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Stereoselective Synthesis of Inositol Mono, Bis and Trisphosphate Analogues From 6-Deoxy-D-Inositol Precursors.

Mauro Vieira de Almeida, Didier Dubreuil[1]*, Jeannine Cleophax, Catherine Verre-Sebrié, Muriel Pipelier[1], Guillaume Prestat[1], Georges Vass and Stéphane D. Gero.

Institut de Chimie des Substances Naturelles, C.N.R.S., 91198 Gif-sur-Yvette Cedex, France.

[1]*Laboratoire de Synthèse Organique associé au C.N.R.S., UMR 6513, Faculté des Sciences et des Techniques, 2 rue de la Houssinière BP 92208, 44322 Nantes Cedex 03, France.*

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Abstract : The synthesis of opticaly pure deoxy-*myo*-inositol mono, bis and trisphosphate analogues is described from 4-*O*-benzyl-2,3-di-*O*-cyclohexylidene-6-deoxy-*myo*-inositol and corresponding 1,5 epimer *chiro*-inositol. These precursors, which derive from galactose, are used to accede to a variety of cyclitol intermediates employing protection/deprotection sequence. The phosphorylation procedure was performed to produce free and original substituted phosphate derivatives aimed to be incorporated through the lipidic cell membrane for *in vivo* evaluation.

It is now well established that receptor stimulated hydrolysis of inositol phospholipids is a common mechanism for transmembrane signalling when cells respond to external stimuli, such as hormones, neurotransmitters, antigens, light, growth factors, insulin, etc.[1] Phosphatidylinositol 1,4-bisphosphate (PIP_2) is a major inositol lipid hydrolysed by activated phospholipase C-β *via* G-protein , resulting into the simultaneous generation of two "second messengers", the D-*myo*-inositol-1,4,5-trisphosphate [Ins(1,4,5)P_3] and the diacylglycerol (DG). Ins(1,4,5)P_3 interacts at N-terminal binding site of a tetrameric receptor to trigger mobilization of Ca^{2+} from non-mitochondrial stores and DG stimulates protein phosphorylation *via* the activation of protein kinase C. These "second messengers" and their metabolites control and modulate vital physiological processes by their independent, additive and synergetic effects. Ins(1,4,5)P_3 was deactivated *via* two different pathways to D-*myo*-inositol-1,4-bisphosphate [Ins(1,4)P_2] and then to D-*myo*-inositol-4-monophosphate [Ins(4)P] or to D-*myo*-inositol-1,3,4,5-tetrakisphosphate [Ins(1,3,4,5)P_4] which was subsequently degraded to D-*myo*-inositol-1,3,4-trisphosphate [Ins(1,3,4)P_3]. Therefore, it is conceivable that inhibitors of the key enzymes of the phosphoinositide cascade, as phosphatases or kinases, could be of medicinal interest and also invaluable tools to elucidate the individual role of the metabolites in the regulation of cell functions.[2] Recently, we have described the synthesis of protected deoxy cyclitols from D-galactose (**Retrosynthesis**).[3] These chiral derivatives have been regarded as suitable precursors of deoxy-D-*myo*-inositol mono, bis, tris and tetrakisphosphate analogues.

*Fax (33) 2 51 12 54 12; E-mail: dubreuil@chimie.univ-nantes.fr or cleophax@icsn.cnrs-gif.fr

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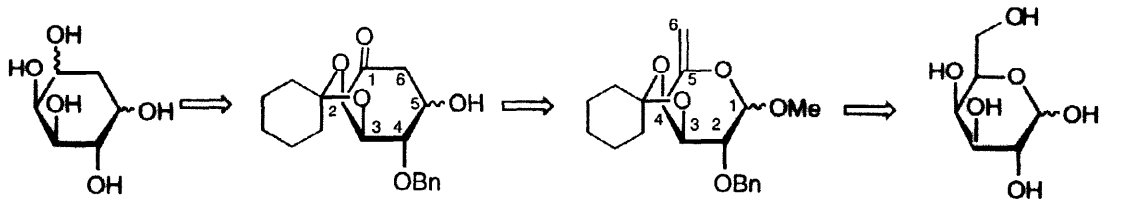


