Asymmetric Total Synthesis of D-*myo*-Inositol 1,2,4,5-Tetrakisphosphate and Its P-2-(*O*-Aminopropyl) Derivative

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The second messenger D-*myo*-inositol 1,4,5-trisphosphate (Ins(1,4,5)P_3 or IP_3) interacts stereospecifically with membrane receptors (IP_3R) to promote the release of Ca^{2+} from intracellular stores.[1−3] Receptor proteins that recognize IP_3 and mediate its role in calcium mobilization have been characterized and purified.[4−6] Considerable effort has been invested in defining the affinity of various IP_3R subtypes and of IP_3 analogues.[7−15] Recently, an IP_3-gated calcium channel protein from rat olfactory cilia was found to respond to a 100-fold lower concentration of Ins(2,4,5)-IP_3 relative to Ins(1,4,5)P_3.[16] Moreover, affinity probes based on P-1-tethered IP_3 showed only very modest utility in characterizing these unique vertebrate olfactory IP_3Rs (D. L. Kalinoski, D. Restrepo, and G. D. Prestwich, unpublished results). Thus, we devised a hybrid ligand possessing both Ins(1,4,5)P_3 and Ins(2,4,5)P_3 recognition elements. A new affinity probe was also prepared in which the tethering group was attached via a phosphodiester linkage from the P-2 phosphate moiety. We now describe the synthesis of optically-active D-*myo*-Ins(1,2,4,5)P_4 and its *P*-2-(*O*-aminopropyl)phosphodiester derivatives.

The synthesis of 2-*O*-(3-aminopropyl)Ins(1,2,4,5)P_4 (**17**) uses a modification[11] of the Ferrier rearrangement[17] to establish the optically-pure D-*myo*-inositol skeleton. The synthetic strategy is summarized in Scheme 1. Thus, selective protection of C-4 and C-6 hydroxyls of the methyl α-D-glucopyranoside (**1**) provides α-D-glucopyranoside **2**.[18] Selective monobenzylation (37% isolated yield) of the C-2 hydroxyl[19] and then *p*-methoxybenzylation (PMB) of the remaining hydroxyl group (75% yield) gave the fully protected ether **4**. Regioselective ring opening of the *p*-methoxybenzylidene acetal using DIBAL-H at −40 °C then gave compound **5** in 77% yield with >90% regioselectivity.[20] Swern oxidation[21] of the alcohol gave an aldehyde (>90% hydrated), which was converted (Ac_2O, CH_3CN, K_2CO_3)[22] in 79% yield directly to the 2-enol acetate **6**. Ferrier rearrangement[17] of **6** using mercuric acetate, followed by treatment with aqueous sodium chloride resulted in the desired axial 2-hydroxyl stereochemistry in 59% yield. Stereoselective reduction of the carbonyl group with sodium triacetoxyborohydride[23] provided the *myo*-inositol skeleton **8** in 55% yield. Basic hydrolysis provided the triol **9** (90%), which was converted (2,2-dimethoxypropane, TsOH) into the thermodynamically stable[24] ketal **10** as the main isomer (85% yield). Benzylation of the remaining free hydroxyl group in the presence of NaH and benzyl bromide in DMF offered the fully protected ether **11** in 80% yield. Removal of the isopropylidene group under very mild conditions (catalytic amount of *p*-TsOH acid in aqueous acetone) at rt for 48 h gave the 1,2-diol in 77% yield.[24] The structure of diol **12** was confirmed by comparison of the proton NMR spectra and melting point and mixed melting point with its racemic form[25] synthesized from 1,2:4,5-bis-*O*-isopropylidene-*myo*-inositol by a known route.[24] Stannylation[26] of two adjacent hydroxyls in the presence of Bu_4NI in toluene followed by *p*-methoxybenzylation resulted in 1,4,5-tri-*O*-*p*-methoxybenzyl-3,6-di-*O*-benzyl-*myo*-inositol (**13**) with the 2-hydroxyl unprotected to allow access to the enantiomerically pure P-2-modified Ins(1,2,4,5)P_4 analogue.

Phosphitylation of the hydroxy group of **13** with benzyloxy 3-(*N*-carboxyamino)propyl diisopropylamino phosphite[13] in the presence of 1*H*-tetrazole and followed by *m*-CPBA oxidation gave the fully protected aminopropyl-tethered inositol **14** in 87% yield. Although DDQ[25] was unable to remove the three PMB groups, ceric ammonium nitrate[27] in acetonitrile/water (9:1) gave the triol **15** in 43% yield. Phosphitylation and oxidation of the triol **15** described for **14** gave the fully protected derivative **16** (76% yield). Hydrogenolysis removed the benzyl groups to provide the optically-active, *P*-2-(*O*-aminopropyl)-tethered D-*myo*-Ins(1,2,4,5)P_4 derivative **17** in essentially quantitative yield after ion-exchange chromatography on Chelex (sodium form).[13]

For biological comparisons, synthesis of untethered optically-pure D-*myo*-1,2,4,5-IP_4 was performed using the

