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Synthesis of 2-*C*-branched derivatives of D-mannose: 2-*C*-aminomethyl-D-mannose binds to the human C-type lectin DC-SIGN with affinity greater than an order of magnitude compared to that of D-mannose

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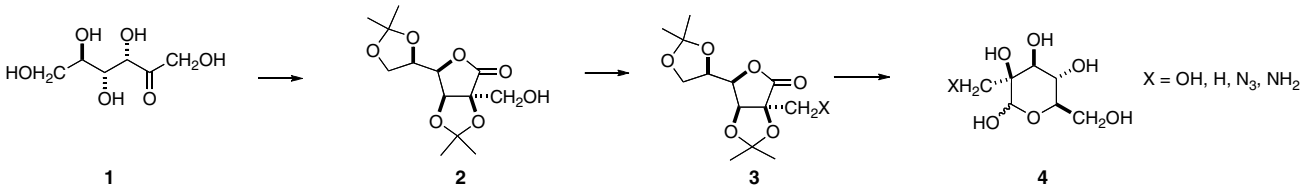
Abstract—2-*C*-Substituted branched D-mannose analogues are novel monosaccharides, readily obtained from a Kiliani-acetonation sequence on D-fructose, followed by subsequent functional group manipulation. 2-*C*-Azidomethyl-D-mannose and 2-*C*-aminomethyl-D-mannose bind to the C-type lectin DC-SIGN (CD209) with significantly greater affinity than mannose. In particular, 2-*C*-aminomethyl-D-mannose exhibits a comparative 48-fold increase in binding as determined using a surface plasmon resonance-based competition assay. DC-SIGN is an important cell-surface type II transmembrane protein that interacts with blood group antigens, endogenous glycoproteins such as ICAM-3, and also deadly pathogens such as the human immunodeficiency and hepatitis C viruses. The effective use of small compounds to block target binding by mannose-selective C-type lectins at sub-millimolar concentrations has not been shown previously; thus, these data represent a very attractive thoroughfare to novel antiviral and immunomodulatory drug development.

1. Introduction

Rare and new monosaccharides, such as D-tagatose, D-psicose and L-glucose, are low calorie sugar alternatives, showing promise as healthy dietary alternatives.[1] Izumoring is a biotechnological concept, which allows the green production of any of the isomeric hexoses.[2,3] Additionally rare sugars show a range of other beneficial biological properties, including 2-deoxy-L-ribose,[4] L-glucose,[5] and D-allose.[6] D-Tagatose[7] and D-psicose[8] have a significant number of chemotherapeutic properties. No examples of studies on the chemotherapeutic potential of carbon branched monosaccharides have been reported; this paper indicates that free monosaccharides bearing a carbon substituent may also have significant potential as novel bioactive agents.

Monosaccharides with a carbon branch at C-2 are readily available from aldoses by a sequential Amadori



Scheme 1.

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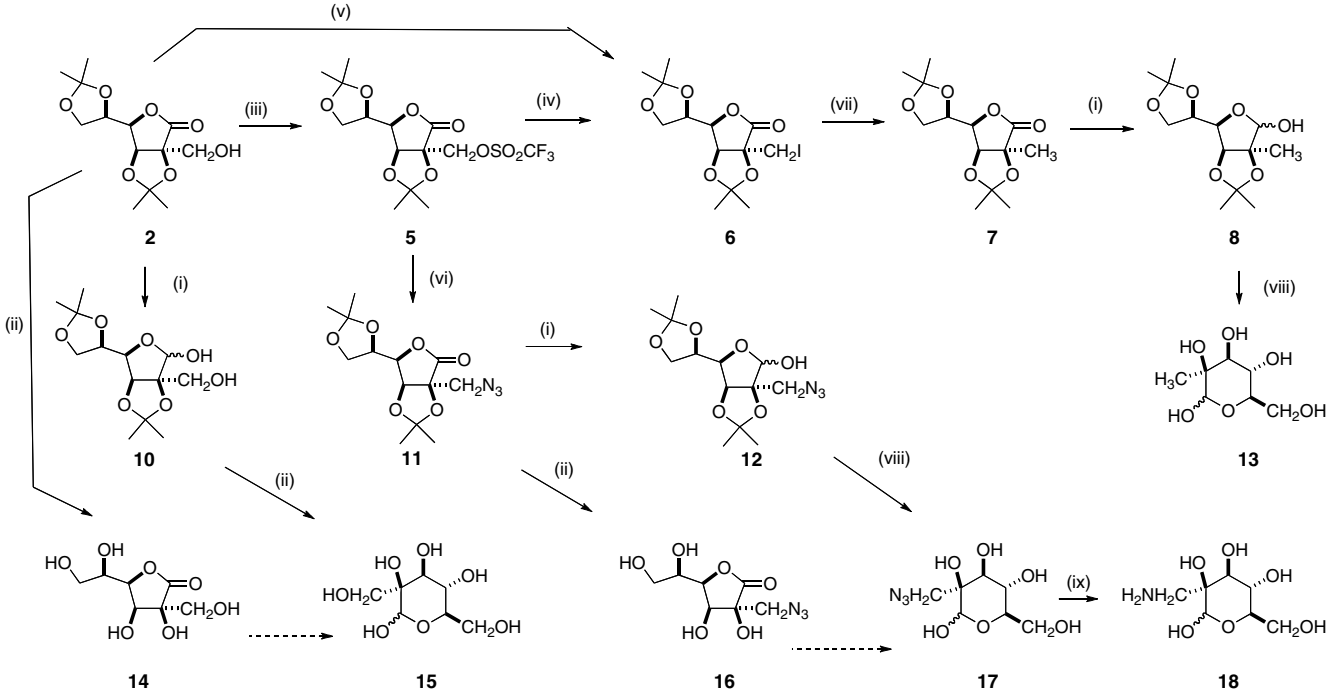
rearrangement followed by treatment with aqueous calcium hydroxide[9] or by a Kiliani ascension of ketoses.[10] For example, D-fructose **1** on reaction with sodium cyanide followed by treatment with acetone and acid allows the isolation of the thermodynamic diacetonide **2** in 51% yield (Scheme 1).[11] This paper describes the synthesis of 2-*C*-branched D-mannose derivatives via functionalisation of the free primary alcohol in **2** to give a range of functional groups CH_2X **3** followed by reduction and deprotection to **4**; aldoses are mixtures of furanose and pyranose forms—this paper provides the first NMR analysis of such branched free sugars. Furthermore, through the capacity to test these compounds for binding to DC-SIGN—a human C-type lectin that binds to mannoside ligands—this paper has indicated the potential for *C*-branched monosaccharide compounds to exert biological activity via a new mechanism. This mechanism involves the direct blockade of a glycan binding protein with the potential of modulating effects within the functional glycome. DC-SIGN is a membrane protein expressed on the surface of specialized cells of the immune system, such as dendritic cells and macrophages, which interacts with oligomannose and the fucosylated glycans present on the surface of host glycoproteins and certain pathogens.[12] In particular, DC-SIGN interacts with the envelope glycoproteins of lethal viruses such as HIV, Ebola and HCV, facilitating the trafficking of viral particles from the periphery to key sites rich in target cells, thus greatly enhancing viral infectivity.[13] Therefore blockade of DC-SIGN binding to viral targets represents a valuable pharmacological drive. In addition, the natural function of DC-SIGN remains obscure, although it has been shown to interact with blood group antigens and heavily glycosylated adhesion molecules such as ICAM-3—the latter a molecule expressed on the surface of T lymphocytes.[14] Targeted blockade of DC-SIGN using a small compound could have value for unlocking the evolved function of this lectin and its physiological role.

