Synthesis of L-*scyllo*-inositol 1,2,4-trisphosphate, *scyllo*-inositol 1,2,4,5-tetrakisphosphate and phosphorothioate and DL-2-deoxy-2-fluoro-*myo*-inositol 1,4,5-trisphosphate: optical resolution of DL-1-*O*-allyl-3,6-di-*O*-benzyl-4,5-*O*-isopropylidene-*scyllo*-inositol

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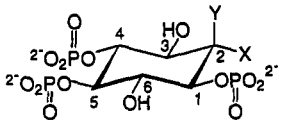
Routes for the synthesis of *scyllo*-inositol tris- and tetrakis-phosphates and 2-deoxy-2-fluoro-*myo*-inositol 1,4,5-trisphosphate from *myo*-inositol have been devised. For DL-*scyllo*-inositol 1,2,4-trisphosphate, DL-1-*O*-allyl-3,6-di-*O*-benzyl-4,5-*O*-isopropylidene-*scyllo*-inositol was prepared from the triflate of DL-1-*O*-allyl-3,6-di-*O*-benzyl-4,5-*O*-isopropylidene-*myo*-inositol by inversion at C-2. Removal of the isopropylidene group and phosphorylation gave the protected trisphosphate. Deblocking with sodium in liquid ammonia afforded racemic *scyllo*-inositol 1,2,4-trisphosphate. DL-1-*O*-Allyl-3,6-di-*O*-benzyl-4,5-*O*-isopropylidene-*scyllo*-inositol was resolved into its enantiomers by means of the crystalline 2-*O*-camphanate ester. The structure of one diastereoisomer, 1D-*O*-allyl-3,6-di-*O*-benzyl-2-*O*-[(−)-camphanoyl]-4,5-*O*-isopropylidene-*scyllo*-inositol was determined by single-crystal X-ray crystallography. 1D-(+)-1-*O*-Allyl-3,6-di-*O*-benzyl-4,5-*O*-isopropylidene-*scyllo*-inositol was used to prepare 1L-(−)-*scyllo*-inositol 1,2,4-trisphosphate in a fashion analogous to the racemic modification. DL-1-*O*-Allyl-3,6-di-*O*-benzyl-*scyllo*-inositol was isomerised to the (*Z*)-prop-1-enyl derivative. The propenyl group was then removed to give the *meso*-1,4-di-*O*-benzyl-*scyllo*-inositol. Phosphitylation followed by oxidation or sulfoxidation gave the fully protected tetrakis-phosphate or -phosphorothioate, respectively. After deblocking and purification, *scyllo*-inositol 1,2,4,5-tetrakisphosphate and *scyllo*-inositol 1,2,4,5-tetrakisphosphorothioate were obtained.

DL-1-*O*-Allyl-3,6-di-*O*-benzyl-4,5-*O*-isopropylidene-*scyllo*-inositol was isomerised to the 1-*O*-[(*Z*)-prop-1-enyl] derivative which was converted into the 2-*O*-triflate. Displacement of the triflate using tetrabutylammonium fluoride proceeded with inversion of configuration to give DL-3,6-di-*O*-benzyl-2-deoxy-2-fluoro-4,5-*O*-isopropylidene-1-*O*-([(*Z*)-prop-1-enyl]-*myo*-inositol. Removal of propenyl and isopropylidene groups afforded DL-3,6-di-*O*-benzyl-2-deoxy-2-fluoro-*myo*-inositol, which was phosphitylated and the product oxidised to give the fully protected 2-fluoro trisphosphate. Deprotection furnished DL-2-deoxy-2-fluoro-*myo*-inositol 1,4,5-trisphosphate. These compounds will be useful probes for investigation of the polyphosphoinositide pathway of cellular signalling.

Introduction

D-*myo*-Inositol 1,4,5-trisphosphate [Ins(1,4,5)P_3], **1** is a second messenger which releases Ca^{2+} from intracellular stores[1,2] *via* an isolated,[3] cloned[4] and sequenced[5] receptor which, when reconstituted, mediates Ca^{2+} release in response to Ins(1,4,5)P_3.[6] Major challenges are now the elucidation of the structural basis for interaction of Ins(1,4,5)P_3 with its receptor and the metabolic enzymes, Ins(1,4,5)P_3 3-kinase and 5-phosphatase, and additionally the rational design of agonists, antagonists and enzyme inhibitors. Recent progress in inositol phosphate chemistry[7,8] has been reviewed.

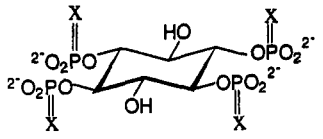


1; X = H, Y = OH
2; X = Y = H
3; X = OH, Y = H

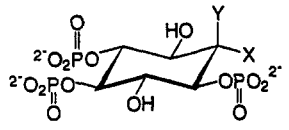
Structure of Ins(1,4,5)P_3 and 2-position-modified analogues

In order to investigate the role of the unique axial hydroxy group at C-2 in Ins(1,4,5)P_3 we were interested in compounds modified at this position. 2-Modified Ins(1,4,5)P_3 analogues including 2-deoxy-*myo*-inositol 1,4,5-trisphosphate **2**[9] and fluoro derivatives[10–14] have already been shown to have interesting properties. A first target was *scyllo*-inositol 1,2,4-trisphosphate [*scyllo*-Ins(1,2,4)P_3] **3**, the *scyllo*-analogue of Ins(1,4,5)P_3. Here the hydroxy group at C-2 [C-5 in *scyllo*-Ins(1,2,4)P_3, which has been numbered according to the IUPAC rules for cyclitols] is in an equatorial rather than an axial position relative to Ins(1,4,5)P_3. We were also interested in *scyllo*-inositol 1,2,4,5-tetrakisphosphate [*scyllo*-Ins(1,2,4,5)P_4] **4** and its phosphorothioate analogue *scyllo*-inositol 1,2,4,5-tetrakisphosphorothioate [*scyllo*-Ins(1,2,4,5)-PS_4] **5**, since both compounds would mimic the phosphate arrangement in Ins(1,3,4,6)P_4, which has been shown to be active[15] and to act as a partial agonist at the Ins(1,4,5)P_3 receptor.[16] We wanted to establish a potential role of the axial hydroxy group (which is replaced by an equatorial one in the *scyllo*-inositol analogues) in this partial agonist behaviour. A second motive to prepare these *scyllo*-inositol derivatives was that racemic *myo*-inositol 1,2,4,5-tetrakisphosphate was found to be a potent inhibitor of 5-phosphatase with a K_i of 2.9 μM.[17] We reasoned both that the symmetrical *scyllo*-inositol analogue of this compound might show enhanced inhibitory properties similar to those of Ins(1,2,4,5)P_4 and that affinity for the enzyme may be increased by replacing the phosphate groups with phosphorothioates.[18] Isosteric replacement of a hydroxy group with fluorine has led to fluorinated *myo*-inositol analogues,[19–26] derivatives,[27] inositol phosphate

analogues[11,28–32] and lipids including 2-deoxy-2-fluoro-1-phosphatidyl-*scyllo*-inositol[33] and 3-deoxy-3-fluoro-1-phosphatidylinositol.[34] D-3-Deoxy-3-fluoro-*myo*-inositol inhibits cell growth[22] and 5-deoxy-5-fluoro-*myo*-inositol is incorporated into phospholipid by PtdIns synthase,[35] although 5-deoxy-5,5-difluoro-*myo*-inositol is a much poorer substrate.[24] We previously reported the synthesis[10] and biological evaluation of the fluorinated inositol phosphate analogues, 2-deoxy-2-fluoro-*scyllo*-inositol 1,4,5-trisphosphate **6**,[11] 2-deoxy-2,2-difluoro-Ins(1,4,5)P_3 **7**[11] and 3-deoxy-3-fluoro-Ins(1,4,5)P_3[36] with the Ca^{2+}-releasing receptor and metabolic enzymes 5-phosphatase and 3-kinase. L-2-Deoxy-2,2-difluoro-Ins-(1,4,5)P_3 was found to be a potent inhibitor of 3-kinase and 5-phosphatase.[14]



Structure of *scyllo*-inositol 1,2,4,5-tetrakisphosphate and phosphorothioate



Analogues of Ins(1,4,5)P_3 fluorinated at the 2-position

We report here the synthesis of a number of novel probes for investigation of the polyphosphoinositide pathway of cellular signalling, together with the optical resolution of a key protected precursor based upon an X-ray crystal structure analysis of the camphanate derivative of one enantiomer.

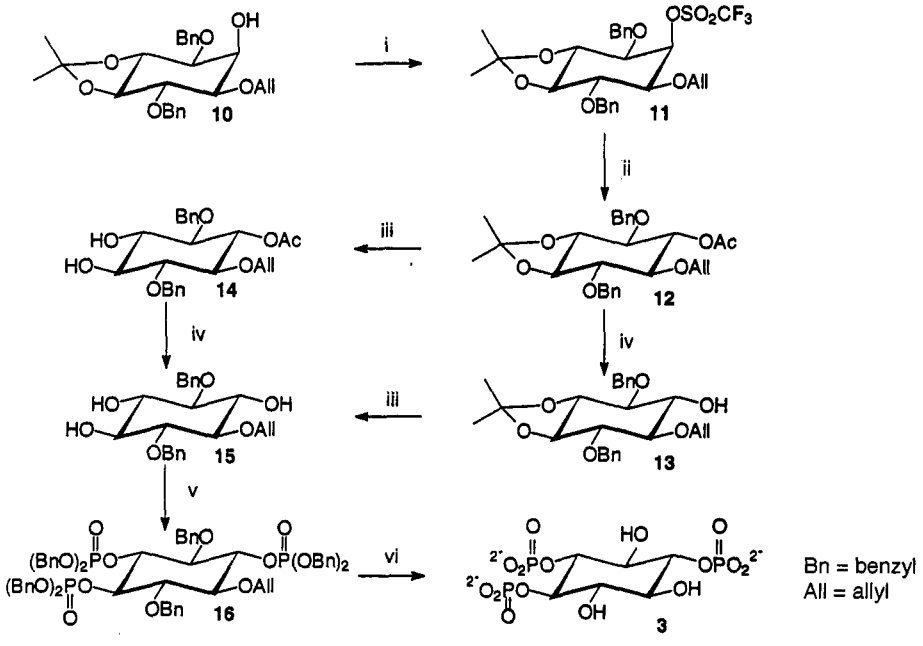
A preliminary account of a part of this work has appeared[37] and the compounds described herein have been biologically evaluated as Ca^{2+}-mobilising agonists and enzyme inhibitors.[38]

Results and discussion

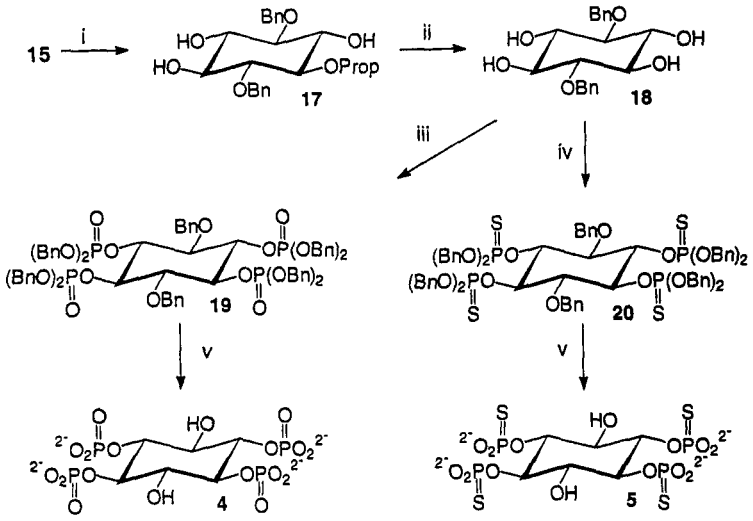
DL-1-*O*-Allyl-3,6-di-*O*-benzyl-4,5-*O*-isopropylidene-*myo*-inositol **10** (Scheme 1) was considered a suitably protected intermediate with a free 2-hydroxy group for the preparation of *scyllo*- and 2-fluoro-inositol phosphates and was synthesized from *myo*-inositol as described.[39] Inversion at C-2 in compound **10** to the corresponding racemic *scyllo*-inositol derivative **13**† was accomplished by first forming the 2-trifluoromethylsulfonic ester (triflate) **11** using trifluoromethanesulfonic (triflic) anhydride (Tf_2O)–pyridine. Owing to its instability (triflates have been reported to decompose on silica gel columns) compound **11** was only partially purified and characterised. The triflate moiety was then displaced with caesium acetate in dimethylformamide (DMF), in a reaction which proceeded with inversion of configuration at the 2-position, to give DL-2-*O*-acetyl-1-*O*-allyl-3,6-di-*O*-benzyl-4,5-*O*-isopropylidene-*scyllo*-inositol **12**. ^{1}H NMR spectroscopy showed the signal for the C-2 proton at δ 5.15 (easily assigned to 2-H by its downfield shift due to acetate-substitution) as a triplet with J = 8.8 Hz, arising from axial–axial coupling to 1-H and 3-H. The 2-H_{eq}–C–C–1(3)-H_{ax} arrangement found in *myo*-inositol derivatives usually results in J values in the order of 2–5 Hz. Removal of acetate and isopropylidene protecting groups by saponification and acid hydrolysis, respectively, then gave racemic 1-*O*-allyl-3,6-di-*O*-benzyl-*scyllo*-inositol **15** either *via* intermediate **13** or **14**. The synthesis of 1-*O*-allyl-3,6-di-*O*-benzyl-4,5-*O*-isopropylidene-*scyllo*-inositol **13** had previously been reported by Gigg *et al.*,[39] who obtained this compound as a by-product (30% yield, the major product in this reaction was compound **10**) in the reduction of the corresponding 2-inosose derivative with sodium borohydride.