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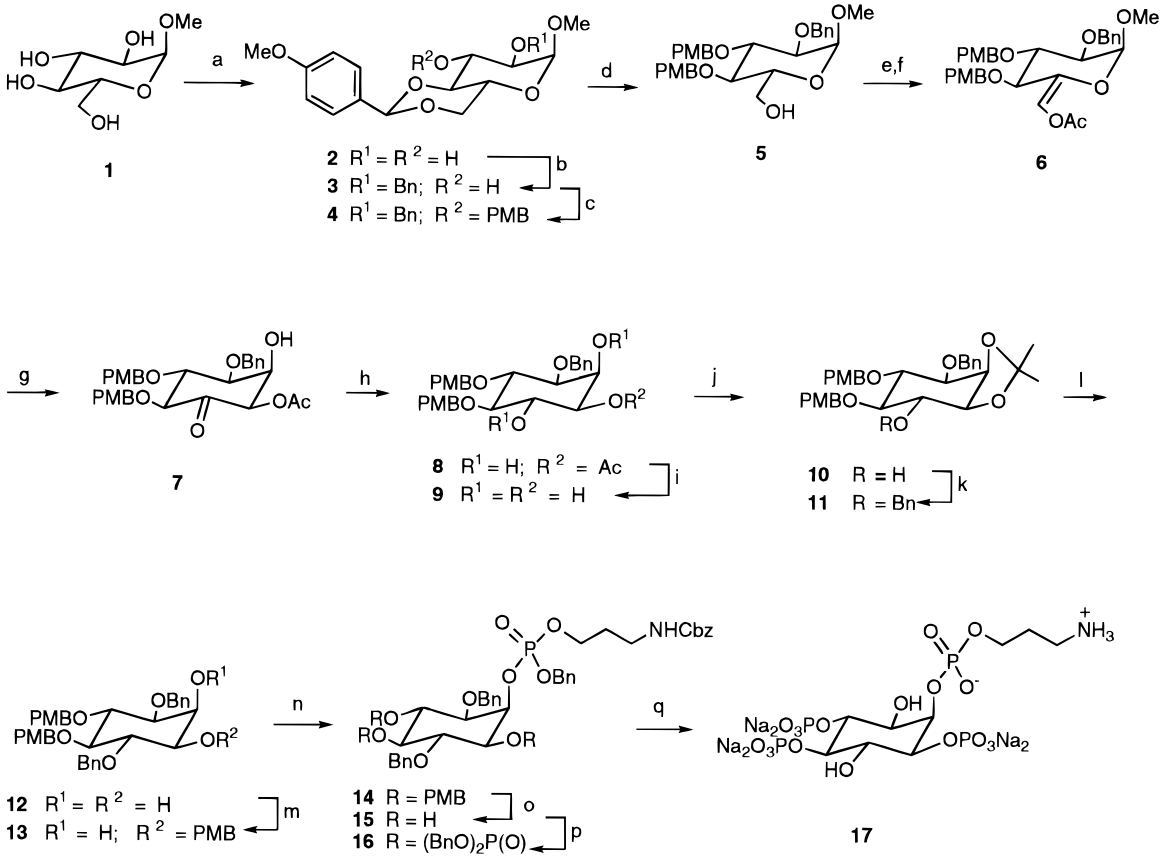
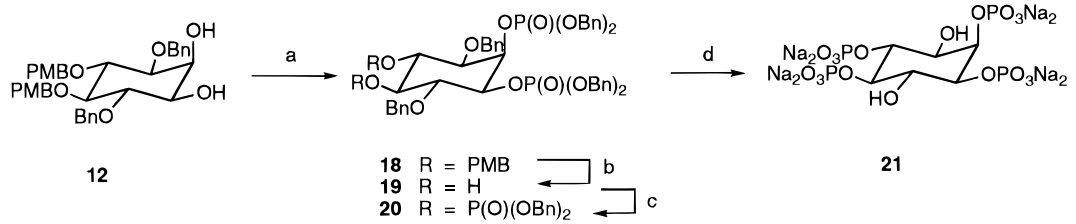
Scheme 1[a]



[a] (a) 4-Methoxybenzaldehyde dimethyl acetal, *p*-TsOH, 50 °C, DMF, 2.5 h; (b) BnBr, CH_2Cl_2, *n*-Bu_4NHSO_4, 10% aqueous NaOH, reflux, 48 h; (c) NaH, PMB-Cl, DMF, 2 h; (d) DIBAL-H, CH_2Cl_2, −40 °C; (e) oxalyl chloride, DMSO, Et_3N, CH_2Cl_2, −78 °C to rt; (f) Ac_2O, K_2CO_3, CH_3CN, 80 °C, 8 h; (g) (1) $Hg(OAc)_2$, 4:1 acetone:water, rt, 30 min, (2) saturated NaCl, rt, 24 h; (h) $NaBH(OAc)_3$, HOAc, CH_3CN, rt, 30 min; (i) NaOH, MeOH, reflux, 2 h; (j) 1% TsOH, 2,2-dimethoxypropane, acetone, rt, 72 h; (k) NaH, BnBr, DMF, rt, 1 h; (l) 1% TsOH, acetone−water (50:1), rt, 48 h; (m) Bu_2SnO, *n*-Bu_4NI, toluene, reflux, 3 h, then PMB-Cl, 1.5 h; (n) (1) (BnO)P(N-*i*-Pr_2)(OC_3H_6NHCbz), 1*H*-tetrazole, CH_2Cl_2, rt, (2) *m*-CPBA, −40 °C to rt, 1 h; (o) $(NH_4)_2Ce(NO_3)_6$, acetonitrile−water (9:1), 2 h; (p) (1) $(BnO)_2$PN-*i*-Pr_2, 1*H*-tetrazole, CH_2Cl_2, rt, (2) *m*-CPBA, −40 °C to rt, 1 h; (q) 10% Pd−C, H_2, 5 atm, 95% EtOH, rt, 6.5 h.

