**, assignments of the anomeric proton signals became impossible and those of the anomeric carbon atoms tentative at best.

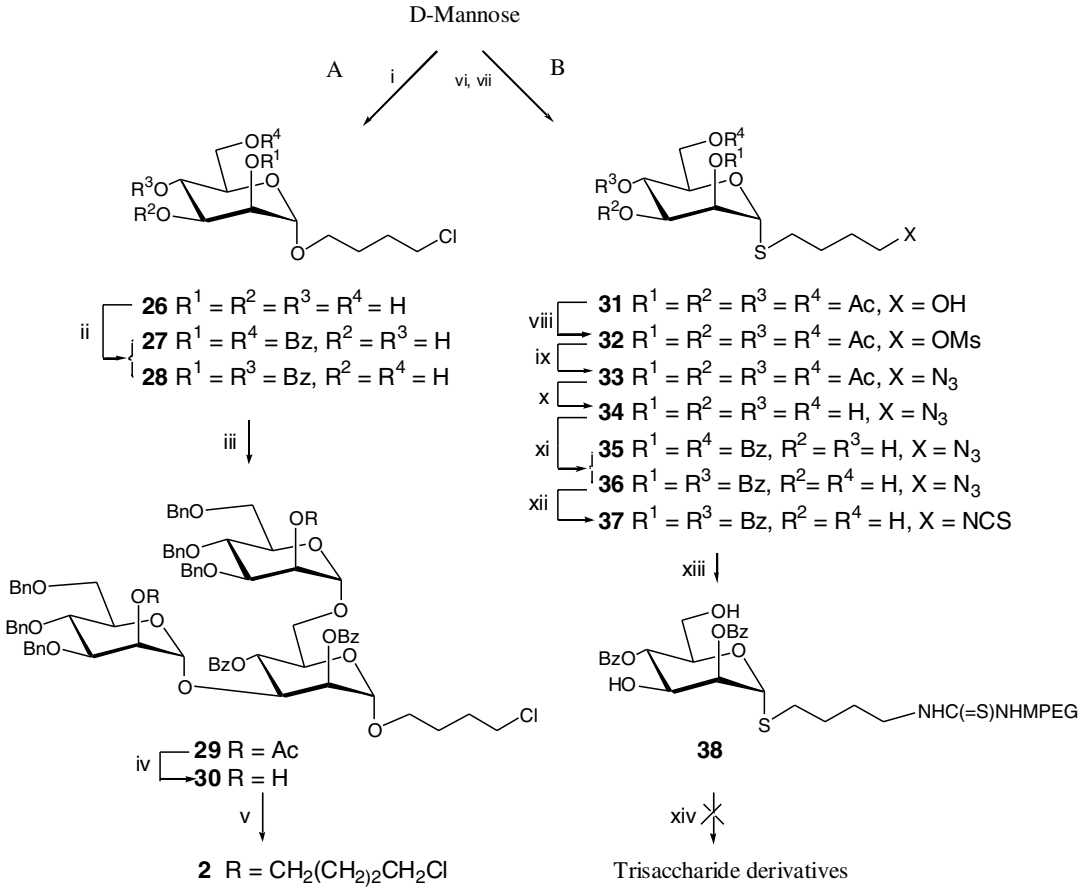
Yields of supported materials were, as in the (1→6)-linked series, ~95% in the initial steps and smaller with growing chain-length. Recovery of product after three glycosylation and four deacetylation steps, that is, of supported tetrasaccharide thioglycoside **4a**, was 46% based on the amount of acceptor monosaccharide polymer **18a** used. On hydrolysis of the thioglycosidic bond, **4a** released 31% of the theoretical yield of tetrasaccharide **24** as a 2:1 α/β mixture of free sugars, corresponding to a 14% overall yield from the supported monosaccharide thioglycoside **18a**. For unambiguous identification the cleaved product was converted in almost two quantitative reactions via the anomerically mixed trichloroacetimidates **25** to allyl glycoside **4** and its β-anomer.

2.2. Branched pentasaccharide 2

Derivatives of pentasaccharide **2** (R = H) and the analogue with β-D-$GlcNH_2$ in place of the terminal (1→2)-linked α-D-Man moieties have been synthesised by solution[11] and by solid phase support[33] methods, respectively, and appear to represent good synthetic targets for the purpose of comparing the solution and MPEG-support procedures here under examination. The former pentasaccharide was selected for the study. It emerged that while the solution approach proved to be straightforward (Scheme 3A) the same did not apply when two approaches involving MPEG-based polymer supports were attempted.

2.2.1. Solution synthesis of the branched pentasaccharide derivative 2 [R = $CH_2(CH_2)_2CH_2Cl$]. Glycosyl acceptor **28**, having free hydroxyl groups at C-3 and C-6, was prepared from D-mannose in three easily conducted steps. Fischer glycosylation with technical grade 4-chloro-*n*-butanol, which contained traces of HCl, gave the crystalline glycoside **26**. This was treated, without rigorous purification and in one pot, first with trimethyl orthobenzoate in the presence of an acid catalyst, then with aqueous HOAc[34] to furnish the required 2,4-dibenzoate **28** presumably via the 2,3:4,6-di-orthoester with moderate selectivity and in 41.6% overall yield from D-mannose. As a hydrolysis by-product 2,6-dibenzoate **27** was also obtained in 24.6% yield (Scheme 3A).

Glycosylation of 2,4-diester **28** with 3 M equiv of trichloroacetimidate **16** (Scheme 2A) under TMSOTf catalysis gave trisaccharide **29** in 85% yield. Selective removal of the *O*-acetates in the presence of benzoates proceeded smoothly by use of methanolic HCl,[35] and the resulting trisaccharide diol **30**, on glycosylation with 4 M equiv of trichloroacetimidate **8** (Scheme 1), furnished the target pentasaccharide **2** [R = $CH_2(CH_2)_2$-CH_2Cl] in 40% overall yield from the acceptor diol **28** (Scheme 3A).



Scheme 3. Reagents: (i) $HOCH_2(CH_2)_2CH_2Cl$, HCl; (ii) a, $Ph(OMe)_3$, CH_2Cl_2, camphor SO_3H, b, HOAc, H_2O; (iii) **16**, CH_2Cl_2, TMSOTf; (iv) MeOH, HCl; (v) HBr, HOAc, Ac_2O; (vi) a, $NH_2C(S)NH_2$, acetone, b, $Na_2S_2O_3$, CH_2Cl_2, H_2O, c, $ClCH_2(CH_2)_2CH_2OH$, KOH; (vii) **8** CH_2Cl_2, TMSOTf; (viii) MsCl, py; (ix) NaN_3, DMF; (x) NaOMe, MeOH; (xi) a, $Ph(OMe)_3$, CH_2Cl_2, CSA, b, AcOH, H_2O; (xii) $P(OEt)_3$, CS_2, PhMe; (xiii) MPEG-NH_2, CH_2Cl_2; (xiv) **16**, CH_2Cl_2, TMSOTf.

2.2.2. Attempted MPEG-supported synthesis of the branched pentasaccharide derivative 2 (R = H). Primary amines add to isothiocyanates, and this reaction can be used to link appropriate carbohydrate derivatives containing isothiocyanate groups to non-carbohydrate amines via thiourea bridges.[36] In the present work, MPEG-NH_2 and compound **37** were used as the amine and isothiocyanate, respectively. Compound **37** was obtained by standard procedures from thioglycoside **31** by way of intermediates **32**–**36** as indicated in Scheme 3B. Addition of MPEG-NH_2 to isothiocyanate **37** occurred in a straightforward manner to give the thiourea-linked **38.** Glycosylation of diol **38** proved to be difficult, and a satisfactory conversion to a trisaccharide derivative was not achieved even after three treatments with considerable excess of donor **16**. ^{13}C NMR signals of equal height at δ 82.6 and 82.9 indicated the presence in the products of two thioglycosidic compounds, but what they were was not established. Attempts to remove selectively the acetyl groups from O-2 of the newly introduced sugars from this mixture were unsuccessful; exposure to methanolic HCl or to HBF_4 resulted in degradation, and this finding acted as a further disincentive to the pursuit of this approach.

Attempts to make diol **38** by desilylation of its di-TBDMS derivative were unsuccessful, and consequently the possible route to **2** by way of the diether of the *O*–$C_6H_4(p)$-SS–$C_6H_4(p)$–OMPEG analogue of thioglycoside **38** was not pursued.

3. Summary and conclusions

The three target oligosaccharide glycoside derivatives **2** [R = $CH_2(CH_2)_2CH_2Cl$], **3** and **4** have been synthesised by conventional solution methods in acceptable yields of 40%, 39% and 42%, respectively, notwithstanding the restrictive requirement to choose protecting groups and general reaction conditions, which were compatible with parallel syntheses of the three compounds on MPEG. Glycosylations with a slight excess of donor and ca. 0.1 mol equiv of promoter were fast, and the chromatographic purifications, although necessary at every step, were uncomplicated.

On MPEG, a several fold excess of donor and ca. 0.3 mol equiv of promoter were used, but as a rule repeated glycosylation was not necessary. As a precautionary measure all reactions were allowed to proceed for slightly longer than in the corresponding solution syntheses. The crude products obtained after cleavage from the support were chromatographed at least twice to give target compounds uncontaminated by β-linked isomers or shorter homologues.

Under these conditions the MPEG-supported synthesis of the α-(1→6)-linked tetrasaccharide derivative **3** was satisfactory, the yield being nearly the same (34%) as in the corresponding conventional synthesis. This could be accounted for by the obvious loss of material by incomplete precipitation of the polymer-bound tetra-, and to some extent even the tri-saccharide derivatives, and gradual deterioration of the polyether backbone (or gradual loss of carbohydrate substrate from the support). The MPEG-based method required less bench time and, had the recrystallisations of all the intermediates been omitted (they were carried out in the present work mainly to improve the quality of the NMR spectra) the procedures would be very time-efficient.

In the α-(1→2)-linked series a 14% yield of isolated, cleaved tetrasaccharide, obtained as the free sugar corresponding to glycoside **4**, was very disappointing. Yields of the supported material at the tri- and tetra-saccharide stages (52% for **22a** and 46% for **4a**) were slightly lower than those achieved in a very similar synthesis by Douglas et al.,[20] who did not report yields of cleaved products.

A new approach to 3,6-branched pentasaccharides involving two double glycosylations was successfully applied to the preparation of compound **2** [R = $CH_2(CH_2)_2CH_2Cl$] in solution, but it failed when attempted using the MPEG approach. The successful synthesis of a 3,6-branched mannotrioside on Merrifield's resin with an alkylthio linker and further extension of the branches with a chemical GlcNAc donor to give a pentasaccharide, all glycosylation repeated once, has been reported by Schmidt's group.[33] They isolated the protected tri- and penta-saccharide after detachment from the support in 38% and 20% yield, respectively. Our failure to achieve a closely related synthesis on MPEG reflects the incompatibility of this support with a number of common reagents and reaction conditions, which in turn severely restricts the choice of suitable protecting groups.