Phosphitylation of triol **15** with bis(benzyloxy)(diisopropylamino)phosphine–1*H*-tetrazole followed by oxidation with Bu^{t}OOH furnished trisphosphate **16**. The ^{1}H-coupled ^{31}P NMR spectrum for this fully protected trisphosphate showed a quartet integrating for 2 P at δ_P −1.72 (J = 7.9 Hz) and a second quartet with the same coupling constant which integrated for 1 P at δ_P −1.47. Treatment with sodium–liquid ammonia deblocked both the benzyl and allyl[40] protecting groups in one step. Ion-exchange chromatography on Q Sepharose Fast Flow using a gradient of triethylammo-

† In this paper, protected *scyllo*-inositol derivatives have not been numbered systematically, but rather to show the relationship to the protected *myo*-inositol derivatives for which they were used or from which they were derived.



Scheme 1 Synthesis of DL-*scyllo*-inositol 1,2,4-trisphosphate **3**. *Reagents and conditions:* (i) triflic anhydride, pyridine, CH_2Cl_2, −78 °C; (ii) caesium acetate, dry DMF; (iii) M HCl, MeOH, reflux; (iv) M NaOH, MeOH, reflux; (v) (a) Pr$^i_2$NP(OBn)$_2$, tetrazole, CH_2Cl_2; (b) 70% *tert*-BuOOH; (*vi*) (*a*) Na–liq. NH_3; (b) water.



Scheme 2 Synthesis of *scyllo*-inositol 1,2,4,5-tetrakisphosphate **4** and *scyllo*-inositol 1,2,4,5-tetrakisphosphorothioate **5**. *Reagents and conditions:* (i) KOBu^t, DMSO, 50 °C; (ii) M HCl, MeOH, reflux; (iii) (a) $Pr^i_2NP(OBn)_2$, tetrazole, CH_2Cl_2; (b) 70% *tert*-BuOOH; (iv) (a) $Pr^i_2NP(OBn)_2$, tetrazole, CH_2Cl_2; (b) S_8, pyridine; (v) (a) Na–liq. NH_3; (b) water.

nium hydrogen carbonate gave pure racemic *scyllo*-inositol 1,2,4-trisphosphate **3**.

The synthesis of *scyllo*-inositol 1,2,4,5-tetrakisphosphate [*scyllo*-Ins(1,2,4,5)P_4] **4** and *scyllo*-inositol 1,2,4,5-tetrakisphosphorothioate [*scyllo*-Ins(1,2,4,5)PS_4] **5** was straightforward (Scheme 2). DL-1-*O*-Allyl-3,6-di-*O*-benzyl-*scyllo*-inositol **15** was isomerised to the (*Z*)-prop-1-enyl derivative **17** using Bu^{t}OK–dimethyl sulfoxide (DMSO). The propenyl group was then removed by acidic hydrolysis to give the *meso*-1,4-di-*O*-benzyl-*scyllo*-inositol **18**. Phosphitylation with bis(benzyloxy)-(diisopropylamino)phosphine followed either by oxidation or sulfoxidation of the product gave the fully protected tetrakisphosphate **19** or phosphorothioate **20** respectively. Both compounds showed a single peak in the ^{31}P NMR spectrum (at δ_P −3.71 and 67.77, typical for phosphate and thiophosphate triesters, respectively), which was to be expected for these symmetrical derivatives. Deprotection and purification as above afforded *scyllo*-Ins(1,2,4,5)P_4 **4** and *scyllo*-Ins(1,2,4,5)-PS_4 **5**, respectively.

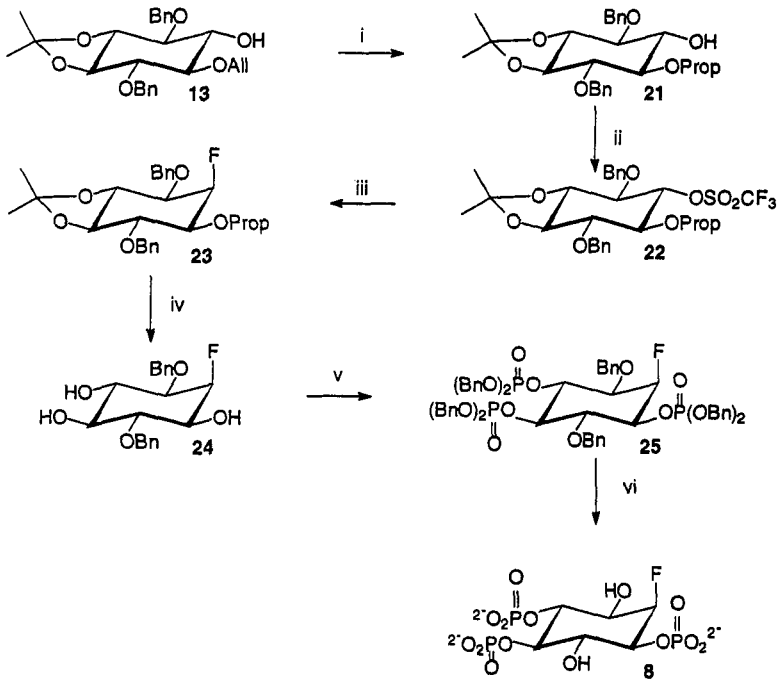
A number of fluorinated analogues of Ins(1,4,5)P_3 have been prepared, including the 2-fluorinated compounds 2-deoxy-2-fluoro-*scyllo*-Ins(1,4,5)P_3 **6** and 2-deoxy-2,2-difluoro-Ins(1,4,5)P_3 **7**. A synthesis of 2-deoxy-2-fluoro-Ins(1,4,5)P_3 [2-F-Ins(1,4,5)P_3] **8**, the 2-fluoro analogue most similar to Ins(1,4,5)P_3, has however not been reported as yet. In view of the interesting properties that the enantiomers of both compounds **6** and **7** displayed, we reasoned that 2-F-Ins(1,4,5)P_3, which resembles Ins(1,4,5)P_3 more closely, might show improved characteristics when compared with the former analogues.

DL-1-*O*-Allyl-3,6-di-*O*-benzyl-4,5-*O*-isopropylidene-*scyllo*-inositol **13** was used as starting material for the preparation of 2-F-Ins(1,4,5)P_3. An earlier attempt to synthesize 2-F-Ins(1,4,5)P_3 from the same precursor by fluorination with (diethylamino)sulfur trifluoride (DAST) failed[28] because the reaction unexpectedly proceeded with retention of configuration, yielding the same 2-deoxy-2-fluoro-*scyllo*-inositol derivative as that obtained by DAST fluorination of the corresponding *myo*-inositol derivative. This may be an indication of steric effects or neighbouring-group participation in the reaction mechanism, as DAST fluorinations usually give an inverted product and, for the reaction of 1,2,3,4,5-penta-*O*-benzyl-*scyllo*-inositol with DAST,[19] the fluorinated product did indeed have the *myo*-inositol configuration.

DL-1-*O*-Allyl-3,6-di-*O*-benzyl-4,5-*O*-isopropylidene-*scyllo*-inositol **13** was isomerised to the 1-*O*-[(*Z*)-prop-1-enyl] derivative **21** (Scheme 3) with Bu^{t}OK–DMSO, and this was converted into the *scyllo*-inositol triflate derivative **22** by treatment with Tf_2O in pyridine. Displacement of the sulfonate at the 2-position using dry tetrabutylammonium fluoride (TBAF) gave racemic 3,6-di-*O*-benzyl-2-deoxy-2-fluoro-4,5-*O*-isopropylidene-1-*O*-[(*Z*)-prop-1-enyl]-*myo*-inositol **23**. Observation of $^2J_{HF}$ = 50.0 Hz and $^3J_{HF}$ = 29.5 Hz in the ^1H NMR and the ^{19}F NMR spectra of this compound confirmed the presence of an axial 2-fluorine atom. The $^2J_{HF}$ and $^3J_{HF}$ coupling constants measured are in good agreement with those reported for 2-deoxy-2-fluoro-*myo*-inositol,[19] where $^2J_{HF}$ and $^3J_{HF}$ were found to be 52.1 and 28.8 Hz, respectively. Inositol derivatives bearing equatorial fluorine atoms (*e.g.*, 1-deoxy-1-fluoro-*scyllo*-inositol[10,33]) show similar coupling constants for $^2J_{HF}$ (50.4 Hz), but $^3J_{HF}$ is much smaller (12.6 Hz) than that found in compounds with an axial fluorine. Additional proof for the *myo*-inositol-like nature of the product obtained are the ^3J coupling constants (2.4 Hz) found in the coupling between 2-H and 1(3)-H, indicative of an equatorial–axial arrangement of the respective protons.

Removal of the acid-labile propenyl and isopropylidene protecting groups from compound **23** gave the fluorinated triol **24**, which was phosphitylated with bis(benzyloxy)-(diisopropylamino)phosphine and oxidised to yield trisphosphate **25**. Deprotection with sodium–liquid ammonia gave racemic 2-F-Ins(1,4,5)P_3 **8**. Marecek and Prestwich[28] had reported that 2-fluoro-*scyllo*-Ins(1,4,5)P_3 underwent slow defluorination ($t_{\frac{1}{2}} \approx 2$ weeks) at pH 13 and 50 °C, whereas the difluoro analogue was stable under these conditions. No defluorination was observed during the deblocking of compound **25**. The ^1H-coupled ^{31}P NMR spectrum of compound **8** showed three equal doublets at δ_P 1.96, 1.56 and 0.37 with J = 10.1, 6.7 and 10.1 Hz respectively, and the ^{19}F spectrum showed a dt at δ_F −211.8 with $J_{2\text{-H,F}}$ 51.3 Hz and $J_{1(3)\text{-H,F}}$ 29.7 Hz.