2. Results and discussion

2.1. Synthesis of 2-*C*–CH_2X–D-mannoses

D-Fructose **1** was treated with sodium cyanide followed by acetonation to give key intermediate **2** in 51% yield as previously described.[11] For 2-*C*-hydroxymethyl-D-mannose **15**, reduction of **2** by diisobutylaluminum hydride (DIBALH) gave lactols **10** in 70% yield (Scheme 2). Removal of the ketal protecting groups in **10** by treatment with aqueous trifluoroacetic acid afforded the unprotected sugar **15** in 90% yield. In an alternative approach, initial deprotection of the lactone **2** with aqueous trifluoroacetic acid gave branched lactone **14** in 82% yield; however all attempts to reduce the unprotected lactone **14** to **15** were unsuccessful.

The primary alcohol in **2** was modified by nucleophilic substitution reactions. Esterification of **2** with trifluoromethanesulfonic (triflic) anhydride in dichloromethane in the presence of pyridine afforded triflate **5** in 81% yield. Although S_N2 reactions of the triflate **5** might be anticipated to be difficult—since there are three substituents at C-2 including a β-oxygen—reaction of **5** with tetrabutylammonium iodide in THF gave the iodide **6** (61% overall yield from **2**) while with sodium azide in DMF, it formed azide **11** (69% overall yield from **2**). Treatment of **2** with triphenylphosphine, iodine and imidazole in toluene afforded iodide **6** in 85% yield.



Scheme 2. Reagents and conditions: (i) DIBALH, toluene, THF; (ii) CF_3CO_2H, H_2O; (iii) $(CF_3SO_2)_2O$, pyridine, CH_2Cl_2 (iv) Bu_4NI, THF; (v) Ph_3P, I_2, imidazole, toluene; (vi) NaN_3, DMF; (vii) H_2, 10% Pd/C, Et_3N, EtOH; (viii) Dowex® 50WX8-100 (H^+) ion-exchange resin, H_2O, 45 °C; (ix) H_2, Pd black, EtOH, H_2O.

Hydrogenolysis of iodide **6** in the presence of palladium on carbon and triethylamine in ethanol gave the protected 2-*C*-methyl lactone **7** (87% yield); reduction of **7** with DIBALH gave the lactols **8** (94% yield) which on deprotection with Dowex (H^+) in aqueous dioxane yielded 2-*C*-methyl-mannose **13** (88% yield).

DIBALH reduction of the protected azidolactone **11** afforded the lactols **12** (98% yield) from which the protecting acetal groups were removed by Dowex (H^+) (100% yield). Again although the ketals may readily be removed from the azidolactone **11** by aqueous trifluoroacetic acid to give the unprotected lactone **16** (93% yield), a subsequently attempted reduction to the target compound **17** gave intractable mixtures. It is thus experimentally advantageous to obtain the correct oxidation level of the sugar before the protecting groups were removed. This is in contrast to the preparation of the aminomethyl monosaccharide **18**, which was efficiently obtained by hydrogenation of the azidolactols **17** in aqueous ethanol in the presence of palladium black (98% yield). Initial hydrogenation of the protected azidomethyl lactols **12**, followed by subsequent deprotection, gave complex mixtures of the products.

The readily available diacetonide **2** thus allows the functionalized carbon branched monosaccharides to be accessed easily for biological evaluation; it is noteworthy that the nucleophilic displacements of leaving groups at the neopentyl alcohol in **2** proceeded efficiently. Other branched sugars would also be accessible from stereoisomers of **2**.

2.2. NMR of the 2-*C*-substituted mannoses

No studies have previously been published on the structures of 2-*C*-branched sugars in solution which are shown by this study to substantially exist in pyranose forms. 2-*C*-Methyl-**13**, 2-*C*-hydroxymethyl-**15**, 2-*C*-azidomethyl-**17** and 2-*C*-aminomethyl- **18** D-mannose were analyzed by ^1H and ^{13}C NMR spectroscopy; the spectra are shown in Figure 1 and the chemical shifts are given in Tables 1 and 2.

All four compounds occur in both the α- and β-anomers, the proportions of the two being estimated from the relative peak intensities in the 1D ^1H spectra.

2-*C*-Methyl-D-mannose **13**. The complete spin-system of the β-anomer can be followed in the COSY and HMBC spectra. For the α-anomer, very weak COSY peaks can be seen from C1H to C3H, C6H/H′ and $C7H_3$. The rest of the spectrum is strongly coupled and assignments were made based on the proton chemical shifts determined from the HSQC spectrum and simulation of the 1D ^1H spectrum.

2-*C*-Hydroxymethyl-D-mannose **15**. The complete spin-system of the β-anomer can be followed in the COSY and HMBC spectra and the strong C1H to C5H NOE confirms the anomericity. For the α-anomer, the C6H/H′ and C7H/H′ proton and carbon chemical shifts can be identified from the unassigned CH_2 groups in the HSQC, as can the C3H, C4H and C5H from the unassigned CH groups. The specific assignments for these protons were then made by simulation of the 1D ^1H spectrum.

2-*C*-Azidomethyl-D-mannose **17**. The spin-system of the β-anomer can be followed from C4H to C6H/H′ in the COSY spectrum, and the C1, C2 and C7 identified in the HMBC. The strong C1H to C5H NOE confirms the anomericity. For the α-anomer, C1, C2 and C7 were identified in the HMBC, and C6 by elimination as the only remaining CH_2 group in the HSQC. C3H of the β-anomer and the rest of the α-anomer were assigned by simulation of the 1D ^1H spectrum. The ^{13}C assignments for C3 and C5 of the α-form cannot be certain.

2-*C*-Aminomethyl-D-mannose **18**. Both spin systems can be followed from the COSY spectrum and the coupling patterns in the 1D ^1H spectrum. Some of the ^{13}C chemical shifts are too similar between the α and β forms to clearly resolve in the HSQC (such as the two C4s and two C6s), so the assignments are based on the relative intensities in the 1D ^{13}C spectrum.

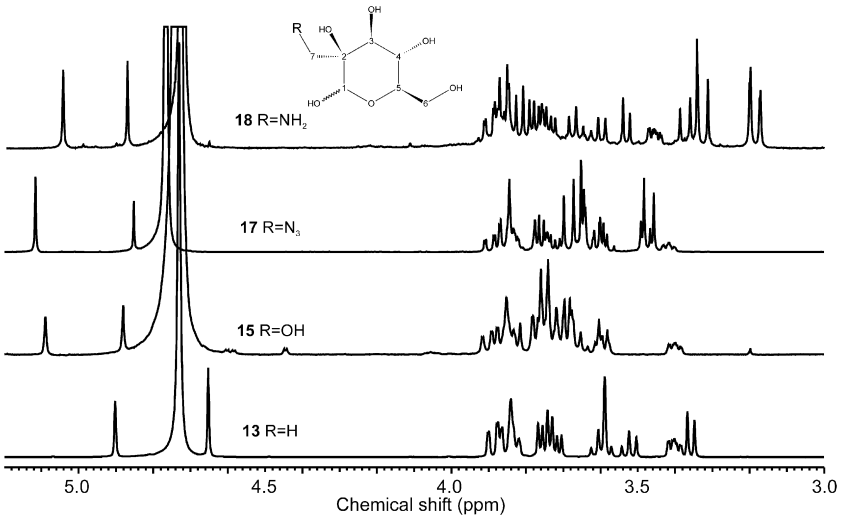


Figure 1. The 1D ^1H NMR spectra of **13** (pH 8.0), **15** (pH 2.8), **17** (pH 1.6), and **18** (pH 5.7) in ^2H_2O and at a temperature of 30 °C. All spectra are referenced to acetone at 2.220 ppm. The numbering scheme used is shown in the insert.