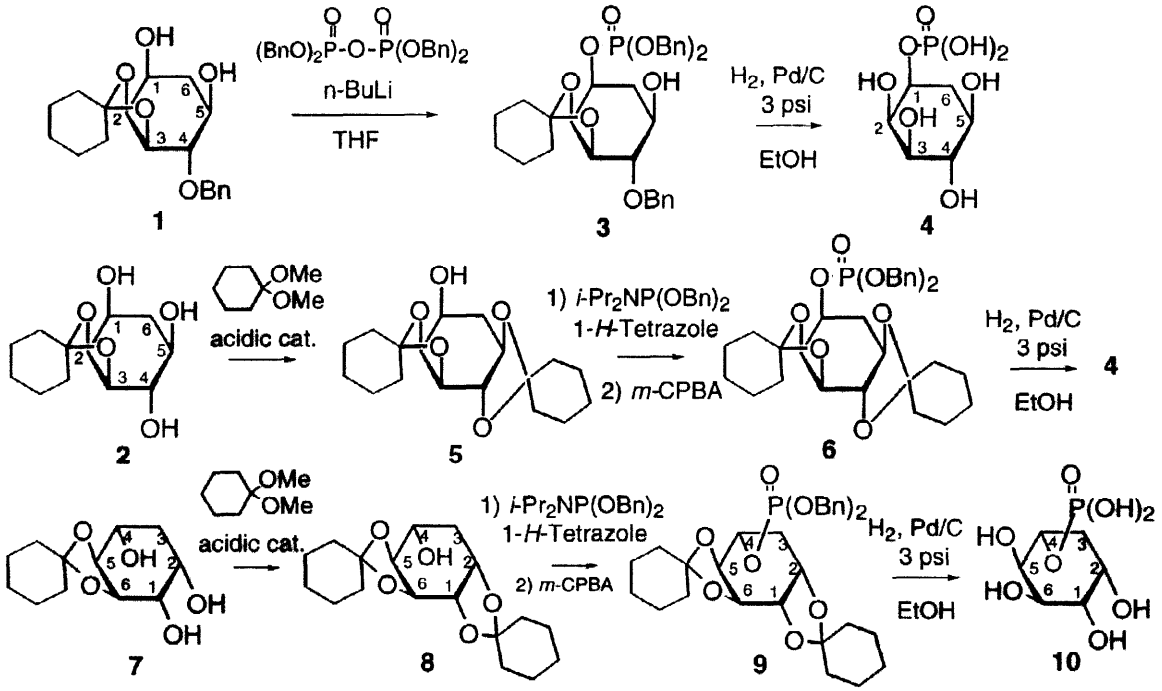
Retrosynthesis

RESULTS AND DISCUSSION

a) Synthesis of deoxy inositol monophosphates: deoxy InsP

Enzyme phosphatase catalyses the hydrolysis of monophosphate esters into free *myo*-inositol including both enantiomers of *myo*-inositol-1-phosphate [Ins(1)P], and *myo*-inositol 4-phosphate [Ins(4)P] and it is believed to be the target for lithium therapy.[4] Recently, 6-*O* -substituted analogues of Ins(1)P were identified as putative inhibitors of inositol monophosphatase. This observation is consistent with the fact that substitution of 6-OH group in inositols by hydrogen or small alkoxy groups gives potent competitive inhibitors which bind with higher affinity than parent substrates.[5] With this considerations in mind, we proposed an easy access to three deoxy cyclitol monophosphate analogues from cyclitol precursors **1**, **2**, **7** and **11**, derived from D-galactose.[3c]

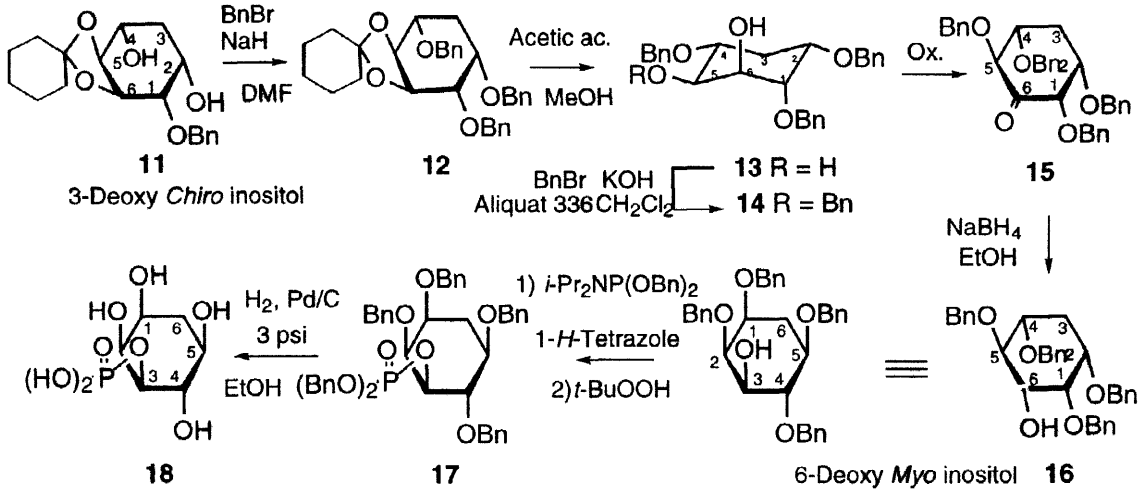
6-Deoxy D-*myo*-inositol-1-monophosphate **4** [6-deoxy Ins(1)P] was prepared from 6-deoxy-D-*myo*-1,5-diol **1** or 1,4,5-triol **2**. The synthesis of 3-deoxy-*chiro*-inositol-4-monophosphate **10** and 6-deoxy-D-*myo*-inositol-3-monophosphate **18** was achieved from *chiro*-inositols **7** and **11** respectively.