In order to obtain optically pure D-*scyllo*-Ins(1,2,4)P_3 **3a**, an intermediate in the synthesis of the racemic material had to be optically resolved. The camphanate approach has so far proved to be the most generally applicable way to obtain optically pure inositol derivatives. Thus, DL-1-*O*-allyl-3,6-di-*O*-benzyl-4,5-*O*-isopropylidene-*scyllo*-inositol **13** was converted into a mixture of diastereomeric camphanates by using camphanoyl chloride and pyridine. It was possible to isolate the two isomers **26a** and **26b** by crystallisation from methanol and diethyl ether respectively. The structure of one isomer, **26a**, was determined by X-ray crystallography using a crystal of approximate



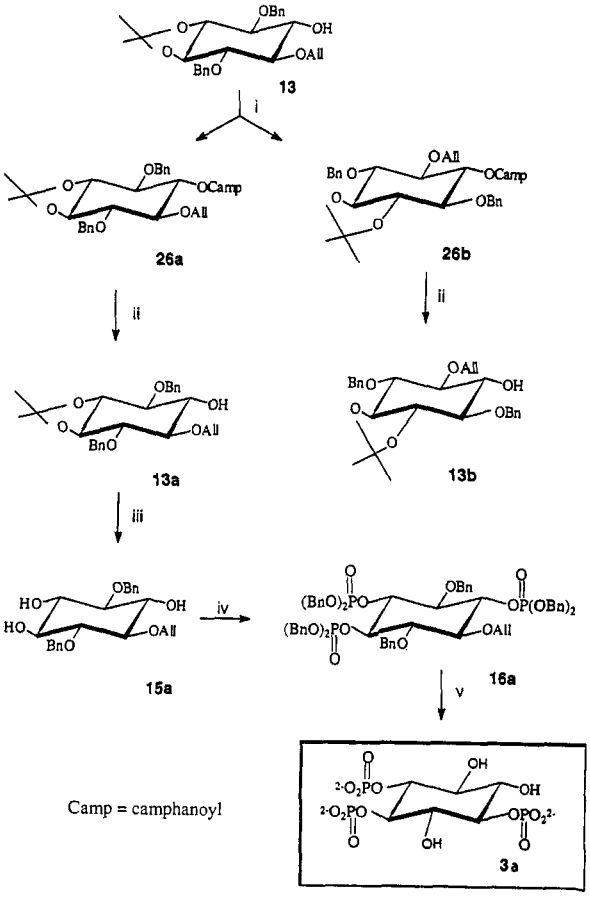
Scheme 3 Synthesis of DL-2-deoxy-2-fluoro-Ins(1,4,5)P_3 **8**. *Reagents and conditions:* (i) KOBu^t, DMSO, 50 °C; (ii) triflic anhydride, pyridine, CH_2Cl_2, −78 °C; (iii) TBAF, THF; (iv) M HCl, MeOH, reflux; (v) (a) $Pr^i_2NP(OBn)_2$, tetrazole, CH_2Cl_2; (b) 70% *tert*-BuOOH; (vi) (a) Na–liq. NH_3; (b) water.

dimensions 0.4 × 0.4 × 0.6 mm for data collection. Removal of the camphanate ester from compound **26a** by saponification furnished enantiomer **13a**, which was converted into optically pure L-*scyllo*-Ins(1,2,4)P_3 **3a** in a fashion analogous to that for the racemic material (Scheme 4).

Crystal data

$C_{36}H_{44}O_9$, M = 620.7, triclinic, a = 6.340(2), b = 11.422(3), c = 12.438(3) Å, α = 100.37(2), β = 94.40(3), γ = 103.27(2)°, U = 855.8 Å^3, space group $P\bar{1}$, Z = 1, D_c = 1.20 g cm^{-3}, μ(Mo-Kα) = 0.90 cm^{-1}, $F(000)$ = 332. Data were measured at room temperature on a CAD4 automatic four-circle diffractometer in the range $2 \leqslant \theta \leqslant 24°$. 2955 Reflections were collected of which 2224 were unique with $I \geqslant 2\sigma(I)$. Data were corrected for Lorentz and polarisation effects but not for absorption. The structure was solved by direct methods and refined using the SHELX[41,42] suite of programs. Hydrogen atoms were included at calculated positions except in the case of H191 and H192 (attached to C19) which were located in an advanced difference Fourier and refined at a fixed distance of 0.98 Å from the parent atom. The hydrogen on C18 was not evident and was therefore not refined. Final residuals after 8 cycles of least squares were R = 0.0381, R_w = 0.0410, for a weighting scheme of w = 1.3987/[$\sigma^2(F)$ + 0.001 498(F)^2]. Max. final shift/esd was 0.000. The max. and min. residual densities were 0.08 and −0.07 e Å^{-3}, respectively. Final fractional atomic coordinates and isotropic thermal parameters, bond distances and angles, and tables of anisotropic temperature factors are available as Supplementary Data at the Cambridge Crystallographic Data Centre.‡ The asymmetric unit is shown in Fig. 1, along with the labelling scheme used. All three different types of protecting group are clearly visible, together with the chiral auxiliary. Bond lengths and angles determined for compound **26a** are generally unexceptional and further discussion from a crystallographic perspective is not merited.

The receptor-binding and Ca^{2+}-release properties of DL-*scyllo*-Ins(1,2,4)P_3 **3**, *scyllo*-Ins(1,2,4,5)P_4 **4**, *scyllo*-Ins(1,2,4,5)-PS_4 **5** and DL-2-F-Ins(1,4,5)P_3 **8** as well as of their interactions with the metabolic enzymes Ins(1,4,5)P_3 3-kinase



Scheme 4 Resolution of compound **13** and preparation of L-*scyllo*-Ins(1,2,4)P_3 **3a**. *Reagents and conditions:* (i) (−)-ω-camphanoyl chloride, pyridine; (ii) M NaOH, MeOH, reflux; (iii) M HCl, MeOH, reflux; (iv) (a) $Pr^i_2NP(OBn)_2$, tetrazole, CH_2Cl_2; (b) 70% *tert*-BuOOH; (v) (a) Na–liq. NH_3; (b) water.

‡ See Instructions for Authors, *Journal*, 1996, Issue 1. Any request to the CCDC for this material should quote the full literature citation and the reference number 207/16.

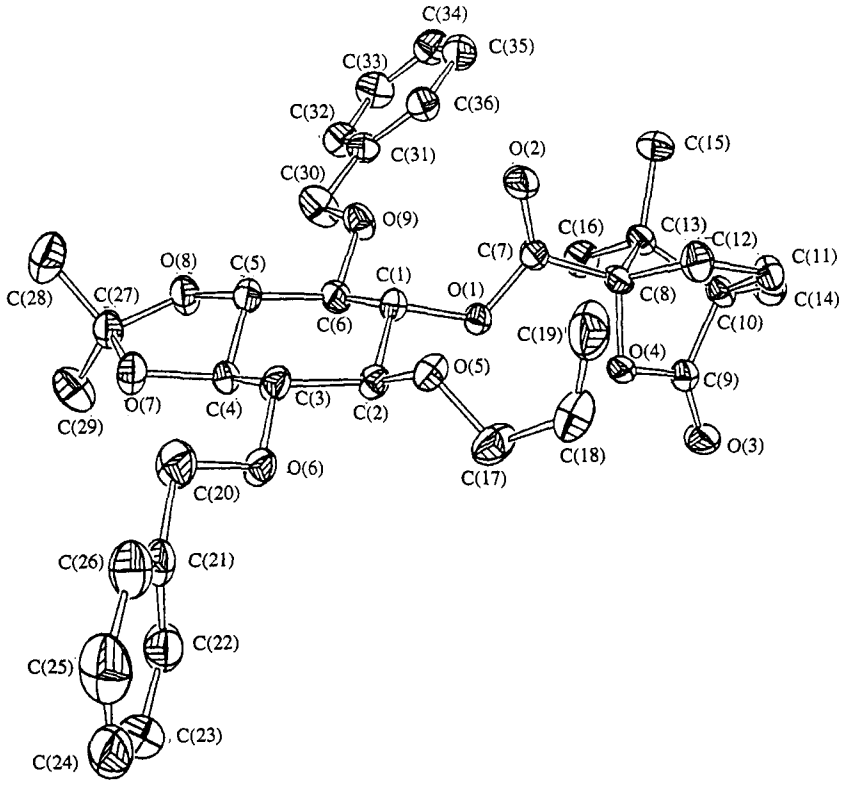


Fig. 1 ORTEP diagram of the X-ray structure of 1D-1-*O*-allyl-3,6-di-*O*-benzyl-2-*O*-[(−)-camphanoyl]-4,5-*O*-isopropylidene-*scyllo*-inositol **26a**. Ellipsoids are represented at the 30% probability level. Crystallographic numbering scheme.

and 5-phosphatase have been examined.[38]§ DL-*scyllo*-Ins(1,2,4)P_3 **3** was found to be a good substrate for 3-kinase (K_i 4.0 μM) and a moderately potent inhibitor of 5-phosphatase (K_i 24.2 μM). *scyllo*-Ins(1,2,4,5)P_4 **4** was a very potent full agonist (EC_{50} = 82.56 nM) at the Ins(1,4,5)P_3 receptor [*cf.* Ins(1,4,5)P_3: EC_{50} = 52 nM], and binding data for this compound {obtained by displacing [^{3}H]-Ins(1,4,5)P_3 from specific sites on bovine adrenal cortex membranes} were equally good (IC_{50} = 14.37 nM, K_i = 10.82 nM).

The fact that, unlike Ins(1,3,4,6)P_4, *scyllo*-Ins(1,2,4,5)P_4 did not show any partial agonist properties is clearly due to the only difference between these two molecules, *i.e.* the stereochemistry of the 2-hydroxy group of Ins(1,3,4,6)P_4, which in *scyllo*-Ins(1,2,4,5)P_4 is equatorial rather than axial. Keeping in mind the alternative binding orientations that have been suggested for Ins(1,3,4,6)P_4,[38] it is evident that the axial pseudo-3(6)-hydroxy group arrangement does indeed contribute significantly to the partial agonist properties of this compound.

scyllo-Ins(1,2,4,5)PS_4 **5** was found to be a good partial agonist at the Ins(1,4,5)P_3 receptor in saponin-permeabilised SH-SY5Y cells, with EC_{50} = 1.6 μM. This compound is able to mobilise some 80% of the Ins(1,4,5)P_3-sensitive Ca^{2+} pool. It is surprising that *scyllo*-Ins(1,2,4,5)PS_4 is a partial agonist, since the parent compound *scyllo*-Ins(1,2,4,5)P_4 does not share this characteristic. From what is currently known, one can only speculate as to why *scyllo*-Ins(1,2,4,5)PS_4 displays this property. Comparison with the now established partial agonists L-*chiro*-Ins(2,3,5)PS_3 and 6-deoxy-Ins(1,4,5)PS_3 [which mobilise 34% and 42% of the Ins(1,4,5)P_3-sensitive Ca^{2+} pool respectively][43] shows that all such compounds are the phosphorothioate analogues of phosphate derivatives which are themselves relatively potent full agonists. It therefore appears that substitution of phosphate groups with phosphorothioate moieties, together with the existence of another perturbation in the molecule relative to Ins(1,4,5)P_3, are major elements in creating compounds with partial agonist characteristics. However, since Ins(1,4,5)PS_3 is a full agonist, it can also be concluded that replacement of the 1-, 4- and 5-phosphate groups with phosphorothioates is not alone sufficient to confer partial agonist properties. Of considerable importance also is the observation that Ins(1,3,4,5)PS_4 is not a partial agonist.

The new fluorinated Ins(1,4,5)P_3 analogue **8** was equipotent to Ins(1,4,5)P_3 in mobilising sequestered Ca^{2+} assuming, not unreasonably, that only the D-isomer displays biological activity [EC_{50} of **8** = 105 nM, EC_{50} of D-Ins(1,4,5)P_3 = 52 nM]. Comparison of the calcium-mobilising properties of compound **8** with those of 2-F-*scyllo*-Ins(1,4,5)P_3 **6** (EC_{50} = 770 nM) and 2,2-F_2-Ins(1,4,5)P_3 **7** (EC_{50} = 410 nM) shows that the closer structural similarity of compound **8** to Ins(1,4,5)P_3 is reflected in its biological activity. The assumption[11,13] that the 2-hydroxy group of Ins(1,4,5)P_3 may interact with the receptor by accepting rather than donating a hydrogen bond thus appears to be consistent with these results: the 2-fluorine atom of compound **8** is still able to accept, but can no longer donate, a hydrogen bond, and the compound is nevertheless as potent as Ins(1,4,5)P_3. Compound **8** is also a good substrate for 3-kinase, acting as a potent competitive inhibitor of the phosphorylation of [^{3}H]-Ins(1,4,5)P_3 by this enzyme [apparent K_i = 3.0 μM; for comparison: 2-F-*scyllo*-Ins(1,4,5)P_3 **6** and 2,2-F_2-Ins(1,4,5)P_3 **7** have K_i-values of 8.8 μM and 11.0 μM, respectively]. Interactions of racemic 2-F-*myo*-Ins(1,4,5)P_3 with 5-phosphatase have also been studied, and the fluorinated derivative was found to be a moderately potent competitive inhibitor of this enzyme (K_i 14.4 μM). It may tentatively be assumed that, by analogy with the difluoro-analogue **7**, the inhibitory effect of *myo*-fluoride DL-**8** is due to the presence of L-2-F-Ins(1,4,5)P_3 in the racemic mixture, whereas the D-isomer is probably a substrate for this enzyme as is D-**7**.