Table 1. ^{1}H Chemical shifts for **13** (pH 8.0), **15** (pH 2.8), **17** (pH 1.6), and **18** (pH 5.7) in $^{2}H_2O$ and at a temperature of 30 °C

Proton	Chemical shift (ppm)							
	2-*C*-Methyl-D-mannose **13**		2-*C*-Hydroxymethyl-D-mannose **15**		2-*C*-Azidomethyl-D-mannose **17**		2-*C*-Aminomethyl-D-mannose **18**	
	α 45%	β 55%	α 45%	β 55%	α 66%	β 33%	α 55%	β 45%
C1H	4.902	4.653	5.088	4.880	5.116	4.852	5.047	4.872
C3H	3.580	3.358	3.690	3.686	3.650	3.612	3.821	3.534
C4H	3.603	3.524	3.652	3.594	3.647	3.582	3.669	3.610
C5H	3.834	3.402	3.836	3.399	3.834	3.417	3.866	3.457
C6H	3.850	3.889	3.864	3.903	3.858	3.897	3.866	3.903
	3.750	3.724	3.762	3.730	3.759	3.728	3.776	3.743
C7H	1.266	1.240	3.69	3.707	3.685	3.605	3.329	3.376
			3.75	3.592	3.470	3.479	3.187	3.190

All chemical shifts are quoted relative to acetone at 2.220 ppm. See Figure 1 for numbering scheme.

Table 2. ^{13}C chemical shifts for **13** (pH 8.0), **15** (pH 2.8), **17** (pH 1.6), and **18** (pH 5.7) in $^{2}H_2O$ and at a temperature of 30 °C

Carbon	Chemical shift (ppm)							
	2-*C*-Methyl-D-mannose **13**		2-*C*-Hydroxymethyl-D-mannose **15**		2-*C*-Azidomethyl-D-mannose **17**		2-*C*-Aminomethyl-D-mannose **18**	
	α 45%	β 55%	α 45%	β 55%	α 66%	β 33%	α 55%	β 45%
C1	97.69	97.39	94.80	94.69	94.51	94.66	94.95	96.03
C2	74.45	74.65	76.14	75.78	76.45	76.61	73.10	72.98
C3	74.59	77.21	72.02	72.67	*72.61*	73.17	73.43	75.32
C4	68.34	68.13	68.24	68.35	68.07	68.22	68.07	67.93
C5	72.85	76.68	72.57	76.54	*72.60*	75.62	72.56	76.84
C6	61.76	61.71	61.70	61.67	61.62	61.62	61.41	61.43
C7	21.33	19.88	64.18	60.95	55.36	51.79	44.26	42.87

All chemical shifts are quoted relative to acetone at 30.90 ppm. See Figure 1 for numbering scheme.

All four compounds show large $^{3}J_{HH}$ coupling constants of 9.0–9.8 Hz between C3H, C4H and C5H. This indicates that these protons are all axial and thus all four compounds adopt the $^{4}C_1$ ring conformation for both anomers. It is interesting to note that compounds **13** and **15** show an α:β anomeric ratio of approximately 45:55, whereas for azide **17**, this is 66:33, suggesting a favourable interaction between the azide group and the C-1 hydroxyl in the α form. A smaller effect is observed for the amine **18**. The ^{1}H and ^{13}C 1D spectra of **17** remain unchanged between pH 1.6 and 8.5, indicating that this effect is pH independent. There is a negligible proportion of any furanose form for any of the compounds present in aqueous solution.

2.3. DC-SIGN binding studies of the 2-*C*-substituted mannose compounds

The surface plasmon resonance competition assay demonstrated the capacity of monosaccharides to disrupt the interaction between DC-SIGN and the gp120 envelope glycoprotein of HIV-1. In reflection of previous solid-phase radioligand assays using microwell plates, competitive inhibition of DC-SIGN binding to gp120 was seen in the presence of monosaccharides such as mannose and fucose at high millimolar concentration.[15] Galactose did not affect binding, as expected, and the $Man_9GlcNAc_2$ oligosaccharide showed significant binding to DC-SIGN at a

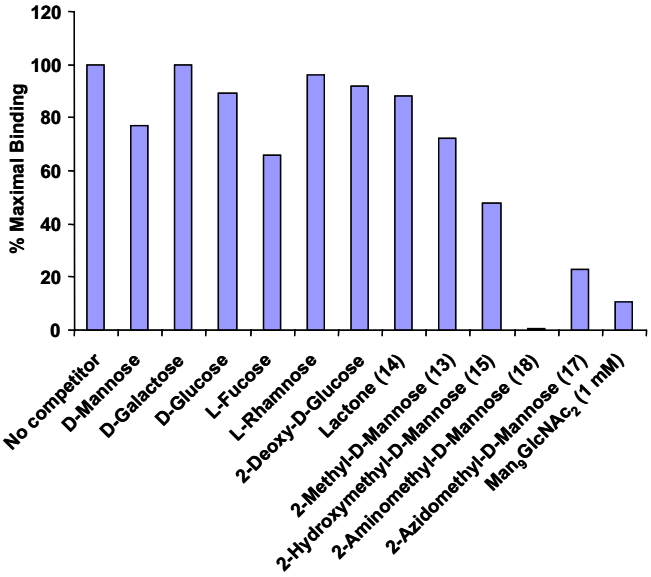


Figure 2. Monosaccharide competition assays I. Natural and synthetic monosaccharides (at 10 mM) were incubated with soluble DC-SIGN prior to injection onto the gp120 glycoprotein-coated surface plasmon resonance chip. The degree of monosaccharide binding was represented as a percentage of the maximal signal obtained from samples run without competitor. The $Man_9GlcNAc_2$ oligosaccharide, a well-characterised ligand for DC-SIGN, was used at a concentration of 1 mM.

concentration of 1 mM, supporting previous data.[15] DC-SIGN contains a glutamate–proline–asparagine–asparagine motif within the carbohydrate recognition domain (CRD), which imparts preference for binding to monosaccharides with the 3-C and 4-C hydroxyls in the equatorial orientation.[16] However, the discrepancies between mannose, glucose and 2-deoxy-glucose indicate factors within the CRD that influence binding at the monosaccharide 2-C position (Fig. 2). 2-*C*-Methyl-D-mannose **15** and 2-*C*-hydroxymethyl-D-mannose **13** showed very slight improvement in DC-SIGN binding. However, the other modifications at the 2-C position showed significant effects on binding, as seen in the case of 2-*C*-aminomethyl-D-mannose **18** and 2-*C*-azidomethyl-D-mannose **17**. Detailed calculation of K_I using a range of 2-*C*-aminomethyl-D-mannose concentrations indicated a value of 0.35 ± 0.03 mM—a value indicating an affinity 48 times greater than that of mannose (K_I for D-mannose $= 17.1 \pm 2.1$ mM) (Fig. 3).

