A New Preparative Synthesis of 1-D-6-*O*-(2-Amino-2-Deoxy-D-glycopyranosyl)-*chiro*-Inositol 1-Phosphate and 1,2-Cyclic Phosphate

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A convenient preparative synthesis of 1-D-6-*O*-(2-amino-2-deoxy-α-D-glycopyranosyl)-*chiro*-inositol 1-phosphate **(III)** and -1,2-cyclic phosphate **(IV)** using D-*chiro*-inositol as starting material is reported. Compound **III** has been previously found to behave as type P inositolphosphoglycans (the putative second messengers of insulin-like growth factor I) in organotypic cultures of chicken embryo. The synthesis of **III** and **IV**, now reported, considerably improves previous synthesis of **III** and permits the effective preparation of these substances for biological investigation.

Introduction

Inositol-containing glycolipids have been reported as substrates for receptor-activated phospholipases that generate intracellular mediators, referred to as inositolphosphoglycans (IPGs). Insulin and insulin-like growth factor *I* (*IGF-I*) are among the best-characterised extracellular ligands.[1] The precise chemical structures of these lipid derived second messengers are still unknown. Two main structural groups have been proposed on the basis of their chemical composition,[2,3] which show different biological activities and tissue distribution:[4] the family of *myo*-inositol containing IPGs (type-A IPGs) and the family of *chiro*-inositol containing IPGs (type-P IPGs). The already existing partial structural data[5] suggest structural similarities with the glycosyl-phosphatidylinositol anchors (GPI anchors)[6] but the determination of the structure of these mediators is rather difficult due the scarcity of active material that can be obtained from biological sources.

In an attempt to disclose the minimal structural requirements for biological activity of these IPG mediators, a number of substructures containing some of the structural motifs thought to be present in these second messengers have been synthesised by us[7] and by others.[8] These synthetic substructures include 1-D-6-*O*- (2-amino-2-deoxy-α-D-glucopyranosyl)-*myo*-inositol 1-phosphate **I**, the corresponding 1,2 cyclic phosphate **II**, and 1-D-6-*O*-(2-amino-2-deoxy-α-D-glucopyranosyl)-*chiro*-inositol 1-phosphate **III** (Figure 1). Interestingly enough, compound **II** behaved as type-A IPGs in exhibiting proliferative effects in organotypic cultures of chicken embryo, whereas compound **III** behaved as type-P IPGs, inducing differentiation in the same biological system.[9] These organotypic cultures have been used as a model in previous studies with *IGF-I*[10,12] as it has been

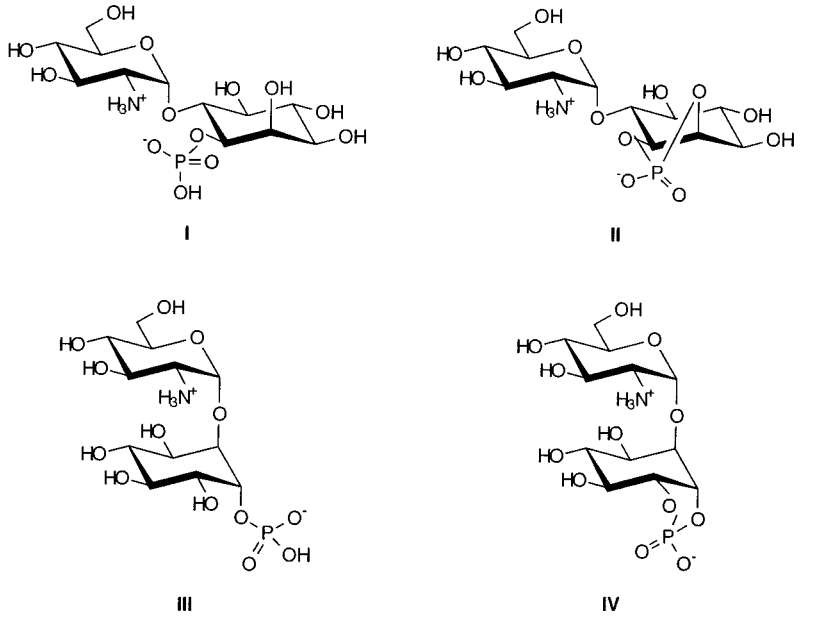

Figure 1. Synthetic substructures containing the structural motifs of IPG mediators

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shown that *IGF-I* controls growth and differentiation in the developing inner ear[10,15] of chicken embryo through the generation of IPG mediators.

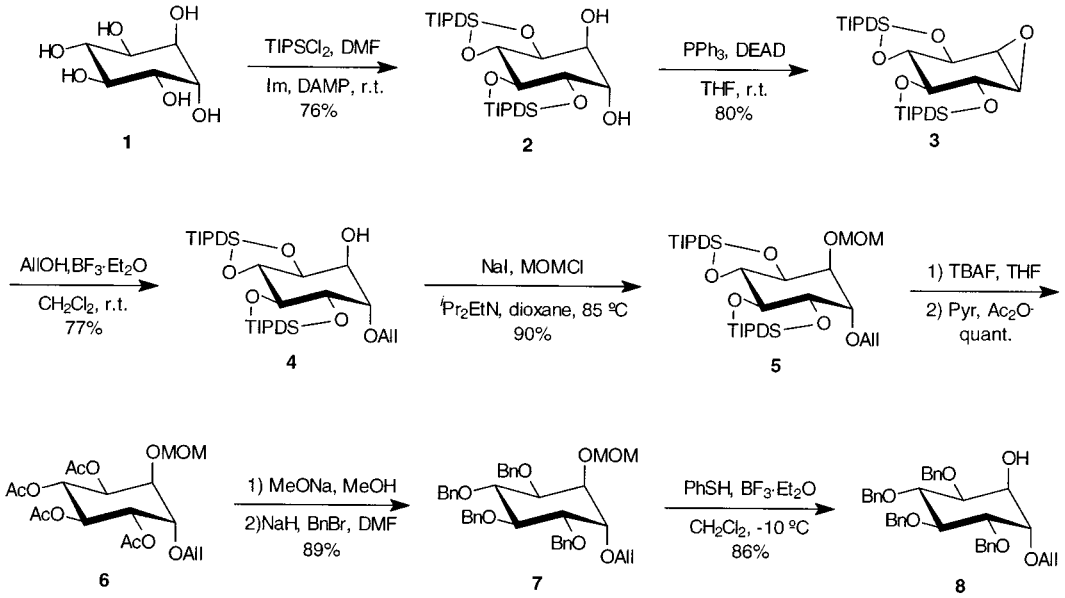
We previously synthesised compounds **I** and **II** using effective strategies starting from *myo*-inositol,[7,13] but compound **III** was prepared after constructing the D-*chiro*-inositol moiety by following a multistep procedure from methyl-α-D-glucopyranoside.[14] As a consequence of the significant biological activity of **III** it was necessary for us to develop a more convenient preparation of this compound. Since both D- and L-*chiro*-inositol are now commercially available, the new synthesis of **III** was envisaged starting from D-*chiro*-inositol. Such an approach seemed more attractive and effective than those involving multistep sequences for the construction of the cyclitol moiety using either *myo*-inositol[15] or other precursors,[14,16,17] This approach presents the additional advantage that both D- and L-*chiro*-inositol-containing IPG-like molecules can be obtained by the same synthetic route, a consideration which may be of some biological significance.[18]