Clearly, these new Ins(1,4,5)P_3 mimics possess considerable potential as pharmacological tools for investigation of the polyphosphoinositide pathway of cellular signalling.

§ Note that in ref. 38 the active enantiomer of *scyllo*-Ins(1,2,4)P_3 was presumed to have the D-configuration; in fact it formally has the L-configuration, although it is related to D-*myo*-inositol 1,2,4-trisphosphate only by inversion of configuration at the C-2 position of this natural messenger.

Experimental

General methods

Chemicals were purchased from Aldrich, Fluka and Lancaster. Diethyl ether was dried over sodium wire and distilled. Dichloromethane, triethylamine and DMF were dried over calcium hydride, distilled and stored over 4 Å molecular sieves. Pyridine was dried by refluxing with sodium hydroxide pellets, followed by distillation, and stored over 5 Å sieves. TLC and high-performance TLC (HPTLC) was performed on pre-coated plates (Merck TLC aluminium sheets silica 60 F_{254}, Art. no. 5554 and Merck HPTLC plates silica 60 F_{254}, Art. no. 5635). Spots were visualised by spraying of plates with phosphomolybdic acid in methanol followed by heating. Flash chromatography refers to the method of Still *et al.*[44] and was carried out using Sorbsil C60 silica gel. Light petroleum refers to the fraction with distillation range 60–80 °C.

NMR spectra (proton frequency 90, 270 or 400 MHz) were referenced to internal $SiMe_4$ (^1H), $CDCl_3$ (^{13}C) and external 85% H_3PO_4 (^{31}P), respectively. *J*-Values are given in Hz. Mp (uncorrected) were determined using a Reichert-Jung Thermo Galen Kofler block. Microanalysis was carried out by the University of Bath microanalysis service. Mass spectra were recorded at the EPSRC Mass Spectrometry Service Centre, Swansea, and at the University of Bath. Optical rotations were measured using an Optical Activity Ltd. AA-10 polarimeter, and $[\alpha]_D$-values are given in 10^{-1} deg cm^2 g^{-1}. Ion-exchange chromatography was performed on an LKB-Pharmacia Medium Pressure Ion Exchange Chromatograph using Q Sepharose Fast Flow with gradients of triethylammonium hydrogen carbonate (TEAB) as eluent. Column fractions containing inositol polyphosphate analogues were assayed for total phosphate by a modification of the Briggs test[45] as described.[46] Compounds containing phosphorothioates were additionally assayed by a modification of the Ellman test[47] for sulfhydryl groups as described.[46]

(Diisopropylamino)dichlorophosphine was prepared by the method of Tanaka *et al.*[48] by adding two mole equivalents of diisopropylamine to a solution of PCl_3 in dry diethyl ether at −78 °C. The crude product (δ_P 166.4) was purified by distillation under reduced pressure, and reaction with two mole equivalents of benzyl alcohol in the presence of two mole equivalents of triethylamine afforded bis(benzyloxy)-(diisopropylamino)phosphine[49] (δ_P 145.24) which could be purified by flash chromatography.

DL-1-*O*-Allyl-3,6-di-*O*-benzyl-4,5-*O*-isopropylidene-2-*O*-trifluoromethylsulfonyl-*myo*-inositol 11

A solution of 1-*O*-allyl-3,6-di-*O*-benzyl-4,5-*O*-isopropylidene-*myo*-inositol **10**[39] (8.604 g, 19.5 mmol) and dry pyridine (10 ml) in dry dichloromethane (50 ml) was cooled to −78 °C. Triflic anhydride (6 ml, 10 g, 35.4 mmol) was added dropwise to the mixture. After the addition, cooling was removed and the mixture was stirred for 2 h at room temperature, after which TLC [light petroleum–diethyl ether (1 : 1)] showed complete conversion of the starting material (R_f 0.39) to a product (R_f 0.62). The reaction mixture was quenched with water (10 ml), diluted with dichloromethane (50 ml), and washed successively with saturated aq. $NaHCO_3$, water and brine (100 ml each). The organic layer was dried over magnesium sulfate and the solvents were evaporated off *in vacuo*. Toluene was added to the orange syrup in order to remove the remaining pyridine, which was then evaporated off to give **11** as a yellow solid in quantitative yield, which was not purified further; $\delta_H(CDCl_3$; 270 MHz) 1.45 (3 H, s, CH_3), 1.49 (3 H, s, CH_3), 3.39 (1 H, t, *J* 10.1, 5-H), 3.43 [1 H, dd, *J* 8.8 and 3.1, 3(1)-H], 3.68 [1 H, dd, *J* 10.5 and 2.8, 1(3)-H], 3.86 [1 H, dd, *J* 9.9 and 9.0, 4(6)-H], 3.88 [1 H, dd, *J* 10.4 and 9.3, 6(4)-H], 4.10–4.25 (2 H, m, $CH_2CH{=}CH_2$), 4.75 and 4.78 (2 H, AB, J_{AB} 12.5, CH_2Ph), 4.76 and 4.88 (2 H, AB, J_{AB} 11.5, CH_2Ph), 5.23 (1 H, t, *J* 2.9, 2-H), 5.17–5.32 (2 H, m, $CH_2CH{=}CH_2$), 5.99 (1 H, ddt, *J* 17.3, 10.35 and 5.9, $CH_2CH{=}CH_2$) and 7.25–7.37 (10 H, m, CH_2Ph); $\delta_C(CDCl_3$; 68 MHz) 26.87 (q, CH_3), 71.86, 73.06 and 73.54 (3 t, 2 × CH_2Ph and $CH_2CH{=}CH_2$), 73.57, 76.88, 77.70, 78.08, 78.93 and 85.84 (6 d, 6 × inositol ring C), 112.40 [s, $(CH_3)_2C$], 118.21 (t, $CH_2CH{=}CH_2$), 125.28 (CF_3), 127.52, 127.58, 127.78, 128.23, 128.36, 128.59 and 129.01 (7 d, CH_2Ph), 133.78 (d, $CH_2CH{=}CH_2$) and 137.15 and 138.28 (2 s, CH_2Ph); *m/z* (+ve ion FAB) 573 [$(M + H)^+$, 2%], 502 [$(M - HCF_3)^+$, 13] and 91 (100).

DL-2-*O*-Acetyl-1-*O*-allyl-3,6-di-*O*-benzyl-4,5-*O*-isopropylidene-*scyllo*-inositol 12

A mixture of crude sulfonate **11** (5.73 g, 10 mmol) and caesium acetate (2.78 g, 15 mmol) in dry DMF (150 ml) was stirred at room temperature for 2 h after which TLC [light petroleum–diethyl ether (1 : 1)] showed conversion of the starting material (R_f 0.62) into a product (R_f 0.70). The solution was concentrated *in vacuo* and the residue was partitioned between water and chloroform (100 ml each). The organic layer was dried over magnesium sulfate and evaporated. The crude product was purified by chromatography [diethyl ether–light petroleum (1 : 1)] to give acetate **12** (4.39 g, 9.1 mmol, 91%), mp 108–110 °C (from hexane) [Found: C, 69.5; H, 7.1. Calc. for $C_{28}H_{34}O_7$ (482.57): C, 69.69; H, 7.10%]; $\delta_H(CDCl_3$; 270 MHz) 1.47 (6 H, s, 2 × CH_3), 2.03 (3 H, s, $COCH_3$), 3.45 (1 H, dd, *J* 9.3 and 8.2, C-H), 3.49–3.63 (3 H, m, C-H), 3.71 (1 H, dd, *J* 9.2 and 8.2, C-H), 4.05 and 4.29 (2 H, AB, dt, J_{AB} 12.6, *J* 5.7 and 1.5, $CH_2CH{=}CH_2$), 4.61 and 4.85 (2 H, AB, J_{AB} 12.4, CH_2Ph), 4.71 and 4.88 (2 H, AB, J_{AB} 11.7, CH_2Ph), 5.08–5.22 (2 H, m, $CH_2CH{=}CH_2$), 5.15 (1 H, t, *J* 8.8, 2-H), 5.81 (1 H, ddt, *J* 17.2, 10.4 and 5.6, $CH_2CH{=}CH_2$) and 7.24–7.39 (10 H, m, CH_2Ph); $\delta_C(CDCl_3$; 68 MHz) 20.92 (q, $COCH_3$), 26.95 (q, CH_3), 72.07, 72.98 and 74.11 (3 t, CH_2Ph and $CH_2CH{=}CH_2$), 74.66, 76.77, 78.56, 78.88, 79.34 and 81.61 (6 d, inositol ring C), 112.58 [s, $(CH_3)_2C$], 116.77 (t, $CH_2CH{=}CH_2$), 127.44, 127.50, 127.70, 128.18 and 128.22 (5 d, CH_2Ph), 134.67 (d, $CH_2CH{=}CH_2$), 138.30 (s, CH_2Ph) and 169.70 (s, CO); *m/z* (+ve ion FAB) 483 [$(M + H)^+$, 4.5%] and 91 (100); *m/z* (−ve ion FAB) 635 [(M + NBA)$^-$, 60%], 485 (50) and 212 (100).

DL-1-*O*-Allyl-3,6-di-*O*-benzyl-4,5-*O*-isopropylidene-*scyllo*-inositol 13

A mixture of acetate **12** (3.19 g, 6.6 mmol) in methanol (50 ml) and 1 M aq. sodium hydroxide (6 ml) was heated under reflux for 30 min after which TLC [light petroleum–diethyl ether (1 : 1)] showed complete conversion of the starting material (R_f 0.70) into a product (R_f 0.63). The solution was allowed to cool and was neutralised with solid carbon dioxide. The solvent was evaporated off and the residue was partitioned between water and chloroform. The organic phase was dried over magnesium sulfate and evaporated to give compound **13** as a solid (2.46 g, 5.6 mmol, 85%), mp 101–103 °C (from hexane) (lit.,[38] 105–106 °C) [Found: C, 70.7; H, 7.3. Calc. for $C_{26}H_{32}O_6$ (440.54): C; 70.89; H, 7.32%]; $\delta_H(CDCl_3$; 270 MHz) 1.46 (6 H, s, 2 × CH_3), 2.73 (1 H, d, *J* 2.4, D_2O ex, OH), 3.39 (1 H, t, *J* 8.5, C-H), 3.51–3.64 (5 H, m, 5 × C-H), 4.28 and 4.39 (2 H, AB, dt, J_{AB} 12.5, 5.8 and 1.5, $CH_2CH{=}CH_2$), 4.72 and 4.90 (2 H, AB, J_{AB} 11.7, CH_2Ph), 4.72 and 4.93 (2 H, AB, J_{AB} 11.7, CH_2Ph), 5.16 (1 H, dq, *J* 10.3 and 1.4, *Z*-$CH_2CH{=}CH_2$), 5.26 (1 H, dq, *J* 17.2 and 1.5, *E*-$CH_2CH{=}CH_2$), 5.94 (1 H, ddt, *J* 17.2, 10.4 and 5.7, $CH_2CH{=}CH_2$) and 7.24–7.40 (10 H, m, CH_2Ph); $\delta_C(CDCl_3$; 68 MHz) 26.92 (q, CH_3) 72.65, 72.95 and 74.37 (3 t, CH_2Ph and $CH_2CH{=}CH_2$), 75.90, 78.43, 78.49, 79.11, 79.37 and 83.39 (6 d, inositol ring C), 112.29 [s, $(CH_3)_2C$], 116.96 (t, $CH_2CH{=}CH_2$), 127.44, 127.60, 127.66, 127.83, 128.22 and

128.31 (6 d, CH_2Ph), 134.99 (d, $CH_2CH{=}CH_2$) and 138.27 and 138.43 (2 s, CH_2Ph); *m/z* (+ve ion FAB) 441 [$(M + H)^+$, 8%] and 91 (100); *m/z* (−ve ion FAB) 593 [$(M + NBA)^-$, 100%] and 440 (M^-, 30).

