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Chain-growth polyglycosylation: synthesis of linker-equipped mannosyl oligomers

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ABSTRACT

Direct syntheses of acetylated poly-mannosides can be achieved in one-step starting from a fully acetylated thioglycoside mannosyl donor using a polymerization-type strategy under the correct conditions. Under conditions that allow polymer growth from non-reducing to reducing end (N→R), different acceptor alcohols can be used as the 'terminating acceptors' to install different linkers at the reducing terminus. The efficiency is dependent on substituents of the linker, its length, temperature and choice of Lewis acid activator.

1. Introduction

As part of glycoproteins, so-called 'high mannose' glycans play fundamental roles in cells such as during protein biosynthesis (e.g., modulating folding, transport, enzymatic degradation) and in interactions (e.g., mediating adhesion, signalling).[1–4] These contain oligosaccharidic mannosyl 'arms' or 'caps', often mannobioside (α-D-Man-(1→2)-D-Man) and mannotrioside (α-D-Man-(1→2)-α-D-Man-(1→2)-D-Man) that are also determinants of pathogenicity of viruses such as the human immunodeficiency virus 1 (HIV-1),[5,6] bacteria (*Mycobacterium tuberculosis*) and protozoan parasites of the genus *Leishmania*.[7] Direct synthetic access to these fragments can be more difficult since the hydroxyl at C-2 is not accessible[8–12] by regioselective glycosylations.[13] Consequently, published syntheses typically require a minimum of 6–8 steps to form the mannosidic union between two moieties, often with bulky, atom-inefficient *O*-benzyl protected disaccharides.[14–19] A low yielding enzymatic access (3%) has also been reported.[20]

Glycosyl donors with 2-*O*-acyl protection give di-/tri-oxolenium ions upon activation (Fig. 1). These can, in principle, react directly with a nucleophile to form glycoside or reaction may proceed via an orthoester intermediate.[21] Orthoesters can also be rearranged to the desired glycosides or act as glycosyl donors themselves.[22,23] Under certain circumstances, they can also open in such a fashion that the 2-*O*-acyl group is transferred to the acceptor alcohol. Both of these latter reactions are often undesired side-reactions during glycosylations leading to, for example, *O*-acetyl migration (*trans*-acetylation) from the donor to the acceptor.[24–26] Nevertheless, we considered that such orthoesters, if generated from the corresponding thioglycosides in situ, theoretically showed a strong synthetic potential for sequential glycosylation at position 2-OH given that they and corresponding tri/dioxoleniums might act as *both* donors and acceptors (electrophiles and nucleophiles). Control of these ambident glycosyl moieties to show both of these characters under the appropriate conditions, might therefore allow a possible polymerization-type manifold. In principle, such a manifold could be made to 'grow'[27] sufficiently by requiring auto-activation to allow the formation of poly-mannosides of controllable length before termination by a chosen reducing end alcohol that possesses only acceptor ability. Here, we investigate the influence of the choice of Lewis acid, donor–acceptor ratio and scope of alcohol 'terminators' in first attempts to test this novel concept of what might be termed 'chain growth'[27] polyglycosylation. In addition, an initial systematic study of parameters offers first insights into the proposed mechanism involved in the generation of an intermediate O-2 mannosyl acceptor and observed acetyl transfer to alcohol.

2. Results and discussion

2.1. Testing polymannosylation

Acetylated thioglycoside **1** was chosen as a model glycosyl donor; such and similar donors have been reported to occasionally

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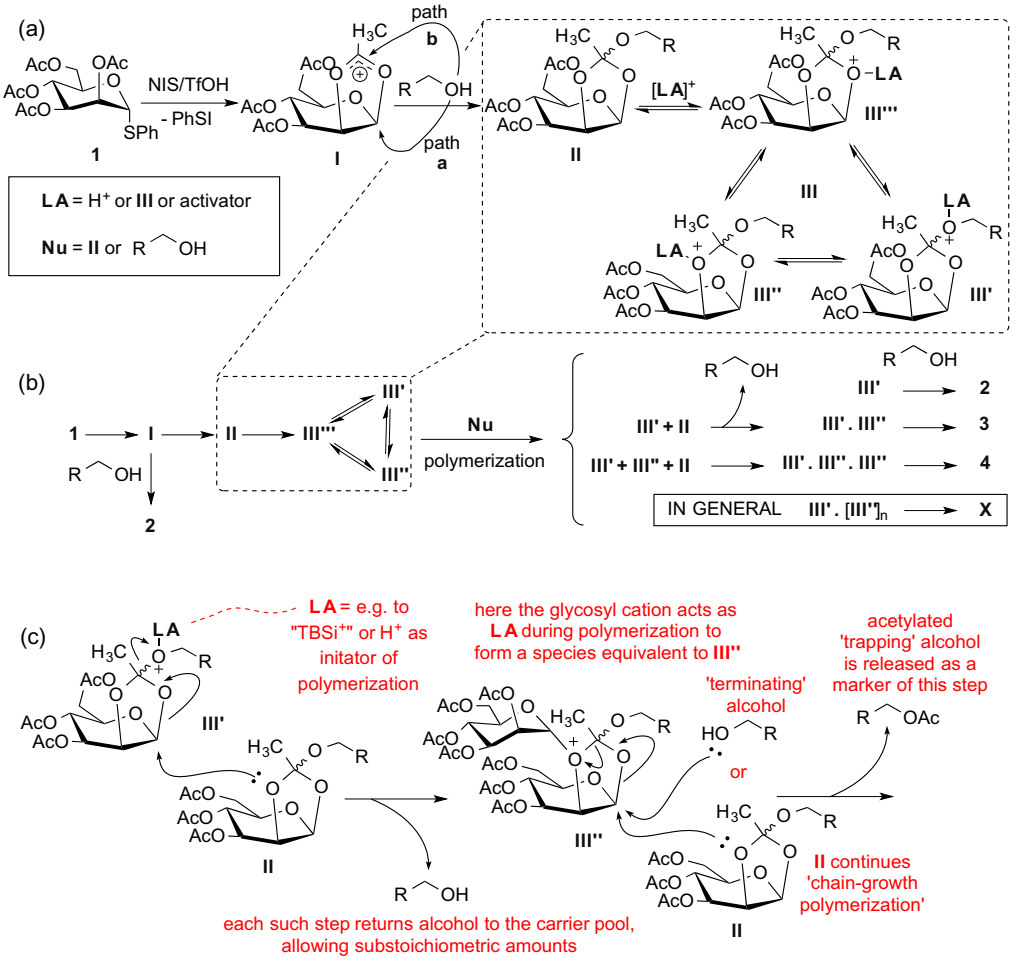


Figure 1. Suggested mechanism for 'chain growth polyglycosylation' leading to the formation of the mannoside oligomers from fully acetylated thioglycoside donor **1**. (a) The initiation and the stabilization of key ambident intermediate **III**; (b) the overall polyglycosylation manifold leading to oligosaccharides (dotted box corresponds to dotted box in panel (a)); (c) an example of one polymerization cycle. **LA** = Lewis acid; **Nu** = nucleophile.