4. Experimental

4.1. Materials and general methods

MPEG [$HOCH_2CH_2(OCH_2CH_2)_nOMe$, *n* 80–160, average MW 5000] was obtained from Fluka Chemie AG, Buchs, Switzerland, and MPEG-amine [$H_2NCH_2CH_2$-$(OCH_2CH_2)_nOMe$, *n* 80–160, average MW ~5200] from Shearwater Corporation, Huntsville, Al, USA. All stoichiometric calculations are based on MW 5000 for both polymers.

For glycosylations, acceptors and donors were dried separately at 1 mmHg and 20 °C for 2 h shortly before use. All glycosylation reactions were carried out in the presence of powdered 4 Å molecular sieves in dry CH_2Cl_2 under argon.

Column chromatography was performed on Silica Gel 230–400 mesh (Scharlau) and TLC on Silica Gel HF-254 with detection by UV or by charring with ethanolic H_2SO_4 (5% v/v) containing anisaldehyde (2%). Optical rotations were measured in CH_2Cl_2 solutions at 20 °C with a Perkin–Elmer 241 automatic polarimeter. Melting points were determined with a Reichert micro hot-stage apparatus and are uncorrected.

Unless otherwise stated, ^{1}H NMR, ^{13}C NMR, ^{1}H–^{1}H COSY and ^{1}H–^{13}C COSY spectra were recorded with a Bruker Avance NMR spectrometer equipped with a 5mm O.D. Quad nuclear probe at 300 MHz (for ^{1}H) or 75.5 MHz (for ^{13}C) on solutions in $CDCl_3$ or D_2O. For $CDCl_3$ solutions chemical shifts are reported in parts per million from Me_4Si (δ_H, δ_C) as internal reference; D_2O solutions contained acetone as a secondary standard (δ_H 2.217, δ_C 33.17). ^{1}H NMR spectra at 500 MHz were recorded with a Varian UNITY 500 instrument. Aromatic resonances and resonances for acetyl, allyl, benzyl, benzoyl, TBDMS and carbonyl groups were observed as required but are usually not listed. For polymer-supported compounds, selected assignments were made by comparison with the spectra of the corresponding solution-synthesised compounds. The polymer methylene protons were observed as required, but are not recorded.

MALDI TOF mass spectra (MALDI HRMS) were acquired using an Applied Biosystems Voyager-DE PRO mass spectrometer (Foster City, CA) in positive ion reflector mode with an acceleration voltage of 20,000 V, a grid voltage of 75%, 0.002% on the guide wire and delay time 180 ns. 2,5-Dihydroxybenzoic acid in H_2O was used as the matrix. All other high resolution mass spectra (HRMS) were measured with a VG70-250S double focusing magnetic sector mass spectrometer or with a Mariner 8105 electrospray TOF mass spectrometer with ionisation effected by use of a caesium ion gun.

4.2. Benzyl α-D-mannopyranoside (5)

Prepared by the method of Winnik et al.,[27] compound **5** had mp 131–131.5 °C, $[\alpha]_D$ +73.5 (*c* 1.3, water); lit.[27] mp 131–2 °C, $[\alpha]_D$ +74 (*c* 1.3, water). ^{1}H and ^{13}C NMR data are given in Table 1.

4.3. Benzyl 2,3,4-tri-*O*-benzoyl-6-*O*-*tert*-butyldimethylsilyl-α-D-mannopyranoside (6)

Benzyl α-D-mannopyranoside (**5**) (50 g, 0.185 mol) and *tert*-butyldimethylsilyl chloride (31 g, 0.205 mol) were stirred in pyridine (500 mL) at 20 °C for 48 h. Benzoyl chloride (96 mL, 120 g, 0.85 mol) was then added with cooling. After being stirred for another 24 h the reaction mixture was poured onto ice and the solid formed after several hours was collected by filtration (57 g). The filtrate was extracted with CH_2Cl_2 (3×), the extracts were washed with brine (2×), dried over $MgSO_4$ and evaporated to a thick brown syrup, which crystallised on trituration with EtOH (12.5 g). The mother liquors were concentrated and purified by flash chromatography (8:1 light petroleum–EtOAc) to furnish a further quantity (38.2 g) of crystalline product. The combined crystals were recrystallised from EtOH to give the fully protected derivative **6** (107.7 g, 0.154 mol, 83.2%): mp 125–6 °C, $[\alpha]_D$ −76.6 (*c* 1.85, CH_2Cl_2). ^1H and ^{13}C NMR data are given in Table 1. Anal. Calcd for $C_{40}H_{44}O_9Si$: C, 68.94; H, 6.37. Found: C, 68.83; H, 6.38.

4.4. 2,3,4-Tri-*O*-benzoyl-6-*O*-*tert*-butyldimethylsilyl-α-D-mannopyranose (7)

Benzyl glycoside **6** (18.5 g, 26.5 mmol) and ammonium formate (20 g, 317 mmol) were stirred with palladium on charcoal (10%, 10 g) in refluxing MeOH (500 mL) for 1 h. The catalyst and solvent were removed without delay by filtration through Celite and evaporation, respectively. The resulting syrup was partitioned between CH_2Cl_2 and brine. The organic phase was dried ($MgSO_4$) and evaporated. The semi-solid residue crystallised on trituration with light petroleum to give the title compound (15.2 g, 25.0 mmol, 94%). Recrystallised from light petroleum the free sugar **7** had mp 145–6 °C, $[\alpha]_D$ −143.8 (*c* 1.16, CH_2Cl_2). ^1H and ^{13}C NMR data are given in Table 1. Anal. Calcd for $C_{33}H_{38}O_9Si$: C, 65.33; H, 6.31. Found: C, 65.45; H, 6.46.

4.5. 2,3,4-Tri-*O*-benzoyl-6-*O*-*tert*-butyldimethylsilyl-α-D-mannopyranosyl trichloroacetimidate (8)

To a stirred solution of free sugar **7** (10 g, 16.5 mmol) in dry CH_2Cl_2 (150 mL) at 0 °C dry K_2CO_3 (22 g, 160 mmol) was added, followed by Cl_3CCN (8.3 mL, 82.5 mmol). After being stirred for 16 h at 20 °C, the mixture was filtered through Celite, the filtrate was reduced to a small volume and flash chromatographed (10:1 light petroleum–EtOAc) to give **8** as a colourless foam (11.8 g, 15.6 mmol, 94%): $[\alpha]_D$ −79.0 (*c* 0.92, CH_2Cl_2). ^1H and ^{13}C NMR data are given in Table 1. Anal. Calcd for $C_{35}H_{38}C_{13}NO_9Si$: C, 55.97; H, 5.10; Cl, 14.16; N, 1.86. Found: C, 56.25; H, 5.27; Cl, 14.18; N, 1.84.

4.6. Phenyl 2,3,4-tri-*O*-benzoyl-6-*O*-*tert*-butyldimethylsilyl-α-D-mannopyranoside (9)

A solution of trichloroacetimidate **8** (1.5 g, 1.95 mmol) and phenol (300 mg, 3.2 mmol) in dry CH_2Cl_2 (40 mL) was stirred with powdered 4 Å molecular sieves (250 mg) for 5 min at 20 °C under argon. After cooling to −40 °C, TMSOTf (20 μL, 0.1 mmol) was added, the cooling bath was removed and stirring was continued for 15 min. The reaction was quenched by the addition of Et_3N (30 μL, 0.21 mmol). Filtration through glass fibre, removal of the solvent by rotary evaporation and purification of the syrupy residue thus obtained by flash chromatography (10:1→5:1 light petroleum–EtOAc) gave glycoside **9** (1.21 g, 1.77 mmol, 91%) as a white foam: $[\alpha]_D$ −68.2 (*c* 0.95, CH_2Cl_2). ^1H and ^{13}C NMR data are given in Table 1. Anal. Calcd for $C_{39}H_{42}O_9Si$: C, 68.60; H, 6.20. Found: C, 68.31; H, 6.02.

4.7. Phenyl 2,3,4-tri-*O*-benzoyl-α-D-mannopyranoside (10)

A solution of silyl ether **9** (0.77 g, 1.12 mmol) in aqueous HOAc (85%, 20 mL) was heated at 85 °C for 90 min. The solvents were removed by evaporation and the oily residue was purified by flash chromatography (4:1 light petroleum–EtOAc) to furnish phenyl glycoside **10** (0.61 g, 1.07 mmol, 96%) as a white foam: $[\alpha]_D$ −45.1 (*c* 1.03, CH_2Cl_2). ^1H and ^{13}C NMR data are given in Table 1. HRMS, *m*/*z*: Calcd for $C_{33}H_{29}O_9$ $[M+H]^+$ 569.1816. Found 569.1825.

4.8. Phenyl (2,3,4-tri-*O*-benzoyl-6-*O*-*tert*-butyldimethylsilyl-α-D-mannopyranosyl)-(1→6)-2,3,4-tri-*O*-benzoyl-α-D-mannopyranoside (11)

Phenyl 2,3,4-tri-*O*-benzoyl-α-D-mannopyranoside (**10**) (1.2 g, 2.1 mmol) was glycosylated with trichloroacetimidate **8** (2.6 g, 3.4 mmol) in dry CH_2Cl_2 (40 mL) using TMSOTf (50 μL, 0.27 mmol) as promoter, as described above for the preparation of compound **9**. Purification of the foam thus obtained by flash chromatography (4:1 light petroleum–EtOAc) gave disaccharide **11** (2.21 g, 1.90 mmol, 90%) as a white foam: $[\alpha]_D$ −73.3 (*c* 2.0, CH_2Cl_2). ^1H and ^{13}C NMR data are given in Tables 2 and 3, respectively. HRMS, *m*/*z*: Calcd for $C_{66}H_{64}O_{17}Si$ $[M]^+$ 1156.3909. Found 1156.3913. Calcd for $C_{66}H_{65}O_{17}Si$ $[M+H]^+$ 1157.3991. Found 1157.3885.

4.9. Phenyl (2,3,4-tri-*O*-benzoyl-α-D-mannopyranosyl)-(1→6)-2,3,4-tri-*O*-benzoyl-α-D-mannopyranoside (12)

A solution of silyl ether **11** (0.80 g, 0.69 mmol) in aqueous HOAc (85%, 20 mL) was heated at 85 °C for 90 min. The solvents were removed by evaporation and the oily residue was purified by flash chromatography (4:1 light

petroleum–EtOAc) to furnish the disaccharide acceptor **12** (0.685 g, 0.655 mmol, 95%) as a white foam: $[\alpha]_D$ −79.0 (*c* 1.32, CH_2Cl_2). ^1H and ^{13}C NMR data are given in Tables 2 and 3, respectively. HRMS, *m/z*: Calcd for $C_{60}H_{50}O_{17}$ $[M]^+$ 1042.3048. Found 1042.3014. Calcd for $C_{60}H_{51}O_{17}$, $[M+H]^+$ 1043.3126. Found 1043.3096.