DL-2-*O*-Acetyl-1-*O*-allyl-3,6-di-*O*-benzyl-*scyllo*-inositol 14

A solution of compound **12** (3.57 g, 7.4 mmol) in methanol–1 M HCl (9:1, 100 ml) was heated under reflux for 20 min after which TLC [ethyl acetate–hexane (1:1)] showed complete conversion of the starting material (R_f 0.85) into a product (R_f 0.44). The mixture was allowed to cool and was neutralised with $NaHCO_3$. The solution was concentrated and then partitioned between water and chloroform (100 ml each). The organic phase was dried over magnesium sulfate and evaporated to give diol **14** (2.7 g, 6.1 mmol, 83%), mp 142–143 °C (from ethyl acetate–hexane) [Found: C, 67.8; H, 6.8. Calc. for $C_{25}H_{30}O_7$ (442.51): C, 67.86; H, 6.83%]; $\delta_H(CDCl_3$; 270 MHz) 2.00 (3 H, s, $COCH_3$), 2.87 (1 H, d, *J* 2.4, D_2O ex, OH), 3.03 (1 H, d, *J* 2.4, D_2O ex, OH), 3.32–3.57 (5 H, m, 1-, 3-, 4-, 5-, 6-H) 4.09 and 4.28 (2 H, AB, dt, J_{AB} 12.5, 5.7 and 1.5, $CH_2CH{=}CH_2$), 4.67 and 4.76 (2 H, AB, J_{AB} 11.6, CH_2Ph), 4.76 and 4.87 (2 H, AB, J_{AB} 11.1, CH_2Ph), 5.10 (1 H, t, *J* 9.7, 2-H), 5.14 (1 H, dq, *J* 10.4 and 1.3, *Z*-$CH_2CH{=}CH_2$), 5.22 (1 H, dq, *J* 17.2 and 1.5, *E*-$CH_2CH{=}CH_2$), 5.84 (1 H, ddt, *J* 17.2, 10.4 and 5.7, $CH_2CH{=}CH_2$) and 7.24–7.37 (10 H, m, CH_2Ph); $\delta_C(CDCl_3$; 68 MHz) 20.98 (q, $COCH_3$), 73.66 (2 d, 2 × inositol ring C), 73.76, 79.98, 80.34 and 81.80 (4 d, inositol ring C), 74.15, 74.70 and 75.38 (3 t, 2 × CH_2Ph and $CH_2CH{=}CH_2$), 116.96 (t, $CH_2CH{=}CH_2$), 127.70, 127.76, 127.89, 127.99, 128.44 and 128.51 (6 d, CH_2Ph), 134.46 (d, $CH_2CH{=}CH_2$), 138.11 and 138.21 (2 s, CH_2Ph) and 169.90 (s, CO); *m/z* (+ve ion FAB) 443 [$(M + H)^+$, 4%] and 91 (100); *m/z* (−ve ion FAB) 883 [$(2M - H)^-$, 19%], 553 [$(M + NBA)^-$, 100] and 441 [$(M - H)^-$, 50].

DL-1-*O*-Allyl-3,6-di-*O*-benzyl-*scyllo*-inositol 15

(a) A solution of compound **13** (1.414 g, 3.21 mmol) in 80% acetic acid (50 ml) was heated under reflux for 15 min, cooled and diluted with water (50 ml). After the mixture had been kept at −20 °C overnight the precipitate was collected to give triol **15** (1.155 g, 2.88 mmol, 90%).

(b) A mixture of diol **14** (2.24 g, 5.06 mmol) in methanol (100 ml) and 1 M aq. sodium hydroxide (12 ml) was heated under reflux for 30 min after which TLC [ethyl acetate–hexane (2:1)] showed complete conversion of the starting material (R_f 0.63) into a product (R_f 0.40). The solution was cooled, and neutralised with solid carbon dioxide. The solvent was evaporated off and the residue was partitioned between water and ethyl acetate. The organic phase was dried over magnesium sulfate and evaporate to give triol **15** as a solid (1.76 g, 4.4 mmol, 87%), mp 180 °C (from ethyl acetate) [Found: C, 69.0; H, 7.05. Calc. for $C_{23}H_{28}O_6$ (400.47): C; 68.98; H, 7.05%]; $\delta_H([^2H_6]$DMSO; 270 MHz) 3.05–3.21 (5 H, m, 5 × C-H), 3.32–3.37 (1 H, m, D_2O ex gives t, *J* 9.0, C-H), 4.20 and 4.32 (2 H, AB, br d, J_{AB} 12.8, 5.5, $CH_2CH{=}CH_2$), 4.71 and 4.81 (2 H, AB, J_{AB} 11.4, CH_2Ph), 4.79 and 4.82 (2 H, AB, J_{AB} 11.5, CH_2Ph), 5.00 (1 H, d, *J* 3.7, D_2O ex, OH), 5.04–5.06 (2 H, m, D_2O ex gives 1 H, d, *J* 10.4, *Z*-$CH_2CH{=}CH_2$, OH), 5.13 (1 H, d, *J* 5.5, D_2O ex, OH), 5.21 (1 H, dq, *J* 17.4 and 1.65, *E*-$CH_2CH{=}CH_2$), 5.94 (1 H, ddt, *J* 17.2, 10.4 and 5.5, $CH_2CH{=}CH_2$) and 7.23–7.45 (10 H, m, CH_2Ph); $\delta_C([^2H_6]$-DMSO; 68 MHz) 73.40, 73.82 and 74.21 (3 t, CH_2Ph and $CH_2CH{=}CH_2$), 73.75, 73.92, 74.34, 82.48, 82.54 and 82.90 (6 d, inositol ring C), 115.60 (t, $CH_2CH{=}CH_2$), 127.11, 127.21, 127.60, 127.69, 127.99 and 128.08 (6 d, CH_2Ph), 136.42 (d, $CH_2CH{=}CH_2$) and 139.53 and 139.79 (2 s, CH_2Ph); *m/z* (+ve ion FAB) 401 [$(M + H)^+$, 4%] and 91 (100); *m/z* (−ve ion FAB) 553 [$(M + NBA)^-$, 80%] and 399 [$(M - H)^-$, 100].

DL-1-*O*-Allyl-3,6-di-*O*-benzyl-*scyllo*-inositol-2,4,5-tris(dibenzyl phosphate) 16

A solution of bis(benzyloxy)(diisopropylamino)phosphine (19.9 g, 26 mmol) in dichloromethane (50 ml) was added to a solution of triol **15** (1 g, 2.5 mmol) and tetrazole (3.15 g, 45 mmol) in dichloromethane (50 ml). The mixture was stirred at room temperature for 1 h. Water (2 ml) in tetrahydrofuran (THF) (20 ml) was added and the mixture was stirred for a further 30 min. 2,6-Lutidine (2 ml) followed by *tert*-butyl hydroperoxide (20 ml) was then added and the mixture was stirred overnight. The solution was washed with saturated aq. sodium hydrogen carbonate (2 × 100 ml) and dried over magnesium sulfate. The solvents were evaporated off and the residue was chromatographed on silica gel with 0–100% ethyl acetate in hexane. The product was recrystallised from ethanol to give trisphosphate **16** (1.294 g, 1.5 mmol, 58%), TLC [ethyl acetate–hexane (1:1)] R_f 0.38; mp 103–105 °C [Found: C, 66.1; H, 5.7. Calc. for $C_{65}H_{67}O_{15}P_3$ (1181.16): C, 66.10; H, 5.72%]; $\delta_H(CDCl_3$; 270 MHz) 3.58 (1 H, t, *J* 8.4, C-H), 3.67 (1 H, t, *J* 8.4, C-H), 3.73 (1 H, t, *J* 8.4, C-H), 4.12–4.26 (2 H, m, $CH_2CH{=}CH_2$), 4.55–5.15 (21 H, m, 8 × CH_2Ph, CH_2-$CH{=}CH_2$, 3 × C-H), 5.78 (1 H, ddt, *J* 17.2, 10.4 and 5.5, $CH_2CH{=}CH_2$), 6.97–7.35 (40 H, m, 8 × CH_2Ph); $\delta_C(CDCl_3$; 68 MHz) 69.18, 69.28, 69.38 and 69.44 (4 t, 4 × PO-CH_2Ph), 73.56, 73.79 and 74.31 (3 t, 2 × CH_2Ph and $CH_2CH{=}CH_2$), 77.71, 78.14, 78.98, 79.08, 79.43 and 79.66 (6 d, 6 × inositol ring C), 117.09 (t, $CH_2CH{=}CH_2$), 127.11, 127.24, 127.79, 127.99, 128.09 and 128.25 (6 d, CH_2Ph), 134.25 (d, $CH_2CH{=}CH_2$) and 135.71, 137.85 and 137.91 (3 s, CH_2Ph); $\delta_P(CDCl_3$; 162 MHz; ^{1}H-decoupled) −1.75 (2 P) and −1.52 (1 P); $\delta_P(CDCl_3$; 162 MHz; ^{1}H-coupled) −1.72 (2 P, q, *J* 7.9) and −1.47 (1 P, q, *J* 7.9); *m/z* (+ve ion FAB) 1181 (1%), 1090 [$(M - C_7H_7)^+$, 0.3], 181 (12) and 91 (100); *m/z* (−ve ion FAB) 1088 [$(M - C_7H_7 - 2H)^-$, 7%], 998 (2.5), 277 (100) and 187 (16).

DL-*scyllo*-Inositol 1,2,4-trisphosphate 3

Ammonia was condensed into a three-neck flask at −78 °C. An excess of sodium was added to dry the liquid ammonia, which was then distilled into a second three-neck flask and kept at −78 °C. Sodium was added until the solution remained blue. Compound **16** (311 mg, 263 μmol) was dissolved in dry 1,4-dioxane (2 ml) and the solution was added to the sodium–liquid ammonia mixture. After being stirred for 15 min the reaction was quenched with ethanol. Ammonia and ethanol were evaporated off and the crude product was purified by ion-exchange chromatography on Pharmacia Q Sepharose Fast Flow with a gradient of TEAB buffers (0.1–1 M), pH 8.0, as eluent. The triethylammonium salt of compound **3** eluted at between 320 and 460 mM (yield 240 μmol, 91%); $\delta_H(D_2O$; pH = 8; 270 MHz) 3.48–3.52 (2 H, m, 2 × C-H), 3.58 (1 H, t, *J* 9.3, C-H) and 3.84–4.07 (3 H, m, 3 × C-H); $\delta_P(D_2O$; pH = 8; 162 MHz; ^{1}H-decoupled) 0.50 (1 P) and 0.72 (2 P); *m/z* (+ve ion FAB) 623 [$(M + 2Et_3NH)^+$, 1.1%], 522 [$(M + Et_3NH)^+$, 1.4] and 102 (Et_3NH^+, 100); *m/z* (−ve ion FAB) 839 [$(2M)^-$, 15%], 419 (M^-, 100) and 97 (10) [Found: *m/z*, 418.9550. Calc. for $C_6H_{14}O_{15}P_3$: $(M - H)^-$, 418.9546].