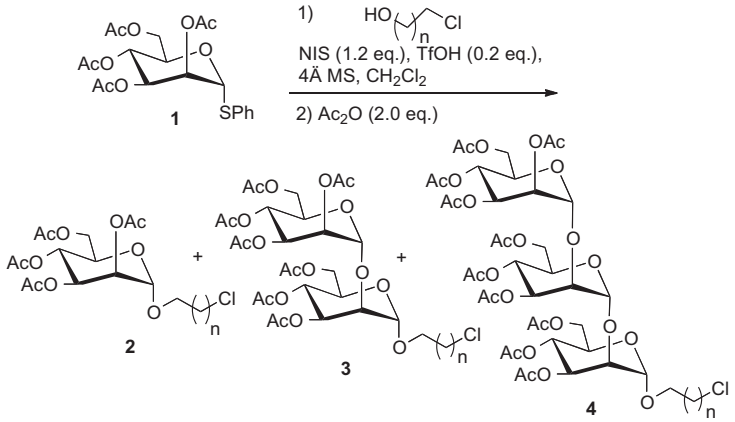
produce 2-OH products[28] or transacetylated acceptors alcohols.[29] We reasoned that this implied orthoester formation and that this donor was therefore be a putative source of the key intermediates needed for the suggested approach. As expected, glycosylations with an excess of primary alcohol acceptor (2 equiv), such as typically used for discrete monomannoside syntheses, gave good yields of the monosaccharide (Table 1). However, we were delighted to see that glycosylation by thioglycoside **1** of functionalized alkyl alcohols (ROH) under varied NIS/TfOH activator conditions (Table 1 and Scheme 1) gave not only the expected monosaccharide products but also varying amounts of disaccharide and trisaccharide; this was a vital early sign that the polymerization manifold was potentially accessible.

2.2. Mechanistic analysis of the synthesis of mannoside oligomers

A plausible, mechanistic explanation for the formation of the α-1→2 linkage in oligomers (considered here for mannobiosides) involves an orthoester **II** (Fig. 1). In the case of thioglycoside, intermediate **II** may be formed from an attack of the acceptor alcohol not at the anomeric centre (pathway a) but the central carbon of the initially generated dioxolenium ion **I** (pathway b) (Fig. 1a). This process is then likely reversible as the orthoester **II** can then be attacked by the Lewis acid at the OR group or at O-1 leading to the formation of intermediate **III**, which can collapse back to **II**. If the incoming alcohol attacks the anomeric carbon of **I** or **III** then that leads to the formation of glycoside, for example, monosaccharide **2**. When sufficient stabilization is provided through Lewis acid coordination in intermediate **III**, we posited that a 'chain growth polyglycosylation' (Fig. 1b and c) might be initiated leading to the formation of disaccharide (**3**) and trisaccharide (**4**) *et cetera*. This idea was further supported by the observation by us and others[22,23,30,31] of acetylated acceptor alcohol as a marker of glycoside formation (Fig. 1c).

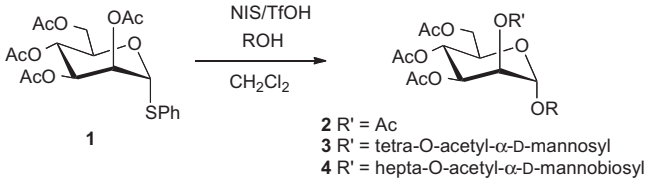
Next, this hypothesis was tested experimentally by the addition of varying equivalents of alcohol (as the putative terminator of polyglycosylation, Fig. 1b and c). This had a direct and marked effect on the formation of increased amount of mannoside oligomers (Table 1). Polyglycosylation would be terminated by the addition of such an acceptor alcohol to the anomeric centre of the intermediate **III**″, thereby eliminating the acetylated alcohol. Alternatively, this intermediate can be attacked by another orthoester **II** leading to the formation of higher mannosides (e.g., trisaccharide product **4**) through a polymerization that continues to 'grow'. When the acceptor alcohol employed in the reaction was reduced this second path is relatively favoured. Distributions obtained were also consistent with this mechanism: even with majority di- or tri-saccharide as product, traces of tetramannoside were also observed by MS when less than stoichiometric quantities of acceptor alcohol were employed for the glycosylation reaction. The rate of each individual pathway is directly determined by the amounts

Table 1
Systematic study with different chloroalcohols and temperatures[a]



Entry	ROH[a]	Equivalent	*T* (°C)	2 (%)	3 (%)	4 (%)
1	**a** *n* = 1	2.0	25	87	n.d.	n.d.
2		1.05	4	80	13	n.d.
3	**b** *n* = 2	2.0	25	98	n.d.	n.d.
4		2.0	4	91	<5	n.d.
5		1.05	4	43	48	n.d.
6		1.05	−2	25	69	n.d.
7	**c** *n* = 4	2.0	25	97	n.d.	n.d.
8		2.0	4	89	10	n.d.
9		1.05	−2	20	70	<5
10		0.8[b]	−2	<5	51	20
11		0.5[b]	−2	<5	31	26[c]
12		0.3[b]	−2	<5	22	36[c]

[a] All reactions performed with 1 mmol of **1**. Yields are calculated based on glycosyl donor 'feedstock' **1** not on alcohol acceptor.
[b] 5–15% tetra-*O*-acetyl-1-deoxy-*N*-succinimido-α-D-mannopyranoside isolated.
[c] Small amount of tetramannoside **5c** also observed in mass spectrum; n.d. not detected.



Scheme 1. 'Polyglycosylation' giving direct access to mannoside oligomers.

Table 2
Influence of the Lewis acid on reactions with 5-chloropentanol[a]

Entry	Lewis acid	2c (%)	3c (%)	4c (%)
1	TfOH	25	75	<5%
2	TMSOTf	10	75	<5%
3	TIPSOTf	20	70	<5%
4	TBDMSOTf	5	80	10%
5	$BF_3{\cdot}Et_2O$[b]	96	3	n.d.

[a] With 1.05 equiv of 5-chloropentanol at −2 °C in CH_2Cl_2 and 1 mmol of **1**, 0.2 equiv of Lewis acid, 1.2 equiv of NIS and addition of Ac_2O after 1 h.
[b] 1.1 equiv used.

of acceptor alcohol and key components and conditions such as Lewis acid, the coordination properties of Lewis acid and the reaction temperatures.

2.3. Variation of conditions

Following this initial survey that was supportive of the proposed manifold, more detailed studies employed versatile chloroalcohols which would allow a variety of subsequent modifications through divergent elaboration (e.g., azide introduction,[32] elimination,[33] or displacement by thiols). Furthermore, they allowed investigation of the influence of chain length (ethyl, propyl and pentyl) which directly affects the electronic properties of the attacking alcohol. All reactions were performed on a millimolar scale in dry dichloromethane and in the presence of molecular sieves. The addition of acetic anhydride after 1 h served to quench and to acetylate any residual acceptor alcohols which co-eluted with the products during purification; additionally this served to reprotect any side products formed by unwanted deacetylation. Table 1 depicts the results of a first study using three alcohols of various chain lengths and varying temperatures. The use of 2.0 equiv clearly favoured formation of the monosaccharides **2** and only at lower temperatures (4 °C) small amounts of disaccharides **3** were found (entries 4, 8). The use of reduced, 1.05 equivalents of alcohol led to predominant formation of **3**. Moreover, a further decrease in temperature to −2 °C (entries 6, 9) gave an even higher ratio in favour of mannobiosides with formation of small quantities of trimannosides **4**. Further reducing the equivalent of alcohol (entries 10, 11, 12) further increased the amount of trimannoside formation and in some cases tetramannosides **5** were also observed (entries 11, 12).

These results with different alcohol equivalents support the existence of a mechanistic pathway generating the intermediates of **III** (**III**′ or **III**″) which react rapidly with alternative nucleophiles (e.g., alcohol or nucleophile). Thus, the terminating alcohol plays an important role: propyl and longer alcohols seem to be of the minimum length required to achieve significant mannobioside formation. This can be attributed to a reduced nucleophilicity,

consistent with the eventual competition by even the poor nucleophile succinimide at lower levels of alcohol (Table 1).