4.10. Phenyl (2,3,4-tri-*O*-benzoyl-6-*O*-*tert*-butyldimethylsilyl-α-D-mannopyranosyl)-(1→6)-(2,3,4-tri-*O*-benzoyl-α-D-mannopyranosyl)-(1→6)-2,3,4-tri-*O*-benzoyl-α-D-mannopyranoside (13)

The disaccharide acceptor **12** (0.40 g, 0.38 mmol) was glycosylated with trichloroacetimidate **8** (0.55 g, 0.72 mmol) using TMSOTf (12 μL, 0.07 mmol) as promoter, as described above for the preparation of compound **9**. Purification of the crude product by flash chromatography (3:1 light petroleum–EtOAc) gave trisaccharide **13** (0.57 g, 0.35 mmol, 92%) as a white foam: $[\alpha]_D$ −60.0 (*c* 1.27, CH_2Cl_2). ^1H and ^{13}C NMR data are given in Tables 2 and 3, respectively. HRMS, *m/z*: Calcd for $C_{93}H_{86}O_{25}Si$ $[M]^+$ 1630.5227. Found 1630.5261. Calcd for $C_{93}H_{87}O_{25}Si$ $[M+H]^+$ 1631.5306. Found 1631.5394.

4.11. Phenyl (2,3,4-tri-*O*-benzoyl-α-D-mannopyranosyl)-(1→6)-(2,3,4-tri-*O*-benzoyl-α-D-mannopyranosyl)-(1→6)-2,3,4-tri-*O*-benzoyl-α-D-mannopyranoside (14)

A solution of silyl ether **13** (0.50 g, 0.306 mmol) in aqueous HOAc (85%, 20 mL) was heated at 85 °C for 150 min. The solvents were removed by evaporation, and the oily residue was purified by flash chromatography (2:1 light petroleum–EtOAc) to furnish the trisaccharide acceptor **14** (0.38 g, 0.25 mmol, 82%) as a white foam: $[\alpha]_D$ −54.8 (*c* 2.0, CH_2Cl_2). ^1H and ^{13}C NMR data are given in Tables 2 and 3, respectively. HRMS, *m/z*: Calcd for $C_{87}H_{72}O_{25}$ $[M]^+$ 1516.4363. Found 1516.4378. Calcd for $C_{87}H_{73}O_{25}$ $[M+H]^+$ 1517.4441. Found 1517.4427.

4.12. Phenyl (2,3,4-tri-*O*-benzoyl-6-*O*-*tert*-butyldimethylsilyl-α-D-mannopyranosyl)-(1→6)-(2,3,4-tri-*O*-benzoyl-α-D-mannopyranosyl)-(1→6)-(2,3,4-tri-*O*-benzoyl-α-D-mannopyranosyl)-(1→6)-2,3,4-tri-*O*-benzoyl-α-D-mannopyranoside (15)

4.12.1. By solution synthesis. Trisaccharide **14** (330 mg, 0.217 mmol) was glycosylated with trichloroacetimidate **8** (330 mg, 0.43 mmol) using TMSOTf (8 μL, 0.044 mmol) as promoter, as described above for the preparation of compound **9**. Purification of the crude product by flash chromatography (2:1 light petroleum–EtOAc) gave the tetrasaccharide derivative **15** (351 mg, 0.167 mmol, 77%) as a white foam: $[\alpha]_D$ −46.6 (*c* 0.91, CH_2Cl_2). ^1H and ^{13}C NMR data are given in Tables 2 and 3, respectively. HRMS, *m/z*: Calcd for $C_{120}H_{108}O_{33}Si$ $[M]^+$ 2104.6542. Found 2104.6685. Calcd for $C_{120}H_{109}O_{33}Si$ $[M+H]^+$ 2105.6620. Found 2105.6742.

4.12.2. By MPEG-supported synthesis. To a stirred mixture of **15a** (200 mg, 0.0275 mmol, see later) and $NiCl_2{\cdot}6H_2O$ (285 mg) in EtOH (95%, 8 mL) a hot solution of boric acid (140 mg) in aqueous EtOH (80%, 4.5 mL) was added. Stirring was continued at 40 °C while a solution of $NaBH_4$ (40 mg, ~1 mmol) in aqueous EtOH (90%, 3.3 mL) was added over a period of 20 min.[30] After being stirred for another 30 min, the reaction mixture was filtered through glass fibre to remove the suspended black material that had formed, the filtrate was evaporated and the residue was partitioned between CH_2Cl_2 and water. The dried organic phase was reduced to a small volume, ether (50 mL) was added, the precipitate was removed by filtration and the oil obtained on evaporation of the filtrate was purified by flash chromatography (2:1 light petroleum–EtOAc) to furnish **15** (38.7 mg, 0.018 mmol, 67%). ^1H and ^{13}C NMR data were identical to those of the solution synthesised material.

4.13. Phenyl (2,3,4-tri-*O*-benzoyl-α-D-mannopyranosyl)-(1→6)-(2,3,4-tri-*O*-benzoyl-α-D-mannopyranosyl)-(1→6)-(2,3,4-tri-*O*-benzoyl-α-D-mannopyranosyl)-(1→6)-2,3,4-tri-*O*-benzoyl-α-D-mannopyranoside (3)

4.13.1. By solution synthesis. A solution of silyl ether **15** (100 mg, 0.047 mmol) in aqueous HOAc (85%, 5 mL) was heated at 85 °C for 2 h. The solvents were removed by evaporation and the oily residue was purified by flash chromatography (2:1 light petroleum–EtOAc) to furnish target **3** (76 mg, 0.039 mmol, 82%) as a white foam: $[\alpha]_D$ −53.1 (*c* 0.93, CH_2Cl_2). ^1H and ^{13}C NMR data are given in Tables 2 and 3. HRMS, *m/z*: Calcd for $C_{114}H_{94}O_{33}$ $[M]^+$ 1990.5574. Found 1990.5677. Calcd for $C_{114}H_{95}O_{33}$ $[M+H]^+$ 1991.5754. Found 1991.5707. Calcd for $C_{114}H_{94}O_{33}Na$ $[M+Na]^+$ 2013.5580. Found 2013.5575.

4.13.2. By MPEG-supported synthesis. Compound **3a** (200 mg, 0.0280 mmol, see later) was desulfurised with $NiCl_2{\cdot}6H_2O$ (285 mg), boric acid (140 mg) and $NaBH_4$ (40 mg, 1 mmol) in aqueous EtOH as described above for compound **15a** to furnish, after extractive work-up and chromatography, **3** (36.1 mg, 0.018 mmol, 64.7%). ^1H and ^{13}C NMR data were identical to those of the solution synthesised material.

4.14. 4-(2,3,4-Tri-*O*-benzoyl-6-*O*-*tert*-butyldimethylsilyl-α-D-mannopyranosyloxyphenyl)-4′-(MPEG-oxyphenyl)-disulfide (9a)

A solution of 4-hydroxyphenyl-4′-MPEG-oxyphenyl disulfide[28] (5.23 g, 1.0 mmol) and trichloroacetimidate

8 (2.3 g, 3.0 mmol) in CH_2Cl_2 (15 mL) were stirred with powdered 4 Å molecular sieves (200 mg) at 0 °C under argon. $BF_3{\cdot}OEt_2$ (378 μL, 3 mmol) was added and stirring was continued at 20 °C. After 1.5 h the reaction was quenched by the addition of Et_3N (0.8 mL, 5.7 mmol) at 0 °C, followed after 5 min by ice-cold dry diethyl ether (150 mL). The precipitate was removed by filtration, washed with ether (50 mL), isopropanol (3 × 50 mL) to remove triethylammonium salts[28] and ether again (50 mL), to give, after recrystallisation from absolute EtOH and thorough drying in vacuo, the title compound **9a** (5.54 g, 0.95 mmol, 95%). ^1H and ^{13}C NMR data are given in Table 1.

4.15. 4-(2,3,4-Tri-*O*-benzoyl-α-D-mannopyranosyloxyphenyl)-4′-(MPEG-oxyphenyl)-disulfide (10a)

A solution of **9a** (5.2 g, 0.89 mmol) in aqueous HOAc (85%, 30 mL) was heated at 85 °C for 2 h. After cooling the reaction mixture to 5 °C, ice-cold dry diethyl ether (250 mL) was added with vigorous shaking. The solid precipitate was removed by filtration, washed with diethyl ether (3 × 25 mL) and recrystallised from absolute EtOH to give, after thorough drying in vacuo, product **10a** (4.95 g, 0.86 mmol, 97%). ^1H and ^{13}C NMR data are given in Table 1.

4.16. 4-[(2,3,4-Tri-*O*-benzoyl-6-*O*-*tert*-butyldimethylsilyl-α-D-mannopyranosyl)-(1→6)-(2,3,4-tri-*O*-benzoyl-α-D-mannopyranosyloxyphenyl)]-4′-(MPEG-oxyphenyl)-disulfide (11a)

A solution of the MPEG supported acceptor **10a** (4.0 g, 0.70 mmol) and trichloroacetimidate **8** (2.3 g, 3.0 mmol) in CH_2Cl_2 (15 mL) was stirred with powdered 4 Å molecular sieves (200 mg) at 0 °C under argon. TMSOTf (40 μL, 0.2 mmol) was added and stirring was continued at 20 °C. After 1.5 h the reaction was quenched and worked-up as described above for the preparation of compound **9a**, to give the title compound **11a** (4.15 g, 0.66 mmol, 95%). ^1H and ^{13}C NMR data are given in Tables 2 and 3, respectively.

4.17. 4-[(2,3,4-Tri-*O*-benzoyl-α-D-mannopyranosyl)-(1→6)-(2,3,4-tri-*O*-benzoyl-α-D-mannopyranosyloxyphenyl)]-4′-(MPEG-oxyphenyl)-disulfide (12a)

Compound **11a** (2.5 g, 0.40 mmol) was desilylated by heating in aqueous HOAc (85%, 12 mL) at 85 °C for 4 h as described above for the preparation of compound **10a**, to give product **12a** (2.36 g, 0.38 mmol, 95%). ^1H and ^{13}C NMR data are given in Tables 2 and 3, respectively.

4.18. 4-[(2,3,4-Tri-*O*-benzoyl-6-*O*-*tert*-butyldimethylsilyl-α-D-mannopyranosyl)-(1→6)-(2,3,4-tri-*O*-benzoyl-α-D-mannopyranosyl)-(1→6)-(2,3,4-tri-*O*-benzoyl-α-D-mannopyranosyloxyphenyl)]-4′-(MPEG-oxyphenyl)-disulfide (13a)

The MPEG supported disaccharide acceptor **12a** (1.75 g, 0.28 mmol) was glycosylated with trichloroacetimidate **8** (0.64 g, 0.85 mmol) as donor and TMSOTf (15 μL, 0.08 mmol) as promoter as described above for the preparation of compound **11a**, to give the title compound **13a** (1.73 g, 0.25 mmol, 90%). ^1H and ^{13}C NMR data are given in Tables 2 and 3, respectively.

4.19. 4-[(2,3,4-Tri-*O*-benzoyl-α-D-mannopyranosyl)-(1→6)-(2,3,4-tri-*O*-benzoyl-α-D-mannopyranosyl)-(1→6)-(2,3,4-tri-*O*-benzoyl-α-D-mannopyranosyloxyphenyl)]-4′-(MPEG- oxyphenyl)-disulfide (14a)

Compound **13a** (2.5 g, 0.40 mmol) was desilylated by heating in aqueous HOAc (85%, 12 mL) at 85 °C for 4 h as described above for the preparation of compound **10a**, to give the title compound **14a** (1.32 g, 0.20 mmol, 89%). ^1H and ^{13}C NMR data are given in Tables 2 and 3, respectively.