We now report a new synthetic sequence for the preparation of **III** and the corresponding 1,2 cyclic phosphate **IV**, in which the cyclitol moiety has been prepared from D-*chiro*-inositol by an efficient multigram procedure and effectively glycosylated using the trichloacetimidate method.

Results and Discussion

The synthesis of 1-D-6-*O*-(2-amino-2-deoxy-α-D-glucopyranosyl)-*chiro*-inositol 1-phosphate **III** starting from the C_2 symmetric D-*chiro*-inositol **1** implies the preparation of a derivative with the positions 1 and 6 differentiated for further phosphorylation and glycosylation. Thus an efficient strategy for the protection of all the equatorial positions of the cyclitol is needed, excluding the utilisation of standard acetal protection methodology, which affords the equatorial 3,4-diol.[19] On the other hand, we have demonstrated that acid catalysed *trans* diaxial ring opening of protected conduritol B epoxide is an effective approach to *chiro* inositol derivatives with positions 1 and 6 differentiated.[14] The reported synthesis of optically pure conduritol epoxide starting from L-*chiro*-inositol in an expeditive three-step approach[20] seemed to be a convenient route toward these epoxides. However, in the conditions reported,[20] the key protection with the bifunctional protecting agent 1,3-dichloro-1,1,3,3,-tetraisopropyldisiloxane (TIPDSCl2)[21] in pyridine occurs in unacceptably low yields. We found, however, that using different experimental conditions[22] the yield of the silylation reaction could be greatly increased.

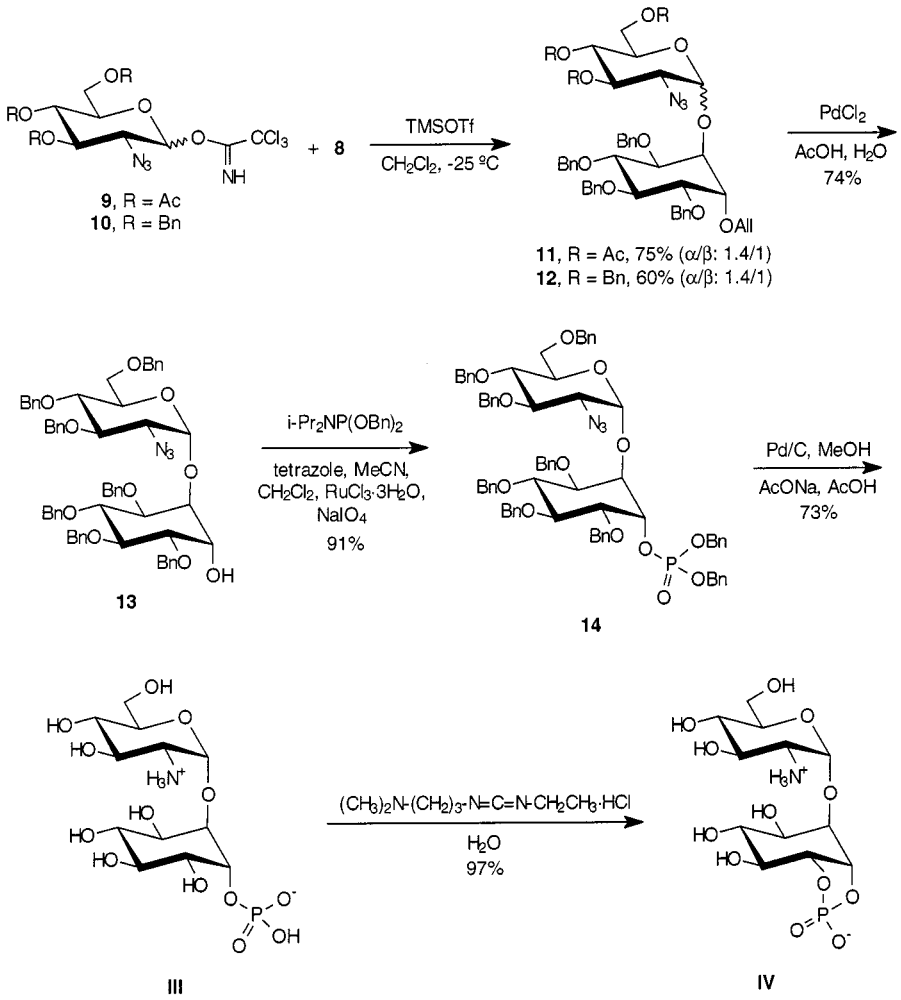
Thus, treatment of D-*chiro*-inositol **1** with 2.1 equiv. of $TIPDSCl_2$ in dimethylformamide in the presence of imidazol and 4-(dimethylamino)pyridine (DAMP) gave compound **2** in 76% yield (Scheme 1), thus achieving the protection of all the four equatorial hydroxyl groups of D-*chiro* inositol in a single high yielding step. Whereas the C_2 symmetry properties of **2** may probably permit its selective monoprotection, monophosphorylation or even a selective monoglycosylation, we preferred to follow our previous route and to obtain building block **4** by ring opening of the epoxide **3**. Treatment of **2** with triphenylphosphane in presence of diethylazodicarboxylate (DEAD) in tetrahydrofuran as previously reported,[20] afforded the fully protected 1-D-1,2-anhydro-*myo*-inositol derivative **3** in 80% yield. Acid-catalysed *trans*-diaxial opening[14,23] of the epoxide ring of **3** with allyl alcohol in the presence of boron trifluoride·diethyl ether afforded the desired D-*chiro*-inositol derivative **4** with position 6 free and position 1 selectively protected. As could be anticipated, all attempts to glycosylate **4** using various glycosyl donors under different conditions failed. This result is