2.4. Variation of activator

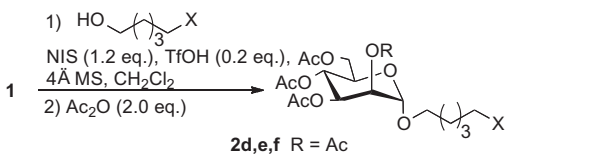
To investigate the influence of the coordinating initiator (Fig. 1c), various Lewis acids were also tested. This also tested the hypothesis of a necessary coordinating effect of a stabilizing counterion. Four triflate-based Lewis acids and $BF_3{\cdot}Et_2O$—the commonly used Lewis acids for activation of *N*-iodosuccinimide[32]—were tested under identical conditions with chloropentanol as terminating nucleophile. As shown in Table 2, the use of TBDMSOTf gave increased polymerization over the other triflate-based acids; allowing the isolation of a small amount (10%) of the mannotrioside **4c**. This may be a result of bulk allowing greater lifetime of all of the intermediates of type **III** or may reflect differential reactivity of intermediates **III′** or **III″**; for example, greater bulk of **LA** (Fig. 1c) might relatively favour initiating reaction of **III′**.

The direct involvement of any silyl electrophile remains unclear; no intermediate 2-*O*-silyl species were isolated or detected in the reaction solutions by mass spectrometry. The use of $BF_3{\cdot}Et_2O$ led almost exclusively to formation of monosaccharide **2c**. Even at lower temperatures no disaccharide formation was found (data not shown), hence, its use would prevent polymerization if monosaccharides were desired targets. Again this favouring of directly productive glycosylation without polymerization may reflect the accessibility of BF_3 as a Lewis acid to **II** and hence greater relative reactivity of **III″** where the less accessible O-2 is coordinated.

2.5. Synthesis of mannoside oligomers equipped with diverse linkers

Finally, to further test the suitability of this synthetic approach for preparing diverse mannooligosaccharides the reactions were repeated with nitrogen-containing linkers (Table 3). Linkers containing amines or masked amines, such as these, find widespread use in immobilization to solid surfaces and conjugation to biomolecules. An efficient approach to higher mannosides with these linkers would provide rapid access to these biologically-relevant glycans, for use in for example, immobilization to array surfaces,[34] preparation of glycoprotein conjugates,[19,35] and affinity-based chromatography for identifying mannose-binding proteins. The successful outcome of these reactions confirmed the versatility of this approach. The reactions with the Cbz- and Boc-protected aminopentanols gave mannobiosides **3d**, **e** as the major products and traces of trisaccharides **4d**, **e** were only detected by mass spectrometric analysis. Azidopentanol gave only 15% of the monosaccharide **2f** and 17% mannotrioside **4f** in addition to the major mannobioside **3f**.

Table 3
Synthesis of mannobiosides with different masked-amine linkers[a]



2d,e,f R = Ac
3d,e,f R = tetra-O-acetyl-α-D-mannosyl
4d,e,f R = hepta-O-acetyl-α-D-mannobiosyl

Entry	Alcohol	2 (%)	3 (%)	4 (%)
1	**d** X = NHCbz	23	65	<5%
2	**e** X = NHBoc	10	75	<5%
3	**f** X = N_3	15	41	17

[a] With 1.05 equiv of 5-chloropentanol at −2 °C in CH_2Cl_2 and 1 mmol of **1**.

3. Conclusions

We have been able to develop a direct protocol leading to mannobiosides and higher oligosaccharides from the acetylated thiomannoside **1**. We suggest a chain-growth mechanism that involves polyglycosylation via an orthoester intermediate and associated Lewis acid-activated species, which are generated during glycosylations with 2-*O*-acetyl protecting groups, reacting in an intermolecular fashion to generate 1→2 linkages. This process is dependent on the type and molar equivalents of the alcohol, the Lewis acid and reaction temperatures. The minimum length acceptors requirement for efficient disaccharide formation are propyl alcohols as 'terminating acceptors' and the reaction seems to be flexible in the nature of functional groups that these alcohols may carry, thus allowing direct installation of 'linkers'. The use of triflate-based Lewis acids as promoters/initiators allows this polyglycosylation whereas $BF_3{\cdot}Et_2O$ can be used to avoid it. Initial results suggest that this protocol allows selective reaction to the corresponding higher mannosides by varying the equivalents of the terminating alcohol and lowering reaction temperatures.

The results we have obtained are consistent with a mechanism (Fig. 1) that, to the best of our knowledge, has not previously been readily exploited in preparative glycosylation chemistry: chain-growth polyglycosylation that proceeds from the non-reducing to reducing (N-to-R) terminus. Whilst other elegant strategies[36–40] for polyglycosides have been explored using anhydrosugars and glycosyl halides, these have relied upon direct step-growth polymerization, which as a general polymerization strategy tends to allow less control and in the case of some polyglycosylations requires extreme conditions of heat or vacuum. As well as potentially allowing greater chain control, the resulting termination by a derivatizable group is an advantage, as for chain growth polymerizations generally. We also suggest that the many suggested (pre)equilibria are essential in allowing a productive outcome based on an ambident reactivity of orthoester **II** as both nucleophile and electrophile (via **III**); it may also be that this process is selective for one diastereomer of **II** (and **III**) over another.

We did not observe exact stoichiometry between recovered acetylated alcohol, formed as a result of alcohol 'trapping' acetyl (Fig. 1c), and polyglycosylation product as a result of 'termination' by alcohol. Other nucleophiles may therefore also play the role of 'trapper' in the mechanism; indeed in crude product mixtures we do observe putative succinimidyl conjugates. Alternative mechanisms can also be envisaged; for example, in situ generation of intermediates with free OH-2 might also be considered. Interestingly, the formation of equivalent species and resulting transglycosylations has also been reported for gluco- and galacto-sides[41] that might also be interpreted through such a mechanism. Hence, the potential for use of this process might be even more widespread. We are therefore currently investigating if such a poly(self-)glycosylation process can also be developed for other sugars, although the mannobiosides are by far the most biologically-relevant 1→2 linked disaccharides. Another possibility is to use in situ generated terminating sugar acceptors for a one-step synthesis delivering mixed oligosaccharides bearing an alternative residue in the reducing terminus. With careful control, even 'block polyglycosylations' giving runs of one sugar type and then another type might be considered. Strategies towards these ends and towards control of higher oligosaccharides are also currently being investigated.

4. Experimental section

4.1. General methods

Chemicals were purchased from Sigma–Aldrich and used as supplied, unless otherwise stated. Anhydrous solvents were

purchased from Fluka or Acros. All other solvents were used as supplied (analytical or HPLC grade) without prior purification. 'Petrol' refers to the fraction of light petroleum ether boiling in the range of 40–60 °C. All reactions using anhydrous conditions were performed using flame-dried apparatus under an atmosphere of argon or nitrogen. Proton nuclear magnetic resonance (^{1}H NMR) spectra were recorded on a Mercury VX 400 (400 MHz) spectrometer, as indicated. Carbon nuclear magnetic resonance (^{13}C NMR) spectra were recorded on a Varian Mercury VX 400 (100.6 MHz) spectrometer, as indicated. NMR spectra were fully assigned using COSY, HSQC and HMBC correlation experiments. All chemical shifts are quoted on δ scale in ppm using residual solvent as the internal standard. Coupling constants (J) are reported in Hz. Low resolution mass spectra (LRMS) were recorded on a Waters Micromass LCT premier TOF spectrometer using electrospray ionization (ESI-MS). High resolution mass spectra (HRMS) were recorded on a Bruker MicroTOF ESI mass spectrometer. Thin layer chromatography (TLC) was carried out using Merck aluminium backed sheets coated with 60F254 silica gel. Visualization of the silica gel plates was achieved using a UV lamp (λ_{max} = 254 nm) and/or by dipping the plate in ceric ammonium molybdate (CAM) stain followed by heating.