4.20. 4-[(2,3,4-Tri-*O*-benzoyl-6-*O*-*tert*-butyldimethylsilyl-α-D-mannopyranosyl)-(1→6)-(2,3,4-tri-*O*-benzoyl-α-D-mannopyranosyl)-(1→6)-(2,3,4-tri-*O*-benzoyl-α-D-mannopyranosyl)-(1→6)-(2,3,4-tri-*O*-benzoyl-α-D-mannopyranosyloxyphenyl)]-4′-(MPEG-oxyphenyl)-disulfide (15a)

The MPEG supported trisaccharide acceptor **14a** (0.75 g, 0.11 mmol) was glycosylated with trichloroacetimidate **8** (0.25 g, 0.33 mmol) as donor and TMSOTf (6 μL, 0.03 mmol) as promoter as described above for the preparation of compound **11a**, to give the tetrasaccharide derivative **15a** (0.69 g, 0.095 mmol, 86%). ^1H and ^{13}C NMR data are given in Tables 2 and 3, respectively.

4.21. 4-[(2,3,4-Tri-*O*-benzoyl-α-D-mannopyranosyl)-(1→6)-(2,3,4-tri-*O*-benzoyl-α-D-mannopyranosyl)-(1→6)-(2,3,4-tri-*O*-benzoyl-α-D-mannopyranosyl)-(1→6)-(2,3,4-tri-*O*-benzoyl-α-D-mannopyranosyl-oxyphenyl)]-4′-(MPEG-oxyphenyl)-disulfide (3a)

Compound **15a** (550 mg, 0.075 mmol) was desilylated by heating in aqueous HOAc (85%, 10 mL) at 85 °C for 4 h as described above for the preparation of compound **10a**, to give the title compound **3a** (465 mg, 0.065 mmol, 87%). ^1H and ^{13}C NMR data are given in Tables 2 and 3, respectively.

4.22. Allyl 2-*O*-acetyl-3,4,6-tri-*O*-benzyl-α-D-mannopyranoside (17)

Allyl alcohol (0.5 mL, 425 mg, 7.3 mmol) was glycosylated with trichloroacetimidate **16**[25,26] (1.4 g, 2.2 mmol) in dry CH_2Cl_2 (15 mL) using TMSOTf (50 μL, 0.27 mmol) as promoter, as described above for the preparation of compound **9**. After removal of the β-anomer by flash chromatography (2:1 light petroleum–EtOAc) **17** (0.86 g, 1.61 mmol, 73%) was obtained as a thick syrup: $[\alpha]_D$ +32.3 (*c* 1.58, CH_2Cl_2); lit.[5] $[\alpha]_D$ +30.5 (*c* 0.5, $CHCl_3$); lit.[31] $[\alpha]_D$ +34.5 (*c* 3.37, $CHCl_3$). The ^1H NMR spectrum was consistent with published data;[31] NMR data are given in Table 4.

4.23. Allyl 3,4,6-tri-*O*-benzyl-α-D-mannopyranoside (18)

2-*O*-Acetate **17** (500 mg, 0.94 mmol) was deacetylated by treatment with DBU (80 mg, 0.52 mmol) in dry MeOH[20,32] (8 mL) for 16 h at 20 °C. The reaction solution was acidified by addition of a few drops of HOAc before removal of the solvent and purification of the oily residue by flash chromatography (3:1 light petroleum–EtOAc) to give **18** (451 mg, 0.92 mmol, 98%) as a foam: $[\alpha]_D$ +59.5 (*c* 1.09, CH_2Cl_2); lit.[5] $[\alpha]_D$ +56 (*c* 1.5, $CHCl_3$); lit.[31] $[\alpha]_D$ +60.2 (*c* 1.23, $CHCl_3$). The ^1H NMR spectrum was consistent with published data;[31] NMR data are given in Table 4.

4.24. Allyl (2-*O*-acetyl-3,4,6-tri-*O*-benzyl-α-D-mannopyranosyl)-(1→2)-3,4,6-tri-*O*-benzyl-α-D-mannopyranoside (19)

Compound **18** (0.83 g, 1.69 mmol) was glycosylated with trichloroacetimidate **16** (1.6 g, 2.5 mmol) in dry CH_2Cl_2 (15 mL) using TMSOTf (10 μL, 0.05 mmol) as promoter, as described above for the preparation of compound **9**. After purification by flash chromatography (6:1→4:1 light petroleum–EtOAc) the 2′-O-acetylated disaccharide **19** (1.51 g, 1.56 mmol, 92%) was obtained as a foam: $[\alpha]_D$ +25.0 (*c* 1.01, CH_2Cl_2); lit.[5] $[\alpha]_D$ +25.5 (*c* 2, $CHCl_3$). ^1H and ^{13}C NMR data are given in Tables 5 and 6, respectively. HRMS, *m*/*z*: Calcd for $C_{59}H_{68}NO_{12}$ $[M+NH_4]^+$ 982.4736. Found 982.4768.

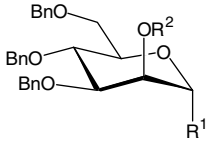
4.25. Allyl (3,4,6-tri-*O*-benzyl-α-D-mannopyranosyl)-(1→2)-3,4,6-tri-*O*-benzyl-α-D-mannopyranoside (20)

2′-*O*-Acetate **19** (1.35 g, 1.40 mmol) was deacetylated by treatment with DBU (150 mg, 1.0 mmol) in dry MeOH (20 mL) as described above for the preparation of compound **18**. Purification by flash chromatography (3:1 light petroleum–EtOAc) gave **20** (1.24 g, 1.34 mmol, 96%) as a foam: $[\alpha]_D$ +32.2 (*c* 2.61, CH_2Cl_2); lit.[5] $[\alpha]_D$ +35 (*c* 0.54, $CHCl_3$). ^1H and ^{13}C NMR data are given in Tables 5 and 6, respectively. HRMS, *m*/*z*: Calcd for $C_{57}H_{66}NO_{11}$ $[M+NH_4]^+$ 940.4530. Found 940.4585.

4.26. Allyl (3,4,6-tri-*O*-benzyl-α-D-mannopyranosyl)-(1→2)-(3,4,6-tri-*O*-benzyl-α-D-mannopyranosyl)-(1→2)-3,4,6-tri-*O*-benzyl-α-D-mannopyranoside (22)

Compound **20** (0.90 g, 0.98 mmol) was glycosylated with trichloroacetimidate **16** (0.96 g, 1.5 mmol) in dry CH_2Cl_2 (15 mL) using TMSOTf (8 μL, 0.04 mmol) as promoter, as described above for the preparation of compound **9**. Purification by flash chromatography (5:1 light petroleum–EtOAc) gave the 2″-O-acetylated trisaccharide **21** (1.19 g, 0.85 mmol, 87%), which was not fully characterised (^1H and ^{13}C NMR data are given in Tables 5 and 6, respectively). A portion of this material (0.18 g, 0.128 mmol) was deacetylated by treatment

Table 4. ^1H and ^{13}C NMR chemical shifts (δ) of the monosaccharide precursors of (1→2)-linked oligosaccharide derivatives[a,b]



Compound	R^1	R^2	H-1	H-2	H-3	H-4	H-5	H-6a	H-6b	C-1	C-2	C-3	C-4	C-5	C-6
17[c]	OAll	Ac	4.89	5.39	4.01	3.89	3.82	3.80	3.70	96.9	68.1	78.2	74.4	71.5	68.9
17a	SC_6H_4OMPEG	Ac	5.34	5.58	—	—	—	—	—	87.1 ($J_{C\text{-}1,H\text{-}1}$ 179 Hz)	—	78.5	74.8	72.5	67.6[d]
18[c]	OAll	H	4.94	4.05	3.90	3.85	3.80	3.76	3.70	98.8	68.7	80.6	74.7	71.5	69.3
18a	SC_6H_4OMPEG	H	5.42	—	—	—	—	—	—	88.4	—	79.9	74.4	72.1	67.1[d]

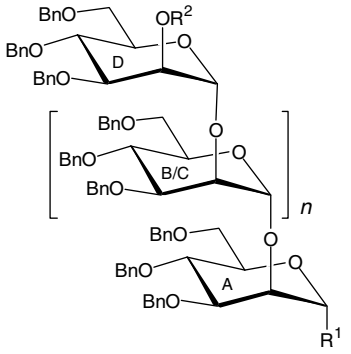
[a] Resonances for acetyl, allyl and benzyl groups were present as required but are not reported. Coupling constants were recorded as follows: $J_{1,2}$ 1.8, $J_{2,3}$ 3.2 ± 0.2, $J_{3,4} = J_{4,5}$ 10.0 ± 0.2, $J_{5,6a}$ 3.8 ± 0.6, $J_{5,6b}$ 2.1 ± 0.1, $J_{6a,6b}$ 12.5 ± 0.5.

[b] For MPEG-supported compounds, selected assignments were made by comparison with the spectra of the corresponding solution-synthesised compounds. In their ^1H NMR spectra doublets for H-3_{Ar} at δ 6.76 and singlets for OMe at δ 3.38 and in their ^{13}C NMR spectra signals for OMe at δ 58.9 were also observed but are not reported.

[c] For ^1H NMR see Ref. 24.

[d] Tentative assignment.

Table 5. ^{1}H NMR chemical shifts (δ) for (1→2)-linked oligosaccharide derivatives[a,b]



Compound	*n*	Sugar ring	R^1	R^2	H-1	H-2	H-3	H-4	H-5	H-6a	H-6b
19	0	A	OAll	—	4.92	4.03	3.94				
		D	—	Ac	5.09	5.54	3.98		3.65–3.90		
19a	0	A	SC_6H_4OMPEG	—	5.44	—	—	—	—	—	—
		D	—	Ac	5.04	5.49	—	—	—	—	—
20[d]	0	A	OAll	—	4.95	4.05	3.95		3.65–3.85		
		D	—	H	5.14	4.12	3.87		3.65–3.85		
20a	0	A	SC_6H_4OMPEG	—	5.49	—	—	—	—	—	—
		D	—	H	5.12	—	—	—	—	—	—
21[d]	1	A	OAll	—	4.87	3.91			3.40–3.85		
		B	—	—	5.11	4.01			3.40–3.85		
		D	—	Ac	4.96	5.45			3.40–3.85		
21a	1	A	SC_6H_4OMPEG	—	5.30	—	—	—	—	—	—
		B	—	—	5.19[c]	—	—	—	—	—	—
		D	—	Ac	5.04[c]	5.50	—	—	—	—	—
22[d]	1	A	OAll	—	4.95	3.99			3.55–3.88		
		B	—	—	5.10[c]	4.11			3.55–3.88		
		D	—	H	5.22[c]	4.11			3.55–3.88		
22a	1	A	SC_6H_4OMPEG	—	5.49	—	—	—	—	—	—
		B	—	—	5.23[c]	—	—	—	—	—	—
		D	—	H	5.10[c]	—	—	—	—	—	—
23[d]	2	A	OAll	—	4.97	3.89			3.44–4.02		
		B	—	—	5.21[c]	4.08			3.44–4.02		
		C	—	—	5.17[c]	4.08			3.44–4.02		
		D	—	Ac	5.02	5.54	4.0		3.44–3.98		
4[d]	2	A	OAll	—	4.98	3.99			3.46–3.96		
		B	—	—	5.22[c]	~4.1			3.46–3.96		
		C	—	—	5.21[c]	~4.1			3.46–3.96		
		D	—	H	5.11[c]	~4.1			3.46–3.96		

[a] The resonances for acetyl, allyl and benzyl groups were present as required but are not reported. As a rule, only H-1 and H-2 could be assigned. Coupling constants were recorded as follows: $J_{1,2}$ 1.7 ± 0.2, $J_{2,3}$ 3.0 ± 0.3, $J_{3,4}$ 9.2 ± 0.2.