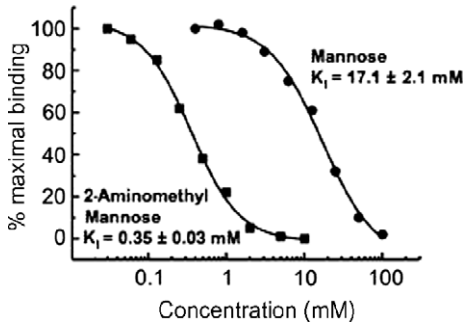


Figure 3. Monosaccharide competition assays II. Quantitation of relative DC-SIGN binding to D-mannose and 2-aminomethyl-D-mannose via determination of K_I values from competition curves.

3. Conclusion

Efficient syntheses of 2-*C*-substituted mannoses have been reported using a key divergent intermediate available on a large scale directly from fructose by green environmentally friendly reactions. The structure of the free sugars, though a complex mixture of four or five species,[17] has been elucidated and may help to understand features about the structures in aqueous solution and thus their biological activity. The capacity to block the binding activity of C-type lectins using a small compound at significant sub-millimolar concentrations is unprecedented. Glycan-binding proteins, such as DC-SIGN, represent highly significant therapeutic targets, and the ability to efficiently generate effective inhibitory compounds from bulk and cost-effective precursors opens up an exciting thoroughfare to a new perspective of drug development—that of directly targeting the expressed glycome.

4. Experimental

All commercial reagents were used as supplied. Tetrahydrofuran and *N*,*N*-dimethylformamide were purchased dry from the Aldrich chemical company in sure-seal bottles. Methanol and pyridine were purchased dry from the Fluka chemical company in sure-seal bottles over molecular sieves. All other solvents were used as supplied (Analytical or HPLC grade), without prior purification. Reactions were performed under an atmosphere of nitrogen or argon, unless stated otherwise. Thin layer chromatography (TLC) was performed on aluminium sheets coated with 60 F_{254} silica. Sheets were visualised using a spray of 0.2% w/v cerium(IV) sulphate and 5% ammonium molybdate in 2 M sulphuric acid. Flash chromatography was performed on Sorbsil C60 40/60 silica. Melting points were recorded on a Kofler hot block and are uncorrected. Optical rotations were recorded on a Perkin–Elmer 241 polarimeter with a path length of 1 dm. Concentrations are quoted in g $100\ mL^{-1}$. Elemental analyses were performed by the microanalysis service of the Inorganic Chemistry Laboratory, Oxford. Infrared spectra were recorded on a Perkin–Elmer 1750 IR Fourier Transform spectrophotometer using thin films on NaCl plates (thin film). Only the characteristic peaks are quoted. Low resolution mass spectra (*m*/*z*) were recorded on VG MassLab 20–250, Micromass BIOQ-II, Micromass Platform 1, Micromass TofSpec 2E, or Micromass Autospec 500 OAT spectrometers and high resolution mass spectra (HRMS *m*/*z*) on a Micromass Autospec 500 OAT spectrometer. Techniques used were electrospray (ESI), chemical ionization (CI NH_3), or atmospheric pressure chemical ionization (APCI). Nuclear magnetic resonance (NMR) spectra were recorded on Bruker AMX 500 (^{1}H: 500 MHz and ^{13}C: 125.7 MHz) and Bruker DPX 400 and DQX 400 spectrometers (^{1}H: 400 MHz and ^{13}C: 100.6 MHz) in the deuterated solvent stated. For spectra recorded in D_2O, acetone was used as an internal reference. Residual signals from other solvents were used as an internal reference. NMR spectra for **13, 15**, **17** and **18** were recorded on a Varian UnityINOVA 500 (^{1}H—500 MHz: ^{13}C—125 MHz) spectrometer, in D_2O, with a probe temperature of 30 °C. Chemical shifts were measured relative to internal standards (^{1}H—acetone at 2.220 ppm: ^{13}C—acetone at 30.90 ppm). Two-dimensional COSY, HSQC and HMBC spectra were used to aid the assignment of ^{1}H and ^{13}C spectra. NOESY spectra were recorded with a 400 ms mixing time. 1D ^{1}H NMR spectral simulations were performed using the program gNMR (Cherwell Scientific Publishing). All chemical shifts (δ) are quoted in ppm and coupling constants (*J*) in Hz.

Monosaccharide binding to DC-SIGN was performed via a competition assay using a BIAcore 2000 surface plasmon resonance instrument (BIAcore, Uppsala). All sensorgrams were recorded at 25 °C with a flowrate of 5 μl per minute. Streptavidin-coated sensorchips (BIAcore, Uppsala) were coated with biotinylated recombinant HIV gp120 glycoprotein at a coupling density of 1000 response units (RU), with reference flowcells simply bearing streptavidin as a source of control, non-glycosylated nonspecific protein. Binding was monitored in HEPES buffered saline containing 5 mM $CaCl_2$ and flowcells were regenerated in HEPES buffered saline containing 3 mM EDTA. Samples, in 100 μl volumes, were prepared as 1 μg per mL soluble recombinant DC-SIGN in the presence of test monosaccharide at a standardized concentration of 10 mM. Binding was monitored in real-time and binding values determined

as peak RU corrected against the signal from the control flowcell. Maximal binding was determined as the peak absolute RU recorded for DC-SIGN binding to the gp120-coated flowcell in the absence of monosaccharide and relative binding in the presence of monosaccharide was expressed as a percentage of this value for each test sample. For more detailed competition curves, serial dilutions of monosaccharide inhibitor were made and several sensorgrams recorded over a range of monosaccharide concentrations.