4.2. General procedures

4.2.1. General procedures for glycosylations of monosaccharides (GP1)

The thiophenylmannoside **1** (440 mg, 1.00 mmol, 1.0 equiv) and the corresponding alcohol (2.0 equiv) in CH_2Cl_2 (5 mL) were stirred at 25 °C in the presence of 4 Å MS (500 mg) for 30 min. *N*-iodosuccinimide (270 mg, 1.2 mmol, 1.2 equiv) and trifluoromethane sulfonic acid (30.0 mg, 17.7 mL, 200 mmol, 0.2 equiv) were slowly added and stirring continued for 1 h. In situ acetylation of free alcohols is achieved by addition of acetic anhydride (0.2–0.3 mL, 2.0–3.0 equiv). After 30 min the molecular sieves were filtered using a pad of Celite® and then washed with EtOAc. Filtrate was then transferred into a separatory funnel and washed with ice-cold 10% $Na_2S_2O_5$ solution (20 mL), phases separated and the water layer reextracted with additional EtOAc (50 mL). The combined organic layers were washed with ice-cold brine (40 mL), dried (Na_2SO_4) and concentrated under vacuum. Column chromatography (20 g silica, 3:1→2:1 petrol/EtOAc) gave the monosaccharides as colourless materials.

4.2.2. General procedure for glycosylation towards mannobiosides (GP2)

The thiophenylmannoside **1** (440 mg, 1.00 mmol, 1.0 equiv) and the corresponding alcohol (1.05 equiv) in CH_2Cl_2 (5 mL) were stirred at 25 °C in the presence of 4 Å MS (500 mg) for 30 min. After cooling to −2 °C with an ice/NaCl bath *N*-iodosuccinimide (270 mg, 1.2 mmol, 1.2 equiv) and trifluoromethanesulfonic acid (30.0 mg, 17.7 mL, 200 mmol, 0.2 equiv) were slowly added and stirring continued at this temperature for 1 h. The reaction mixture was allowed to warm to 25 °C and then acetic anhydride (0.2–0.3 mL, 2.0–3.0 equiv) added. Work-up was analogous to GP1 and column chromatography was performed with a gradient 3:1 → 1:1 petrol/EtOAc to yield the mono, di and trisaccharides as colourless materials.

4.3. Synthesis and characterization

4.3.1. 2-Chloroethyl 2,3,4,6-tetra-*O*-acetyl-α-D-mannopyranoside (2a)

The chloride was prepared according to GP1 giving the title (**2a**) compound as colourless solid (332 mg, 0.864 mmol, 87%). TLC: R_f = 0.47 (60% EtOAc/petrol); $[\alpha]_D^{20}$ +44.5 (c 0.65, $CHCl_3$); ^{1}H NMR (400 MHz, $CDCl_3$) δ ppm 5.34 (dd, J = 10.0, 3.3 Hz, 1H, H-3), 5.33–5.25 (m, 2H, H-2, H-4), 4.86 (d, J = 1.0 Hz, 1H, H-1), 4.26 (dd, J = 12.2, 5.5 Hz, 1H, H-6$_b$), 4.12 (m, 2H, H-5, H-6$_a$), 3.91 (td, J = 11.2, 5.5 Hz, 1 H, H-1$_b$′), 3.81 (td, J = 10.9, 5.3 Hz, 1H, H-1$_a$′), 3.67 (t, J = 5.6 Hz, 2 H, H-2′), 2.15, 2.09, 2.04, 1.99 (4 × s, 12H, 4 × $COCH_3$); ^{13}C NMR (100 MHz, $CDCl_3$) δ ppm 170.65, 170.06, 169.81, 169.73 (4 × $COCH_3$), 97.79 (C-1), 69.41 (C-2), 68.82 (C-3), 68.90 (C-1′), 68.60 (C-5), 66.00 (C-4), 62.39 (C-6), 42.35 (C-2′), 20.64, 20.67, 20.70, 20.84 (4 × $COCH_3$); HRMS (ESI): m/z = 433.08624 $[M+Na]^+$; calculated $C_{16}H_{23}ClNaO_{10}$ (433.08720).

4.3.2. 2-Chloroethyl 2,3,4,6-tetra-*O*-acetyl-α-D-mannopyranosyl-(1→2)-3,4,6-tri-*O*-acetyl-α-D-mannopyranoside (3a)

The synthesis was performed according to GP2 yielding the monosaccharide **2a** (330.1 mg, 0.804 mmol, 80%), the disaccharide **3a** (44.2 mg, 63.2 μmol, 13%) as colourless solids. Data for **3a**, TLC: R_f = 0.25 (60% EtOAc/petrol); $[\alpha]_D^{20}$ +22.7 (c 1.90, $CHCl_3$); ^{1}H NMR (400 MHz, $CDCl_3$) δ ppm 5.38 (dd, J = 10.0, 3.3 Hz, 1H, H-3′), 5.31 (t, J = 9.9 Hz, 1H, H-4), 5.29–5.22 (m, 3H, H-3, H-2′, H-4′), 5.00 (d, J = 1.3 Hz, 1H, H-1), 4.92 (d, J = 1.1 Hz, H-1′), 4.25–4.09 (m, 5 H, H-5, H-6$_{a,b}$, H-6$_{a',b'}$), 4.07 (dd, J = 4.8, 1.3 Hz, 1H, H-2), 4.01 (ddd, J = 8.7, 3.9, 2.3 Hz, 1H, H-5), 3.93 (td, J = 11.1, 5.5 Hz, 1H, H-1$_b$″), 3.78 (td, J = 11.0, 5.4 Hz, 1H, H-1$_a$″), 3.66 (t, J = 5.5 Hz, 2H, H-2″), 2.13, 2.12, 2.07, 2.06, 2.03, 2.02, 1.99 (7 × s, 21H, 7 × $COCH_3$); ^{13}C NMR (100 MHz, $CDCl_3$) δ ppm 169.53, 169.88, 169.85, 169.92, 170.41, 170.67, 170.91 (7 × $COCH_3$), 99.05 (C-1′), 98.55 (C-1), 76.59 (C-2), 70.14 (C-3), 69.69 (C-2′), 69.14, 68.96 (C-5, C-5′), 68.49 (C-1″), 68.38 (C-3′), 66.41, 66.11 (C-4, C-4′), 62.17, 62.62 (C-6, C-6′), 42.56 (C-2″), 20.85, 20.74, 20.71, 20.66, 20.61 (7 × $COCH_3$); HRMS (ESI): m/z = 721.16945 $[M+Na]^+$; calculated $C_{28}H_{39}ClNaO_{18}$ (721.17171).