Scheme 1

not surprising taking into account the high steric hindrance induced by the large TIPDS group which makes an approach of the glycosyl donor to an unreactive axial hydroxyl group difficult. Accordingly, protecting group manipulation was needed, and two different sequences were used to obtain the more reactive acceptor **8**: a) protection of the hydroxyl group of **4** with methoxymethyl chloride (MOMCl) in the presence of NaI using Hünig's base[24] to give the fully protected 1-D-*chiro*-inositol **5** in 90% yield, which was then desilylated and perbenzylated to afford **7** in low yield; b) one-pot desilylation-acetylation followed by Zemplén deacetylation and benzylation to give **7** in 89% yield. Compound **8** was then obtained after removal of the MOM group using thiophenol in the presence of boron trifluoride·diethyl ether[25] in 86% yield.

Previous experience has shown that the trichloroacetimidate method[26] using 2-azido-2-deoxy-glucose as glycosyl donor is the method of choice for glycosylating the poorly reactive hydroxyl groups of cyclitols in general, and the axially oriented hydroxyl groups in particular. The two glycosyl donors **9** and **10** were obtained from the readily available 1,3,4,6-tetra-*O*-acetyl-2-azido-2-deoxy-D-glucose[27] as previously reported.[7] Condensation of trichloroacetimidate **9** with **8** in dichloromethane at room temperature using TMSOTf as a promoter gave the desired disaccharide **11** in 75% yield as an α/β mixture (1.4:1) (Scheme 2). The use of the more reactive glycosyl donor **10** gave the disaccharide **12** in 60% yield in the same α/β ratio. All attempts to improve the stereoselectivity of the reaction by decreasing the reaction temperature or by using pure β-trichloroacetimidate **10β** had little effect on product distribution. Nevertheless, disaccharide **12α** was easily separated from the reaction mixture. Removal of the allyl group was carried out in one step using palladium chloride in acetic acid-water[28] and gave **13** in 74% yield. Phosphorylation using the phosphoroamidite method[29] gave **14**, which after hydrogenolysis yielded the desired compound **III**. Finally, the transformation of **III** into **IV** was performed in water by ring closure using a water-soluble carbodiimide. This last transformation occurred in quantitative yield, in contrast to the 20% yield reported in the *myo*-inositol series for the similar transformation of **I** into **II**.[30]



Scheme 2

Conclusion

The pseudodisaccharide **III** has previously been synthesised by us.[14] To further investigate the relationship between the natural type P-IPGs and **III**, a more effective synthetic approach to this molecule was needed that avoided the previous multistep sequence to building block **8** and afforded **III** in sufficient amount for biological investigation. This has now been achieved starting from D-*chiro*-inositol (**1**) using the synthetic sequence in Scheme 1 that proceeds through a key bis- protection of the *trans*-equatorially oriented hydroxyl group in a good yield. The difficult glycosylation of the axially-oriented hydroxyl group in **8** proceeds with reasonable yield and selectivity using the trichloroacetimidate method (Scheme 2) to give the pseudodisaccharides **11** and **12**, from which both pseudodisaccharide **III** and the corresponding cyclic phosphate **IV** have been prepared effectively.

This new synthetic approach to **III** and **IV** presents the additional advantage that both D- and L-*chiro*-inositol containing IPG-like molecules may be prepared depending on the configuration of *chiro*-inositol used as starting material, and this can be of some importance in future investigations on the key enzymatic cleavage of glycolipid precursors to generate IPG-mediators.[18]

Experimental Section

General Remarks: All reactions were conducted under an atmosphere of dry argon using oven-dried glassware and freshly distilled and dried solvents. THF and diethyl ether were distilled from sodium benzophenone ketyl. Dichloromethane and acetonitrile were distilled from calcium hydride. – TLC was performed on silica gel GF_{254} (Merck) with detection by charring with phosphomolybdic acid/EtOH. – For flash chromatography, silica gel (Merck 230–400 mesh) was used. Columns were eluted with positive air pressure. Chromatographic eluents are given as volume to volume ratios (v/v). – NMR spectra were recorded with a Bruker Avance DPX_{300} (^{1}H, 300 MHz), Bruker Avance DRX_{400} (^{1}H, 400 MHz), and Bruker Avance DRX_{500} (^{1}H, 500 MHz) spectrometers. Chemical shifts are reported in ppm, and coupling constants are reported in Hz. Routine spectra were referenced to the residual proton or carbon signals of the solvent. – High-resolution mass spectra were recorded on a Kratos MS-80RFA 241-MC apparatus. – Optical rotations were determined with a Perkin–Elmer 341 polarimeter. – Elemental analyses were recorded on a Leco CHNS-932 apparatus. The organic extracts were dried over anhydrous sodium sulfate and concentrated in vacuo.

2,3:4,5-Bis-*O*-(tetraisopropyldisiloxane-1,3-diyl)-D-*chiro*-inositol(2): D-*Chiro*-inositol (1 g, 6.55 mmol) was dissolved in dimethyl formamide (11 mL) with imidazole (1.5 g, 22 mmol) and a catalytic amount of 4-(dimethylamino)pyridine (274 mg, 2.22 mmol) and treated with $TIPDSCl_2$ (4.6 mL, 13.89 mmol) dropwise. After 3.5 h the solution was diluted with ethyl acetate and washed successively with saturated ammonium chloride, water, and brine, dried over sodium sulfate, concentrated, and the residue purified by column chromatography (hexane/ethyl acetate 20:1) to give **2** (2.79 g, 76%) as a noncrystalline monohydrate. – R_f (Hex/Ac 15:1) = 0.38. – $[\alpha]_D$ = –5.9 (c = 0.87, $CHCl_3$). – ^{1}H NMR ($CDCl_3$, 500 MHz): δ = 0.90–1.11 (m, 56 H, *i*Pr), 2.58 (s, 2 H, OH), 3.83–3.87 (m, 2 H), 3.95–3.98 (m, 2 H), 4.03–4.06 (bd, 2 H). – ^{13}C NMR ($CDCl_3$, 125 MHz): δ = 11.9, 12.0, 12.8, 12.9, 17.1, 17.2, 17.2, 17.3, 17.4, 17.4, 17.5, 71.8, 74.9, and 76.5. – HRMS calcd. for $C_{30}H_{65}O_8Si_4$ (M^+) 665.375658; found 665.371069. – $C_{30}H_{64}O_8Si_4 \cdot H_2O$ (678.392): calcd. C 52.74, H 9.73; found C 52.70, H 9.56.