Scheme 1

The selective phosphorylation of the *myo* inositol diol **1** using the pyrophosphate method,[6] in the presence of tetrabenzylpyrophosphate and *n*-Buli, afforded the dibenzyl monophosphate **3** in 65% yield (**Scheme 1**). The total deprotection of the intermediate **3** was achieved in one-pot procedure by catalytic hydrogenolysis using palladium on charcoal (Pd/C 10%) to give the 6-deoxy Ins(1)P **4** isolated as a bis-TRIS salt. The acidity of the free phosphate group, resulting from the release of the benzyl substituents, induced subsequent *in situ* hydrolysis of the ketal protecting group. The same monophosphate **4** was also prepared from the *myo*-inositol **2**. The treatment of the triol **2** with 1,1-dimethoxy cyclohexane in the presence of acid catalyst led to the 6-deoxy-bis-cyclohexylidene inositol **5** in 99% yield. The phosphinylation of **5** using bis(benzyloxy)(diisopropylamino)phosphine and 1-*H*-tetrazole, followed by oxidation of P(III) to P(V) with *t*-BuOOH (phosphoramidite-oxidation method)[7] yielded the dibenzylphosphate intermediate **6**. Hydrogenolysis of compound **6**, in the presence of a catalytic amount of Pd/C 10%, furnished the Ins(1)P **4** in 70 % overall yield. Following a similar reaction sequence, the 3-deoxy-*chiro*-inositol-4-monophosphate **10** was accessible from the *chiro*-inositol **7**, *via* the intermediates **8** and **9**, in 75% overall yield.

The synthesis of the 6-deoxy Ins(3)P **18** was carried out from the *myo*-inositol intermediate **16** obtained in five steps from the 3-deoxy-*chiro* -inositol **11** (**Scheme 2**).