Scheme 2[a]



[a] (a) (1) $(BnO)_2$PN-*i*-Pr_2, 1*H*-tetrazole, CH_2Cl_2, rt, (2) *m*-CPBA, −40 °C to rt, 1 h; (b) $(NH_4)_2Ce(NO_3)_6$, acetonitrile−water (9:1), 2 h; (c) (1) $(BnO)_2$PN-*i*-Pr_2, 1*H*-tetrazole, CH_2Cl_2, rt, (2) *m*-CPBA, −40 °C to rt, 1 h; (d) 10% Pd−C, H_2, 5 atm, 95% EtOH, rt, 6.5 h.

intermediate compound **12** (Scheme 2). Phosphitylation of the diol **12** under similar conditions gave the 1,2-bisphosphate **18** in 80% yield. Removal of the two PMB groups with $(NH_4)_2Ce(NO_3)_6$ resulted in free 4,5-hydroxyls **19** in 64% yield. Further phosphitylation gave fully protected 1,2,4,5-tetrakisphosphate **20** in 90% yield. Hydrogenolytic cleavage of all benzyl groups gave optically-pure *myo*-inositol 1,2,4,5-tetrakisphosphate **21** in essentially quantitative yield. The racemic form of this material has been previously show to have a high affinity for the IP_3 receptor[14] and to mobilize Ca^{2+} in SH-SY5Y cultured cells[14] with approximately 2−3-fold lower potency relative to Ins(1,4,5)P_3. Biological tests of these derivatives in olfactory IP_3 receptor studies will be reported in due course.

Experimental Section

^1H, ^{13}C, and ^{31}P NMR spectra were recorded in $CDCl_3$ on a QE-300 or AC-250 NMR spectrometer and are reported relative to δ(TMS) = 0 ppm. When necessary, solvents and reagents were dried using standard procedures.

Methyl 4,6-*O*-(*p*-methoxybenzylidene)-α-D-glucopyranoside (2) was prepared according to Johansson et al.[18] and recrystallized from EtOAc; mp 197−198 °C.

Methyl 2-*O*-Benzyl-4,6-*O*-(*p*-methoxybenzylidene)-α-D-glucopyranoside (3). To a solution of methyl 4,6-*O*-(*p*-methoxybenzylidene)-α-D-glucopyranoside (**2**, 26.5 g) in 2.65 L of CH_2Cl_2 were added 10.6 g of tetra-*n*-butylammonium hydrogen sulfate and 31.7 mL of benzyl bromide followed by 220 mL of 5% sodium hydroxide. The mixture was refluxed for 48 h. The organic phase was separated, washed with water, dried ($MgSO_4$), and concentrated to a white syrup (120 g). After trituration with hexane and EtOAc in hexane (2:1) to remove unreacted starting

material, the crude product was purified on 1.5 kg of silica gel, using a *tert*-butyl methyl ether−hexane eluent (1:1.5 to 1:1) to give 15.7 g of pure and 9.0 g of partially purified benzyl ether **3** (yield 37%); mp 128−129 °C. ^{1}H NMR (300 MHz, $CDCl_3$) δ: 7.4−7.2 (m, 7H), 6.8 (d, 2H), 5.4 (s, 1H), 4.83−4.69 (m, 2H), 4.61 (d, J = 3.6 Hz, 1H), 4.25 (dd, J = 4.5, 9.8 Hz, 1H), 4.15 (t, J = 9.3 Hz, 1H), 3.83−3.73 (s+m, 4H), 3.71 (dd, J = 10.2 Hz, 1H), 3.51−3.45 (m, 2H), 3.38 (s, 3H), 2.58 (bs, 1H) ppm. ^{13}C NMR (63 MHz, $CDCl_3$) δ: 161.1, 137.3, 129.2, 128.8, 128.2, 127.7, 113.7, 101.9, 98.7, 81.2, 79.6, 76.5, 73.4, 70.3, 69.0, 62.0, 55.4 ppm.

Methyl 2-*O*-Benzyl-3-(*p*-methoxybenzyl)-4,6-*O*-(*p*-methoxybenzylidene)-α-D-glucopyranoside (4). Methyl 3-*O*-benzyl-4,6-*O*-(*p*-methoxybenzylidene)-α-D-glucopyranoside (**3**, 11 g, 27.2 mmol) in 100 mL of DMF was added to a suspension of oil-free sodium hydride (2.127 g, 88.6 mmol, previously washed three times with THF) in 15 mL of DMF followed by addition of 4-methoxybenzyl chloride (6.2 mL, 45.8 mmol) in 3 mL of DMF. The mixture was stirred at 50 °C for 2 h. The usual workup (R_f = 0.63, 50% EtOAc−hexane) gave 10.86 g (76%) of white solid **4**; mp 144−146 °C. ^{1}H NMR (250 MHz, $CDCl_3$) δ: 7.43−7.25 (m, 9H), 6.90, 6.83 (2d, 4H), 5.50 (s, 1H), 4.87−4.63 (m, 4H), 4.58 (d, J = 3.6 Hz, 1H), 4.23 (dd, J = 4.3 Hz, 1H), 4.07 (t, 1H), 3.83−3.75 (s+m, 7H), 3.62−3.43 (m, 3H), 3.37 (s, 3H) ppm. ^{13}C NMR (63 MHz, $CDCl_3$) δ: 162.0, 161.1, 137.7, 129.7, 128.4, 128.1, 127.9, 127.4, 113.7, 113.5, 101.2, 99.3, 82.1, 79.2, 78.3, 75.0, 73.7, 70.3, 69.0, 62.4, 55.2 ppm. Anal. Calcd for $C_{30}H_{34}O_8$: C, 68.95; H, 6.55. Found: C, 68.85; H, 6.19.