4.3.3. 3-Chloropropyl 2,3,4,6-tetra-*O*-acetyl-α-D-mannopyranoside (2b)

The chloride **2b** was prepared according to GP1 yielding the title compound as colourless oil (415 mg, 0.977 mmol, 98%). TLC: R_f = 0.49 (60% EtOAc/petrol); $[\alpha]_D^{20}$ +48.7 (c 1.65, $CHCl_3$); ^{1}H NMR (400 MHz, $CDCl_3$) δ ppm 5.29 (dd, J = 9.9, 3.5 Hz, 1H, H-3), 5.27–5.20 (m, 2H, H-2, H-4), 4.81 (d, J = 1.0 Hz, 1H, H-1), 4.26 (dd, J = 12.2, 5.4 Hz, 1H, H-6$_b$), 4.11 (dd, J = 12.4, 2.3 Hz, 1H, H-6$_a$), 3.97 (m, 1H, H-1$_b$′), 3.90 (ddd, J = 9.9, 7.3, 5.1 Hz, 1H, H-5), 3.70–3.60 (m, 2H, H-3′), 3.57 (td, J = 10.1, 5.6 Hz, 1H, H-1$_a$′), 2.06–2.02 (m, 2H, H-2′), 2.14, 2.09, 2.03, 1.98 (4 × s, 12H, 4 × $COCH_3$); ^{13}C NMR (100 MHz, $CDCl_3$) δ ppm 170.65, 170.04, 169.91, 169.76 (4 × $COCH_3$), 97.57 (C-1), 69.44 (C-2), 69.02 (C-3), 68.61 (C-5), 66.02 (C-4), 64.39 (C-1′), 62.39 (C-6), 41.41 (C-3′), 31.88 (C-2′), 20.84, 20.69, 20.66 (4 × $COCH_3$); HRMS (ESI): m/z = 447.10169 $[M+Na]^+$; calculated $C_{17}H_{25}ClNaO_{10}$ (447.10285).

4.3.4. 3-Chloropropyl 2,3,4,6-tetra-*O*-acetyl-α-D-mannopyranosyl-(1→2)-3,4,6-tri-*O*-acetyl-α-D-mannopyranoside (3b)

The mannobioside **3b** was prepared following GP2 to result in a colourless solid (246 mg, 0.345 mmol, 69%) along with the monosaccharide **2b** (104 mg, 0.245 mmol, 25%). Data for **3b**, TLC: R_f = 0.35 (65% EtOAc/petrol); $[\alpha]_D^{20}$ +32.5 (c 0.95, $CHCl_3$); ^{1}H NMR (400 MHz, $CDCl_3$) δ ppm 5.40 (dd, J = 10.0, 3.3 Hz, 1H, H-3′), 5.32 (t, J = 9.9 Hz, 1H, H-4), 5.30–5.22 (m, 3H, H-3, H-2′, H-4′), 4.95 (d, J = 1.0 Hz, 1H, H-1), 4.92 (d, J = 1.3 Hz, 1 H, H-1′), 4.07–4.27 (m, 5H, H-5, H-6$_{a,b}$, H-6$_{a',b'}$), 4.02 (m, 1H, H-2), 3.97–3.86 (m, 2H, H-5, H-1$_b$″), 3.65 (t, J = 6.1 Hz, 2H, H-3″), 3.58 (td, J = 9.9, 5.7 Hz, 2H, H-1$_a$″), 2.15, 2.13, 2.08, 2.04, 2.00 (7 × s, 21H, 7 × $COCH_3$), 2.05 (m, 2H, H-2″); ^{13}C NMR (100 MHz, $CDCl_3$) δ ppm 170.98, 170.56, 169.81, 169.74, 169.43 (7 × $COCH_3$), 99.15 (C-1′), 98.31 (C-1), 76.96 (C-2), 70.24 (C-3), 69.73 (C-2′), 69.16, 68.70 (C-5, C-5′), 68.33 (C-3′), 66.37, 66.09 (C-4, C-4′), 64.39 (C-1″), 62.56, 62.14 (C-6, C-6′), 41.42 (C-3″), 31.92 (C-2″), 20.86, 20.72, 20.71, 20.66,

20.63 (7 × $COCH_3$),. HRMS (ESI): m/z = 735.18477 $[M+Na]^+$; calculated $C_{29}H_{41}ClNaO_{18}$ (735.18736).

4.3.5. 5-Chloropentyl 2,3,4,6-tetra-*O*-acetyl-α-D-mannopyranoside (2c)

The synthesis was performed according to GP1 yielding the monosaccharide **2c** (438 mg, 0.968 mmol, 97%) as colourless oil. TLC: R_f = 0.55 (60% EtOAc/petrol); $[\alpha]_D^{20}$ + 41.5 (*c* 1.20, $CHCl_3$); ^1H NMR (400 MHz, $CDCl_3$) δ ppm 5.33 (dd, *J* = 10.0, 3.3 Hz, 1H, H-3), 5.26 (t, *J* = 9.9 Hz, 1H, H-4), 5.22 (dd, *J* = 3.2, 1.6 Hz, 1H, H-2), 4.79 (d, *J* = 1.2 Hz, 1H, H-1), 4.27 (dd, *J* = 12.2, 5.2 Hz, 1H, H-6_b), 4.09 (dd, *J* = 12.2, 2.1 Hz, 1H, H-6_a), 3.97 (ddd, *J* = 7.3, 5.1, 2.0 Hz, 1H, H-5), 3.69 (td, *J* = 9.7, 6.8 Hz, 1H, H-$1_{b'}$), 3.54 (t, *J* = 6.6 Hz, 2H, H-5′), 3.45 (td, *J* = 9.7, 6.3 Hz, 1 H, H-$1_{a'}$), 2.14, 2.09, 2.03, 1.98 (4 × s, 12H, 4 × $COCH_3$), 1.80 (td, *J* = 13.9, 6.8 Hz, 2H, H-4′), 1.62 (qd, *J* = 13.6, 7.2, 6.8 Hz, 2H, H-2′), 1.51 (m, 2H, H-3′); ^{13}C NMR (100 MHz, $CDCl_3$) δ ppm 170.65, 170.11, 169.93, 169.77 (4 × *C*OCH_3), 97.54 (C-1), 69.62 (C-2), 69.06 (C-3), 68.43 (C-5), 68.10 (C-1′), 66.20 (C-4), 62.50 (C-6), 44.74 (C-5′), 32.19 (C-4′), 28.51 (C-2′), 23.43(C-3′), 20.87, 20.71, 20.67 (4 × *C*OCH_3); HRMS (ESI): m/z = 475.13304 $[M+Na]^+$; calculated $C_{19}H_{29}ClNaO_{10}$ (475.13415).

4.3.6. 5-Chloropentyl 2,3,4,6-tetra-*O*-acetyl-α-D-mannopyranosyl-(1→2)-3,4,6-tri-*O*-acetyl-α-D-mannopyranoside (3c)