4.1. 2,3:5,6-Di-*O*-isopropylidene 2-*C*-hydroxymethyl-D-mannofuranose 10

Diisobutylaluminum hydride (1.5 M) in toluene (0.75 mL, 1.12 mmol) was added dropwise to a stirred solution of lactone **2** (200 mg, 0.69 mmol) in anhydrous dichloromethane (2 mL) at −78 °C. The reaction mixture was stirred for 2 h at −78 °C under an atmosphere of nitrogen. The reaction was quenched with water (20 mL) and allowed to warm to room temperature. Saturated aqueous sodium tartrate solution (2 mL) was added, the reaction stirred for 1 h after which the product was then extracted with diethyl ether (2 × 20 mL). The combined organic phases were dried over $MgSO_4$, filtered and concentrated in vacuo. The resulting residue was purified by flash column chromatography (cyclohexane/EtOAc 3:2) affording the protected lactol **10** (0.14 g, 70%) as a colourless oil; A/B = 2:1 (from integration of ^{1}H NMR signals); $[\alpha]_D^{21} = +11.5$ (*c* 1.0, MeOH) Lit.[18] $[\alpha]_D$ +11.2 (*c* 1.0, MeOH); $R_f = 0.20$ (cyclohexane/EtOAc 3:2); ν_{max} (NaCl), 3418 cm^{-1} (OH); δ_H (400 MHz, $CDCl_3$): 1.37, 1.41, 1.45, 1.46 (12H, 4 × s, 2 × C(CH_3)$_2^A$), 1.37, 1.44, 1.48, 1.56, (12H, 4 × s, 2 × C(CH_3)$_2^B$), 2.24 (1H, br s, H–CH_2O*H*^{B}). 2.86 (1H, br s, H–CH_2O*H*^{A}), 3.51 (1H, dd, $J_{3,4} = 2.8$ Hz, $J_{4,5} = 8.3$ Hz, H-4^B), 3.78 (2H, br s, H-2′a^B, H-2′b^B), 3.82 (1H, d, $J_{2'a,2'b} = 11.8$ Hz, H-2′a^A), 3.89 (1H, d, $J_{1,OH} = 2.3$ Hz, OH^A), 3.93 (1H, d, $J_{1,OH} = 11.7$ Hz, OH^B), 3.98 (1H, d, $J_{2'a,2'b} = 11.8$ Hz, H-2′b^A), 4.02–4.12 (4H, m, H-6a^A, H-6b^A, H-6a^B, H-6b^B), 4.15 (1H, dd, $J_{3,4} = 2.8$ Hz, $J_{4,5} = 7.6$ Hz, H-4^A), 4.35–4.43 (2H, m, H-5^A, H-5^B), 4.65 (1H, d, $J_{3,4} = 2.8$ Hz, H-3^B), 4.66 (1H, d, $J_{3,4} = 2.8$ Hz, H-3^A), 4.91 (1H, d, $J_{1,OH} = 11.7$ Hz, H-1^B), 5.36 (1H, d, $J_{1,OH} = 2.3$ Hz, H-1^A); δ_C (100 MHz, $CDCl_3$): 25.1, 26.8, 26.9, 27.0 (C(CH_3)$_2^A$), 25.2, 26.9, 27.0, 27.3 (C(CH_3)$_2^A$), 62.7 (C-2′^{B}), 63.6 (C-2′^{A}), 66.6 (C-6^A), 67.1 (C-6^B), 72.9 (C-5^B), 73.1 (C-5^B), 76.4 (C-4^B), 81.0 (C-4^A), 81.9 (C-3^B), 82.8 (C-3^A), 89.5 (C-2^B), 93.6 (C-2^A), 97.6 (C-1^B), 103.8 (C-1^A), 109.3, 113.8 (2 × *C*Me$_2^A$), 109.4, 114.1 (2 × *C*Me$_2^B$); HR ESI-MS: found *m*/*z* 289.1284 $[M-H]^+$, calcd for $C_{13}H_{21}O_7$: 289.1287; Elemental Analysis: Found: C, 53.75; H, 7.65. $C_{13}H_{21}O_7$ requires: C, 53.78; H, 7.64.

4.2. 2-*C*-Hydroxymethyl-D-mannopyranose 15

A solution of protected lactol **10** (60 mg, 0.20 mmol) in trifluoroacetic acid/water (1:1, 2 mL) was stirred for 3 h at room temperature. The reaction mixture was concentrated in vacuo and the residue co-evaporated with toluene (3 × 2 mL). The resulting residue was dissolved in water (5 mL) and washed with cyclohexane (2 mL) and ethyl acetate (2 mL) and the aqueous layer was concentrated in vacuo to afford 2-*C*-hydroxymethyl-D-mannopyranose **15** (39 mg, 90%) as a colourless oil; $[\alpha]_D^{21} = +27.4$ (*c* 1.0, H_2O); ν_{max} (Ge), 3357 (br, OH); The NMR of compound **15** is discussed above; HR ESI-MS: found *m*/*z* 209.0658 $[M-H]^+$, calcd for $C_7H_{12}O_7$: 209.0661.

4.3. 2-*C*-Hydroxymethyl-D-mannono-1,4-lactone 14

A solution of diacetonide **2** (100 mg, 0.50 mmol) in trifluoroacetic acid/water (1:1, 4 mL) was stirred overnight at room temperature. The reaction mixture was concentrated in vacuo and the residue co-evaporated with toluene (3 × 2 mL). The resulting residue was dissolved in water and washed with cyclohexane (2 mL) and ethyl acetate (2 mL). The aqueous extract was then concentrated in vacuo to afford the unprotected lactone **14** (59 mg, 82%) as a colourless oil; $[\alpha]_D^{21} = +10.0$ (*c* 0.65, MeOH); ν_{max} (Ge), 1771 cm^{-1} (CO), 3390 cm^{-1} (OH); δ_H (D_2O, 400 MHz): 3.63 (1H, dd, $J_{5,6a} = 5.2$ Hz, $J_{6a,6b} = 12.5$ Hz, H-6a), 3.72 (2H, s, H-2′a, H-2′b), 3.76 (1H, dd, $J_{5,6b} = 3.2$ Hz, $J_{6a,6b} = 12.5$ Hz, H-6b), 3.96 (1H, br s, H-5), 4.35 (1H, d, $J_{3,4} = 2.0$ Hz, H-3), 4.50 (1H, dd, $J_{3,4} = 2.0$ Hz, $J_{4,5} = 8.8$ Hz, H-4); δ_C (D_2O, 100 MHz): 62.9 (C-6), 63.1 (C-2′), 68.5 (C-5), 71.0 (C-3), 79.2 (C-2), 79.6 (C-4), 178.4 (C-1); HR ESI-MS: found *m*/*z* 207.0497 $[M-H]^+$, calcd for $C_7H_{11}O_7$: 207.0505.

4.4. 2,3:5,6-Di-*O*-isopropylidene-2-*C*-trifluoromethanesulfonyloxymethyl-D-mannono-1,4-lactone 5

Triflic anhydride (438 μL, 2.60 mmol) was added dropwise to a stirred solution of the protected lactone **2** (500 mg, 1.73 mmol) and pyridine (425 μL, 5.20 mmol) in dry dichloromethane (4.5 mL) at −30 °C. The reaction mixture was stirred under an atmosphere of nitrogen; after 30 min, TLC analysis (cyclohexane/EtOAc 1:1) revealed the conversion of the starting material (R_f 0.41) to one major product (R_f 0.68). The reaction mixture was diluted with dichloromethane (20 mL) and washed with aqueous hydrochloric acid (0.1 M, 25 mL). The organic residue was dried ($MgSO_4$), filtered and concentrated in vacuo and the residue purified by flash column chromatography (cyclohexane/EtOAc 6:1) to afford the triflate **5** (590 mg, 81%) as a pink oil; $[\alpha]_D^{21} = +22.2$ (*c* 1.0, $CHCl_3$); ν_{max} (NaCl), 1795 cm^{-1} (CO); δ_H (400 MHz, $CDCl_3$): 1.39, 1.42, 1.47, 1.48 (12H, 4 × s, 2 × C(CH_3)$_2$), 4.06–4.18 (2H, m, H-6a, H-6b), 4.35 (1H, dd, $J_{3,4} = 3.3$ Hz, $J_{4,5} = 8.3$ Hz, H-4), 4.45 (1H, ddd, $J_{4,5} = 8.3$ Hz, $J_{5,6a} = 3.7$ Hz, $J_{5,6b} = 6.2$ Hz, H-5), 4.67 (1H, d, $J_{2'a,2'b} = 11.1$ Hz, H-2′a), 4.76 (1H, d, $J_{2'a,2'b} = 11.1$ Hz, H-2′b), 4.88 (1H, d, $J_{3,4} = 3.3$ Hz, H-3); δ_C (100 MHz, $CDCl_3$): 25.0, 25.9, 26.8, 26.9 (C(CH_3)$_2$), 66.3 (C-6), 70.7 (C-2′), 72.2 (C-5), 77.6 (C-3), 78.1 (C-4), 83.5 (C-2), 110.1, 115.3 (2 × *C*Me$_2$), 171.5 (C-1).