1-D-1,2-Anhydro-3,4:5,6-bis-*O*-(tetraisopropyldisiloxane-1,3-diyl)-*myo*-inositol (3): Compound **2** (2.54 g, 3.89 mmol) was dissolved in THF (64 mL) with triphenylphosphane (5.07 g, 19.14 mmol) and treated at room temperature with DEAD (2.7 mL, 17.22 mmol) dropwise. After 7 h the solvent was evaporated and the residue was purified by column chromatography [hexane, hexane/dichloromethane, (10:1–4:1)] to give pure **3** (2 g, 80%) as a colourless glass. – R_f (Hex/Ac 40:1) = 0.38. – $[\alpha]_D$ = +10.2 (c = 1.23, $CHCl_3$). – ^{1}H NMR ($CDCl_3$, 500 MHz): δ = 0.89–1.09 (m, 56 H, *i*Pr),3.14 (d, J = 3.7 Hz, 1 H), 3.33 (dd, J = 1.9 Hz, J = 2.8 Hz, 1 H), 3.52 (dd, J = 9.9 Hz, J = 7.1 Hz, 1 H), 3.61 (dd, J = 7.7 Hz, J = 9.8 Hz, 1 H), 3.97 (d, J = 7.1 Hz, 1 H), 4.04 (dd, J = 1.8 Hz, J = 7.7 Hz, 1 H). – ^{13}C NMR ($CDCl_3$, 125 MHz): δ = 12.1, 12.7, 12.7, 12.8, 13.0, 17.0, 17.1, 17.1, 17.1, 17.2, 17.2, 17.3, 17.3, 17.3, 17.4, 17.5, 17.6, 56.2, 57.4, 73.4, 74.5, 75.4, 79.0. – HRMS calcd. for $C_{30}H_{63}O_7Si_4$ (M^+) 647.365094; found 647.362811.

1-*O*-Allyl-3,4:5,6-bis-*O*-(tetraisopropyldisiloxane-1,3-diyl)-D-*chiro*-inositol (4): Compound **3** (2.2 g, 3.4 mmol) was dissolved in dichloromethane (30 mL) with allyl alcohol (1.5 mL, 22.1 mmol) then treated at room temperature with boron trifluoride·diethyl ether (1.5 mL, 11.9 mmol). After 3 h the reaction was stopped with triethylamine, the solvent evaporated and the residue purified by column chromatography (hexane/ethyl acetate, 70:1) to give pure **4** (1.84 g, 77%) as a syrup. – R_f (Hex/Ac 20:1) = 0.20. – $[\alpha]_D$ = –8.0 (c = 0.55, $CHCl_3$). – ^{1}H NMR ($CDCl_3$, 500 MHz): δ = 0.85–1.20 (m, 56 H, *i*Pr), 2.56 (s, 1 H, OH), 3.76–3.80 (m, 2 H), 3.90–3.93 (m, 3 H), 3.99 (dd, J = 2.9 Hz, J = 9.2 Hz, 1 H), 4.12 (dd, J = 13.3 Hz, J = 5.8 Hz, 1 H, allyl), 4.34 (dd, J = 4.9 Hz, J = 13.3 Hz, 1 H, allyl), 5.11 (dd, J = 1.3 Hz, J = 10.4 Hz, 1 H, allyl), 5.21 (dd, J = 1.6 Hz, J = 17.2 Hz, 1 H, allyl), 5.82–5.90 (m, 1 H, allyl). – ^{13}C NMR ($CDCl_3$, 125 MHz): δ = 11.9, 12.0, 12.2, 12.7, 12.8, 12.8, 12.9, 17.1, 17.2, 17.3, 17.3, 17.4, 17.4, 17.5, 17.5, 71.8, 73.7, 74.7, 75.2, 76.3, 78.6, 116.1, 135.5. – HRFABMS calcd. for $C_{33}H_{69}O_8Si_4$: 705.4406958; found 705.411590.

6-*O*-Allyl-1-*O*-methoxymethyl-2,3:4,5-bis-*O*-(tetraisopropyldisiloxane-1,3-diyl)-D-*chiro*-inositol (5): MOMCl (1.125 mL, 14.8 mmol) in dioxane (3.5 mL) was treated with sodium iodide (1.78 g, 11.8 mmol) and the mixture was stirred for ten minutes at room temperature. Compound **4** (1.67 g, 2.36 mmol) dissolved in dioxane (1.5 mL) with *N*,*N*-diisopropylethylamine (2.76 mL, 16.34 mmol) was added to the above solution and the mixture was heated at 85° C overnight. The solution was cooled to room temperature, diluted with dichloromethane, washed successively with saturated aqueous sodium bicarbonate, water, and brine, and concentrated. The residue was purified by column chromatography (hexane/ethyl acetate, 50:1) to give pure **5** (1.57 g, 90%) as a syrup. – R_f (Hex/Et_2O, 50:1) = 0.21. – $[\alpha]_D$ = –36.2 (c = 0.71, $CHCl_3$). – ^{1}H NMR ($CDCl_3$, 500 MHz): δ = 0.89–1.07 (m, 56 H, *i*Pr), 3.34 (s, 3 H, MOM), 3.63 (t, J = 3.6 Hz, 1 H), 3.79–3.88 (m, 4 H), 3.95 (dd, J = 2.7 Hz, J = 8.9 Hz, 1 H), 4.08–4.12 (m, 1 H, allyl), 4.31–4.36 (m, 1 H, allyl), 4.74 (dd, 2 H, MOM), 5.11 (dd, J = 1.5 Hz, J = 10.4 Hz, 1 H, allyl), 5.20–5.24 (m, 1 H, allyl), 5.82–5.90 (m, 1 H, allyl). – ^{13}C NMR ($CDCl_3$, 125 MHz): = δ 12.0, 12.1, 12.2, 12.7, 12.7, 12.8, 12.8, 17.1, 17.3, 17.4, 17.4, 17.4, 17.4, 17.5, 55.3, 73.3, 74.4, 75.0, 76.9, 77.0, 79.0, 97.7, 115.9, 135.5. – HRFABMS

calcd. for $C_{35}H_{72}NaO_9Si_4$ (M^+) 771.415118; found 771.414410. – $C_{35}H_{72}O_9Si_4$: calcd. C 56.10, H 9.68; found C 55.61, H 9.98.