The mannobioside was prepared following GP2 to give two colourless syrups of the mannobioside **3c** (259 mg, 0.350 mmol, 70%) along with the corresponding monosaccharide **2c** (90 mg, 0.2 mmol, 20%). Data for **3c**, TLC: R_f = 0.30 (50% EtOAc/petrol); $[\alpha]_D^{20}$ +30.8 (*c* 1.60, $CHCl_3$); ^1H NMR (400 MHz, $CDCl_3$) δ ppm 5.39 (dd, *J* = 10.0, 3.3 Hz, 1H, H-3′), 5.26 (t, *J* = 9.8 Hz, 1H, H-4), 5.22–5.28 (m, 3H, H-3, H-2′, H-4′), 4.91 (s_{br}, 2H, H-1, H-1′), 4.06–4.25 (m, 5H, H-5, H-$6_{a,b}$, H-$6_{a',b'}$), 4.00 (dd, *J* = 2.4, 1.9 Hz, 1H, H-2), 3.89 (ddd, *J* = 9.2, 3.9, 2.4 Hz, 1H, H-5), 3.69 (td, *J* = 9.5, 6.5 Hz, 1H, H-$1_{b''}$), 3.53 (t, *J* = 6.6 Hz, 2H, H-5″), 3.44 (td, *J* = 9.8, 6.4 Hz, 1H, H-$1_{a''}$), 2.13, 2.12, 2.06, 2.02, 2.01, 1.99 (6 × s, 21H, 7 × $COCH_3$), 1.78 (m, 2H, H-4″), 1.62 (qd, *J* = 13.1, 6.5 Hz, 2H, H-2″), 1.51 (m, 2H, H-3″); ^{13}C NMR (100 MHz, $CDCl_3$) δ ppm 170.86, 170.43, 169.81, 169.73, 169.45 (7 × *C*OCH_3), 99.11 (C-1′), 98.23 (C-1), 77.06 (C-2), 70.26 (C-3), 69.71 (C-2′), 69.07 (C-5′), 68.50 (C-5), 68.33 (C-3′), 68.07 (C-1″), 66.18, 66.34 (C-4, C-4′), 62.19, 62.50 (C-6, C-6′), 44.70 (C-5″), 32.15 (C-4″), 28.58 (C-2″), 23.44 (C-3″), 20.82, 20.69, 20.62 (7 × *C*OCH_3); HRMS (ESI): m/z = 763.21603 $[M+Na]^+$; calculated $C_{31}H_{45}ClNaO_{18}$ (763.21866).

4.3.7. 5-Chloropentyl 2,3,4,6-tetra-*O*-acetyl-α-D-mannopyranosyl-(1→2)-3,4,6-tri-*O*-acetyl-α-D-mannopyranosyl-(1→2)-3,4,6-tri-*O*-acetyl-α-D-mannopyranoside (4c)

The mannobioside was prepared following GP2 (0.8 equiv of 5-chloropentanol) to give two colourless syrups of the mannobioside **3c** (189 mg, 0.254 mmol, 51%) along with the corresponding mannose trisaccharide **4c** (62 mg, 60 μmol, 20%). Data for **4c**, TLC: R_f = 0.18 (50% EtOAc/petrol); $[\alpha]_D^{20}$ +28.8 (*c* 0.80, $CHCl_3$); ^1H NMR (400 MHz, $CDCl_3$): δ ppm 5.40 (dd, *J* = 9.8, 3.3 Hz, 1H, H-3″), 5.34–5.25 (m, 6H, H-3, H-3′, H-2″, H-4, H-4′, H-4″), 5.11 (d, *J* = 1.3 Hz, 1H, H-1″), 4.95 (s_{br}, 2H, H-1, H-1′), 4.23 (dd, *J* = 12.0, 4.3 Hz, 2H, H-$6_{a'}$, H-$6_{a''}$), 4.20–4.10 (m, 7H, H-2′, H-5′, H-5″, H-$6_{a,b}$, H-$6_{b'}$, H-$6_{b''}$), 4.02 (s_{br}, 1H, H-2), 3.92 (s_{br}, 1H, H-5), 3.72 (td, *J* = 9.6, 6.6 Hz, 1H, H-$1_{a'''}$), 3.55 (t, *J* = 6.6 Hz, 2H, H-5‴), 3.45 (td, *J* = 9.6, 6.3 Hz, 1H, H-$1_{b'''}$), 2.15, 2.13, 2.12, 2.09, 2.07, 2.06, 2.04, 2.03, 2.00 (9s, 30 H, 10 × $COCH_3$), 1.85–1.78 (m, 2H, H-2‴), 1.68–1.60 (m, 2H, H-4‴), 1.55–1.46 (m, 2H, H-3‴); ^{13}C NMR (100 MHz, $CDCl_3$) δ ppm 170.91, 170.75, 170.16, 170.05, 169.81, 169.56, 169.45 (10 × *C*OCH_3), 99.81 (C-1″), 99.40 (C-1′), 98.23 (C-1), 77.30 (C-2), 76.71 (C-5″), 70.46 (C-3), 69.71 (C-2′), 69.63 (C-2″), 69.41 (C-3″), 69.28 (C-5′), 68.60 (C-5), 68.43 (C-3′), 68.11 (C-1‴), 66.48, 66.34, 66.25 (C-4″, C-4′, C-4), 62.51, 62.25, 62.11 (C-6″, C-6′, C-6), 44.87 (C-5‴), 32.15 (C-4‴), 29.58 (C-2‴), 23.44 (C-3‴), 20.88, 20.71, 20.63 (10 × $COCH_3$); HRMS (ESI): m/z = 1051.29976 $[M+Na]^+$; calculated $C_{43}H_{61}ClNaO_{26}$ (1051.30318).

4.3.8. 5-(Benzyloxycarbonylamino)pentyl 2,3,4,6-tetra-*O*-acetyl-α-D-mannopyranoside (2d) & 5-(benzyloxycarbonylamino) pentyl 2,3,4,6-tetra-*O*-acetyl-α-D-mannopyranosyl-(1→2)-3,4,6-tri-*O*-acetyl-α-D-mannopyranoside (3d)

The mannobioside was prepared following GP2 to give two colourless syrups of the corresponding monosaccharide **2d** (130 mg, 0.229 mmol, 23%) along with mannobioside **3d** (277 mg, 0.324 mmol, 65%). Data for **2d**, TLC: R_f = 0.43 (50% EtOAc/Petrol); $[\alpha]_D^{20}$ +32.2 (*c* 1.05, $CHCl_3$); ^1H NMR (400 MHz, $CDCl_3$) δ ppm 7.35–7.25 (m, 5H, H-Ph), 5.34 (dd, *J* = 10.0, 3.3 Hz, 1H, H-3), 5.27 (t, *J* = 9.9 Hz, 1H, H-4), 5.23 (dd, *J* = 3.3, 1.7 Hz, 1H, H-2), 5.10 (s, 2H, CH_2Ph), 4.86 (s_{br}, 1H, NH), 4.80 (d, *J* = 1.3 Hz, 1H, H-1), 4.29 (dd, *J* = 12.2, 5.3 Hz, 1H, H-6_b), 4.12 (m, 1H, H-6_a), 3.98 (ddd, *J* = 9.4, 5.2, 2.3 Hz, 1H, H-5), 3.69 (td, *J* = 9.5, 6.3 Hz, 1H, H-$1_{b'}$), 3.45 (td, *J* = 9.5, 6.3 Hz, 1H, H-$1_{a'}$), 3.21 (dd, *J* = 9.2, 6.5 Hz, 2H, H-5′), 2.16, 2.10, 2.04, 1.99 (4s, 12H, 4 × $COCH_3$), 1.70–1.48 (m, 4H, H-2′, H-4′), 1.40 (m, 2H, H-3′); ^{13}C NMR (50 MHz, $CDCl_3$) δ ppm 170.64, 170.02, 169.91, 169.73 (4 × *C*OCH_3), 156.42 (NH*C*O), 136.51, 128.44, 128.07 (*C*-Ph), 97.49 (C-1), 69.67 (C-2), 69.06 (C-3), 68.31 (C-5), 68.22 (C-1′), 66.51 (*C*H_2Ph), 66.27 (C-4), 62.54 (C-6), 40.83 (C-5′), 29.71 (C-4′), 28.86 (C-2′), 23.31(C-3′), 20.86, 20.75, 20.69 (4 × *C*OCH_3); HRMS (ESI) m/z 590.21761 $[M+Na]^+$; calculated $C_{27}H_{37}NNaO_{12}$ 590.22135.