4.5. 2,3:5,6-Di-*O*-isopropylidene-2-*C*-iodomethyl-D-mannono-1,4-lactone 6

Method A: Triflic anhydride (1.0 mL, 5.60 mmol) was added dropwise to a mixture of diacetonide **2** (1.0 g, 3.47 mmol) in dry dichloromethane (20 mL) and pyridine (1.0 mL, 11.2 mmol) at −30 °C. After stirring for 1 h at

−30 °C, TLC analysis (cyclohexane/EtOAc 4:1) revealed the presence of one major product (R_f 0.3). The reaction mixture was diluted with dichloromethane (50 mL) and washed with aqueous hydrochloric acid (1 M, 50 mL). The aqueous layers were further extracted with dichloromethane (3 × 20 mL). The combined organic extracts were washed with brine (2 × 15 mL), dried over $MgSO_4$, filtered and concentrated in vacuo to afford the triflate **5** which was used without further purification. A solution of the triflate **5** and tetrabutylammonium iodide (5.0 g, 13.5 mmol) in THF (40 mL) was stirred at room temperature. After 14 h, the solvent was evaporated and the residue was partitioned between diethyl ether (50 mL) and water (50 mL). The organic layer was washed with an aqueous solution of sodium thiosulfate (1 M, 50 mL), dried ($MgSO_4$), filtered and concentrated in vacuo. The resulting residue was purified by flash chromatography (cyclohexane/EtOAc 4:1) to afford the iodide **6** (1.38 g, 62%); $[\alpha]_D^{22} = +31.8$ (*c*, 1.09, $CHCl_3$); ν_{max} (NaCl): 1791 cm^{-1} (CO); δ_H (400 MHz, $CDCl_3$): 1.39, 1.46, 1.48, 1.50 (12H, 4 × s, 2 × –C(CH_3)$_2$), 3.38 (1H, d, $J_{2'a,2'b} = 10.4$ Hz, H-2′a), 3.50 (1H, d, $J_{2'a,2'b} = 10.4$ Hz, H-2′b), 4.07–4.17 (2H, m, H-6a, H-6b), 4.43–4.49 (2H, m, H-4, H-5), 4.07 (1H, d, $J_{3,4} = 3.2$ Hz, H-3); δ_C (100 MHz, $CDCl_3$): 0.4 (–CH_2I), 24.8, 26.3, 26.6 (2 × –C(CH_3)$_2$), 65.9 (C-6), 72.1, 78.3, 80.4 (C-3, C-4, C-5), 84.7 (C-2), 109.5, 114.1 (2 × CMe_2), 172.1 (C-1); HR ESI-MS: found *m/z* 421.0123 $[M+Na]^+$, calcd for $C_{13}H_{19}IO_6Na$ 421.0119.

Method B: Imidazole (630 mg, 9.24 mmol), triphenylphosphine (1.75 g, 6.67 mmol), and iodine (1.69 g, 6.67 mmol) were added to a stirred solution of alcohol **2** (740 mg, 2.57 mmol) in toluene (12 mL) and stirred at 85 °C for 1.5 h. The reaction mixture was allowed to cool to room temperature, concentrated in vacuo and then partitioned between CH_2Cl_2 (30 mL), and saturated sodium hydrogen carbonate solution (30 mL) and the resulting solution was extracted with CH_2Cl_2 (3 × 30 mL). The combined organic layers were washed with water (2 × 30 mL), dried ($MgSO_4$), filtered and concentrated in vacuo. The resulting residue was purified by flash column chromatography (cyclohexane/EtOAc 6:1→3:1) to afford the iodide **6** as a colourless oil (863 mg, 85%) which was identical by ^{1}H NMR to the compound prepared in method A.

4.6. 2,3:5,6-Di-*O*-isopropylidene-2-*C*-methyl-D-mannono-1,4-lactone 7

Iodolactone **6** (3.30 g, 8.29 mmol) in ethanol (30 mL) and triethylamine (1.4 mL, 9.99 mmol) were stirred under hydrogen for 3 days in the presence of 10% palladium on carbon (300 mg); the reaction mixture was filtered through Celite and the filtrate was evaporated in vacuo. The residue was dissolved in dichloromethane (30 mL), washed with saturated aqueous sodium thiosulphate solution (30 mL), water (2 × 30 mL), dried over $MgSO_4$, filtered and concentrated in vacuo. The residue was purified by flash column chromatography (cyclohexane/EtOAc 3:1→1:1) to afford the protected methyl lactone **7** (1.97 g, 87%); mp 111–113 °C (cyclohexane/Et_2O); $[\alpha]_D^{22} = +48.8$ (*c* 0.9, $CHCl_3$); ν_{max} (NaCl), 1787 cm^{-1} (CO); δ_H (400 MHz, $CDCl_3$): 1.38, 1.42, 1.44, 1.46 (12H, 4 × s, 2 × –C(CH_3)$_2$), 1.56 (3H, s, CH_3), 4.06 (1H, dd, $J_{5,6a} = 4.0$ Hz, $J_{6a,6b} = 9.2$ Hz, H-6a), 4.14 (1H, dd, $J_{5,6a} = 5.6$ Hz, $J_{6a,6b} = 9.2$ Hz, H-6b), 4.26 (1H, dd, $J_{3,4} = 3.2$ Hz, $J_{4,5} = 8.0$ Hz, H-4), 4.39–4.43 (1H, m, H-5), 4.46 (1H, d, $J_{3,4} = 3.2$ Hz, H-3); δ_C (100 MHz, $CDCl_3$): 18.0 (CH_3), 25.0, 26.7, 26.8, 26.9 (2 × –C(CH_3)$_2$), 66.4 (C-6), 72.5 (C-5), 77.2 (C-4), 80.4 (C-3), 82.9 (C-2), 109.8, 113.4 (2 × CMe_2), 176.0 (C-1); HR ESI-MS: found *m/z* 295.1155 $[M+Na]^+$, calcd for $C_{13}H_{20}O_6Na$ 295.1152; $C_{13}H_{20}O_6$ requires C, 57.34; H, 7.40. Found: C, 57.35; H, 7.41.