2,3,4,5-Tetra-*O*-acetyl-6-*O*-allyl-1-*O*-methoxymethyl-D-*chiro*-inositol (6): To a solution of **5** (1.17 g, 1.56 mmol, 1 equiv.) in THF (22 mL) at room temperature was added TBAF (1M solution in THF, 6.25 mL, 6.25 mmol, 4 equiv.). After 2 h the reaction mixture was cooled to 0 °C, treated with pyridine (40 mL) and acetic anhydride (18 mL). The mixture was stirred for 18 h at room temperature, poured into a mixture of ice/water and extracted with EtOAc. The organic layer was washed with brine and dried over sodium sulfate. The residue was purified by column chromatography to obtain **6** as a syrup (675 mg, quantitative). – R_f (Hex/Ac, 2:1) = 0.34. – $[\alpha]_D$ = +31.9 (c = 0.75, $CHCl_3$). – ^{1}H NMR ($CDCl_3$, 300 MHz): δ = 1.99 (s, 6 H, Ac), 2.03 (s, 3 H, Ac), 2.04 (s, 3 H, Ac), 3.39 (s, 3 H, MOM), 3.91 (dd, J = 3.1 Hz, J = 4.3 Hz, 1 H), 4.07–4.13 (m, 3 H, 2 allyl), 4.62 (dd, 2 H, MOM), 5.16–5.30 (m, 4 H, 2allyl, H-5, H-2), 5.40–5.47 (m, 2 H, H-3, H-4), 5.79–5.90 (m, 1 H, allyl). – ^{13}C NMR ($CDCl_3$, 500 MHz): δ = 20.6, 20.8, 20.9, 70.3, 70.4, 70.6, 71.2, 72.6, 73.7, 75.1, 97.4, 117.9, 134.0, 169.1, 169.3, 169.4. – HRMS calcd. for $C_{19}H_{29}O_{11}$ (M^+): 433.170987; found 433.170514. – $C_{19}H_{28}O_{11}$: calcd. C 52.77, H 6.53; found C 52.61, H 6.65.

6-*O*-Allyl-2, 3,4,5-tetra-*O*-benzyl-1-*O*-methoxymethyl-D-*chiro*-inositol (7). - From Compound 5: Compound **5** (318 mg, 0.425 mmol) was dissolved in THF (4.25 mL) then treated with TBAF (1 M solution in THF, 0.85 mL, 0.85 mmol). After 30 min, DMF (7 mL) was added followed by sodium hydride (136 mg, 3.4 mmol) and benzyl bromide (303 mL, 2.55 mmol). As no benzylation reaction took place, the mixture was filtered over silica gel and benzylated in the above conditions at 45 °C for 18 h. The mixture was diluted with ethyl acetate, washed with saturated aqueous ammonium chloride, water, and brine, concentrated and the residue was purified by column chromatography (hexane/ethyl acetate, 6:1) to give pure **7** (102 mg, 38%). **– From Compound 6:** To a solution of **6** (650 mg, 1.50 mmol) in MeOH (30 mL) was added a 0.95M solution of NaOMe in MeOH (1.3 mL, 1.20 mmol, 0.8 equiv.) at room temperature. After 30 min the solvent was evaporated and the crude coevaporated twice with toluene. The crude was dissolved in DMF (21 mL) with NaH (481 mg, 12.03 mmol, 8 equiv.) and treated with benzyl bromide (1.07 mL, 9.03 mmol, 6.0 equiv.). The reaction was stirred overnight and stopped with methanol, diluted with EtOAc, washed with water, brine and dried over sodium sulfate. The residue was purified by column chromatography (hexane/diethyl ether, 1:2) to give pure **7** as a syrup (834 mg, 89%). – R_f (Hex/Ac, 10:1) = 0.12. – $[\alpha]_D$ = –49.1 (c = 0.99, $CHCl_3$). – ^{1}H NMR ($CDCl_3$, 500 MHz): δ = 3.18 (s, 3 H, MOM), 3.68–3.70 (m, 1 H), 3.78–3.88 (m, 5 H), 3.93–3.98 (m, 1 H, allyl), 4.12–4.16 (m, 1 H, allyl), 4.54–4.91 (m, 10 H, CH_2Ph, MOM), 5.11–5.13 (m, 1 H, allyl), 5.16–5.20 (m, 1 H, allyl), 5.78–5.83 (m, 1 H, allyl), 7.22–7.35 (m, 20 H, ArH). – ^{13}C NMR ($CDCl_3$, 125 MHz): δ = 55.6, 72.2, 73.3, 73.4, 74.0, 75.7, 75.9, 79.3, 79.8, 82.1, 82.1, 97.3, 117.0, 127.4, 127.4, 127.6, 127.7, 127.9, 127.9, 128.0, 128.0, 128.1, 128.3, 128.3, 128.3, 128.4, 135.0, 138.5, 138.7, 139.1, 139.2. – $C_{39}H_{44}O_7$ (624.781): calcd. C 74.97, H 7.10; found C 75.49, H 7.05.