Data for **3d**, TLC: R_f = 0.25 (50% EtOAc/petrol); $[\alpha]_D^{20}$ +24.1 (*c* 1.90, $CHCl_3$); ^1H NMR (400 MHz, $CDCl_3$) δ ppm 7.36–7.27 (m, 5H, H-Ph), 5.39 (dd, *J* = 10.0, 3.3 Hz, 1H, H-3′), 5.34–5.20 (m, 4H, H-3, H-4, H-2′, H-4′), 5.07 (s_{br}, 2H, CH_2Ph), 4.90 (s_{br}, 3H, H-1, H-1′, NH), 4.22–3.98 (m, 6H, H-2, H-5′, H-$6_{a,b}$, H-$6_{a',b'}$), 3.89 (dd, *J* = 5.0, 3.6 Hz, 1H, H-5), 3.67 (td, *J* = 9.3, 6.5 Hz, 1H, H-$1_{b''}$), 3.41 (td, *J* = 8.9, 6.3 Hz, 1H, H-$1_{a''}$), 3.18 (t, *J* = 12.1, 6.4 Hz, 2H, H-5″), 2.13, 2.11, 2.06, 2.02, 2.01, 1.99 (6 × s, 21H, 7 × COCH_3), 1.61 (td, *J* = 13.0, 6.4 Hz, 2H, H-4″), 1.52 (td, *J* = 14.4, 7.3 Hz, 2 H, H-2″), 1.37 (m, 2H, H-3″); ^{13}C NMR (100 MHz, $CDCl_3$) δ ppm 170.91, 170.44, 169.85, 169.78, 169.45 (7 × *C*OCH_3), 156.42 (NH*C*O), 136.51, 128.45, 128.05 (5 × C-Ph), 99.09 (C-1′), 98.16 (C-1), 77.09 (C-2), 70.24 (C-3), 69.69 (C-2′), 69.05 (C-5′), 68.43 (C-5), 68.33 (C-3′), 68.16 (C-1″), 66.33 (C-4, C-4′), 66.21 (*C*H_2Ph), 62.46, 62.20 (C-6, C-6′), 40.82 (C-5″), 29.52 (C-4″), 28.88 (C-2″), 23.35 (C-3″), 20.80, 20.68, 20.61 (7 × CO*C*H_3); HRMS (ESI): m/z = 878.30198 $[M+Na]^+$; calculated $C_{39}H_{53}NNaO_{20}$ (878.30586).

4.3.9. 5-(*tert*-Butoxycarbonylamino)pentyl 2,3,4,6-tetra-*O*-acetyl-α-D-mannopyranoside (2e) & 5-(*tert*-butoxycarbonyl amino)pentyl 2,3,4,6-tetra-*O*-acetyl-α-D-mannopyranosyl-(1→2)-3,4,6-tri-*O*-acetyl-α-D-mannopyranoside (3e)

The mannobioside was prepared following GP2 to give two colourless syrups of the corresponding monosaccharide **2e** (53 mg, 0.100 mmol, 10%) along with mannobioside **3e** (307 mg, 0.374 mmol, 75%). Data for **2e**, TLC: R_f = 0.38 (50% EtOAc/Petrol); $[\alpha]_D^{20}$ +38.3 (*c* 1.20, $CHCl_3$); ^1H NMR (400 MHz, $CDCl_3$) δ ppm 5.31–5.23 (m, 2H, H-2, H-4), 4.80 (d, *J* = 1.1 Hz, 1H, H-1), 4.58 (s_{br}, 1H, NH), 4.28 (dd, *J* = 12.2, 5.2 Hz, 1H, H-6_b), 4.14 (m, 1H, H-6_a), 3.97 (ddd, *J* = 9.3, 5.2, 2.1 Hz, 1H, H-5), 3.68 (td, *J* = 9.7, 6.3 Hz, 1H, H-$1_{b'}$), 3.42 (td, *J* = 9.5, 6.3 Hz, 1H, H-$1_{a'}$), 3.11 (t, *J* = 6.8, 2H, H_2-5′), 2.15, 2.11, 2.03, 1.99 (4s, 12H, 4 × $COCH_3$), 1.55 (m, 2H, H-2′), 1.48 (q, *J* = 7.6 Hz, 2H, H-4′), 1.42 (s_{br}, 9H, C(CH_3)$_3$), 1.41–1.33 (m, 2H, H-3′); ^{13}C NMR (100 MHz, $CDCl_3$) δ ppm 170.52, 170.18, 169.88, 169.55 (4 × *C*OCH_3), 156.22 (NH*C*O), 97.64 (C-1), 79.09 (*C*(CH_3)$_3$), 69.72 (C-2), 69.61 (C-3), 68.40 (C-5), 68.22 (C-1′), 66.24 (C-4), 62.54 (C-6), 40.46 (C-5′), 32.16 (C-2′), 29.79 (C-4′), 28.41 (C(*C*H_3)$_3$), 23.00 (C-3′), 20.86, 20.75, 20.69

(4 × $COCH_3$); HRMS (ESI) m/z 556.23212 $[M+Na]^+$; calculated $C_{24}H_{39}NNaO_{12}$ 556.23700.

Data for **3e**, TLC: R_f = 0.20 (1:1 = EtOAc/petrol); $[\alpha]_D^{20}$ +26.9 (*c* 1.65, $CHCl_3$); ^{1}H NMR (400 MHz, $CDCl_3$): δ = 5.37 (dd, *J* = 10.0, 3.4 Hz, 1H, H-3′), 5.33–5.19 (m, 3 H, H-3, H-4, H-2′, H-4′), 4.92 (s_{br}, 3H, H-1, H-1′, NH), 4.22–3.98 (m, 6H, H-2, H-5, H-$6_{a,b}$, H-$6_{a',b'}$), 3.88 (dd, *J* = 5.1, 3.6 Hz, 1H, H-5), 3.68 (td, *J* = 9.5, 6.3 Hz, 1H, H-1_b″), 3.42 (td, *J* = 9.6, 6.3 Hz, 1H, H-1_a″), 3.11 (t, *J* = 6.8, 2H, H_2-5″), 2.12, 2.10, 2.06, 2.02, 2.01, 1.98 (6 × s, 21H, 7 × $COCH_3$), 1.55 (m, 2H, H-2″), 1.47 (q, *J* = 7.5 Hz, 2H, H-4″), 1.42 (s_{br}, 9H, $C(CH_3)_3$), 1.37 (m, 2H, H-3″); ^{13}C NMR (100 MHz, $CDCl_3$): δ ppm 170.88, 170.42, 169.91, 169.75, 169.43 (7 × $COCH_3$), 156.30 (NHCO), 99.10 (C-1′), 98.22 (C-1), 79.19 ($C(CH_3)_3$), 77.09 (C-2), 70.24 (C-3), 69.74 (C-2′), 69.03 (C-5′), 68.44 (C-5), 68.35 (C-3′), 68.19 (C-1″), 66.32, 66.21 (C-4, C-4′), 62.45, 62.21 (C-6, C-6′), 40.50 (C-5″), 32.16 (C-2″), 29.82 (C-4″), 28.36 ($C(CH_3)_3$), 22,89 (C-3″), 20.80, 20.68, 20.61 (7 × $COCH_3$); HRMS (ESI): m/z = 844.31846 $[M+Na]^+$; Calculated $C_{36}H_{55}NNaO_{20}$ (844.32151).