[b] For MPEG-supported compounds, the anomeric protons and acetylated C-2 protons were assigned by comparison with the spectra of the corresponding solution-synthesised compounds. All other protons were obscured by the signals for benzylic and polyether methylene groups. Singlets for OMe at δ 3.38 doublets for H-3_{Ar} at δ 6.65–676 were observed but are not reported.

[c] Assignments may be reversed.

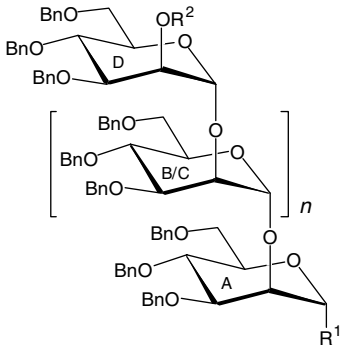
[d] Spectrum recorded at 500 MHz.

with DBU (50 mg, 9.33 mmol) in dry MeOH (10 mL) as described above for the preparation of compound **18**. Purification by flash chromatography (4:1→3:1 light petroleum–EtOAc) gave **22** (0.16 g, 0.118 mmol, 92%) as a foam: $[\alpha]_D$ +33.2 (*c* 1.67, CH_2Cl_2). ^{1}H and ^{13}C NMR data are given in Tables 5 and 6, respectively. HRMS, *m/z*: Calcd for $C_{84}H_{90}O_{16}$ $[M]^+$ 1354.6229. Found 1354.6141. Calcd for $C_{84}H_{90}O_{16}Na$ $[M+Na]^+$ 1377.6127. Found 1377.5983. Calcd for $C_{84}H_{90}O_{16}K$ $[M+K]^+$ 1393.5866. Found 1393.5852.

4.27. Allyl (3,4,6-tri-*O*-benzyl-α-D-mannopyranosyl)-(1→2)-(3,4,6-tri-*O*-benzyl-α-D-mannopyranosyl)-(1→2)-(3,4,6-tri-*O*-benzyl-α-D-mannopyranosyl)-(1→2)-(3,4,6-tri-*O*-benzyl-α-D-mannopyranoside (4)

4.27.1. By solution synthesis. Compound **22** (200 mg, 0.15 mmol) was glycosylated with trichloroacetimidate **16** (190 mg, 0.30 mmol) in dry CH_2Cl_2 (5 mL) using TMSOTf (5 μL, 0.025 mmol) as promoter, as described above for the preparation of compound **9**. Purification

Table 6. ^{13}C NMR chemical (δ) shifts for (1→2)-linked oligosaccharide derivatives[a,b]



Compound	*n*	Sugar ring	R^1	R^2	C-1	C-2	C-3	C-4	C-5	C-6
19	0	A	OAll	—	98.3	75.3	80.1	72.3,72.3, 75.1	74.8,	69.5[c]
		D	—	Ac	100.0	69.2	78.6			69.7[c]
19a	0	A	SC_6H_4OMPEG	—	87.9	—	—	—	—	—
		D	—	Ac	99.6	—	—	—	—	—
20	0	A	OAll	—	98.4	75.3	80.1[c]	71.9, 72.3, 75.1	74.7,	69.5[d]
		D	—	H	101.4	68.9	80.3[c]			69.7[d]
20a	0	A	SC_6H_4OMPEG	—	87.9, $J_{C\text{-}1,H\text{-}1}$ 179 Hz		—	—	—	—
		D	—	H	101.3, $J_{C\text{-}1,H\text{-}1}$ 180 Hz		—	—	—	—
21	1	A	OAll	—	98.5	~76	80.0[c]	—	—	70.0[d]
		B	—	—	101.1	~76	79.8[c]	—	—	69.8[d]
		D	—	Ac	99.9	69.2	78.6[c]	—	—	69.3[d]
21a	1	A	SC_6H_4OMPEG	—	87.9	—	—	—	—	—
		B	—	—	100.7[c]	—	—	—	—	—
		D	—	Ac	99.2[c]	—	—	—	—	—
22	1	A	OAll	—	98.5	75.6	80.4[d]	75.3, 74.7, 72.7, 72.3, 72.3, 72.0		69.9[e]
		B	—	—	101.4[c]	75.6	79.9[d]			69.7[e]
		D	—	H	101.2[c]	69.0	79.9[d]			69.4[e]
22a	1	A	SC_6H_4OMPEG	—	88.3	—	—	—	—	—
		B	—	—	101.8[c]	—	—[b]	—	—	—
		D	—	H	101.0[c]	—	—		—	—
23	2	A	OAll		98.5	—	79.8[d]	—	—	70.1[e]
		B	—	—	101.5[c]	—	79.8[d]	—	—	70.0[e]
		C	—	—	101.2[c]	—	79.6[d]	—	—	69.8[e]
		D	—	Ac	99.8[c]	69.1	78.7[d]	—	—	69.4[e]
4	2	A	OAll	—	98.5	76.1[d]	80.6[e]	75.4, 75.3, 75.3, 74.8, 72.8, 72.6, 72.2, 72.1		70.0[e]
		B	—	—	101.4[c]	76.1[d]	79.8[e]			70.0[f]
		C	—	—	101.4[c]	75.6[d]	79.8[e]			69.8[f]
		D	—	H	101.5[c]	69.0	79.6[e]			69.3[f]

[a] The resonances for acetyl, allyl and benzyl groups were present as required but are not reported.
[b] For MPEG-supported compounds, the anomeric carbons were assigned by comparison with the spectra of the corresponding solution-synthesised compounds. The signals of all other carbohydrate carbon atoms were obscured by the massive polyether methylene peak. Signals for MeO at δ 58.9 were also observed but are not reported.
[c–f] Assignments may be reversed.

by flash chromatography (4:1 light petroleum–EtOAc) gave the 2‴-O-acetylated tetrasaccharide **23** (241 mg, 0.13 mmol, 88%), which was not characterised [^{13}C NMR ($CDCl_3$) δ 101.5, 101.2, 99.7 (C-1′, -1″, -1‴), 98.4 (C-1)]. Deacetylation of most of this material (227 mg, 0.124 mmol) by treatment with DBU (100 mg, 0.65 mmol) in dry MeOH (20 mL), as described above for the preparation of **22**, and purification of the crude product by flash chromatography (4:1 light petroleum–EtOAc) gave tetrasaccharide **4** (182 mg, 0.102 mmol, 82%) as a foam: $[\alpha]_D$ +27.4 (*c* 1.6, CH_2Cl_2). ^{1}H and ^{13}C NMR data are given in Tables 5 and 6, respectively. HRMS, *m*/*z*: Calcd for $C_{111}H_{118}O_{21}$ $[M]^+$ 1786.8166. Found: 1786.8103.

4.27.2. By MPEG-supported synthesis. To a stirred solution of **4a** (300 mg, 0.044 mmol, see later) in aqueous acetone (85%, 9 mL) NBS (60 mg, 0.34 mmol) and 2,6-di-*tert*-butylpyridine (one drop, ~10 mg, 0.05 mmol) were added.[37] After stirring for 2 h the solvents were removed, CH_2Cl_2 (2 mL) was added, followed by cold ether (100 mL). The precipitate was filtered off and washed well with more ether. The combined filtrate and washings were evaporated and the residue was purified by flash chromatography (2:1 light petroleum–EtOAc) to yield as the major product (3,4,6-tri-*O*-benzyl-α-D-mannopyranosyl)-(1→2)-(3,4,6-tri-*O*-benzyl-α-D-mannopyranosyl)-(1→2)-(3,4,6-tri-*O*-benzyl-α-D-mannopyranosyl)-(1→2)-(3,4,6-tri-*O*-benzyl-α/β-D-mannopyranose **24** (24 mg, 0.0138 mmol, 31%) as a 2:1 mixture of anomers. ^{1}H NMR ($CDCl_3$, 500 MHz): δ 5.29 (br s, H-1α), 5.23, 5.21, 5.12 (3 d, $J_{1,2}$ ~ 1 Hz, H-1′α, -1″α, -1‴α), 4.38 ($J_{1,2}$ ~ 1 Hz, H-1β), 5.18, 5.13, 5.02 (3 d, $J_{1,2}$ ~ 1 Hz, H-1′β, -1″β, -1‴β), 3.30 (d, $J_{2,3}$ 3 Hz, $J_{3,4}$ 9 Hz, H-3β); ^{13}C NMR ($CDCl_3$): δ 101.9, 101.6, 101.4 (C-1′β, -1″β, -1‴β), 101.5, 101.4, 101.4 (C-1′α, -1″α, -1‴α), 94.0 ($J_{C\text{-}1,H\text{-}1}$ 160 Hz, H-1β), 93.8, $J_{C\text{-}1,H\text{-}1}$ 171 Hz, C-1α), 81.7 (C-3β), 80.3, 80.1, 78.5 (C-3′β, -3″-β, -3‴β), 80.5, 79.9, 79.4, 79.1 (C-3α, -3′α, -3″α, -3‴α). HRMS, *m*/*z*: Calcd for $C_{108}H_{114}CsO_{21}$ $[M+Cs]^+$ 1879.6907. Found 1879.6877.

To a stirred solution of this compound (15 mg, 0.0126 mmol) in dry CH_2Cl_2 (1 mL) at 0 °C dry K_2CO_3 (50 mg) was added, followed by Cl_3CCN (15 μL). After being stirred for 16 h at 20 °C, the mixture was flash chromatographed (4:1 light petroleum–EtOAc) to give (3,4,6-tri-*O*-benzyl-α-D-mannopyranosyl)-(1→2)-(3,4,6-tri-*O*-benzyl-α-D-mannopyranosyl)-(1→2)-(3,4,6-tri-*O*-benzyl-α-D-mannopyranosyl)-(1→2)-(3,4,6-tri-*O*-benzyl-α/β-D-mannopyranosyl trichloroacetimidate (16 mg, 0.00845 mmol, 98.5%) **25** as a colourless syrup. ^{1}H NMR ($CDCl_3$): δ 8.61 s, NH minor anomer), 8.48 (s, NH major anomer), 6.27, 6.27, 5.62, 5.43 (4 br s, H-1, -1′, -1″, -1‴, minor anomer), 5.28, 5.26, 5.17, 5.12 (4 d, $J_{1,2}$ ~ 1 Hz, H-1, -1′, -1″, -1‴, major anomer); ^{13}C NMR ($CDCl_3$): δ 101.1, 99.9, 99.9, 99.8 (C-1, -1′, -1″, -1‴, major anomer), 97.9, 95.8, 95.8, 94.4 (C-1, -1′, -1″, -1‴, minor anomer).