1-*O*-Allyl-2,3,4,5-tetra-*O*-benzyl-D-*chiro*-inositol (8): Compound **7** (730 mg, 1.17 mmol, 1.0 equiv) dissolved in dichloromethane (11.7 mL) was treated with thiophenol (0.156 mL, 1.52 mmol) and boron trifluoride·diethyl ether (0.148 mL, 1.17 mmol) at –10 °C. After 30 min the reaction was quenched with saturated aqueous sodium bicarbonate, extracted with dichloromethane, dried over sodium sulfate and evaporated. The residue was purified by flash column chromatography (hexane/ethyl acetate, 7:1) to give pure **8** (584 mg, 86%) as a syrup. – R_f (Hex/Ac, 5:1) = 0.27. – $[\alpha]_D$ = –1.4 (c = 0.95, $CHCl_3$). – ^{1}H NMR ($CDCl_3$, 500 MHz): δ = 2.40 (s, 1 H, OH), 3.73–3.92 (m, 5 H), 3.99–4.03 (m, 2 H, H-6, allyl), 4.18–4.22 (m, 1 H, allyl), 4.62–4.91 (m, 8 H, CH_2Ph), 5.13 (dd, J = 1.4 Hz, J = 10.4 Hz, 1 H, allyl), 5.18–5.22 (m, 1 H, allyl), 5.80–5.86 (m, 1 H, allyl), 7.23–7.35 (m, 20 H, ArH). – ^{13}C NMR ($CDCl_3$, 125 MHz): δ = 68.2, 72.6, 73.3, 73.3, 75.6, 75.8, 75.9, 79.9, 80.3, 80.8, 81.6, 82.0, 116.9, 127.4, 127.5, 127.5, 127.7, 127.7, 127.7, 127.7, 127.9, 127.9, 127.9, 128.0, 128.3, 128.3, 128.3, 128.3, 128.4, 128.4, 128.5, 135.1, 138.2, 138.7, 139.1, 139.0, 139.1. – $C_{37}H_{40}O_6$ (550.727): calcd. C 76.53, H 6.94; found C 76.64, H 6.61.

1-*O*-(3,4,6-Tri-*O*-acetyl-2-azido-2-deoxy-α-D-glucopyranosyl)-6-*O*-allyl-2,3,4,5-tetra-*O*-benzyl-D-*chiro*-inositol (11): A mixture of compound **8** (197 mg, 0.339 mmol), compound **9** (490 mg, 1.04 mmol) and 4 Å molecular sieves in CH_2Cl_2 (3 mL) was stirred at room temperature for 45 min, then TMSOTf (0.21 M solution in CH_2Cl_2, 160 μL, 0.034 mmol) was added. After 1 h the reaction mixture was quenched with Et_3N, filtered over a pad of celite and evaporated to dryness. Column chromatography (cyclohexane/EtOAc, 3:1) afforded 133 mg (44%) of **11α** and 93 mg (31%) of **11β**.

11α: ^{1}H NMR ($CDCl_3$, 500 MHz): δ = 1.99 (s, 3 H, Ac), 2.00 (s, 3 H, Ac), 2.08 (s, 3 H, Ac), 3.55 (dd, J = 3.7 Hz, J = 10.4 Hz, 1 H, H-2′), 3.58 (dd, J = 2.0 Hz, J = 10.7 Hz, 1 H, H-6′), 3.70–3.71 (m, 1 H), 3.81–3.91 (m, 5 H, H-6′), 3.97–4.01 (m, 1 H, allyl), 4.20–4.24 (m, 1 H, allyl), 4.33–4.37 (m, 1 H, H-5′), 4.78 (d, J = 3.6 Hz, 1 H, H-1′), 4.59–4.94 (m, 8 H, CH_2Ph), 4.98 (t, J = 10.2 Hz, J = 9.5 Hz, 1 H, H-4′), 5.14–5.17 (m, 1 H, allyl), 5.18–5.22 (m, 1 H, allyl), 5.37 (t, J = 9.4 Hz, J = 10.2 Hz, 1 H, H-3′), 5.78–5.85 (m, 1 H, allyl), 7.24–7.36 (m, 20 H, ArH). – ^{13}C NMR ($CDCl_3$, 125 MHz): δ = 68.2, 72.6, 73.3, 73.3, 75.6, 75.8, 75.9, 79.9, 80.3, 80.8, 81.6, 82.0, 116.9, 127.4, 127.5, 127.5, 127.7, 127.7, 127.7, 127.7, 127.9, 127.9, 127.9, 128.0, 128.3, 128.3, 128.3, 128.3, 128.4, 128.4, 128.5, 135.1, 138.2, 138.7, 139.1, 139.0, 139.1.

11β: ^{1}H NMR ($CDCl_3$, 500 MHz): δ = 1.99 (s, 3 H, Ac), 2.00 (s, 3 H, Ac), 2.08 (s, 3 H, Ac), 3.33 (dd, J = 8.1 Hz, J = 10.0 Hz, 1 H, H-2′), 346–3.50 (m, 1 H, H-5′), 3.74–3.79 (m, 1 H), 3.80–3.86 (m, 3 H), 3.91–3.98 (m, 1 H, allyl), 406–4.16 (m, 5 H, H-6a′, H6b′, allyl), 4.53 (d, J = 8.0 Hz, 1 H, H-1′), 4.58 (d, 1 H, CH_2Ph), 4.65 (dd, 2 H, CH_2Ph), 4.78–4.91 (m, 7 H, H-3′, H-4′, CH_2Ph), 5.09–5.19 (m, 2 H, allyl), 5.75–5.83 (m, 1 H, allyl), 7.22–7.35 (m, 20 H, ArH).

6-*O*-Allyl-1-*O*-(3,4,6-tri-*O*-benzyl-2-azido-2-deoxy-α-D-glucopyranosyl)-2,3,4,5-tetra-*O*-benzyl-D-*chiro*-inositol (12α). – From Compound 11α: To a solution of **11α** (140 mg, 0.156 mmol) in methanol (3.1 mL) was added sodium methylate (0.95 M, 0.165 mL) at room temperature. The reaction mixture was stirred for 10 min and then neutralised with Amberlite resin IR-120 (H^+). After filtration and evaporation of the solvent, the residue was dissolved in DMF (2.3 mL), and treated with NaH (60% in oil, 39 mg, 0.97 mmol, 6 equiv.) and benzyl bromide (87 μL, 0.73 mmol). The reaction mixture was stirred for 1 h, quenched with MeOH at 0 °C and diluted with dichloromethane. The organics were washed with saturated aqueous sodium bicarbonate solution, dried over sodium sulfate, evaporated under vacuum and the residue purified by flash column chromatography to give pure **12α** (161 mg, 99%). – **From Compounds 8 and 10:** A mixture of compound **8** (73 mg, 0.125 mmol) and compound **10** (49 mg, 0.078 mmol) was coevaporated two times with toluene, dissolved in dichloromethane (1 mL) and treated with TMSOTf (0.1 M solution in CH_2Cl_2, 50 μL) at –25° C. After 30 minutes at –25° C, the mixture was allowed to warm during 10 minutes, then stopped by addition of triethylamine

and evaporated under vacuo. The residue was purified by flash column chromatography [hexane/ethyl acetate, (7:1–3:1)] to give 35 mg (35%) of **12α** and 25 mg (25%) of **12β** as syrups.