DL-3,6-Di-*O*-benzyl-1-*O*-[(*Z*)-prop-1-enyl]-*scyllo*-inositol 17

A solution of compound **15** (443 mg, 1.106 mmol) and freshly sublimed potassium *tert*-butoxide (570 mg, 5 mmol) in dry DMSO (20 ml) was stirred for 3 h at 50 °C after which TLC [ethyl acetate–hexane (2:1)] showed complete conversion of the starting material (R_f 0.67) into a product (R_f 0.73). Water (50 ml) was added and the mixture was extracted with diethyl ether (2 × 50 ml). The organic phase was dried over magnesium sulfate and evaporated and the crude product was purified by flash chromatography [diethyl ether–hexane (1:1)] to give *title compound* **17** (430 mg, 97%), mp 140 °C (from aq. ethanol) [Found: C, 68.6; H, 7.1. Calc for $C_{23}H_{28}O_6$ (400.47): C; 68.98;

H, 7.05%]; δ_H([$^2H_6$]DMSO; 270 MHz) 1.53 (3 H, d, *J* 6.8, CH_3), 3.07–3.15 (1 H, m, C-H), 3.22–3.29 (3 H, m, C-H), 3.41–3.49 (2 H, m, C-H), 4.18 (1 H, quint, *J* 6.6, CH=C*H*CH_3), 4.66 and 4.75 (2 H, AB, J_{AB} 11.2, CH_2Ph), 4.79 and 4.79 (2 H, AB, J_{AB} 12.1, CH_2Ph), 5.04 (1 H, br s, D_2O ex, OH), 5.13 (1 H, br s, D_2O ex, OH), 5.26 (1 H, d, *J* 5.3, D_2O ex, OH), 6.21 (1 H, dd, *J* 6.4 and 1.5, *CH*=CHCH$_3$) and 7.21–7.45 (10 H, m, CH_2*Ph*); δ_C([$^2H_6$]DMSO; 68 MHz) 9.24 (q, CH=CH*C*H_3), 72.43, 73.50, 73.79, 81.45, 82.52 and 85.05 (6 d, 6 × inositol ring C), 73.60 and 73.92 (2 t, *C*H_2Ph), 96.24 (d, CH=*C*HCH$_3$), 126.89, 127.05, 127.47, 127.54 and 127.80 (5 d, CH_2*Ph*), 139.05 and 139.47 (2 s, CH_2*Ph*) and 148.13 (d, *C*H=CHCH$_3$); *m*/*z* (+ve ion FAB) 401 [(M + H)$^+$, 4%], 181 (8) and 91 (100); *m*/*z* (−ve ion FAB) 799 [(2 M − H)$^-$, 13%], 566 (49), 553 [(M + NBA)$^-$, 77], 399 [(M − H)$^-$, 100] and 341 (38).

1,4-Di-*O*-benzyl-*scyllo*-inositol 18

Compound **17** (190 mg, 474 μmol) was heated under reflux in a mixture of methanol–1 M HCl (5:1) for 15 min after which TLC [ethyl acetate–hexane (2:1)] showed complete conversion of the starting material (R_f 0.73) into a product (R_f 0.43). The solvent was evaporated off and the residue was collected to give tetraol **18** (124 mg, 344 μmol, 73%), mp 309–310 °C (from ethanol) with a phase transition between 240 and 260 °C [Found: C, 66.6; H, 6.7. Calc. for $C_{20}H_{24}O_6$ (360.41): C, 66.65; H, 6.71%]; δ_H([$^2H_6$]DMSO; 270 MHz) 3.03–3.07 (2 H, m, 1-, 4-H), 3.18–3.21 (4 H, m, 2-, 3-, 5-, 6-H), 4.76 (2 H, AB s, CH_2Ph), 4.96 and 4.97 (2 H, AB d, CH_2Ph) and 7.21–7.41 (10 H, m, CH_2*Ph*); δ_C([$^2H_6$]DMSO; 68 MHz) 73.79 (2 t, 2 × *C*H_2Ph), 74.27 (4 d, 4 × C-OH), 82.90 (2 d, 2 × C-O-benzyl), 127.14 (2 d, CH_2*Ph*), 127.69 and 128.05 (2 × 2 d, CH_2*Ph*) and 139.89 (2 s, CH_2*Ph*).

3,6-Di-*O*-benzyl-*scyllo*-inositol 1,2,4,5-tetrakis(dibenzyl phosphate) 19

Compound **18** (307 mg, 852 μmol) and tetrazole (1.19 g, 17 mmol) were stirred at room temperature in dry DMF (15 ml). Bis(benzyloxy)(diisopropylamino)phosphine was added to the mixture and stirring was continued for 1 h, after which ^{31}P NMR showed a single peak at δ_P 141. 2,6-Lutidine (2 ml) followed by *tert*-butyl hydroperoxide (6 ml), was then added and stirring was continued overnight. The solution was partitioned between chloroform and saturated aq. sodium hydrogen carbonate (50 ml each). The organic phase was dried over magnesium sulfate, the solvents were evaporated off, and the residue was chromatographed on silica gel with 0–100% ethyl acetate in hexane. The product was recrystallised from ethanol to give compound **19** (885 mg, 632 mmol, 74%), R_f [hexane–ethyl acetate (1:1)] 0.46; mp 110–111 °C (from methanol) [Found: C, 65.2; H, 5.45. Calc. for $C_{76}H_{76}O_{18}P_4$ (1401.32): C, 65.14; H, 5.47%]; δ_H($CDCl_3$; 270 MHz) 4.01 (2 H, t, br, 3-, 6-H), 4.66 (4 H, br s, 1-, 2-, 4-, 5-H), 4.73–5.02 (10 H, m, CH_2Ph) and 7.06–7.30 (60 H, m, CH_2*Ph*); δ_C($CDCl_3$; 68 MHz) 69.51 and 73.14 (2 t, *C*H_2Ph), 77.55 and 79.73 (2 d, 6 × inositol ring C), 127.37, 127.47, 127.86, 128.12, 128.25 and 128.35 (6 d, CH_2*Ph*) and 135.55, 135.64, 135.77 and 137.53 (4 s, CH_2*Ph*); δ_P($CDCl_3$; 36 MHz; ^{1}H-decoupled) −3.71; *m*/*z* (+ve ion FAB) 1401 [(M + H)$^+$, 0.5%] and 1310 [(M + H − C_7H_7)$^+$, 0.2]; *m*/*z* (−ve ion FAB) 1309 [(M − C_7H_7)$^-$, 2.3%], 1218 [(M − 2 C_7H_7)$^-$, 0.6] and 277 [(OP(O)(OC_7H_7)$_2$)$^-$, 100]

scyllo-Inositol 1,2,4,5-tetrakisphosphate 4

Ammonia was condensed into a three-neck flask at −78 °C. An excess of sodium was added to dry the liquid ammonia, which was then distilled into a second three-neck flask and kept at −78 °C. Sodium was added until the solution remained blue. Compound **19** (252 mg, 180 μmol) was dissolved in dry 1,4-dioxane (4 ml) and the solution was added to the sodium–liquid ammonia mixture. After being stirred for 15 min the reaction was quenched with ethanol. Ammonia and ethanol were evaporated off and the crude product was purified by ion-exchange chromatography on Pharmacia Q Sepharose Fast Flow with a gradient of TEAB buffers (0.1–1 M), pH 8.0, as eluent. The triethylammonium salt of compound **4** eluted between 560 and 650 mM (74 μmol, 41%); δ_H(D_2O; 270 MHz) 3.48 (2 H, t, *J* 9.3, 3-, 6-H) and 3.87 (4 H, dt, *J* 12.2 and 9.3, 1-, 2-, 4-, 5-H); δ_C(D_2O; 100 MHz) 73.61 (2 C) and 77.09 (4 C); δ_P(D_2O; 36 MHz; ^{1}H-coupled) 2.83 (d, *J* 8.3); *m*/*z* (+ve ion FAB) 804 (1%), 703 (2.5) and 102 (100); *m*/*z* (−ve ion FAB) 998 (4.5%), 555 (4.5) and 499 [(M − H)$^-$, 100] [Found: *m*/*z*, 498.9191. Calc. for $C_6H_{15}O_{18}P_4$: (M − H)$^-$, 498.9209].

3,6-Di-*O*-benzyl-*scyllo*-inositol 1,2,4,5-tetrakis(*O*,*O*-dibenzyl thiophosphate) 20

Compound **18** (522 mg, 1.45 mmol) and tetrazole (2.02 g, 29 mmol) were stirred at room temperature in dry DMF (25 ml). Bis(benzyloxy)(diisopropylamino)phosphine was added to the mixture and the mixture was stirred for 1 h, when ^{31}P NMR showed a single peak at δ_P 141. Pyridine and an excess of sulfur were then added and stirring was continued for 10 min. The solution was separated between chloroform and saturated aq. sodium hydrogen carbonate (50 ml each). The organic phase was dried over magnesium sulfate, the solvents were evaporated off, and the residue was chromatographed on silica gel [hexane–diethyl ether (3:1)]. The product was recrystallised from ethanol to give compound **20** (1.8 g, 1.23 mmol, 85%), R_f [hexane–diethyl ether (1:1)] 0.63; mp 91–93 °C (from ethyl acetate–ethanol) [Found: C, 61.9; H, 5.2. Calc. for $C_{76}H_{76}O_{14}P_4S_4$ (1465.57): C, 62.29; H, 5.23%]; δ_H($CDCl_3$; 270 MHz) 4.15 (2 H, t, br, 3-, 6-H), 4.58 (4 H, br s, 1-, 2-, 4-, 5-H), 4.75–5.23 (10 H, m, CH_2Ph) and 7.08–7.23 (60 H, m, CH_2*Ph*); δ_C($CDCl_3$; 68 MHz) 70.03 and 72.59 (2 t, *C*H_2Ph), 77.97 and 79.92 (2 d, inositol ring C), 127.43, 127.99, 128.05, 128.12, 128.22 and 128.34 (6 d, CH_2*Ph*) and 135.56, 135.64, 135.71 and 137.53 (4 s, CH_2*Ph*); δ_P($CDCl_3$; 36 MHz; ^{1}H-decoupled) 67.77; *m*/*z* (+ve ion FAB) 1465 [M$^+$, 0.1%], 1035 (0.1), 945 (0.2) and 91 (100); *m*/*z* (−ve ion FAB) 1373 [(M − C_7H_7)$^-$, 9%], 1282 [(M − 2 C_7H_7)$^-$, 3], 1249 [(M − 2 OC_7H_7)$^-$, 4] and 293 [OP(S)(OC_7H_7)$_2^-$, 100]

scyllo-Inositol 1,2,4,5-tetrakisphosphorothioate 5

Ammonia was condensed into a three-neck flask at −78 °C. An excess of sodium was added to dry the liquid ammonia, which was then distilled into a second three-neck flask and kept at −78 °C. Sodium was added until the solution remained blue. Compound **20** (205 mg, 140 μmol) was dissolved in dry 1,4-dioxane (3 ml) and the solution was added to the sodium–liquid ammonia mixture. After being stirred for 15 min the reaction was quenched with ethanol. Ammonia and ethanol were evaporated off and the crude product was purified by ion-exchange chromatography on Pharmacia Q Sepharose Fast Flow with a gradient of TEAB buffers (0.1–1 M), pH 8.0, as eluent. The triethylammonium salt of compound **5** eluted between 850 and 950 mM (25 μmol, 18%); δ_H(D_2O; 270 MHz) 3.46 (2 H, t, *J* 9.1, 3-, 6-H) and 3.83 (4 H, dt, *J* 11.8 and 9.1, 1-, 2-, 4-, 5-H); δ_C(D_2O; 68 MHz) 80.41 (2 C) and 84.97 (4 C); δ_P(D_2O; 36 MHz) 43.48 (d, *J* 11.0); *m*/*z* (−ve ion FAB) 563 [(M − H)$^-$, 95%], 529 (18), 188 (47), 154 (32), 113 (28) and 95 [(PSO_2)$^-$, 100] [Found: *m*/*z*, 562.8273. Calc. for $C_6H_{15}O_{14}P_4S_4$: (M − H)$^-$, 562.8295].