The mixed trichloroacetimidates (15 mg, 0.0079 mmol) were stirred in dry CH_2Cl_2 (1.5 mL) with powdered 4 Å molecular sieves (30 mg) and allyl alcohol (100 μL) at 0 °C under argon. TMSOTf (2 μL) was added and stirring was continued at 20 °C. After 1.5 h the reaction was quenched with Et_3N (5 μL), the solvents were evaporated and the residue was purified by flash chromatography to give successively compound **4** (2 mg), a 2:1 mixture of compound **4** and its β-anomer (10 mg) and the β-anomer (2 mg, altogether 14 mg, 0.00783 mmol, 98.7%).

α-Anomer: ^{1}H and ^{13}C NMR data identical to those of the solution synthesised compound (Tables 5 and 6). MALDI HRMS, *m*/*z*: Calcd for $C_{111}H_{118}O_{21}Na$ $[M+Na]^+$ 1809.8063. Found 1809.7731.

β-Anomer: ^{1}H NMR ($CDCl_3$): δ 5.24, 5.17, ~5.1, 4.92 (4 br s, H-1, -1′, -1″, -1‴, ^{13}C NMR ($CDCl_3$): δ 101.4, 101.1, 100.4, 99.88 (C-1, -1′, -1″, -1‴). MALDI HRMS, *m*/*z*: Calcd for $C_{111}H_{118}O_{21}Na$ $[M+Na]^+$ 1809.8063. Found 1809.7728.

4.28. *S*-(4-MPEG-oxyphenyl) 2-*O*-acetyl-3,4,6-tri-*O*-benzyl-1-thio-α-D-mannopyranoside (17a)

A solution of 4-MPEG-oxythiophenol[21] (4.0 g, 0.78 mmol) and trichloroacetimidate **16** (4.0 g, 6.3 mmol) in CH_2Cl_2 (20 mL) was stirred with powdered 4 Å molecular sieves (200 mg) at 0 °C under argon. $BF_3{\cdot}OEt_2$ (500 μL, 4 mmol) was added and stirring was continued at 20 °C. After 1.5 h the reaction was quenched by addition of Et_3N (1.0 mL, 5.7 mmol) at 0 °C, followed after 5 min by dry diethyl ether (250 mL). The solid precipitate was removed by filtration, washed with ether (50 mL), isopropanol (3 × 50 mL) to remove triethylammonium salts[28] and ether again (50 mL), to give, after recrystallisation from absolute EtOH and thorough drying in vacuo, the title compound **17a** (4.15 g, 0.74 mmol, 94%). ^{1}H and ^{13}C NMR data are given in Table 4.

4.29. *S*-(4-MPEG-oxyphenyl) 3,4,6-tri-*O*-benzyl-1-thio-α-D-mannopyranoside (18a)

A solution of **17a** (2.8 g, 0.505 mmol) in dry MeOH (40 mL) containing DBU (250 mg) was kept at 20 °C for 48 h. After it was cooled to 0 °C, cold ether (200 mL) was added. The precipitate was collected by filtration, washed well with ether and recrystallised from absolute EtOH to give the title compound **18a** (2.6 g, 0.47 mmol, 93%). ^{1}H and ^{13}C NMR data are given in Table 4.

4.30. *S*-(4-MPEG-oxyphenyl) (2-*O*-acetyl-3,4,6-tri-*O*-benzyl-α-D-mannopyranosyl)-(1→2)-3,4,6-tri-*O*-benzyl-1-thio-α-D-mannopyranoside (19a)

A solution of compound **18a** (2.5 g, 0.45 mmol) and trichloroacetimidate **16** (1.15 g, 1.8 mmol) in dry CH_2Cl_2 (20 mL) was stirred with powdered 4 Å molecular sieves (200 mg) at 0 °C under argon. TMSOTf (30 μL, 0.16 mmol) was added and stirring was continued at 20 °C. After 1.5 h the reaction mixture was processed as described above for the preparation of **17a**, to give the supported disaccharide glycoside **19a** (2.51 g, 0.42 mmol, 93%). NMR data for the anomeric protons and H-2′ and for the anomeric carbon atoms are given in Tables 5 and 6, respectively.

4.31. *S*-(4-MPEG-oxyphenyl) (3,4,6-tri-*O*-benzyl-α-D-mannopyranosyl)-(1→2)-3,4,6-tri-*O*-benzyl-1-thio-α-D-mannopyranoside (20a)

A solution of 2′-*O*-acetate **19a** (2.0 g, 0.332 mmol) was treated with DBU (250 mg) in dry MeOH (40 mL) as described above for the preparation of **18a**, to give compound **20a** (1.91 g, 0.320 mmol, 96%). NMR data for the anomeric protons and carbon atoms are given in Tables 5 and 6, respectively.

4.32. *S*-(4-MPEG-oxyphenyl) (2-*O*-acetyl-3,4,6-tri-*O*-benzyl-α-D-mannopyranosyl)-(1→2)-(3,4,6-tri-*O*-benzyl-1-thio-α-D-mannopyranosyl)-(1→2)-3,4,6-tri-*O*-benzyl-α-D-mannopyranoside (21a)

Compound **20a** (1.91 g, 0.32 mmol) was glycosylated with trichloroacetimidate **16** (0.85 g, 1.33 mmol) and TMSOTf (20 μL, 0.11 mmol) as promoter as described above for the preparation of **19a**, to give a mixture (1.96 g) of trisaccharide derivative **21a** and starting material **20a**. This mixture was glycosylated again using trichloroacetimidate **16** (535 mg, 0.84 mmol) and following the same procedure, to give product **21a** (1.62 g, 0.25 mmol, 89% over both glycosylations). NMR data for the anomeric protons and H-2″ and for the anomeric carbon atoms are given in Tables 5 and 6, respectively.

4.33. *S*-(4-MPEG-oxyphenyl) (3,4,6-tri-*O*-benzyl-α-D-mannopyranosyl)-(1→2)-(3,4,6-tri-*O*-benzyl-α-D-mannopyranosyl)-(1→2)-3,4,6-tri-*O*-benzyl-1-thio-α-D-mannopyranoside (22a)

A solution of 2″-*O*-acetate **21a** (1.0 g, 0.155 mmol) was treated with DBU (200 mg) in dry MeOH (40 mL) as described above for the preparation of **18a** to give the title compound **22a** (0.89 g, 0.139 mmol, 90%). NMR data for the anomeric protons and carbon atoms are given in Tables 5 and 6, respectively.

4.34. *S*-(4-MPEG-oxyphenyl) (2-*O*-acetyl-3,4,6-tri-*O*-benzyl-α-D-mannopyranosyl)-(1→2)-(3,4,6-tri-*O*-benzyl-α-D-mannopyranosyl)-(1→2)-(3,4,6-tri-*O*-benzyl-α-D-mannopyranosyl)-(1→2)-3,4,6-tri-*O*-benzyl-1-thio-α-D-mannopyranoside (23a)

Compound **22a** (0.75 g, 0.117 mmol) was glycosylated with trichloroacetimidate **16** (0.30 g, 0.47 mmol) and TMSOTf (10 μL, 0.05 mmol) as promoter as described above for the preparation of **19a**, to give a mixture (0.73 g) of trisaccharide derivative **23a** and starting material **22a**. This mixture (0.45 g) was glycosylated again using trichloroacetimidate **16** (180 mg, 0.28 mmol) and repeating the procedure, to give the title compound **23a** (405 mg, 0.059 mmol, 82% over both glycosylations).

4.35. *S*-(4-MPEG-oxyphenyl) (3,4,6-tri-*O*-benzyl-α-D-mannopyranosyl)-(1→2)-(3,4,6-tri-*O*-benzyl-α-D-mannopyranosyl)-(1→2)-(3,4,6-tri-*O*-benzyl-α-D-mannopyranosyl)-(1→2)-3,4,6-tri-*O*-benzyl-1-thio-α-D-mannopyranoside (4a)

A solution of 2‴-*O*-acetate **23a** (0.5 g, 0.073 mmol) was treated with DBU (100 mg) in dry MeOH (20 mL) as described above for the preparation of **18a**, to give compound **4a** (0.42 g, 0.062 mmol, 85%).

4.36. 4-Chlorobutyl α-D-mannopyranoside (26)

A mixture of D-mannose (20 g, 110 mmol) and 4-chlorobutanol (tech., containing traces of HCl, 150 mL) was stirred at 20 °C for 7 d. Most of the unreacted 4-chlorobutanol was removed under vacuum to give the crude glycoside, contaminated with 4-chlorobutanol, as a brown syrup (45 g). This material was used in the next step without further treatment. Purification of a small sample by chromatography (4:1 EtOAc–MeOH) and crystallisation from EtOAc gave the pure mannoside **26**, mp 106–107 °C, $[\alpha]_D$ +68.6 (*c* 1.17, MeOH). ^{1}H and ^{13}C NMR data are given in Tables 7 and 8, respectively. Anal. Calcd for $C_{10}H_{19}ClO_6$: C, 44.37; H, 7.07, Cl, 13.10. Found: C, 44.31; H, 6.92; Cl, 12.97.

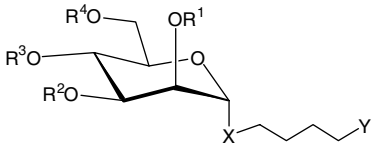
4.37. 4-Chlorobutyl 2,6-di-*O*-benzoyl-α-D-mannopyranoside (27) and 4-chlorobutyl 2,4-di-*O*-benzoyl-α-D-mannopyranoside (28)

A solution of crude glycoside **26** (43.8 g, obtained from 0.108 mol D-mannose, contaminated with 4-chlorobutanol, see above) in CH_2Cl_2 (400 mL) was stirred with trimethyl orthobenzoate (100 mL, 149 g, 0.80 mol) and camphor sulfonic acid (300 mg) under argon at 20 °C for 16 h. Aqueous acetic acid (1 L, 1:1) was added and stirring was continued for 5 h. The organic layer was removed, diluted with CH_2Cl_2 (600 mL), washed successively with water, aqueous $NaHCO_3$ and water, dried ($MgSO_4$) and evaporated to an almost colourless oil. Flash chromatography (4:1 light petroleum–EtOAc) furnished the required 2,4-di-*O*-benzoate **28** (21.5 g, 45.0 mmol, 41.6% from D-mannose) as a colourless syrup, followed by 2,6-di-*O*-benzoate **27** (12.7 g, 26.6 mmol, 24.6% from D-mannose), which crystallised from EtOH.