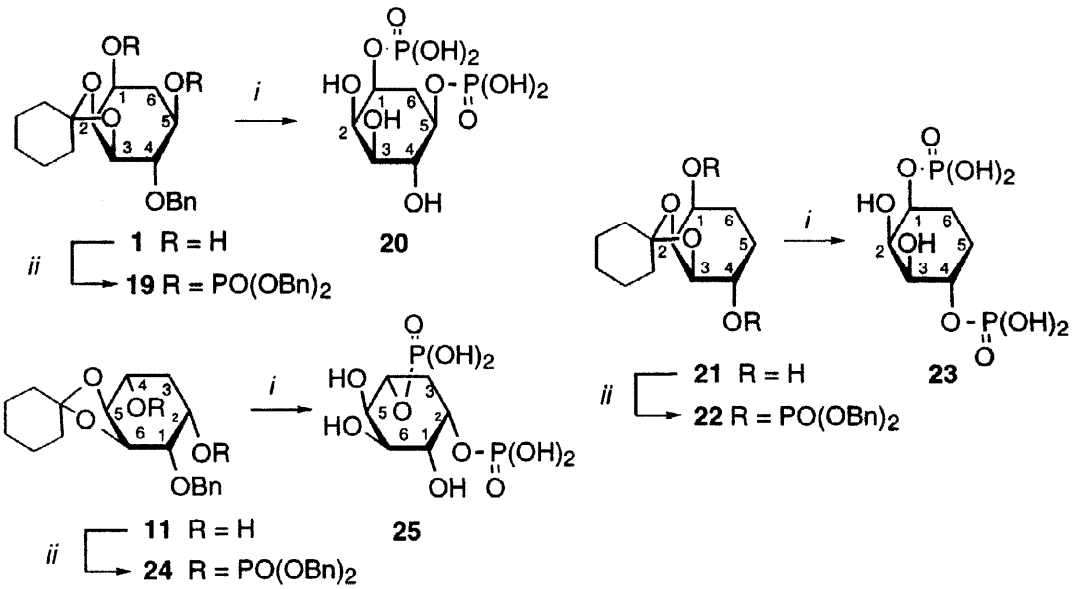
Scheme 2

The dibenzylation of compound **11**, using benzyl bromide and sodium hydride in DMF, gave the tri-*O*-benzyl derivative **12** in 93% yield. Hydrolysis of the cyclohexylidene acetal, under mild acidic conditions, afforded the *chiro*-inositol **13** which presented two free hydroxyls at 5 and 6 positions. Phase transfer benzylation of **13**[8] occured selectively, in 80% yield, at the equatorial position of the *cis* hydroxyls. The conversion of the *chiro*-inositol ring into the desired *myo*-inositol structure was accomplished in two steps by epimerisation of the axial free hydroxyl of **14**. First, the oxidation of alcool **14**, by the tetra-n-propylammonium per-ruthenate (10 %) in the presence of *N*-oxy-4-methylmorpholine,[9] furnished the 3-deoxy inosose **15** in 98% yield. The stereoselective reduction of the ketone **15** to the equatorial configuration led to the 1,2,4,5-tetra-*O*-benzyl 3-deoxy D-*myo* inositol **16** in 75% yield. Finally, the phosphorylation of **16** according to the

phosphoramidite-oxidation method, followed by hydrogenolysis of the protected phosphate **17** in the presence of Pd/C 10%, produced the 6-deoxy Ins(3)P **18** which was isolated as bis-TRIS salt.

b) Synthesis of deoxy inositol bisphosphates: deoxy $InsP_2$

During the course of our study on the synthesis of deoxy cyclitol from galactose,[3c] obtention of other carbocyclic diols gave us the opportunity to easily prepare *myo*-inositol bisphosphate analogues. Ins(1,4)P_2 have been recently reported to be allosteric activators of the enzyme 6-phosphofructo-1-kinase[10] and to activate the enzyme DNA polymerase α.[11] (**Scheme 3**).



general procedure : *i)* 1) diol, *i*-$Pr_2NP(OBn)_2$, 1-*H*-Tetrazole, MeCN, 1h.; 2) *t*-BuOOH, CH_2Cl_2
ii) H_2, 2-3psi, Pd/C 10%, EtOH