DL-3,6-Di-*O*-benzyl-4,5-*O*-isopropylidene-1-*O*-[(*Z*)-prop-1-enyl]-*scyllo*-inositol 21

A solution of compound **13** (3.16 g, 7.2 mmol) and freshly sublimed potassium *tert*-butoxide (4 g, 3.27 mmol) in dry DMSO (50 ml) was stirred for 2 h at 55 °C, after which TLC [hexane–diethyl ether (1:1)] showed complete conversion of the starting material (R_f 0.59) into a product (R_f 0.65). Water (100 ml) was added and the mixture was extracted with diethyl ether (2 × 100 ml). The organic phase was dried over

magnesium sulfate and evaporated, and the crude product was purified by flash chromatography [hexane–diethyl ether (3:1)] to give isomer **21** (3.03 g, 95%), mp 109–110 °C (from hexane) [Found: C, 70.7; H, 7.3. Calc. for $C_{26}H_{32}O_6$ (440.54): C, 70.89; H, 7.32%]; δ_H($CDCl_3$; 270 MHz) 1.46 (6 H, s, 2 × CH_3), 1.61 (3 H, dd, *J* 6.8 and 1.65, OCH=CHCH_3), 2.69 (1 H, d, *J* 2.4, D_2O ex, OH), 3.52–3.75 (6 H, m, 6 × C-H), 4.37 (1 H, quint, *J* 6.8, OCH=C*H*CH_3), 4.71 and 4.93 (2 H, AB, J_{AB} 11.9, CH_2Ph), 4.74 and 4.82 (2 H, AB, J_{AB} 11.7, CH_2Ph), 6.18 (1 H, dd, *J* 6.8 and 1.65, OC*H*=CHCH$_3$) and 7.28–7.39 (10 H, m, CH_2Ph); δ_C($CDCl_3$; 68 MHz) 8.99 (q, OCH=CH*C*H$_3$), 26.63 (q, CH_3) 72.40 and 72.62, (2 t, *C*H$_2$Ph), 75.22, 77.75, 78.04, 78.10, 78.43 and 85.82 (6 d, inositol ring C), 99.38 (d, OCH=*C*HCH$_3$), 112.16 [s, $(CH_3)_2C$], 127.18, 127.37, 127.54, 127.89 and 128.05 (5 d, CH_2Ph), 137.92 (s, CH_2Ph) and 147.00 (d, O*C*H=CHCH$_3$); *m*/*z* (+ve ion FAB) 441 [(M + H)$^+$, 3%] and 91 (100); *m*/*z* (−ve ion FAB) 593 [(M + NBA)$^-$, 100%], 399 [(M − CH=CHCH$_3$)$^-$, 66], 291 (31) and 105 (38).

DL-3,6-Di-*O*-benzyl-4,5-*O*-isopropylidene-1-*O*-[(*Z*)-prop-1-enyl]-2-*O*-trifluoromethylsulfonyl-*scyllo*-inositol 22

A mixture of compound **21** (1.654 g, 3.75 mmol) and dry pyridine (10 ml) in dry dichloromethane (50 ml) was cooled to −78 °C. Triflic anhydride (1.8 ml, 3 g, 10.62 mmol) was added dropwise to this mixture. After the addition, the cooling bath was removed and the mixture was stirred for 2 h at room temperature, after which TLC [diethyl ether–light petroleum (40–60 °C) (1:1)] showed conversion of the starting material (R_f 0.53) to a product (R_f 0.67). The reaction mixture was quenched with water (10 ml), diluted with dichloromethane (50 ml), and washed successively with sat. aq. $NaHCO_3$, water and brine. The organic layer was dried over magnesium sulfate and the solvents were evaporated off *in vacuo*. Toluene was added to the orange syrup in order to remove the remaining pyridine, which was then evaporated off to give title compound **22** as a yellow solid in quantitative yield, *m*/*z* (+ve ion FAB) 863 (1.8%), 650 (1.5), 502 [(M − HCF_3), 1.6], 212 (85) and 91 (100); *m*/*z* (−ve ion FAB) 931 (1%) and 149 (100).

DL-3,6-Di-*O*-benzyl-2-deoxy-2-fluoro-4,5-*O*-isopropylidene-1-*O*-[(*Z*)-prop-1-enyl]-*myo*-inositol 23

TBAF (15 ml of a 1.1 M solution in THF) was added to a solution of crude sulfonate **22** (2.15 g, 3.75 mmol) in THF (20 ml) at room temperature. The colour of the reaction mixture turned to red-brown and TLC [diethyl ether–light petroleum (40–60 °C)] showed conversion of the starting material (R_f 0.67) to a product (R_f 0.56). Stirring was continued for a further 1 h. The solvent was evaporated off and the brown residue was taken up in chloroform. After being washed with water, the organic phase was dried over magnesium sulfate and evaporated, and the crude product was purified by flash chromatography [hexane–diethyl ether (3:1)] to give fluoride **23** (1.143 g, 2.58 mmol, 69%), mp 70–72 °C (from ethanol) [Found: C, 70.4; H, 7.0. Calc. for $C_{26}H_{31}FO_5$ (442.53): C, 70.57; H, 7.06%]; δ_H($CDCl_3$; 270 MHz) 1.47 (3 H, s, CH_3), 1.48 (3 H, s, CH_3), 1.64 (3 H, dd, *J* 7.0 and 1.65, OCH=CHCH_3), 3.41 (1 H, t, *J* 9.6, C-H), 3.54 [1 H, ddd, *J* 29.5, 8.8 and 2.4, 1(3)-H], 3.59 [1 H, ddd, *J* 23.3, 10.4 and 2.2, 3(1)-H], 3.95–4.07 (2 H, m, 2 × C-H), 4.50 (1 H, quint, *J* 6.8, OCH=C*H*CH_3), 4.72 and 4.84 (2 H, AB, J_{AB} 12.3, CH_2Ph), 4.77 and 4.82 (2 H, AB, J_{AB} 12.0, CH_2Ph), 4.91 (1 H, dt, *J* 50.0 and 2.4, 2-H), 6.01 (1 H, ddd, *J* 7.0, 1.65 and 0.7, OC*H*=CHCH$_3$) and 7.21–7.66 (10 H, m, CH_2Ph); δ_C($CDCl_3$; 68 MHz) 9.24 (q, OCH=CH*C*H$_3$), 26.82 and 26.92 [2 q, C(*C*H$_3$)$_2$], 71.52 and 73.37 (2 t, 2 × *C*H$_2$Ph), 74.74 [dd, $J_{C\text{-}C\text{-}F}$ 15.4, C-1(3)], 76.74, 76.81 and 78.95 (3 d, inositol ring C), 81.86 [dd, $J_{C\text{-}C\text{-}F}$ 17.6, C-3(1)], 90.24 (dd, $J_{C\text{-}F}$ 187.3, C-2), 103.18 (d, OCH=*C*HCH$_3$), 112.26 [s, *C*(CH$_3$)$_2$], 127.50, 127.60, 127.73, 127.86, 128.18 and 128.41 (6 d, CH_2Ph), 137.52 and 138.24 (2 s, CH_2Ph) and 144.95 (d, O*C*H=CHCH$_3$); δ_F($CDCl_3$ with reference to $CFCl_3$; 84 MHz) −210.21 [dt, $J_{2\text{-}H\text{-}F}$ 50.0, $J_{1(3)\text{-}H\text{-}F}$ 29.5]; *m*/*z* (+ve ion FAB) 443 [(M + H)$^+$, 6%], 351 (3) and 91 (100); *m*/*z* (−ve ion FAB) 595 [(M + NBA)$^-$, 100%], 488 (20), 401 [(M − CH=CHCH$_3$)$^-$, 14], 172 (69) and 133 (56).

DL-3,6-Di-*O*-benzyl-2-deoxy-2-fluoro-*myo*-inositol 24

Compound **23** (801 mg, 1.81 mmol) was heated under reflux in 60 ml of a mixture of methanol and 1 M HCl (5:1) for 30 min. The reaction was allowed to cool and was then made alkaline with an excess of sodium hydrogen carbonate. The solvents were evaporated off and the residue was extracted with ethyl acetate to give compound **24** (451 mg, 1.24 mmol, 69%), mp 193–194 °C (from ethanol) [Found: C, 66.0; H, 6.4. Calc. for $C_{20}H_{23}FO_5$ (362.40): C, 66.29; H, 6.40%]; δ_H([$^2H_6$] DMSO; 270 MHz) 3.21–3.59 (5 H, m, 5 × C-H), 4.67 (2 H, AB, s, CH_2Ph), 4.78 and 4.82 (2 H, AB, J_{AB} 11.7, CH_2Ph), 4.85 (1 H, d, *J* 54.0, 2-H), 5.09 (1 H, d, *J* 5.1, D_2O ex, OH), 5.16 (1 H, d, *J* 5.0, D_2O ex, OH), 5.32 (1 H, d, *J* 6.0, D_2O ex, OH) and 7.21–7.45 (10 H, m, CH_2Ph); δ_C([$^2H_6$] DMSO; 68 MHz) 69.62 [dd, $J_{C\text{-}C\text{-}F}$ 15.4, C-1(3)], 71.29 (t, *C*H$_2$Ph), 72.14 (d, inositol ring C), 73.73 (t, *C*H$_2$Ph), 74.37 (d, inositol ring C), 77.88 [dd, $J_{C\text{-}C\text{-}F}$ 17.7, C-3(1)], 81.38 (d, inositol ring C), 92.23 (dd, $J_{C\text{-}F}$ 178.5, C-2), 126.92, 127.21, 127.41, 127.47, 127.80 and 128.02 (6 d, CH_2Ph) and 138.76 and 139.50 (2 s, CH_2Ph); δ_F([$^2H_6$] DMSO with reference to $CFCl_3$; 84 MHz) −212.6 [dt, $J_{2\text{-}H\text{-}F}$ 51.3, $J_{1(3)\text{-}H,F}$ 29.3]; *m*/*z* (+ve ion FAB) 361 [(M − H)$^+$, 8%] and 91 (100); *m*/*z* (−ve ion FAB) 528 (70%), 515 [(M + NBA)$^-$, 100], 470 (20), 408 (23), 361 [(M − H)$^-$, 71] and 322 (50).

DL-3,6-Di-*O*-benzyl-2-deoxy-2-fluoro-1,4,5-tris(dibenzyl phosphate)-*myo*-inositol 25

Compound **24** (185 mg, 510 μmol) and tetrazole (763 mg, 10.9 mmol) were stirred at room temperature in dichloromethane (30 ml). Bis(benzyloxy)(diisopropylamino)phosphine was added and stirring was continued for 1 h, after which ^{31}P NMR showed three signals at δ_P 141.74, 141.60 and 141.47. The mixture was cooled to −78 °C and 2,6-lutidine (2 ml) and then *tert*-BuOOH (5 ml, 80% in *tert*-butyl alcohol) were added to the mixture. The mixture was allowed to warm to room temperature and the mixture was stirred for 1 h. The solution was partitioned between chloroform and saturated aq. $NaHCO_3$, the organic phase was dried over magnesium sulfate and the solvents were evaporated off. The residue was chromatographed on silica gel to give compound **25** (441 mg, 386 μmol, 76%), mp 129–131 °C [Found: C, 65.2; H, 5.5. Calc. for $C_{62}H_{62}FO_{14}P_3$ (1143.09): C, 65.15; H, 5.47%]; δ_H($CDCl_3$; 270 MHz) 3.50 (1 H, ddd, *J* 27.0, 9.8 and 1.5, 3-H), 3.98 (1 H, t, *J* 9.6, 6-H), 4.28 (1 H, ddt, *J* 27.0, 9.0 and 1.5, 1-H), 4.50–5.07 (19 H, m, 8 × CH_2Ph, 3 × C-H) and 6.95–7.33 (40 H, m, 8 × CH_2Ph); δ_C($CDCl_3$; 68 MHz) 69.25, 69.48, 72.07, 74.76 (4 t) and 75.54 (dd, $J_{C\text{-}C\text{-}F}$ 17.6, C-3), 76.13 (ddd, $J_{C\text{-}C\text{-}F}$ 17.6, $J_{C\text{-}O\text{-}P}$ 5.2, C-1), 76.99 and 77.19 (2 dd, $J_{C\text{-}O\text{-}P}$ 5.2, C-4 and -5), 78.23 (d, C-6), 87.12 (dd, $J_{C\text{-}F}$ 185.1, C-2), 127.11, 127.21, 127.73, 128.05, 128.22, 128.38, 128.44 and 128.54 (8 d, CH_2Ph) and 135.71, 135.97, 136.81 and 137.78 (4 s, CH_2Ph); δ_P($CDCl_3$; 36 MHz; ^{1}H-decoupled) −1.61 (1 P) and −1.95 (2 P); δ_P(DMSO with reference to $CFCl_3$; 84 MHz) −213.1 [dt, $J_{2\text{-}H,F}$ 51.3, $J_{1(3)\text{-}H,F}$ 29.3]; *m*/*z* (+ve ion FAB) 1142 [(M − H)$^+$, 4.7%], 1052 [(M − CH_2Ph)$^+$, 1.4], 181 (13) and 91 (100); *m*/*z* (−ve ion FAB) 1049 (13%) 1015 (1), 959 (2.5), 277 {[OP(O)(OCH$_2$Ph)$_2$]$^-$, 100} and 187 (14).