2,6-Dibenzoate **27**: mp 76–78 °C, $[\alpha]_D$ +20.9 (*c* 1.5, CH_2Cl_2). ^{1}H and ^{13}C NMR data are given in Tables 7 and 8, respectively. Anal. Calcd for $C_{24}H_{27}ClO_8$: C, 60.19; H, 5.68; Cl, 7.40. Found: C, 59.89; H, 5.77; Cl, 7.31.

2,4-Dibenzoate **28**: $[\alpha]_D$ −32.8 (*c* 1.2, CH_2Cl_2). ^{1}H and ^{13}C NMR data are given in Tables 7 and 8, respectively. HRMS, *m*/*z*: Calcd for $C_{24}H_{28}ClO_8$ $[M+H]^+$ 479.1473. Found 479.1479.

Table 7. ^{1}H NMR Chemical shifts (δ) for the monosaccharide precursors of 3,6-branched oligosaccharide derivatives[a]



Compound	X	Y	R^1	R^2	R^3	R^4	H-1	H-2	H-3	H-4	H-5	H-6a	H-6b
26[b]	O	Cl	H	H	H	H	4.75			3.45–3.85			
27	O	Cl	Bz	H	H	Bz	4.92	5.34	~4.15	3.85–4.0		4.75	4.55
28	O	Cl	Bz	H	Bz	H	5.04	5.39	4.40	5.50	3.93	3.5–3.8	
31[c]	S	OH	Ac	Ac	Ac	Ac	5.26	5.33	5.27	5.29	4.38	4.31	4.11
32[c]	S	OMs	Ac	Ac	Ac	Ac	5.26	5.32	5.24	5.31	4.35	4.31	4.10
33[c]	S	N_3	Ac	Ac	Ac	Ac	5.26	5.33	5.25	5.30	4.36	4.31	l4.10
34[c,d]	S	N_3	H	H	H	H	5.08	3.69	3.4–3.45		3.6–3.65		3.49
35	S	N_3	Bz	H	H	Bz	5.37	5.45	4.09	3.97	4.28	4.79	4.53
36[c,d]	S	N_3	Bz	H	Bz	H	5.49	5.35	4.1–4.2	5.39	4.1–4.2	3.52–3.56	
37[c,d]	S	NCS	Bz	H	Bz	H	5.51	5.41	4.25	5.45	~4.24	3.60–3.62	
38[e]	S	'MPEG'[f]	Bz	H	Bz	H	5.4–5.6		4.3–4.4	5.4–5.6	4.3–4.4	—	—

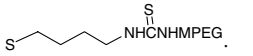
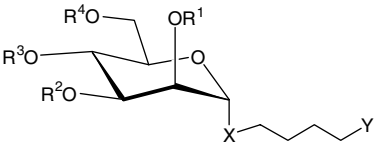
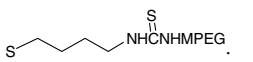
[a] The resonances for acetyl, benzyl, benzoyl and methanesulfonyl groups were present as required but are not reported. Coupling constants were recorded as follows: $J_{1,2}$ 1.4 ± 0.3, $J_{2,3}$ 3.25 ± 0.15, $J_{3,4} = J_{4,5}$ 9.7 ± 0.2, $J_{5,6a}$ 4.7 ± 0.7, $J_{5,6b}$ 2.0 ± 0.4, $J_{6a,6b}$ 12.15 ± 0.15.
[b] Spectrum recorded in CD_3OD.
[c] Spectrum recorded at 500 MHz.
[d] Spectrum recorded in $(CD_3)_2SO$.
[e] The signals for the linker methylene groups were present but are not reported.
[f] 'MPEG' = 

Table 8. ^{13}C NMR chemical shifts (δ) for the monosaccharide precursors of 3,6-branched oligosaccharide derivatives[a]



Compound	X	Y	R^1	R^2	R^3	R^4	C-1	C-2	C-3	C-4	C-5	C-6
26[b]	O	Cl	H	H	H	H	101.6		74.7, 72.7, 72.2, 68.6			62.9
27	O	Cl	Bz	H	H	Bz	98.1	72.8	70.5	68.3[c]	71.3[c]	64.1
28	O	Cl	Bz	H	Bz	H	98.0	73.4	69.1	70.8	71.1	61.9
31	S	OH	Ac	Ac	Ac	Ac	82.9	71.6	69.8	66.8	69.4	62.9
32	S	OMs	Ac	Ac	Ac	Ac	82.4	71.1	69.5	66.3	69.1	62.5
33	S	N_3	Ac	Ac	Ac	Ac	82.8	71.3	69.7	66.6	69.2	62.7
34[d]	S	N_3	H	H	H	H	84.8	71.9	71.6	67.3	74.5	61.1
35	S	N_3	Bz	H	H	Bz	83.1	74.3	71.0	68.5	71.6	64.0
36	S	N_3	Bz	H	Bz	H	83.2	75.0	71.1[c]	70.9	69.7[c]	61.8
37	S	NCS	Bz	H	Bz	H	82.3	74.8	72.5	70.6	67.9	60.9
38[e]	S	'MPEG'[f]	Bz	H	Bz	H	83.0	74.8	—	—	69.0	61.4

[a] The resonances for acetyl, benzoyl, benzyl, hydroxybutyl and methanesulfonyl groups were present as required but are not reported.
[b] Spectrum recorded in CD_3OD.
[c] Assignment may be reversed.
[d] Spectrum recorded in $(CD_3)_2SO$.
[e] The carbon resonances for the linker were present as required but are not reported and a signal for C=S was observed at δ 183 ppm.
[f] 'MPEG' = 

4.38. 4-Chlorobutyl 3,6-di-*O*-(2-*O*-acetyl-3,4,6-tri-*O*-benzyl-α-D-mannopyranosyl)-2,4-di-*O*-benzoyl-α-D-mannopyranoside (29)

4-Chlorobutyl 2,4-di-*O*-benzoyl-α-D-mannopyranoside **28** (1.02 g, 2.1 mmol) was glycosylated with trichloroacetimidate **16** (3.9 g, 6.1 mmol) in dry CH_2Cl_2 (40 mL) using TMSOTf (70 μL, 0.38 mmol) as promoter, as described above for the preparation of compound **9**. Purification of the foam thus obtained by flash chromatography (4:1→2:1 light petroleum–EtOAc) gave trisaccharide derivative **29** (2.54 g,

1.78 mmol, 85%) as a foam: $[\alpha]_D$ +2.8 (*c* 0.96, CH_2Cl_2). ^1H and ^{13}C NMR data are given in Table 9. HRMS, *m/z*: Calcd for $C_{82}H_{87}ClO_{20}$ $[M]^+$ 1426.5479. Found 1426.5348. Calcd for $C_{82}H_{88}ClO_{20}$ $[M+H]^+$ 1427.5558. Found 1427.5426.

4.39. 4-Chlorobutyl 2,4-di-*O*-benzoyl-3,6-di-*O*-(3,4,6-tri-*O*-benzyl-α-D-mannopyranosyl)-α-D-mannopyranoside (30)

A stirred suspension of **29** (2.0 mg, 1.40 mmol) in dry MeOH (20 mL) was treated at 20 °C with methanolic HCl, prepared by addition of acetyl chloride (0.8 mL) to dry MeOH (10 mL) with cooling.[29] After stirring of the mixture at 50 °C for 8 h, then cooling to 0 °C, the acid was neutralised by the addition of pyridine (ca. 3 mL). The solvents were evaporated and the syrupy residue was subjected to flash chromatography (2:1 light petroleum–EtOAc) to give the deacetylated trisaccharide derivative **30** (1.41 g, 1.05 mmol, 75%) as a foam: $[\alpha]_D$ +8.6 (*c* 1.16, CH_2Cl_2). ^1H and ^{13}C NMR data are given in Table 9. HRMS, *m/z*: Calcd for $C_{78}H_{84}ClO_{18}$ $[M+H]^+$ 1343.5346. Found 1343.5330.

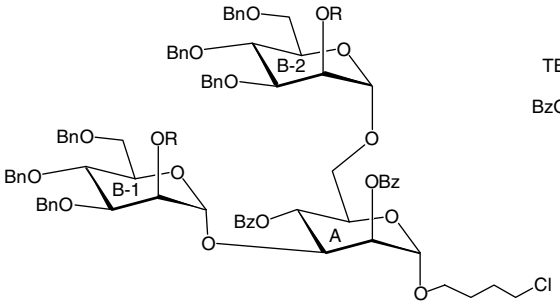
4.40. 4-Chlorobutyl 2,4-di-*O*-benzoyl-3,6-di-*O*-[2,3,4-tri-*O*-benzoyl-6-*O*-*tert*-butyldimethylsilyl-3,4,6-(tri-*O*-benzyl-2-*O*-α-D-mannopyranosyl)-α-D-mannopyranosyl]-α-D-mannopyranoside (2 [R = $CH_2(CH_2)_2CH_2Cl$])

Trisaccharide diol **30** (300 mg, 0.233 mmol) was glycosylated with trichloroacetimidate **8** (690 mg, 0.92 mmol) in dry CH_2Cl_2 (50 mL) using TMSOTf (10 μL, 0.05 mmol) as promoter, as described above for the preparation of compound **9**. Purification of the foam thus obtained by flash chromatography (5:1→2:1 light petroleum–EtOAc) gave the pentasaccharide derivative **2** [R = $CH_2(CH_2)_2CH_2Cl$] (351 mg, 0.139 mmol, 62%) as a white foam: $[\alpha]_D$ −39.0 (*c* 1.44, CH_2Cl_2). ^1H and ^{13}C NMR data are given in Table 9. MALDI HRMS, *m/z*: Calcd for $C_{144}H_{155}ClO_{34}Si_2Na$ $[M+Na]^+$ 2541.9525. Found 2541.9767.