12α: R_f (Hex/Ac, 4:1) = 0.44. – $[\alpha]_D$ = +40.7 (c = 0.11, $CHCl_3$). – ^{1}H NMR ($CDCl_3$, 500 MHz): δ = 3.13 (dd, J = 1.7 Hz, J = 10.9 Hz, 1 H, H-6′), 3.35 (dd, J = 2.3 Hz, J = 10.9 Hz, 1 H, H-6′), 3.48 (dd, J = 3.6 Hz, J = 10.1 Hz, 1 H, H-2′), 3.71 (t, J = 9.5 Hz, 1 H, H-4′), 3.74–3.88 (m, 5 H, H-3′), 3.94–4.02 (m, 3 H, allyl), 4.10–4.13 (m, 1 H, H-5′), 4.18–4.21 (m, 1 H, allyl), 4.24–4.92 (m, 14 H, CH_2Ph), 4.75 (d, J = 3.6 Hz, 1 H, H-1′), 5.14 (dd, J = 1.2 Hz, J = 10.4 Hz, 1 H, allyl), 5.18 (dd, J = 1.5 Hz, J = 17.2 Hz, 1 H, allyl), 5.77–5.85 (m, 1 H, allyl), 7.05–7.48 (m, 35 H, ArH). – ^{13}C NMR ($CDCl_3$, 125 MHz): δ = 63.8, 67.6, 70.7, 72.5, 73.1, 73.3, 73.5, 73.6, 74.2, 74.9, 75.5, 75.8, 76.1, 78.2, 78.3, 80.0, 80.5, 81.8, 82.0, 96.9, 117.3, 127.4, 127.4, 127.5, 127.7, 127.8, 127.9, 128.0, 128.0, 128.0, 128.1, 128.1, 128.1, 128.2, 128.3, 128.3, 128.3, 128.4, 128.4, 128.5, 134.9, 137.8, 137.9, 138.1, 138.6, 138.8, 139.0, 139.0. – HRFABMS calcd. for $C_{64}H_{67}N_3NaO_{10}$ (M + Na^+): 1060.472416; found 1060.476982. – $C_{64}H_{67}N_3O_{10}$ (1037.464): calcd. C 74.04, H 6.50, N 4.05; found C 73.54, H 6.88, N 4.10.

12β: ^{1}H NMR ($CDCl_3$, 500 MHz): δ = 3.29–3.38 (m, 3 H, H-2′, H-3′, H-5′), 3.55 (t, J = 9.2 Hz, 1 H, H-4′), 3.63 (d, 2 H, H-6a′, H-6b′), 3.79–3.86 (m, 3 H), 3.88–3.95 (m, 3 H, allyl), 4.00 (t, J = 3.9 Hz, J = 3.1 Hz, 1 H), 4.09–4.14 (m, 1 H, allyl), 4.45–4.55 (m, 4 H, H-1′, CH_2Ph), 4.60–4.68 (m, 3 H, CH_2Ph), 4.76, 4.92 (m, 8 H, CH_2Ph), 5.05–5.16 (m, 2 H, allyl), 5.73–5.81 (m, 1 H, allyl), 7.14–7.38 (m, 35 H, ArH).

1-*O*-(2-Azido-2-deoxy-3,4,6-tri-*O*-benzyl-α-D-glucopyranosyl)-2,3,4,5-tetra-*O*-benzyl-D-*chiro*-inositol (13): A solution of sodium acetate (89.11 mg, 1.09 mmol, 8 equiv.) in 20:1 acetic acid-water (3.4 mL) was degassed under argon then added to **12α** (141 mg, 0.135 mmol, 1 equiv) and palladium chloride (73.24 mg, 0.407 mmol, 3 equiv.). The reaction mixture was stirred for 6 h 30 min, diluted with dichloromethane, filtered over celite, neutralised with sodium bicarbonate and washed successively with water and brine. The organic layer was dried over sodium sulfate, evaporated under vacuo and the residue was purified by flash column chromatography (hexane/ethyl acetate, 5:1) to give pure **13** (100 mg, 74%) as a syrup. – ^{1}H NMR ($CDCl_3$, 500 MHz): δ = 2.46 (s, 1 H, OH), 3.06 (dd, J = 1.9 Hz, J = 11.0 Hz, 1 H, H-6′), 3.30 (dd, J = 2.4 Hz, J = 11.0 Hz, 1 H, H-6′), 3.47 (dd, J = 3.6 Hz, J = 10.1 Hz, 1 H, H-2′), 3.71 (Ψt, J = 9.2 Hz, J = 10.0 Hz, 1 H, H-4′), 3.77 (Ψt, J = 8.7 Hz, J = 9.3 Hz, 1 H, H-3′), 3.80–3.87 (m, 3 H, H-3, H-4, H-5), 3.94 (dd, J = 9.1 Hz, J = 3.2 Hz, 1 H, H-2), 4.04 (t, J = 3.6 Hz, 1 H, H-1), 4.09–4.12 (m, 1 H, H-5′), 4.16 (Ψt, J = 3.6 Hz, J = 3.2 Hz, 1 H H-6), 4.22 (d, 1 H, CH_2Ph), 4.41–4.47 (2d, 2 H, CH_2Ph), 4.81 (d, J = 3.6 Hz, 1 H, H-1′), 4.63–4.94 (m, 11 H, CH_2Ph), 7.08–7.36 (m, 35 H, ArH). – ^{13}C NMR ($CDCl_3$, 125 MHz): δ = 63.8, 67.2, 67.5, 70.7, 73.0, 73.2, 73.4, 74.8, 74.9, 75.4, 75.7, 76.1, 78.2, 78.3, 80.0, 80.4, 81.2, 81.8, 97.2, 127.3, 127.3, 127.5, 127.6, 127.7, 127.7, 127.8, 127.9, 128.0, 128.1, 128.3, 128.3, 128.4, 128.4, 128.5, 128.5, 128.9, 138.1, 138.8. – HRFABMS calcd. for $C_{61}H_{63}N_3NaO_{10}$ (M + Na^+): 1020.441116; found 1020.448025.