DL-2-Deoxy-2-fluoro-*myo*-inositol 1,4,5-trisphosphate 8

Ammonia was condensed into a three-neck flask at −78 °C. An excess of sodium was added to dry the liquid ammonia, which was then distilled into a second three-neck flask and kept at −78 °C. Sodium was added until the solution remained blue. Compound **25** (79 mg, 69 μmol) was dissolved in dry 1,4-

dioxane (2 ml) and the solution was added to the sodium–liquid ammonia mixture. After being stirred for 15 min the reaction mixture was quenched with ethanol. Ammonia and ethanol were evaporated off, the crude product was dissolved in water, and the solution adjusted to pH 7 with Dowex 50-W ion exchanger (H^+-form). Purification by ion-exchange chromatography on Pharmacia Q Sepharose Fast Flow with a gradient of TEAB buffer (0.1–1 M), pH 8.0, as eluent gave the triethylammonium salt of trisphosphate **8** (elution between 320 and 460 mM) (28.3 μmol, 41%); $\delta_H(D_2O$; 270 MHz) 3.86 (1 H, ddd, *J* 28.5, 9.9 and 2.0, 3-H), 3.90 (1 H, t, *J* 9.5, 6-H), 4.09 [1 H, q, *J* 9.2, 4(5)-H], 4.14 (1 H, ddt, *J* 27.5, 8.4 and 1.7, 1-H), 4.29 [1 H, q, *J* 9.2, 5(4)-H] and 5.10 (1 H, dt, *J* 51.8 and 1.5, 2-H); $\delta_P(D_2O$, 36 MHz; ^{1}H-coupled) 1.96 (1 P, *J* 10.1), 1.56 (1 P, *J* 6.7) and 0.37 (1 P, *J* 10.1); $\delta_F(D_2O$ with reference to $CFCl_3$; 84 MHz) −211.8 [dt, $J_{2\text{-H,F}}$ 51.3, $J_{1(3)\text{-H,F}}$ 29.7]; *m*/*z* (−ve ion FAB) 421 [$(M - H)^-$, 100%], 401 (6), 339 (14), 325 (13), 311 (19), 272 (10) and 93 (10) [Found: *m*/*z*, 420.950. Calc. for $C_6H_{13}FO_{14}P_3$: $(M - H)^-$ 420.950].

1D-1-*O*-Allyl-3,6-di-*O*-benzyl-2-*O*[(−)-camphanoyl]4,5-*O*-isopropylidene-*scyllo*-inositol 26a

A mixture of compound **13** (3.432 g, 7 mmol) and (−)-ω-camphanoyl chloride (6.067 g, 28 mmol) in dry pyridine (50 ml) was stirred for 12 h at room temperature. The solution was cooled in ice–water, water (0.5 ml) was added, and the solution was stirred for another 1 h at room temperature, after which HPTLC [diethyl ether–light petroleum (1:1)] showed two products (R_f 0.53 and 0.49). Diethyl ether (100 ml) and dichloromethane (50 ml) were added and the organic phase was washed successively with saturated aq. potassium chloride, ice-cold M-hydrochloric acid, saturated aq. potassium chloride and saturated aq. sodium hydrogen carbonate (200 ml each) and then was dried ($MgSO_4$). Evaporation of the solvents gave a syrup, which was taken up in methanol (40 ml) and kept at −20 °C overnight. The crystals formed (1.2 g) were filtered off, the mother liquor was concentrated to a volume of 20 ml, and after a further 12 h at −20 °C a second crop of crystals (1 g) was obtained [overall yield 2.2 g (2.6 mmol, 74%)] of the less polar **26a**, mp 157–160 °C (from methanol); $[\alpha]_D^{21}$ −33.7 (*c* 6, $CHCl_3$) [Found: C, 69.7; H, 7.3. Calc. for $C_{36}H_{44}O_9$ (620.74): C, 69.66; H, 7.14%]; $\delta_H(CDCl_3$; 270 MHz) 0.82 (3 H, s, Camph-CH_3), 0.94 (3 H, s, Camph-CH_3), 1.08 (3 H, s, Camph-CH_3), 1.48 and 1.49 (6 H, 2 s, 2 × CH_3), 1.64–1.98 (3 H, m, CH_2), 2.34–2.44 (1 H, m, CH_2), 3.58–3.79 (5 H, m, CH), 4.20 and 4.32 (2 H, AB, dt, J_{AB} 12.8, *J* 5.5 and 1.8, $CH_2CH{=}CH_2$), 4.55 and 4.95 (2 H, AB, J_{AB} 11.9, CH_2Ph), 4.74 and 4.92 (2 H, AB, J_{AB} 11.7, CH_2Ph), 5.13 (1 H, dq, *J* 10.4 and 1.8, *Z*-$CH_2CH{=}CH_2$), 5.22 (1 H, dq, *J* 17.2 and 1.8, *E*-$CH_2CH{=}CH_2$), 5.33 (1 H, t, *J* 9.0, 2-H), 5.85 (1 H, ddt, *J* 17.2, 10.4 and 5.2, $CH_2CH{=}CH_2$) and 7.24–7.41 (10 H, m, CH_2Ph); $\delta_C(CDCl_3$; 68 MHz) 9.57, 16.28, 16.31, 26.87 and 26.90 (5 q), 28.77 and 30.88 (2 t), 54.18 and 54.77 (2 s), 71.84, 72.88 and 73.90 (3 t), 75.46, 76.60, 78.64, 78.90 and 81.27 (5 d), 90.95 (s), 112.66 (s), 116.54 (t), 127.26, 127.29, 127.50, 127.70, 128.07 and 128.22 (6 d), 134.43 (d), 137.90 and 138.19 (2 s) and 166.47 and 178.31 (2 s); *m*/*z* (+ve ion FAB) 621 [$(M + H^+)$, 5%], 563, 473, 455, 221, 181 and 91 (100).

1L-1-*O*-Allyl-3,6-di-*O*-benzyl-2-*O*-[(−)-camphanoyl]-4,5-*O*-isopropylidene-*scyllo*-inositol 26b

The mother liquor left from the crystallisation of compound **26a** was evaporated and the residue was dissolved in hot diethyl ether. After storage of the solution at 4 °C for two days crystals had formed, which were collected to give L-isomer **26b** (1.8 g, 2.1 mmol, 60%), mp 148.5–150 °C (from diethyl ether); $[\alpha]_D^{21}$ +38.8 (*c* 4, $CHCl_3$) [Found: C, 69.5; H, 7.1. Calc. for $C_{36}H_{44}O_9$ (620.74): C, 69.66; H, 7.14%]; $\delta_H(CDCl_3$; 270 MHz) 0.86 (3 H, s, Camph-CH_3), 1.00 (3 H, s, Camph-CH_3), 1.07 (3 H, s, Camph-CH_3), 1.47 (6 H, s, 2 × CH_3), 1.58–1.91 (3 H, m, Camph-CH_2), 2.25–2.35 (1 H, m, Camph-CH_2), 3.55–3.79 (5 H, m, C–H), 4.03 and 4.37 (2 H, AB, dt, J_{AB} 12.5, *J* 5.4 and 1.7, $CH_2CH{=}CH_2$), 4.58 and 4.90 (2 H, AB, J_{AB} 11.7, CH_2Ph), 4.71 and 4.90 (2 H, AB, J_{AB} 11.7, CH_2Ph), 5.10 (1 H, dd, *J* 10.4 and 1.7, *Z*-$CH_2CH{=}CH_2$), 5.19 (1 H, ddt, *J* 17.3 and 1.7, *E*-$CH_2CH{=}CH_2$), 5.31 (1 H, t, *J* 9.1, 2-H), 5.80 (1 H, ddt, *J* 17.3, 10.4 and 5.3, $CH_2CH{=}CH_2$) and 7.23–7.38 (10 H, m, CH_2Ph); $\delta_C(CDCl_3$; 68 MHz) 9.57, 16.41, 16.57 and 26.94 (4 q), 28.88 and 30.86 (2 t), 54.07 and 54.73 (2 s), 72.14, 72.91 and 73.51 (3 t), 75.30, 77.03, 78.43, 79.00, 79.06 and 80.76 (6 d), 90.90 (s), 112.74 (s), 116.65 (t), 127.31, 127.39, 127.58, 127.81, 128.13 and 128.26 (6 d), 134.33 (d), 137.95 and 138.12 (2 s), 166.38 and 178.15 (2 s); *m*/*z* (+ve ion FAB) 621 (M^+, 5%), 563 (5), 181 (15) and 91 (100); *m*/*z* (−ve ion FAB) 773 [$(M + NBA)^-$, 8%], 745 (7), 732 (9), 579 (5), 318 (6) and 197 (100).

1D-(−)-1-*O*-Allyl-3,6-di-*O*-benzyl-4,5-*O*-isopropylidene-*scyllo*-inositol 13a

A mixture of compound **26a** (2 g, 2.4 mmol) in methanol (100 ml) and 1 M aq. sodium hydroxide (12 ml) was heated under reflux for 30 min. The solution was allowed to cool and was neutralised with solid carbon dioxide. The solvent was evaporated off and the residue was partitioned between water and chloroform. The organic phase was dried over magnesium sulfate and evaporated to give title compound **13a** as a solid (0.93 g, 2.1 mmol, 88%), mp 65–67 °C (from hexane); $[\alpha]_D^{21}$ −36 (*c* 0.6, $CHCl_3$) [Found: C, 69.4; H, 7.3. Calc. for $C_{26}H_{32}O_6$ (440.54): C; 70.89; H, 7.32%]; NMR and mass spectra as for racemate **13**.

1L-(+)-1-*O*-Allyl-3,6-di-*O*-benzyl-4,5-*O*-isopropylidene-*scyllo*-inositol 13b

Identical treatment of compound **26b** (2 g, 2.4 mmol) gave title compound **13b** (0.95 g, 2.16 mmol, 90%), mp 64–65 °C (from hexane); $[\alpha]_D^{21}$ +37 (*c* 0.6, $CHCl_3$) [Found: C, 69.9; H, 7.4%]; NMR and mass spectra as for racemate **13**.

1D-(+)-1-*O*-Allyl-3,6-di-*O*-benzyl-*scyllo*-inositol 15a

Enantiomer **15a** was obtained in a similar way to racemate **15**, mp 114–116 °C (from hexane–ethyl acetate); $[\alpha]_D^{21}$ +23.1 (*c* 0.3, $CHCl_3$) [Found: C, 69.2; H, 7.1. Calc. for $C_{23}H_{28}O_6$ (400.47): C, 68.98; H, 7.05%]; NMR and mass spectra as for 15.

1D-(−)-1-*O*-Allyl-3,6-di-*O*-benzyl-*scyllo*-inositol 2,4,5-tris-(dibenzyl phosphate) 16a

Enantiomer **16a** was obtained in a similar way to racemate **16**, mp 95–97 °C; $[\alpha]_D^{21}$ −4.6 (*c* 0.6, $CHCl_3$) [Found: C, 66.0; H, 5.75. Calc. for $C_{66}H_{67}O_{15}P_3$ (1181.16): C, 66.10; H, 5.72%]; NMR and mass spectra as for racemate **16**.

1L-(−)-*scyllo*-Inositol 1,2,4-trisphosphate 3a

Enantiomer **3a** was obtained in a similar way to racemate **3**, $[\alpha]_D^{21}$ −24.1 (*c* 0.4, $CHCl_3$); NMR and mass spectra as for racemate **3** [Found: *m*/*z*, 418.956. Calc. for $C_6H_{14}O_{15}P_3$: $(M - H)^-$, 418.955].

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