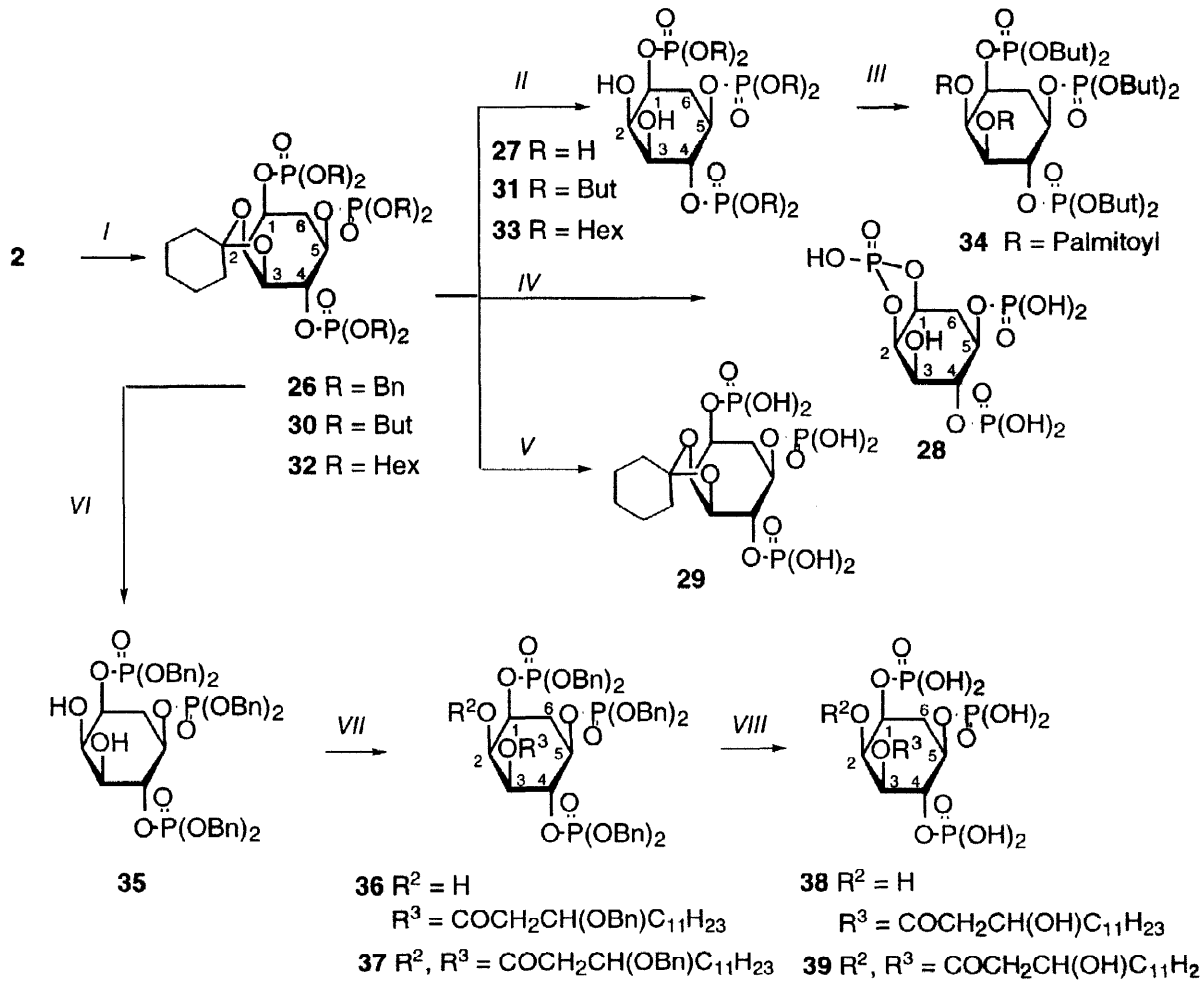
Scheme 3

Thus, using the phosphorylation-deprotection method described above, the 6-deoxy Ins(1,5)P_2 **20** and the 5,6-dideoxy Ins(1,4)P_2 **23** were prepared from the 6-deoxy and 5,6-dideoxy-*myo*-inositols **1** and **21**, respectively. The same procedure starting from the 3-deoxy *chiro* inositol **11** afforded the 3-deoxy-*chiro*-inositol 2,4-bisphosphate **25**.

c) Synthesis of deoxy inositol trisphosphates: deoxy $InsP_3$

As a continuation of our efforts in the research of inositol metabolite analogues, the *myo*-inositol-1,4,5-trisphosphate [Ins(1,4,5)P_3] appeared as one of the most important target submitted to a lot of investigations. This second messenger interacts with a family of receptor-operating calcium channels to mobilize intracellular Ca^{++} stores in many cell types.[12] Its activity is regulated by a specific dephosphorylation by 5-phosphatase into Ins(1,4,)P_2 or by a selective phosphorylation at the 3-hydroxyl by a 3-kinase leading to Ins(1,3,4,5)P_4 derivative. Thus, the inhibition of the specific enzymes seemed attractive in view of the modulation of the Ins(1,4,5)P_3 metabolism. The critical importance of the 4,5-phosphate groups of Ins (1,4,5)P_3 in receptor

binding was recognized in studies that used stereoisomers and positional isomers, while the presence of 1-phosphate further enhanced receptor affinity.[13] The significance of the hydroxyl groups has promoted new interest. The potential for 2,3 and 6 OH to form intermolecular hydrogen bonds with the receptor protein and to fix the conformation of Ins(1,4,5)P_3 in solution, *via* intramolecular hydrogen bonds to the neighbouring phosphate groups, was emphasized.[14] Several ring- and phosphate-modified analogues have been synthesized and progress has been made in understanding the role of phosphate and hydroxyl groups in determining activity of second messengers.[2,15] We already disposed of suitable protected 6-deoxy-D-*myo*- and *chiro* -1,4,5-triols[3c] which were good candidates to produce deoxy D-inositol-trisphosphate derivatives. Thus, the 6-deoxy-D-*myo*-inositol **2**, which was readily converted into the corresponding (dibenzyl)trisphosphate **26** in 75% yield, has been regarded as an interesting intermediate able to generate several $InsP_3$ analogues (**Scheme 4**).