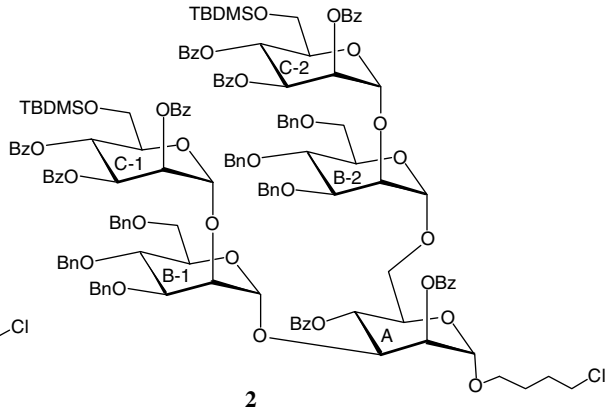
4.41. *S*-(4-Hydroxybutyl) 2,3,4,6-tetra-*O*-acetyl-1-thio-α-D-mannopyranoside (31)

A solution of tetra-*O*-acetyl-α-D-mannopyranosyl bromide (50 g, 0.11 mol) and thiourea (11.0 g, 0.145 mol) in acetone (200 mL) was stirred at 65 °C for 1 h. The

Table 9. H and ^{13}C NMR chemical shifts (δ) for 3,6-branched oligosaccharide derivatives[a]



29 R = Ac
30 R = H



2

Compound	Sugar ring	H-1	H-2	H-3	H-4	H-5	H-6a	H-6b	C-1	C-2	C-3	C-4	C-5	C-6
29	A	~5.0	5.47	~4.4	5.72	~4.0	3.4–3.9		97.5	72.9	76.4	69.2	69.6	69.0[c]
	B-1[b]	5.04	~5.0	3.63		3.4–3.9			100.1	68.9	77.9	{71.8,	72.6,	67.8[c]
	B-2[b]	4.86	5.32	3.86					98.0	68.8	78.7	73.9,	74.4	67.0[c]
30	A	4.98	5.47	~4.4	5.73	4.05	3.88	~3.67	97.8	72.3	76.0	69.7	69.9	67.0
	B-1[b]	5.05	3.67	~3.6		3.45–3.8			102.0	68.7	80.7	{71.5,	72.6,	69.1[c]
	B-2[b]	4.93	3.94	~3.8					99.7	68.4	80.0	74.0,	74.5	68.0[c]
2[h]	A	5.08	5.74	4.52	5.78	4.11	~4.03	~3.70	97.4	72.4	75.7	69.6	69.6	66.8
	B-1[b]	5.30	~3.70	~3.80	4.29	—	—	—	101.1	76.9	79.3	70.5[c]	—	67.9[d]
	B-2[b]	5.04	~4.02	3.96	—	—	—	—	98.8	—	80.1	72.2[c]	—	67.5[d]
	C-1[b]	4.72	5.74	5.91	6.02	4.30[c]	3.63–3.67		99.3	70.5	70.5	66.8[e]	71.9[f]	61.7[g]
	C-2[b]	5.18	5.90	5.83	6.02	4.17[c]	3.85–3.90		99.5	70.5	70.5	66.4[e]	71.7[f]	66.2[g]

[a] Resonances for acetyl, benzoyl, benzyl, chlorobutyl and methanesulfonyl groups were present as required but are not reported.
[b] The resonances in each row are derived from the same sugar moiety (^1H–^1H COSY and ^1H–^{13}C COSY experiments), but the assignments for rings B^1 and B^2, C^1 and C^2 may be reversed.
[c–g] Assignments may be reversed.
[h] Spectra recorded at 500 MHz.

solvent was evaporated and the residue was dissolved in CH_2Cl_2 (150 mL). A solution of sodium metabisulfite (15 g) in water (75 mL) was added and the mixture was stirred at 65 °C for 1 h. After it was cooled to 20 °C, the organic layer was removed, washed with water, dried ($MgSO_4$) and evaporated. A solution of the residue in acetone (150 mL) was stirred overnight at 20 °C with K_2CO_3 (20 g, 0.14 mol), 4-chlorobutanol (85%, 20 mL, 0.14 mol) and sodium iodide (20 mg). The dark, brown-red syrup obtained after removal of the solids and solvent was subjected to flash chromatography (2:1→1:1 light petroleum–EtOAc) to afford the title compound **31** (19.1 g, 0.44 mol, 40%) as a pale orange oil, which solidified on standing. Recrystallisation from light petroleum–EtOAc gave colourless needles of mp 98–99 °C, $[\alpha]_D$ +95.5 (*c* 1.4, CH_2Cl_2). ^1H and ^{13}C NMR data are given in Tables 7 and 8, respectively. Anal. Calcd for $C_{18}H_{28}O_{10}S$: C, 49.53; H, 6.47; S, 7.35. Found: C, 49.13; H, 6.58; S, 7.32.

4.42. *S*-(4-Mesyloxybutyl) 2,3,4,6-tetra-*O*-acetyl-1-thio-α-D-mannopyranoside (32)

A solution of compound **31** (18 g, 41 mmol) and mesyl chloride (5 mL) in pyridine (300 mL) was stirred for 2 h at 20 °C. The solvent was evaporated to leave a brown syrup, which crystallised on trituration with EtOH. Recrystallised from the same solvent mesylate **32** (16.4 g, 31.9 mmol, 78%) had mp 122–123 °C, $[\alpha]_D$ +83.3 (*c* 1.5, CH_2Cl_2). Purification of the mother liquors by flash chromatography (2:1→1:1 light petroleum–EtOAc) gave a further quantity (2.8 g, total 19.2 g, 37 mmol, 91%) of product. ^1H and ^{13}C NMR data are given in Tables 7 and 8, respectively. Anal. Calcd for $C_{19}H_{30}O_{12}S_2$: C, 44.35; H, 5.88; S, 12.46. Found: C, 44.46; H, 6.06, S, 12.17.

4.43. *S*-(4-Azidobutyl) 2,3,4,6-tetra-*O*-acetyl-1-thio-α-D-mannopyranoside (33)

A solution of mesylate **32** (15.0 g, 29 mmol) and sodium azide (5.0 g, 76 mmol) in dry DMF (150 mL) was stirred at 80 °C under argon for 5 h, then left at 20 °C for 16 h. The dark brown syrup obtained on removal of the solvent was triturated with EtOH to give almost colourless crystals of azide **33** (8.9 g, 19.3 mmol, 66%); mp 82–82.5 °C, $[\alpha]_D$ +95.1 (*c* 1.0, CH_2Cl_2). Purification of the mother liquors by flash chromatography (2:1→1:1 light petroleum–EtOAc) gave a further quantity (3.3 g, total 12.2 g, 26.4 mmol, 91%) of product. ^1H and ^{13}C NMR data are given in Tables 7 and 8, respectively. Anal. Calcd for $C_{18}H_{27}N_3O_9S$: C, 46.85; H, 5.90; N, 9.11; S, 6.95. Found: C, 46.87; H, 5.81; N, 9.04; S, 6.96.

4.44. *S*-(4-Azidobutyl) 1-thio-α-D-mannopyranoside (34)

A solution of tetraacetate **33** (10 g, 21.6 mmol) in MeOH 200 mL was treated with methanolic NaOMe (1 M, 4 mL) for 4 h at 20 °C. The solvent was removed and the residue was purified by flash chromatography (10:1 CH_2Cl_2–MeOH) to afford the unprotected thioglycoside **34** (6.2 g, 21.1 mmol, 98%) as an oil; $[\alpha]_D$ +158.5 (*c* 1.53, water). ^1H and ^{13}C NMR data are given in Tables 7 and 8, respectively. HRMS, *m*/*z*: Calcd for $C_{10}H_{20}N_3O_5S$ $[M+H]^+$ 294.1124. Found 294.1118.

4.45. *S*-(4-Azidobutyl) 2,6-di-*O*-benzoyl-1-thio-α-D-mannopyranoside (35) and *S*-(4-azidobutyl) 2,4-di-*O*-benzoyl-1-thio-α-D-mannopyranoside (36)

A solution of glycoside **34** (4.2 g, 14.3 mmol) in CH_2Cl_2 (120 mL) was stirred with trimethyl orthobenzoate (12.5 mL, 18.6 g, 100 mmol) and camphor sulfonic acid (100 mg) under argon at 20 °C for 16 h. Aqueous acetic acid (300 mL, 1:1) was added and stirring was continued for 5 h. The organic layer was removed, diluted with CH_2Cl_2 (250 mL), washed with water, aqueous $NaHCO_3$ and water, dried ($MgSO_4$) and evaporated to an almost colourless oil. Flash chromatography (3:1→1:1 light petroleum–EtOAc) furnished the required 2,4-di-*O*-benzoate **36** (3.60 g, 50%) as a colourless syrup, followed by 2,6-di-*O*-benzoate **35** (3.11 g, 43%), which crystallised from EtOH.

2,6-Dibenzoate **35**: mp 103–4 °C, $[\alpha]_D$ +67.0 (*c* 1.99, CH_2Cl_2). ^1H and ^{13}C NMR data are given in Tables 7 and 8, respectively. Anal. Calcd for $C_{24}H_{27}N_3O_7S$: C, 57.47; H, 5.43; N, 8.38; S, 6.39. Found: C, 57.41; H, 5.43; N, 8.50; S, 6.39.

2,4-Dibenzoate **36**: $[\alpha]_D$ +47.8 (*c* 0.78, CH_2Cl_2). ^1H and ^{13}C NMR data are given in Tables 7 and 8, respectively. Anal. Calcd for $C_{24}H_{27}N_3O_7S$: C, 57.47; H, 5.43; N, 8.38; S, 6.39. Found: C, 57.53; H, 5.45; N, 8.55; S, 6.20. HRMS, *m*/*z*: Calcd for $C_{24}H_{28}N_3O_7S$ $[M+H]^+$ 502.1614. Found 502.1629.

4.46. *S*-(4-Isocyanatobutyl) 2,4-di-*O*-benzoyl-1-thio-α-D-mannopyranoside (37)

A solution of azide **36** (3.1 g, 6.18 mmol) in dry toluene (150 mL) was heated with triethyl phosphite (15 mL) and carbon disulfide (50 mL) at 70 °C under argon for 16 h. To the cooled reaction solution, aqueous $NaHCO_3$ (150 mL) was added and stirring was continued for 1 h. EtOAc (200 mL) was added, the organic phase was separated, washed with brine (2×), dried ($MgSO_4$) and evaporated to a pale yellow syrup. Purification by flash chromatography (2:1 light petroleum–EtOAc) gave product **37** (2.99 g, 5.78 mmol, 93%) as a colourless foam; $[\alpha]_D$ +44.7 (*c* 0.53, CH_2Cl_2). ^1H and ^{13}C NMR data are given in Tables 7 and 8, respectively. Anal.

Calcd for $C_{25}H_{27}NO_7S_2$: 58.01 H, 5.26; N, 2.71; S, 12.39. Found: C 57.62; H 5.29; N 2.85; S, 12.31.

4.47. 1-[4-(2,4-Di-*O*-benzoyl-1-thio-α-D-mannopyranosyl)-butyl]-3-(MPEG-yl)-thiourea (38)

A solution of MPEG-NH_2 (3.0 g, 0.60 mmol) and isothiocyanate **37** (1.2 g, 2.3 mmol) in CH_2Cl_2 (30 mL) was kept at 20 °C for 16 h, then cooled to 5 °C. Ice-cold diethyl ether (150 mL) was added, the solid precipitate was removed by filtration, washed with diethyl ether (3 × 25 mL) and recrystallised from absolute EtOH to give, after thorough drying in vacuo, the supported 1-thiomannoside **38** (3.18 g, 0.576 mmol, 96%). ^1H and ^{13}C NMR data are included in Tables 7 and 8, respectively.

From the filtrate unreacted **37** (0.82 g, 1.59 mmol) was isolated by flash chromatography (2:1 light petroleum–EtOAc).

4.48. Attempted glycosylation of the supported diol 38

The supported monosaccharide diol **38** (3.00 g, 0.54 mmol) was treated with trichloroacetimidate **16** (3.4 g, 5.4 mmol, 10 equiv) in dry CH_2Cl_2 (25 mL) using TMSOTf (60 μL, 0.33 mmol) as promoter, as described above for the preparation of **19a**. The product thus obtained (3.2 g) was retreated twice, using trichloroacetimidate **16** (10 equiv, total 30 equiv) following the same procedure, to give a product (2.85 g, after the three glycosylations, 81.5%, assuming complete glycosylation), which, however, was not the expected compound (NMR evidence, see Section 2.2.2).

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Supplementary data

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