Methyl 2-*O*-Benzyl-3,4-di-*O*-(*p*-methoxybenzyl)-α-D-glucopyranoside (5). To a solution of methyl 2-*O*-benzyl-3-(*p*-methoxybenzyl)-4,6-*O*-(*p*-methoxybenzylidene)-α-D-glucopyranoside (**4**, 2.0 g, 3.83 mmol) in 30 mL of dry CH_2Cl_2 at −40 °C was added dropwise 1 M DIBAL-H (18 mL, 18 mmol) in CH_2Cl_2. After stirring for 1 h, the reaction was quenched with methanol. The mixture was kept at 0 °C, and a 0.5 M $NaHSO_4$ solution was added dropwise to decompose the Al alkoxide complex. The usual workup gave 1.54 g of methyl 2-*O*-benzyl-3,4-di-*O*-(*p*-methoxybenzyl)-α-D-gluocopyranoside (**5**, 77% yield, >90% selectivity). ^{1}H NMR (250 MHz, $CDCl_3$) δ: 7.35−7.20 (m, 9H), 6.88, 6.84 (2 × d, 4H), 4.92−4.63 (m, 4H), 4.55 (d, J = 3.7 Hz, 1H), 3.97 (t, 1H), 3.79 (s, 6H), 3.72−3.63 (m, 3H), 3.5−3.35 (m, 2H), 3.35 (s, 3H) ppm. ^{13}C NMR (63 MHz, $CDCl_3$) δ: 158.6, 148, 131.0, 130.3, 129.7, 128.5, 128.1, 127.9, 113.9, 113.8, 98.2, 81.8, 80.0, 76.5, 75.4, 73.5, 70.7, 61.9, 55.3, 55.2 ppm. Anal. Calcd for $C_{30}H_{36}O_8$: C, 69.38; H, 6.76; O, 23.85. Found: C, 69.46; H, 6.57; O, 23.66.

Methyl (*Z*)-2-*O*-Benzyl-3,4-di-*O*-(*p*-methoxybenzyl)-6-*O*-acetyl-α-D-glucos-5-enopyranoside (6). To a solution of oxalyl chloride (0.234 mL, 0.432 mmol) in 15 mL of CH_2Cl_2, cooled to −78 °C, was added dropwise dry DMSO (0.38 mL, 0.86 mmol). The mixture was stirred at −78 °C for 10 min, and a solution of methyl 2-*O*-benzyl-3,4-di-*O*-(*p*-methoxybenzyl)-α-D-glucopyranoside (**5**, 568 mg, 1.08 mmol) in 5 mL of CH_2Cl_2 was added over 10 min. The cloudy solution was stirred for 30 min, then 1.2 mL of Et_3N was added to produce a clear solution and the mixture was warmed to rt. The usual workup gave an oil used directly in the next step. The oil was dissolved in 13.5 mL of dry CH_3CN, and anhydrous K_2CO_3 (0.918 g, 6.7 mmol) was added. After stirring for 10 min, acetic anhydride (0.6 mL, 6.3 mmol) was added and the mixture was refluxed overnight under nitrogen. The volume was reduced to one-third, and the residue was diluted with water and extracted with ether. The usual workup gave 473 mg of **6** (79% for two steps) (R_f = 0.33, 50% EtOAc in hexane). ^{1}H NMR (250 MHz, $CDCl_3$) δ: 7.4−7.2 (m, 9H), 6.9, 6.5 (2s, 4H, PMB), 4.9−4.6 (m, 8H), 4.0−3.9 (m, 2H), 3.8 (s, 6H), 3.6−3.5 (m, 1H), 3.35 (s, 3H), 2.20 (s, 3H) ppm. ^{13}C NMR (63 MHz, $CDCl_3$) δ: 167.3, 159.2, 153.3, 138.0, 135.1, 130.8, 129.8, 128.5, 128.1,123.0, 113.9, 113.8, 99.8, 81.1, 79.1, 75.4, 74.2, 73.7, 60.4, 56.2, 55.3 ppm. Anal. Calcd for $C_{32}H_{36}O_9$: C, 68.73; H, 6.29. Found: C, 68.66; H, 6.23.

1-*O*-Acetyl-3-*O*-benzyl-4,5-di-*O*-(*p*-methoxybenzyl)-2-deoxy-2-oxo-*myo*-inositol (7). To a solution of methyl (*Z*)-2-*O*-benzyl-3,4-di-*O*-(*p*-methoxybenzyl)-6-*O*-acetyl-α-D-glucos-5-enopyranoside (**6**, 550 mg, 0.97 mmol) in 20 mL of acetone and 8 mL water was added mercuric acetate (3.10 g, 9.9 mmol). The solution was stirred for 45 min and 12 mL of a saturated NaCl solution was added. The mixture was stirred for 24 h. The acetone was evaporated, and the residue was extracted with EtOAc. Purification by flash chromatography (EtOAc in hexane 1:1.5) yielded 320 mg of **7** (59%). ^{1}H NMR (250 MHz, $CDCl_3$) δ: 7.33−7.13 (m, 9H), 6.86, 6.83 (2d, 4H, PMB), 5.16 (bs, 1H), 4.87−4.70 (m, 5H), 4.46 (d, J = 10.8 Hz, 1H), 4.32 (bs, 1H), 4.14−4.10 (m, 2H), 3.81, 3.80 (s,s, 6H), 3.85−3.80 (m, 1H), 2.52 (s, 1H), 2.20 (s, 3H) ppm. ^{13}C NMR (63 MHz, $CDCl_3$) δ: 195.0, 169.7, 159.0, 130.0, 129.7, 128.6, 128.0, 113.8, 83.2, 81.5, 78.9, 75.9, 74.9, 73.4, 73.2, 69.5, 55.3 ppm. FAB HRMS: $C_{31}H_{34}O_9Na$ (MNa) calcd 573.2101, found 573.2124.