4.7. 2,3:5,6-Di-*O*-isopropylidene-2-*C*-methyl-D-manno-furanose 8

Diisobutylaluminum hydride (1.0 M) in toluene (1.30 mL, 1.29 mmol) was added dropwise to a stirred solution of the branched methyl lactone **7** (320 mg, 1.18 mmol) in dry dichloromethane (3.0 mL) at −78 °C. The reaction mixture was stirred for 1.5 h at −78 °C under an atmosphere of nitrogen. TLC analysis (cyclohexane/EtOAc 3:1) revealed conversion of the starting material (R_f 0.7) into one major product (R_f 0.4). The reaction was quenched with methanol (3 mL) and allowed to warm to room temperature. Saturated aqueous sodium tartrate solution (5 mL) was added and the reaction was stirred for 1 h at room temperature. The aqueous phase was extracted with ethyl acetate (3 × 10 mL), dried ($MgSO_4$), filtered and concentrated in vacuo. The residue was purified by flash column chromatography (cyclohexane/EtOAc 3:1→1:1) to afford the methyl lactol **8** (300 mg, 94%) as a crystalline solid; A/B = 3:1 (from integration of ^{1}H NMR signals); mp 96–98 °C; ν_{max} (NaCl), 3484 cm^{-1} (OH); $[\alpha]_D^{22} = +10.1$ (*c* 1.01, $CHCl_3$); δ_H (400 MHz, $CDCl_3$): 1.36, 1.44, 1.47, 1.52 (12H, 4 × s, 2 × C(CH_3)$_2^A$), 1.38, 1.42, 1.47, 1.51 (12H, 4 × s, 2 × C(CH_3)$_2^B$), 1.47 (6H, s, CH_3^A, CH_3^B), 2.73 (1H, d, $J_{1,OH} = 2.7$ Hz, OH^B), 3.48 (1H, dd, $J_{3,4} = 2.9$ Hz, $J_{4,5} = 8.3$ Hz, H-4^A), 3.92 (1H, d, $J_{1,OH} = 11.9$ Hz, OH^A), 4.00–4.14 (5H, m, H-6a^A, H-6b^A, H-4^B, H-6a^B, H-6b^B), 4.35–4.42 (4H, m, H-3^A, H-5^A, H-3^B, H-5^B), 4.66 (1H, d, $J_{1,OH} = 11.9$ Hz, H-1^A), 5.21 (1H, d, $J_{1,OH} = 2.7$ Hz, H-1^B); δ_C (100 MHz, $CDCl_3$): 20.0 (CH_3^B), 21.6 (CH_3^A), 25.2, 25.2, 26.6, 26.9, 26.9, 27.0, 27.3, 27.4 (C(CH_3)$_2^A$, C(CH_3)$_2^B$), 66.8 (C-6^B), 67.2 (C-6^A), 72.8 (C-5^A), 73.1 (C-5^B), 75.7 (C-4^A), 80.1 (C-4^B), 84.8 (C-3^A), 86.1 (C-3^B), 86.5 (C-2^A), 92.1 (C-2^B), 101.2 (C-1^A), 103.4 (C-1^B), 109.1, 112.8 (2 × CMe_2^B), 109.3, 113.2 (2 × CMe_2^A); HR ESI-MS: found *m/z* 297.1304 $[M+Na]^+$, calcd for $C_{13}H_{22}O_6Na$ 297.1309; Elemental Analysis: Found: C, 56.90; H, 8.10. $C_{13}H_{22}O_6$ requires: C, 56.92; H, 8.08.

4.8. 2-*C*-Methyl-D-mannopyranose 13

A mixture of the protected methyl lactol **8** (240 mg, 0.88 mmol) and Dowex® 50WX8-100 (H^+) ion-exchange resin (50 mg) in dioxane/water (1:1, 1.5 mL) was stirred for 16 h at 45 °C, filtered and concentrated in vacuo affording free branched methyl sugar **13** as a white solid (150 mg, 88%); mp 184–186 °C; $[\alpha]_D^{22} = +15.1$ (*c* 1.01, H_2O); The NMR of compound **13** is discussed above; HR ESI-MS: found *m/z* 217.0680 $[M+Na]^+$, calcd for $C_7H_{14}O_6Na$ 217.0683.

4.9. 2-*C*-Azidomethyl-2,3:5,6-di-*O*-isopropylidene-D-mannono-1,4-lactone 11

Triflic anhydride (1.42 mL, 8.41 mmol) was added dropwise to a stirred solution of the protected lactone **2** (1.62 g, 5.61 mmol) and pyridine (1.37 mL, 16.8 mmol) in dry dichloromethane (8.4 mL) at −30 °C. The reaction mixture was stirred under an atmosphere of nitrogen; after 30 min, TLC analysis (cyclohexane/EtOAc 1:1) revealed the conversion of starting material (R_f 0.41) to one major product (R_f 0.68). The reaction mixture was diluted with dichloromethane (50 mL) and washed with aqueous hydrochloric acid (0.1 M, 60 mL). The organic residue was dried over $MgSO_4$, filtered and concentrated in vacuo to afford the triflate **5** which was used without any further purification. Sodium azide (401 mg, 6.17 mmol) was added to a stirred solution of the triflate **5** (2.21 g, 5.61 mmol, assumed quantitative) in dry DMF (5.6 mL) at room temperature. The reaction mixture was stirred for 3 h under an atmosphere of nitrogen, after which time TLC analysis (cyclohexane/EtOAc 1:1) revealed complete conversion of starting material (R_f 0.68) into one major product (R_f 0.62). The reaction mixture was concentrated in vacuo and the residue was dissolved in ethyl acetate (60 mL) and washed with brine (60 mL). The organic layer was dried ($MgSO_4$), filtered and concentrated in vacuo. The resulting residue was purified by flash column chromatography (cyclohexane/EtOAc 3:1) to afford the azide **11** (1.22 g, 69% over two steps); mp 77–78 °C; $[\alpha]_D^{22} = -130$ (*c* 0.7, $CHCl_3$); ν_{max} (NaCl), 1789 cm^{-1} (CO), 2110 cm^{-1} (N_3); δ_H (400 MHz, $CDCl_3$): 1.38, 1.41, 1.47, 1.48 (12H, 4 × s, 2 × C(CH_3)$_2$), 3.58 (1H, d, $J_{2'a,2'b} = 13.0$ Hz, H-2′a), 3.81 (1H, d, $J_{2'a,2'b} = 13.0$ Hz, H-2′b), 4.06 (1H, dd, $J_{5,6a} = 3.4$ Hz, $J_{6a,6b} = 8.5$ Hz, H-6a), 4.14 (1H, dd, $J_{5,6b} = 5.8$ Hz, $J_{6a,6b} = 8.5$ Hz, H-6b), 4.31 (1H, dd, $J_{3,4} = 3.3$ Hz, $J_{4,5} = 8.5$ Hz, H-4), 4.41 (1H, ddd, $J_{4,5} = 8.5$ Hz, $J_{5,6a} = 3.4$ Hz, $J_{5,6b}$ 5.8 Hz, H-5), 4.76 (1H, d, $J_{3,4} = 3.3$ Hz, H-3); δ_C (100 MHz, $CDCl_3$): 25.0, 26.1, 26.9, 27.0 (C(CH_3)$_2$), 50.5 (C-2′), 66.4 (C-6), 72.4 (C-5), 78.1 (C-4), 78.3 (C-3), 84.9 (C-2) 110.0, 114.5 (2 × CMe_2), 173.8 (C-1); HR ESI-MS: found *m*/*z* 336.1166 $[M+Na]^+$, calcd for $C_{13}H_{19}O_6N_3Na$ 336.1166; Elemental analysis: Found: C, 49.74; H, 6.17; N, 13.10. $C_{13}H_{19}N_3O_6$ requires: C, 49.84; H, 6.11; N, 13.41.