6-*O*-(2-Azido-2-deoxy-3,4,6-tri-*O*-benzyl-α-D-glucopyranosyl)-2,3,4,5-tetra-*O*-benzyl-1-*O*-(dibenzyloxyphosphoryl)-D-*chiro*-inositol (14): Compound **13** (42 mg, 0.042 mmol) was dissolved under argon in a 1:1 mixture of dichloromethane/acetonitrile (1 mL) then treated with dibenzyl *N,N*-diisopropylphosphoramidite (42 μL, 0.095 mmol) and tetrazol (13.4 mg, 0.189 mmol). The reaction mixture was stirred for 1.5 h at room temperature, then treated with *tert*-butyl hydroperoxide at 0 °C. After 40 minutes, the solvents were evaporated and the residue purified by column chromatography (hexane/ethyl acetate, 3:1) to give pure **14** (48 mg, 91%) as a syrup. – ^{1}H NMR ($CDCl_3$, 500 MHz): δ = 3.10 (dd, J = 1.7 Hz, J = 11.0 Hz, 1 H, H-6′), 3.36 (dd, J = 2.4 Hz, J = 10.9 Hz, 1 H, H-6′), 3.51 (dd, J = 3.6 Hz, J = 10.1 Hz, 1 H, H-2′), 3.70–3.79 (m, 4 H, H-4′, H-3, H-4, H-5), 3.82 (t, J = 9.8 Hz, J = 9.3 Hz, 1 H, H-3′), 4.03–4.07 (m, 2 H, H-2, H-5′), 4.15 (t, J = 3.6 Hz, 1 H, H-6), 4.28 (d, 1 H, CH_2Ph), 4.45–5.01 (m, 9 H, CH_2Ph, H-1, H-1′), 7.10–7.40 (m, 45 H, ArH). – ^{13}C NMR ($CDCl_3$, 125 MHz): δ = 63.8, 67.6, 69.2, 69.3, 69.3, 69.5, 69.6, 71.0, 72.6, 72.8, 73.5, 74.3, 75.5, 76.1, 77.8, 77.9, 77.9, 78.1, 80.6, 81.0, 81.3, 97.6, 127.3, 127.4, 127.5, 127.6, 127.7, 127.8, 127.8, 127.8, 127.9, 127.9, 128.0, 128.1, 128.2, 128.2, 128.3, 128.3, 128.4, 128.4, 128.4, 128.5, 128.6, 128.6, 130.2, 137.8, 137.9, 138.0, 138.1, 138.3, 138.7, 138.8. – ^{31}P NMR ($CDCl_3$, 202 MHz): δ = –2.18.

6-*O*-(2-Amino-2-deoxy-α-D-glucopyranosyl)-D-*chiro*-inositol 1-Phosphate (III): Compound **14** (57 mg, 0.045 mmol) was dissolved in methanol (5.3 mL), buffer sodium acetate-acetic acid pH 5 (5.3 mL) and stirred vigorously under hydrogen with 10% Pd/C (70 mg). After the reaction was conducted overnight, the catalyst was filtered off and the filtrate was concentrated. The residue was dissolved in water, filtered over a Sep-pack C-18 and passed through a column of Sephadex G-10 (EtOH/H_2O 10%) to give **III** (13.8 mg, 73%). – ^{1}H NMR (D_2O, 500 MHz): δ = 3.41 (dd, J = 3.7 Hz, J = 10.7 Hz, 1 H, H-2′), 3.56–3.65 (m, 2 H, H-4′, H-4), 3.72–3.76 (m, 2 H, H-2, H-3), 3.85 (dd, J = 4.4 Hz, J = 12.5 Hz, 1 H, H-6′), 3.89 (dd, J = 2.5 Hz, J = 12.5 Hz, 1 H, H-6′), 3.95 (dd, J = 9.2 Hz, J = 10.6 Hz, 1 H, H-3′), 4.02 (dd, J = 3.4 Hz, J = 10.4 Hz, 1 H, H-5), 4.11 (ddd, J = 2.5 Hz, J = 4.2 Hz, J = 10.0 Hz, 1 H, H-5′), 4.31 (t, J = 3.8 Hz, 1 H, H-6), 4.51–4.54 (m, 1 H, H-1), 5.40 (d, J = 3.7 Hz, 1 H, H-1′). – ^{31}P NMR (D_2O, 202 MHz): δ = +3.00.

6-*O*-(2-Amino-2-deoxy-α-D-glucopyranosyl)-D-*chiro*-inositol-1,2-cyclic Phosphate (IV): Compound **III** (3.5 mg, 8.31 μmol) was dissolved in H_2O (210 μL) and 1-(3-dimethylaminopropyl)-3-ethylcarbodiimide hydrochloride (41 mg) was added in three times during 2 h 30 min. The reaction mixture was filtered over a Sep-pack C-18 and passed through a column Sephadex G-10 (EtOH/H_2O 10%) to give **IV** (3.4 mg, 97%). – ^{1}H NMR (D_2O, 500 MHz): δ = 3.26–3.28 (m, 1 H, H-2′), 3.57 (t, J = 9.6 Hz, 1 H, H-4′), 3.75 (dd, J = 7.8 Hz, J = 8.9 Hz, 1 H, H-4), 3.84–3.92 (m, 4 H, H-3, H-3′, H-6a′, H-6b′), 3.99 (dd, J = 3.0 Hz, J = 7.7 Hz, 1 H, H-5), 4.05–4.09 (m, 1 H, H-5′), 4.38 (t, J = 3.6 Hz, 1 H, H-6), 449–4.56 (m, 1 H, H-2), 4.75 (t, J = 5.1 Hz, 1 H, H-1), 5.36 (d, J = 3.4 Hz, 1 H, H-1′). – ^{31}P NMR (D_2O, 202 MHz): δ = +15.24.

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