General procedure:*i)* 1) **2**, i-$Pr_2NP(OR)_2$, 1-*H*-Tetrazole, MeCN, 1h.; 2) t-BuOOH, CH_2Cl_2; R = Bn **26**, R= But **30** and R = Hex **32**; *ii)* **for 27**: 1) **26**,H_2, Pd/C 10%, 5 psi, EtOH, 2) TRIS salt ; **for 31** and **33**: 1) **30** and **32**, $BF_3.Et_2O$, MeCN, 12h., r. t.; 2) Dioxane, HCl 37%, 3h., r.t.; *iii)* Palmitic acid, DCC, DMAP, CH_2Cl_2, 6h., r.t.; *iv)* **26**, H_2, Pd/C 10%, 5 psi, EtOH; *v)* **26**, H_2, Pd/C 20%, 2 psi, EtOH; *vi)* **26**, HCl 2M, MeOH, 3h., r.t.; *vii)* **for 36**: **35**, β-(BnO)myristic acid (1.1eq.), DCC (1.1eq.), DMAP cat., CH_2Cl_2, 4h. r. t.; **for 37**: **35**, β-(BnO)myristic acid (2.4eq.), DCC (2.4eq.), DMAP cat., CH_2Cl_2, 4h. r. t.; *viii)* H_2, Pd/C 10%, 5 psi, EtOH, 2) TRIS salt.

Scheme 4

The deprotection of compound **26** was carried out under various hydrogenolysis conditions which allowed to produce original Ins(1,4,5)P_3 analogues. Under medium pressure of hydrogen (5 psi), in the presence of Pd/C 10%, the protected phosphate **26**, dissolved in ethanol (95 %) solution containing a small amount of water, afforded in one pot the 6-deoxy Ins(1,4,5)P_3 **27** isolated as its hexa-TRIS salt. Using a slightly modified experimental procedure, the 6-deoxy-D-Ins-1,2-cyclic-4,5-trisphosphate **28** could be isolated in a quantitative yield, as a white solid, when evaporation of the ethanolic solution to dryness was effected prior to addition of TRIS. The reduction of charge in that product could be correlated with the required ionization state of the 4 and 5-phosphates.[16,17] Alternatively, the hydrogenolysis at lower pressure (2 psi) of **26**, dissolved in ***absolute*** ethanol, left the ketal substituent untouched to give the 2,3-*O*-cyclohexylidene-6-deoxy-Ins(1,4,5)P_3 **29** isolated as hexa-TRIS salt. The relative instability of the ketal group, due to the intrinsic acidity of the free phosphate, resulted in the rapid partial degradation of **29**.

The particular interest of the 6-deoxy-Ins(1,4,5)P_3 in the inhibition of the target enzymes, emphasized by preliminary *in vitro* studies on permeabilized cells,[18] prompted us to attempt the transformation of 6-deoxy-Ins(1,4,5)P_3 derivatives into more lipophilic compounds. The use of such highly hydrophilic polyphosphate derivatives, which expressed important charges, for *in vivo* experiments depended on their ability to be incorporated into the lipidic cell membrane. Thus, the full or partial protection of alcohol and phosphate groups by temporary protecting groups should be helpful to resolve this problem as previously performed in case of AMPc.[19] This hypothesis was supported by the presence in cell membrane of a number of lipases, esterases and phosphatases, able to release such protected analogues on their ionized form in the internal cellular medium.