1-*O*-Acetyl-3-*O*-benzyl-4,5-di-*O*-(*p*-methoxybenzyl)-*myo*-inositol (8). To a solution of 1-*O*-acetyl-3-*O*-benzyl-4,5-di-*O*-(*p*-methoxybenzyl)-2-deoxy-2-oxo-*myo*-inositol (**7**, 170 mg, 0.33 mmol) in 9.5 mL of dry acetonitrile were added sodium triacetoxyborohydride (0.67 g, 3.1 mmol) and 1.6 mL glacial acetic acid. The mixture was stirred for 45 min at rt, and the excess sodium triacetoxyborohydride was destroyed by dropwise addition of 0.5 M $NaHSO_4$. The mixture was extracted with EtOAc, washed successively with 0.5 M $NaHSO_4$ and saturated Na_2HPO_4, dried (Na_2SO_4), and concentrated. The residue was recrystallized from EtOAc−hexane to yield 100 mg of **8** (59%). ^{1}H NMR (250 MHz, $CDCl_3$) δ: 7.32 (d, 4H), 7.26−7.23 (m, 5H), 6.86, 6.84 (d, d, 4H), 4.90−4.63 (m, 6H), 4.26 (bs, 1H), 4.06 (t, 1H), 3.92−3.85 (m, 1H), 3.79 (s, 6H), 3.52 (m, 2H), 3.32 (t, 3H), 2.40 (s, 1H), 2.20 (s, 3H) ppm. ^{13}C NMR (63 MHz, $CDCl_3$) δ: 172.3, 158.7, 130.0, 129.6, 128.1, 128.0, 113.8, 113.6, 82.6, 80.6, 80.1, 75.5, 72.9, 67.1, 55.8 ppm. Anal. Calcd for $C_{31}H_{36}O_9$: C, 67.37; H, 6.57. Found: C, 67.23; H, 6.56.

3-*O*-Benzyl-4,5-di-*O*-(*p*-methoxybenzyl)-*myo*-inositol (9). A mixture of 1-*O*-acetyl-3-*O*-benzyl-4,5-di-*O*-(*p*-methoxybenzyl)-*myo*-inositol (**8**, 2.0 g, 3.62 mmol) and 40 mL of 0.35 M NaOH in methanol was refluxed for 2 h. The usual workup gave 1.662 g of product **9** (90%); mp 144−146 °C. ^{1}H NMR (250 MHz, $CDCl_3$) δ: 7.4−7.2 (m, 9H), 6.9−6.7 (m, 4H), 4.9 (m, 6H), 4.1 (m, 1H), 3.9 (m, 2H), 3.8 (2s, 6H), 3.4 (m, 2H), 3.2 (m, 1H), 2.6 (m, 1H), 2.5 (m, 2H) ppm. ^{13}C NMR ($CDCl_3$, 63 MHz) δ: 137.9, 130.9, 130.8, 129.6, 129.5, 128.5, 127.9, 113.9, 113.8, 82.5, 81.0, 80.1, 77.7, 77.1, 76.6, 75.4, 75.1, 72.6, 72.5, 71.8, 69.3, 55.3, 55.2 ppm. Anal. Calcd for $C_{29}H_{34}O_8$: C, 68.22; H, 6.71. Found: C, 68.00, H, 6.68.

1,2-*O*-Isopropylidene-3-*O*-benzyl-4,5-di-*O*-(*p*-methoxybenzyl)-*myo*-inositol (10). A mixture of triol **9** (1.4 g, 2.745 mmol), 2,2-dimethoxypropane (1.4 g, 13.73 mmol), and 40 mg of TsOH in 20 mL of dry acetone was stirred at rt. After 10 min, no starting material **9** was detected, and three spots appeared in TLC. Stirring was continued for 2 days under the same conditions. The usual workup gave 1.29 g of a colorless oil in 85% yield (R_f = 0.31, EtOAc−hexane 1:1). ^{1}H NMR (300 MHz, $CDCl_3$) δ: 7.4−7.2 (m, 9H), 6.9−6.7 (m, 4H), 4.9−4.6 (m, 6H), 4.3 (m, 1H), 4.1 (m, 1H), 3.9−3.8 (m, 3H), 3.8 (s, 6H, OMe), 3.2 (m, 1H), 2.4 (m, 1H), 1.48, 1.32 (2s, 6H) ppm. ^{13}C NMR ($CDCl_3$, 63 MHz) δ: 129.7, 129.6, 128.5, 128.1, 127.9, 114.0, 113.9, 81.2, 78.2, 76.5, 74.7, 74.5, 74.2, 74.0, 73.3, 55.3, 27.9, 25.7 ppm. FAB HRMS: $C_{32}H_{38}O_8Na$ (MNa) calcd 573.2464, found 573.2467.

1,2-*O*-Isopropylidene-3,6-di-*O*-benzyl-4,5-di-*O*-(*p*-methoxybenzyl)-*myo*-inositol (11). 1,2-*O*-Isopropylidene-3-*O*-benzyl-4,5-di-*O*-(*p*-methoxybenzyl)-*myo*-inositol (**10**, 1.29 g, 2.34 mmol) dissolved in 50 mL of DMF was added to a suspension of oil-free sodium hydride (200 mg, 8.3 mmol) in 15 mL of DMF followed by addition of benzyl bromide (1200 mg, 7.1 mmol) in 1.5 mL of DMF. The mixture was stirred at rt for 2 h. The usual workup gave 1.2 g (yield 80%) of product **11** as a thick oil (R_f = 0.23, 25% EtOAc in hexane). ^{1}H NMR (300 MHz, $CDCl_3$) δ: 7.4−7.2 (m, 14H), 6.85−6.75 (m, 4H), 4.9−4.6 (m, 8H), 4.2 (m, 1H), 4.1 (m, 1H), 3.9 (m, 2H), 3.76 (s, 6H, OMe), 3.65 (m, 1H), 3.36 (m, 1H), 2.4 (m, 1H), 1.48, 1.32 (2s, 6H) ppm. ^{13}C NMR ($CDCl_3$, 63 MHz) δ: 130.0, 129.6, 128.6, 128.2, 127.9, 113.8, 113.6, 81.1, 79.6, 78.9, 77.5, 77.0, 76.8, 76.5, 74.4, 71.8, 71.7, 71.4, 55.3, 27.9, 27.0 ppm.