4.3.10. 5-(Azido)pentyl 2,3,4,6-tetra-*O*-acetyl-α-D-mannopyranoside (2f), 5-(azido)pentyl 2,3,4,6-tetra-*O*-acetyl-α-D-mannopyranosyl-(1→2)-3,4,6-tetra-*O*-acetyl-α-D-mannopyranoside (3f) & 5-(azido)pentyl 2,3,4,6-tetra-*O*-acetyl-α-D-mannopyranosyl-(1→2)-3,4,6-tri-*O*-acetyl-α-D-mannopyranosyl-(1→2)-3,4,6-tri-*O*-acetyl-α-D-mannopyranoside (4f)

The corresponding azido-mannosides were prepared following GP2 to give three colourless syrups of the monosaccharide **2f** (69 mg, 0.15 mmol, 15%), mannobioside **3f** (152 mg, 0.203 mmol, 41%) along with the corresponding mannose trisaccharide **4f** (51 mg, 50 µmol, 17%). Data for **2f**, TLC: R_f = 0.52 (50 % EtOAc/Petrol); $[\alpha]_D^{20}$ +38.7 (*c* 1.10, $CHCl_3$); ^{1}H NMR (400 MHz, $CDCl_3$) δ ppm 5.48 (d, *J* = 2.5 Hz, 1H, H-1), 5.31 (dd, *J* = 10.4, 9.1 Hz, 1H, H-4), 5.15 (dd, *J* = 9.9, 3.9 Hz, 1H, H-3), 4.60 (dd, *J* = 3.9, 2.6 Hz, 1H, H-2), 4.24 (dd, *J* = 12.1, 4.9 Hz, 1H, H-6_a), 4.15 (dd, *J* = 12.1, 2.6 Hz, 1H, H-6_b), 3.69 (ddd, *J* = 9.4, 4.9, 2.6 Hz, 1H, H-5), 3.56–3.44 (m, 2 H, H-1′), 3.27 (t, *J* = 6.8 Hz, 1H, H-5′), 2.13, 2.08, 2.06 (3 × s, 4 × $COCH_3$), 1.66–1.54 (m, 4H, H-2′, H-4′), 1.49–1.38 (m, 2H, H-3′); ^{13}C NMR (100 MHz, $CDCl_3$) δ ppm 170.65, 170.01, 169.84, 169.68 (4 × $COCH_3$), 97.31 (C-1), 76.35 (C-4), 71.31 (C-2), 70.64 (C-5), 70.52 (C-3), 65.45 (C-6), 62.52 (C-1′), 51.34 (C-5′), 28.57 (C-2′), 24.68 (C-4′), 23.35 (C-3′), 20.83, 20.72, 20.64 (4 × $COCH_3$); HRMS (ESI) m/z = 482.17189 $[M+Na]^+$; calculated $C_{19}H_{29}N_3NaO_{10}$ 482.17506.

Data for **3f**, TLC: R_f = 0.42 (50% EtOAc/Petrol); $[\alpha]_D^{20}$ +28.9 (*c* 2.20, $CHCl_3$); ^{1}H NMR (400 MHz, $CDCl_3$) δ ppm 5.41 (dd, *J* = 10.0, 3.3 Hz, 1H, H-3′), 5.34 (t, *J* = 9.6 Hz, 1H, H-4), 5.32–5.25 (m, 3H, H-3, H-2′, H-4′), 4.93 (s_{br}, 2H, H-1, H-1′), 4.25(dd, *J* = 10.8, 4.8 Hz, 2H, H-6′$_{a,b}$), 4.19–4.14 (m, 2H, H-$6_{a,b}$), 4.11 (dd, *J* = 7.0, 2.5 Hz, 1H, H-5′), 4.03 (t, *J* = 2.3 Hz, 1H, H-2), 3.91 (ddd, *J* = 9.3, 3.8, 2.2 Hz, 1H, H-5), 3.73 (td, *J* = 9.6, 6.3 Hz, 1H, H-1_a″), 3.45 (td, *J* = 9.0, 6.3 Hz, 1H, H-1_b″), 3.30 (t, *J* = 6.8 Hz, 2H, H-5″), 2.16, 2.15, 2.09, 2.05, 2.04, 2.02 (6s, 21H, 7 × $COCH_3$), 1.68–1.60 (m, 2H, H-2″, H-4″), 1.51–1.42 (m, 2H, H-3″); ^{13}C NMR (100 MHz, $CDCl_3$) δ ppm 170.48, 169.92, 169.71, 169.54, 169.46 (7 × $COCH_3$), 99.21 (C-1′), 98.23 (C-1), 77.26 (C-2), 70.36 (C-3), 69.81 (C-2′), 69.17 (C-5′), 68.59 (C-5), 68.33 (C-3′), 68.07 (C-1″), 66.48, 66.34 (C-4, C-4′), 62.59, 62.25 (C-6, C-6′), 51.25 (C-5″), 28.96 (C-2″), 28.58 (C-4″), 23.44 (C-3″), 20.92, 20.74, 20.62 (7 × $COCH_3$); HRMS (ESI): m/z = 770.25639 $[M+Na]^+$; calculated $C_{31}H_{45}N_3NaO_{18}$ (770.25903).

Data for **4f**, TLC: R_f = 0.21 (50% EtOAc/Petrol); $[\alpha]_D^{20}$ +27.2 (*c* 1.15, $CHCl_3$); ^{1}H NMR (400 MHz, $CDCl_3$) δ ppm 5.39 (dd, *J* = 9.8, 3.3 Hz, 1H, H-3″), 5.34–5.24 (m, 7H, H-3, H-3′, H-2″, H-2, H-4, H-4′, H-4″), 5.10 (d, *J* = 1.5 Hz, 1H, H-1″), 4.94 (s_{br}, 2H, H-1, H-1′), 4.23 (dd, *J* = 12.3, 4.5 Hz, 2H, H-$6_{a'}$, H-$6_{a''}$), 4.19–4.10 (m, 6H, H-5′, H-5″, H-$6_{a,b}$, H-$6_{b'}$, H-$6_{b''}$), 4.01 (s_{br}, 1H, H-2), 3.91 (ddd, *J* = 9.3, 4.5, 2.2 Hz, 1H, H-5), 3.72 (td, *J* = 9.6, 6.6 Hz, 1H, H-1_a‴), 3.45 (td, *J* = 9.6, 6.3 Hz, 1H, H-1_b‴), 3.30 (t, *J* = 6.8 Hz, 2H, H-5‴), 2.15, 2.13, 2.12, 2.08, 2.06, 2.05, 2.03, 2.00 (8 × s, 30H, 10 × $COCH_3$), 1.68–1.58 (m, 4H, H-2‴, H-4‴), 1.46–1.42 (m, 2H, H-3‴); ^{13}C NMR (100 MHz, $CDCl_3$) δ ppm 170.89, 170.75, 170.44, 170.11, 170.07, 169.74, 169.58, 169.32 (10 × $COCH_3$), 99.83 (C-1″), 99.45 (C-1′), 98.32 (C-1), 77.30 (C-2), 76.81 (C-5″), 70.46 (C-3), 69.91 (C-2′), 69.63 (C-2″), 69.43 (C-3″), 69.23 (C-5′), 68.60 (C-5), 68.43 (C-3′), 68.11 (C-1‴), 66.45, 66.36, 66.21 (C-4″, C-4′, C-4), 62.50, 62.28, 62.15 (C-6″, C-6′, C-6), 51.20 (C-5‴), 28.99 (C-4‴), 28.58 (C-2‴), 23.44 (C-3‴), 20.88, 20.71, 20.63 (10 × $COCH_3$); HRMS (ESI): m/z = 1058.34035 $[M+Na]^+$; calculated $C_{43}H_{61}N_3NaO_{26}$ (1035.34355).

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