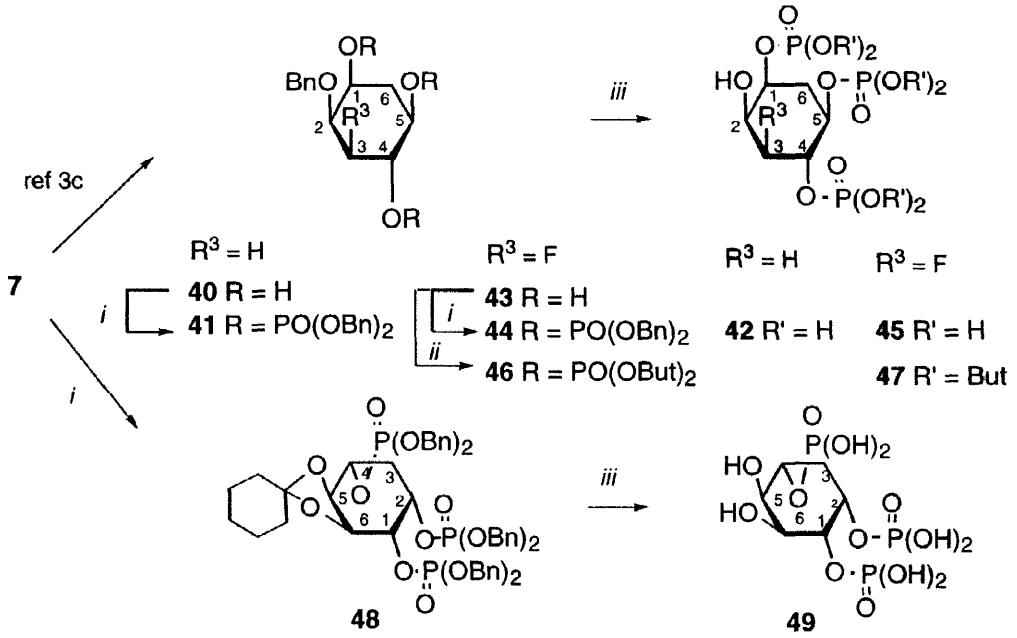
We first envisaged to neutralize the phosphate groups of the 6-deoxy-Ins(1,4,5)P_3 **27** by dibutyl or dihexyl substituents.[20] This was readily realized in 60% yields starting from the triol **2**, using the phosphoramidite method involving the dibutyloxy(diisopropylamino)phosphine or the corresponding dihexyloxy analogue as phosphitylation reagents. The selective hydrolysis of the ketal group from the resulting protected trisphosphates **30** and **32**, under usual conditions (MeOH, HCl), afforded the expected 6-deoxy Ins(1,4,5)-tris(dibutyl)- and tris(dihexyl)-phosphate **31** and **33** in 55% and 80% yields, respectively. The deprotection yield was improved to 80% when a small amount of boron trifluoride etherate in acetonitrile was added prior to the acidic treatment of **30** and **32** with HCl in dioxane.

On the other hand, we considered a more lipophylic analogue of the tris(dibutyl)phosphate analogue of **31** by substitution of the two free hydroxyls by a fatty acid. In the presence of palmitic acid, dicyclohexyl carbodiimide (DCC) and a catalytic amount of dimethylaminopyridine (DMAP), the 6-deoxy-2,3-*O*-palmitoyl-Ins(1,4,5)-tris(dibutyl)phosphate **34** was isolated in 78% yield from **31**. We also anticipated the synthesis of the two lipophilic trisphosphate analogues **38** and **39** following a similar approach starting from the dibenzylphosphates intermediate **26**. These products should help for the determination of the minimum modifications allowed for the incorporation of these analogues through the membrane barrier. Thus, the use of 1,1 eq. of (hydroxybenzyl)myristic acid, DCC and DMAP in CH_2Cl_2, achieved the selective substitution of the equatorial hydroxyl of **26** leading to the 3-*O*-(benzyloxy)myristoyl **36** in 60% yield. The corresponding 2,3-diester **37** was obtained in 84% yield from **26** in the presence of an excess of the latter reagents. Final deprotection of the intermediates **36** and **37**, under the hydrogenolysis conditions applied above, led to the

3-*O*-(hydroxy)myristoyl and 2,3-di-*O*-(hydroxy)myristoyl-6-deoxy-Ins(1,4,5)P_3 **38** and **39** respectively, isolated as hexa-TRIS salt.

Some recent publications described the potent inhibition of the 3-kinase by 2- or 3-deoxy fluorinated Ins(1,4,5)P_3 derivatives; these observations justified the synthesis of 3,6-dideoxy analogues.[21] The 3,6-dideoxy and the 3,6-dideoxy-3-fluoro Ins(1,4,5)P_3 were prepared *via* the 2-*O*-benzyl-3,6-dideoxy-D-*myo*-inositol **40** and the 3,6-dideoxy-3-fluoro-D-*myo*-inositol **43** derived from the 3-deoxy-*chiro* -1,2,4-triol **7**[3c] (**Scheme 5**).