3,6-Di-*O*-benzyl-4,5-di-*O*-(*p*-methoxybenzyl)-*myo*-inositol (12). A mixture of 1,2-*O*-isopropylidene-3,6-di-*O*-benzyl-4,5-di-*O*-(*p*-methoxybenzyl)-*myo*-inositol (**11**, 1.2 g, 1.87 mmol) and 40 mg of TsOH in 50 mL of acetone containing 0.5 mL of water was stirred at rt until TLC showed no remaining starting material (ca. 3 days). The usual workup gave the pure **12** (864 mg of solid, yield 77%), which was recrystallized from EtOAc and hexane; mp 127−129 °C (lit.[25] mp racemic form 130−131 °C). ^{1}H NMR (300 MHz, $CDCl_3$) δ: 7.4−7.2 (m, 14H), 6.9−6.7

(m, 4H), 4.9−4.6 (m, 8H), 4.13 (m, 1H), 3.9 (m, 1H), 3.80 (m, 1H), 3.75 (s, 6H, OMe), 3.40 (m, 1H), 2.55 (s, 1H, OH), 2.45 (d, 1H, OH) ppm. ^{13}C NMR ($CDCl_3$, 63 MHz) δ: 129.6, 129.5, 128.6, 127.9, 113.8, 83.0, 81.3, 77.6, 77.1, 76.5, 75.6, 75.4, 72.6, 71.7, 69.2, 55.3 ppm. Anal. Calcd for $C_{36}H_{40}O_8$: C, 71.98; H, 6.71. Found: C, 71.79; H, 6.63.

3,6-Di-*O*-benzyl-1,4,5-tri-*O*-(*p*-methoxybenzyl)-*myo*-inositol (13). A mixture of 3,6-di-*O*-benzyl-4,5-di-*O*-(*p*-methoxybenzyl)-*myo*-inositol (**12**, 400 mg, 0.67 mmol), dibutyltin oxide (182 mg, 0.73 mmol), and Bu_4NI (369 mg, 1 mmol) in 40 mL of toluene was refluxed for 2 h. Then *p*-methoxybenzyl chloride (230 mg, 1.47 mmol) was added and the mixture was stirred for an additional 2 h. The usual workup gave 400 mg of a solid (83%); mp 122−124 °C. ^{1}H NMR (300 MHz, $CDCl_3$) δ: 7.4−7.2 (m, 16H), 6.9−6.7 (m, 6H), 4.9−4.6 (m, 10H), 4.3 (m, 1H), 4.15 (m, 1H), 3.9 (s, 9H, OMe), 3.40 (m, 1H), 2.50 (s, 1H, OH) ppm. ^{13}C NMR ($CDCl_3$, 63 MHz) δ: 129.6, 128.6, 128.4, 127.9, 127.8, 127.7, 127.6, 113.8, 83.0, 81.3, 79.9, 79.5, 77.1, 75.9, 75.6, 72.8, 72.6, 72.4, 67.7, 55.3 ppm. Anal. Calcd for $C_{44}H_{48}O_9$: C, 73.31; H, 6.72. Found: C, 73.23; H, 6.55.

Benzyl *N*-Cbz-3-amino-1-propyl 2-[3,6-Di-*O*-benzyl-1,4,5-tri-*O*-(*p*-methoxybenzyl)-*myo*-inositil] Phosphate (14). A solution of 3,6-di-*O*-benzyl-1,4,5-tri-*O*-(*p*-methoxybenzyl)-*myo*-inositol (**13**, 160 mg, 0.22 mmol) in 8 mL of CH_2Cl_2 and 1*H*-tetrazole (62 mg, 0.88 mmol) was stirred at rt, while a solution of benzyloxy *N*-Cbz-3-amino-1-propoxy *N*,*N*-diisopropylamino-phosphite (198 mg, 0.44 mmol) in 2 mL of $CH_2Cl_2$was added. The mixture was stirred at rt for 1 h and cooled to −40 °C, *m*-CPBA (200 mg) was added, and the reaction was stirred for 30 min at 0 °C and then 30 min at rt. The mixture was diluted with CH_2Cl_2, washed with 10% aqueous Na_2SO_3, saturated $NaHCO_3$, and water, dried (Na_2SO_4), concentrated, and chromatographed on silica gel using EtOAc−hexane−triethylamine (20:20:1) (R_f = 0.3) to give 210 mg (87%) of compound **14** as a very viscous colorless oil. ^{1}H NMR (250 MHz, $CDCl_3$) δ: 7.4−7.2 (m, 26H, phenyl), 6.9−6.7 (m, 6H, PMB), 5.2−4.6 (m, 16H), 4.1 (m, 2H), 3.9 (m, 2H), 3.85 (s, 9H, OMe), 3.5 (m, 2H), 3.2 (m, 2H), 1.8 (m, 2H) ppm. ^{13}C NMR (63 MHz, $CDCl_3$) δ: 159.3, 130.8, 130.2, 129.7, 129.6, 128.6, 128.5, 128.4, 128.1, 128.0; 127.9, 127.7, 113.9, 113.8, 82.9, 81.0; 78.6, 77.2, 76.7, 75.5, 72.4, 69.1, 66.6, 55.3, 37.2 ppm. ^{31}P NMR (101 MHz, $CDCl_3$): −0.52 ppm. FAB HRMS: $C_{62}H_{68}NO_{14}PNa$ (MNa) calcd 1104.4272, found 1104.4296.