4.10. 2-*C*-Azidomethyl-2,3:5,6-di-*O*-isopropylidene-D-mannofuranose 12

Diisobutylaluminum hydride (1.0 M) in toluene (4.3 mL, 4.29 mmol) was added dropwise to a stirred solution of 2-*C*-azidomethyl-2,3:5,6-di-*O*-isopropylidene-D-mannono-1,4-lactone **11** (1.22 g, 3.90 mmol) in dichloromethane (12 mL) at −78 °C. The reaction was stirred for 1.5 h at this temperature, quenched with methanol (5 mL) and allowed to warm to room temperature. Saturated aqueous sodium tartrate solution (25 mL) was added and the reaction was stirred for 1 h. The product was extracted with dichloromethane (2 × 30 mL), dried over $MgSO_4$, filtered and concentrated in vacuo. The resulting residue was purified by flash column chromatography (cyclohexane/EtOAc 5:1→2:1) to afford lactol **12** (1.20 g, 98%) as a colourless oil; A/B = 5:1 (from integration of ^1H NMR signals); $[\alpha]_D^{21} = -39$ (*c* 1.0, $CHCl_3$); ν_{max} (NaCl), 2106 cm^{-1} (N_3), 3425 cm^{-1} (OH); δ_H (400 MHz, $CDCl_3$): 1.37, 1.45, 1.48, 1.49 (12H, 4 × s, 2 × C(CH_3)$_2^A$), 1.36, 1.44, 1.53, 1.56 (12H, 4 × s, 2 × C(CH_3)$_2^B$), 2.91 (1H, d, $J_{1,OH} = 0.9$ Hz, OH^A), 3.47 (1H, dd, $J_{3,4} = 2.7$ Hz, $J_{4,5} = 8.3$ Hz, H-4^B), 3.54 (1H, d, $J_{2'a,2'b} = 13.2$, H-2′a^A), 3.58 (1H, d, $J_{2'a,2'b} = 11.9$ Hz, H-2′a^B), 3.68 (1H, d, $J_{2'a,2'b} = 13.2$ Hz, H-2′b^A), 3.74 (1H, d, $J_{1,OH} = 11.5$ Hz, OH^B), 3.87 (1H, d, $J_{2'a,2'b} = 11.9$ Hz, H-2′b^B), 4.02–4.12 (5H, m, H-4^A, H-6a^A, H-6b^A, H-6a^B, H-6b^B), 4.35 (1H, ddd, $J_{4,5} = 8.3$ Hz, $J_{5,6a} = 4.5$ Hz, $J_{5,6b} = 6.1$ Hz, H-5^B), 4.39 (1H, ddd, $J_{4,5} = 7.9$ Hz, $J_{5,6a} = 4.4$ Hz, $J_{5,6b} = 6.1$ Hz, H-5^A), 4.51 (1H, d, $J_{3,4} = 3.1$ Hz, H-3^A), 4.59 (1H, d, $J_{3,4} = 2.7$ Hz, H-3^B), 4.83 (1H, d, $J_{1,OH} = 11.5$ Hz, H-1^B), 5.34 (1H, d, $J_{1,OH} = 0.9$ Hz, H-1^A); δ_C (100 MHz, $CDCl_3$): 25.2, 26.6, 26.9, 27.0, 27.0, 27.1, 27.1, 27.5 (C(CH_3)$_2^A$, C(CH_3)$_2^B$), 52.5 (C-2′^{A}), 52.9 (C-2′^{B}), 66.8 (C-6^A), 67.1 (C-6^B), 72.7 (C-5^B), 72.9 (C-5^A), 76.3 (C-4^B), 80.5 (C-4^A), 82.4 (C-3^B), 83.3 (C-3^A), 94.3 (C-2^A). 94.3 (C-2^B), 98.0 (C-1^B), 102.4 (C-1^A), 109.4, 114.7 (2 × CMe_2^A), 109.5, 114.9 (2 × CMe_2^B); HR ESI-MS: found *m*/*z* 314.1346 $[M-H]^+$, calcd for $C_{13}H_{20}N_3O_6$: 314.1352.

4.11. 2-*C*-Azidomethyl-D-mannopyranose 17

A mixture of the protected methyl lactol **12** (1.0 g, 3.18 mmol) and Dowex® 50WX8-100 (H^+) ion-exchange resin (600 mg) in dioxane/water (1:1, 10 mL) was stirred for 48 h at 45 °C, filtered and concentrated in vacuo affording 2-*C*-azidomethyl mannose **17** (750 mg, quant.) as a colourless oil; $[\alpha]_D^{23} = +41.0$ (*c* 1.0, H_2O); ν_{max} (Ge), 2115 cm^{-1} (N_3), 3776 cm^{-1} (OH); The NMR of compound **17** is discussed above; HR ESI-MS: found *m*/*z* 234.0725 $[M-H]^+$, calcd for $C_7H_{12}N_3O_6$ 234.0726.

4.12. 2-*C*-Azidomethyl-D-mannono-1,4-lactone 16

The protected azide **11** (100 mg, 0.40 mmol) in trifluoroacetic acid/water (1:1, 4 mL) was stirred overnight at room temperature. The reaction mixture was concentrated in vacuo and the residue was co-evaporated with toluene (3 × 2 mL). The residue was dissolved in water, and washed with cyclohexane (2 mL) and ethyl acetate (2 mL). The aqueous extract was then concentrated in vacuo to give the azidolactone **16** (70 mg, 93%) as a colourless oil; $[\alpha]_D^{25} = +14.2$ (*c* 0.85, MeOH); ν_{max} (Ge), 1779 cm^{-1} (CO), 2115 cm^{-1} (N_3), 3423 cm^{-1} (OH); δ_H (400 MHz, D_2O): 3.56 (2H, s, H-2′a, H-2′b), 3.61 (1H, dd, $J_{5,6a} = 5.5$ Hz, $J_{6a,6b} = 12.2$ Hz, H-6a), 3.73 (1 H, d, $J_{6a,6b} = 12.2$ Hz, H-6b), 3.94 (1H, br s, H-5), 4.33 (1H, d, $J_{3,4} = 2.9$ Hz, H-3), 4.47 (1H, dd, $J_{3,4} = 2.9$ Hz, $J_{4,5} = 8.8$ Hz, H-4); δ_C (100 MHz, D_2O) 52.9 (C-2′), 62.9 (C-6), 68.4 (C-5), 71.1 (C-3), 78.5 (C-2), 79.4 (C-4), 177.3 (C-1); HR ESI-MS: found *m*/*z* 232.0571 $[M-H]^+$, calcd for $C_7H_{10}N_3O_6$: 232.0570.

4.13. 2-*C*-Aminomethyl-D-mannopyranose 18

A solution of the azide **17** (500 mg, 2.13 mmol) in ethanol/water (1:1, 5 mL) was stirred overnight under an atmosphere of hydrogen in the presence of palladium black (50 mg). Infra red spectroscopy indicated the consumption of all azide starting material. The mixture was filtered

through Celite and washed thoroughly with water. The filtrate and washings were combined and concentrated in vacuo to give the branched 2-*C*-aminomethyl-D-mannopyranose **18** (430 mg, 98%) as a clear oil; $[\alpha]_D^{22} = +3.7$ (*c* 1.0, H_2O); ν_{max} (Ge), 3698 cm^{-1} (OH/NH); The NMR of compound **18** is discussed above; HR ESI-MS: found *m*/*z* 210.0972 $[M+H]^+$, calcd for $C_7H_{16}NO_6$: 210.0972.

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