Benzyl *N*-Cbz-3-amino-1-propyl 2-(3,6-Di-*O*-benzyl-*myo*-inosityl) Phosphate (15). Benzyl *N*-Cbz-3-amino-1-propyl 2-[3,6-di-*O*-benzyl-1,4,5-tri-*O*-(*p*-methoxybenzyl)-*myo*-inosityl] phosphate[13] (**14**, 200 mg, 0.185 mmol) was dissolved in 50 mL of acetonitrile and water (9:1). After addition of 3 equiv of $(NH_4)_2Ce(NO_3)_6$ in three portions, the mixture was stirred at rt for 2 h. After removal of acetonitrile, the residue was extracted with ether. The usual workup gave 58 mg of compound **15** (43%) (EtOAc in hexane 5:1, R_f = 0.43). The ^{31}P NMR spectra showed two peaks (1:1) corresponding to two diastereoisomers. ^{1}H NMR (300 MHz, $CDCl_3$) δ: 7.4−7.2 (m, 20H, phenyl), 5.4 (m, 1H), 5.1−4.8 (m, 10H), 4.2 (m, 1H), 3.9 (m, 2H), 3.6 (m, 2H), 3.2 (m, 2H), 2.9 (m, 2H), 1.8 (m, 2H) ppm. ^{13}C NMR (63 MHz, $CDCl_3$) δ: 159.3, 128.5, 128.1, 127.9, 127.7, 81.6, 78.6, 77.6, 77.1, 76.6, 74.9, 66.5, 55.3, 37.2 ppm. ^{31}P NMR (101 MHz, $CDCl_3$) δ: 0.75, −1.08 (1:1) ppm. FAB MS: 653 (M^+Na − Bn).

Benzyl *N*-Cbz-3-amino-1-propyl 2-(3,6-Di-*O*-benzyl-1,4,5-tris(dibenzylphospho)-*myo*-inositil) Phosphate (16). A solution of benzyl *N*-Cbz-3-amino-1-propyl 2-(3,6-di-*O*-benzyl-*myo*-inositil) phosphate (**15**, 210 mg, 0.293 mmol) in 8 mL of CH_2Cl_2 and 1*H*-tetrazole (246 mg, 3.52 mmol) was stirred at rt and a solution of dibenzyl *N*,*N*-diisopropylphosphamide (610 mg, 1.76 mmol) in 2 mL of CH_2Cl_2 was added. The next procedure was the same as that for compound **14**. Purification by chromatography (EtOAc−CH_2Cl_2 1:1, R_f = 0.32) gave 309 mg (76%) of compound **16** as a syrup. ^{1}H NMR (300 MHz, $CDCl_3$) δ: 7.4−7.2 (m, 50H, phenyl), 5.4 (m, 1H), 5.1−4.5 (m, 23H), 4.2 (m, 1H), 3.4 (m, 2H), 3.2 (m, 2H), 1.8 (m, 2H) ppm. ^{13}C NMR (63 MHz, $CDCl_3$) δ: 159.3, 128.5, 128.1, 127.0, 126.7, 81.6, 78.6, 77.6, 77.1, 76.6, 74.9, 66.5, 55.3, 37.2 ppm. ^{31}P NMR (101 MHz, $CDCl_3$) δ: −0.04, −0.27, −0.53 (1:2:1) ppm. FAB HRMS: $C_{80}H_{83}NO_{20}P_4$-Na (MNa) calcd 1524.4357, found 1524.4299.

2-*O*-(3-Aminopropyl-1-phospho)-*myo*-inositol 1,4,5-Trisphosphate (17). Benzyl *N*-Cbz-3-amino-1-propyl 2-(3,6-di-*O*-benzyl-1,4,5-tris(dibenzylphospho)-*myo*-inosityl) phosphate (**16**, 248 mg, 0.165 mmol) was dissolved in 100 mL of 95% EtOH and 160 mg of 10% Pd−C was added. Hydrogenolysis at an initial hydrogen pressure of ca. 5 atm was allowed to proceed at rt for 6.5 h. The catalyst was filtered off and washed with 30 mL of 2:1 ethanol−water followed by 3 mL of water. The filtrate was brought to pH 8.0 with a few drops of concd ammonium hydroxide, and the solvent was removed *in vacuo*. The residue was dissolved in 3 mL of water and applied to one 5 × 2 cm column of Bio-Rad Chelex 100 resin (sodium form) eluted with 25 mL of water. Evaporation afforded 114 mg (95%) of the sodium salt of **17** as a colorless glass. ^{1}H NMR (300 MHz, D_2O) δ: 4.15−3.80 (m, 7H), 3.65 (d, J = 9.6 Hz, 1H), 3.09 (t, J = 6.9 Hz, 2H), 1.90−1.86 (m, 2H) ppm. ^{13}C NMR (63 MHz, D_2O) δ: 80.7, 78.4, 75.5, 75.1, 74.1, 67.0, 40.5, 40.1, 30.2 ppm. ^{31}P NMR (101 MHz, D_2O) δ: 8.54, 8.08, 7.50, 3.65 (1:1:1:1) ppm. FAB HRMS: $C_9H_{18}NO_{18}P_4Na_6$ (MH^+) calcd 689.8861, found 689.8894.

Tetrabenzyl 1,2-(3,6-di-*O*-benzyl-4,5-di-*O*-(*p*-methoxybenzyl)-*myo*-inosityl) Bisphosphate (18). A solution of 3,6-di-*O*-benzyl-4,5-di-*O*-(*p*-methoxybenzyl)-*myo*-inositol (**12**, 40 mg, 0.067 mmol) in 5 mL of CH_2Cl_2 and 1*H*-tetrazole (38 mg, 0.53 mmol) was stirred at rt and a solution of dibenzyl *N*,*N*-diisopropylphosphamide (92 mg, 0.27 mmol) in 1 mL of CH_2Cl_2 was added. The next procedure was the same as that for compound **14**. Purification by chromatography (70% EtOAc in hexane, R_f = 0.52) gave 60 mg (80%) of compound **18** as a syrup. ^{1}H NMR (250 MHz, $CDCl_3$) δ: 7.4−7.2 (m, 34H, phenyl), 6.85, 6.75 (AB, 4H, PMB), 5.3 (m, 1H), 5.1−4.4 (m, 19H), 3.95 (m, 1H), 3.8 (s, 6H), 3.5 (m, 1H) ppm. ^{13}C NMR (63 MHz, $CDCl_3$) δ: 129.6, 128.7, 128.6, 128.5, 128.4, 128.3, 128.2, 128.1, 127.9, 127.6, 113.8, 82.5, 80.4, 79.7, 78.6, 75.8, 75.5, 75.4, 72.7, 69.5, 69.4, 69.2, 69.1, 67.4, 55.3 ppm. ^{31}P NMR (101 MHz, $CDCl_3$) δ: 0.39, −0.63 (1:1) ppm. FAB HRMS: $C_{64}H_{67}O_{14}P_2$ (MH^+) calcd 1121.4006, found 1121.4042.

Tetrabenzyl 1,2-(3,6-Di-*O*-benzyl-*myo*-inosityl) Bisphosphate (19). Tetrabenzyl 1,2-(3,6-di-*O*-benzyl-4,5-di-*O*-(*p*-methoxybenzyl)-*myo*-inosityl) bisphosphate (**18**, 60 mg, 0.054 mmol) was dissolved in 10 mL of CH_3CN and water (9:1). After addition of 3 equiv of $(NH_4)_2Ce(NO_3)_6$ (90 mg) in three portions, the mixture was stirred at rt for 2 h. The workup as before (75% EtOAc in hexane, R_f = 0.33) gave 30 mg of compound **19** (64%). ^{1}H NMR (300 MHz, $CDCl_3$) δ: 7.4−7.05 (m, 30H, phenyl), 5.3 (d, J = 10 Hz, 1H), 5.1−4.65 (m, 12H), 4.4 (d, J = 11 Hz, 1H), 3.80−3.65 (m, 2H), 3.44 (m, 1H), 3.23 (d, J = 11 Hz, 1H), 2.85 (s, 1H), 2.70 (s, 1H) ppm. ^{13}C NMR (63 MHz, $CDCl_3$) δ: 128.5, 128.4, 128.3, 128.2, 128.1, 127.9, 127.7, 127.6, 127.5, 79.5, 78.9, 76.5, 75.3, 75.2, 74.3, 72.2, 71,89, 69.6, 69.5, 69.4, 69.3 ppm. ^{31}P NMR (101 MHz, $CDCl_3$) δ: 0.33, −0.51 (1:1) ppm. FAB HRMS: $C_{48}H_{51}O_{12}P_2$ (MH^+) calcd 881.2856, found 881.2889.

Octabenzyl 1,2,4,5-(3,6-di-*O*-benzyl-*myo*-inosityl) Tetrakisphosphate (20). A solution of tetrabenzyl 1,2-(3,6-di-*O*-benzyl-*myo*-inosityl) bisphosphate (**19**, 30 mg, 0.034 mmol) in 2 mL of CH_2Cl_2 and 1*H*-tetrazole (20 mg, 0.27 mmol) was stirred at rt, and a solution of dibenzyl *N*,*N*-diisopropylphosphamide (47 mg, 0.14 mmol) in 0.5 mL of $CH_2Cl_2$was added. The next procedure was the same as that for compound **14**. Purification by chromatography (60% EtOAc in hexane, R_f = 0.45) gave 45 mg (90%) of compound **20** as a syrup. ^{1}H NMR (300 MHz, $CDCl_3$) δ: 7.4−6.9 (m, 30H, phenyl), 5.4 (d, J = 9 Hz, 1H), 5.1−4.7 (m, 24H), 4.6(m, 1H), 4.5 (t, J = 10 Hz, 1H), 4.4 (t, J = 10 Hz, 1H), 3.9 (t, J = 9 Hz, 1H), 3.5 (d, J = 9.6 Hz, 1H) ppm. ^{13}C NMR (63 MHz, $CDCl_3$) δ: 128.8, 128.5, 128.3, 128.2, 128.1, 128.0, 127.9, 127.7, 127.6, 79.0, 78.9, 74.6, 72.2, 69.7, 69.6, 69.3 ppm. ^{31}P NMR (101 MHz, $CDCl_3$) δ: 0.27, −0.27, −0.63 (1:2:1) ppm. FAB HRMS: $C_{76}H_{77}O_{18}P_4$ (MH^+) calcd 1401.4060, found 1401.4005.

1,2,4,5-*myo*-Inositol Tetrakisphosphate (21).[14] Octabenzyl 1,2,4,5-(3,6-di-*O*-benzyl-*myo*-inosityl) tetrakisphosphate (**20**, 24 mg, 0.017 mmol) was dissolved in 25 mL of 95% EtOH, and 20 mg of 10% Pd−C was added. The next procedure was the same as that for compound **17**. Compound **21** (8 mg, yield 97%) was obtained as a colorless glass after chromatography (3 × 2 cm column of Bio-Rad Chelex 100 resin, sodium form). ^{1}H NMR (300 MHz, D_2O) δ: 4.2 (q, J = 9 Hz, 1H), 4.0−3.8 (m, 4H), 3.6−3.5 (m, 1H) ppm. ^{13}C NMR (63 MHz, D_2O) δ: 80.8, 79.9, 77.4, 77.1, 74.7, 73.7 ppm. ^{31}P NMR (101 MHz, D_2O) δ: 6.90, 5.88, 5.09 (2:1:1) ppm. FAB MS m/z: 678 (M + 2), 630 (M − 2Na), 608 (M − 3Na + H), 586 (M − 4Na + 2H), 546 (M − 5Na + 3H), 542 (542 (M − 6Na + 4H).

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Supporting Information Available: ^{1}H, ^{13}C, and ^{31}P NMR spectra of compounds **3**−**21** (45 pages). This material is contained in libraries on microfiche, immediately follows this article in the microfilm version of the journal, and can be ordered from the ACS; see any current masthead page for ordering information.

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