The dideoxy triols **40** and **43** were submitted to the phosphorylation-deprotection procedure previously discussed to give the corresponding 1,4,5-trisphosphate **42** and **45** respectively in 50% overall yields. We also achieved the preparation of the lipophilic 2-*O*-benzyl-3,6-dideoxy-3-fluoro-Ins(1,4,5)-tris(dibutyl)phosphate **46** in 65% yield from the 3-fluoro cyclitol **43** using the (dibutyloxy)phosphitylation reagent. Deprotection of the benzylated intermediate **46** under catalytic hydrogenolysis conditions had to be run in EtOAc to be quantitative.



i) 1) **40**, **43** or **7**, i-Pr_2NP(OBn)$_2$, 1-*H*-Tetrazole, MeCN, 1h.; 2) t-BuOOH, CH_2Cl_2; *ii)* i-Pr_2NP(OBut)$_2$, 1-*H*-Tetrazole, MeCN, 1h.; 2) t-BuOOH, CH_2Cl_2; *iii)* **for 42, 45** or **49**: **41**, **44** or **48**, H_2, Pd/C 10%, 5 psi, EtOH, 2) TRIS salt ; **for 47: 46**, H_2, Pd/C 10%, 5 psi, AcOEt, 2) TRIS salt.

Scheme 5

Finally, following the phosphorylation-deprotection procedure, the access to an isomer 3-deoxy-*chiro*-inositol-1,2,4-trisphosphate **49** was also performed in 70% yield from the 3-deoxy-*chiro*-1,2,4-triol **7**. Since it was unclear whether the substitution or the lack of hydroxyl or phosphate group at the inositol moiety was responsible for the properties of inositol phosphates derivatives, these latter trisphosphate analogues represented interesting supports for evaluation.

CONCLUSION AND BIOLOGICAL RESULTS.

From 6-deoxy-inositols derived from galactose,[3c] a range of deoxy analogues of inositol phosphate metabolites were shown easily accessible and are regarded as potential regulators of the phosphoinositide cascade. These cyclitol precursors presented an interesting flexibility in regards to their selective substitutions using simple protection-deprotection procedures. The 6-deoxy analogues of the well known second messenger Ins(1,4,5)P_3 possessing a promising biological effect on permeabilized cells[18] have been transformed into lypophilic forms in order to assume their incorporation through the membrane of intact cells.

Biological evaluation of the activity of deoxy Ins phosphate analogues has been realized by Bayer AG company. In order to illustrate the interest of such deoxy analogues, we resume here the biological activity of the trisphosphate derivatives. Further experimental complements and details will be fully discussed in another context.

In order to assess the specificity of the deoxy analogues for Ins(1,4,5)P_3 mediated actions, the investigations started with Ins(1,4,5)P_3 receptor assay. Purified endoplasmatic reticulum fractions from dog cerebellum and ^{3}H-Ins(1,4,5)P_3 as radioligand, which binds highly selectively to the membranes and are displaced by Ins(1,4,5)P_3 with an IC_{50} of 2 10^{-8} mol/l, were used. 6-Deoxy Ins(1,4,5)P_3 **27** displaced the radioligand with an IC_{50} of 2 µmol/l. But it became obvious that substitutions in position 2 and 3 had no substantial influence on the receptor binding. Good examples for that are compounds 3,6-dideoxy Ins(1,4,5)P_3 **42** and 3,6-dideoxy-3-fluoro Ins(1,4,5)P_3 **45** with IC_{50} value of 9 and 3 µmol/l, respectively. Even esterification of the 2- and 3- positions with long chain fatty acid residues did not reduce dramatically the displacement of the radioligand as shown with analogues **38** and **39** (IC_{50} : 4 and 3 µmol/l). Changes in the stereochemistry of the phosphate groups or esterification of the phosphate groups with butanol or another alcohol resulted in complete inactive compounds in receptor assay as shown with compounds **30**, **31**, **32**, **33**, **34**, **47** and **57**. None of the compounds tested showed inhibitory effects in *in vitro* enzyme assays like protein kinase C, phospholipase A2, 5'-phosphatase of erythrocytes and protein-phosphatase. Unfortunately assays to test these analogues against isolated phospholipase C which is the key enzyme of Ins(1,4,5)P_3 release, could not be performed *in vitro*.

In addition to the above mentioned enzymatic and receptor assays, efforts were made with functional assays at the level of isolated cells and cell cultures.

It is well known that endogenous mediators formyl-methionyl-leucyl-phenylalanine (fMLP) activate white blood cells, especially poly-morpho-nucea-leucosites (PMNL) by an Ins(1,4,5)P_3 mediated intracellular Ca^{2+} release from the endoplasmic reticulum. This activation leads to physiological reactions like O_2^- generation or degranulation of hydrolytic like β-glucuronidase. In the experimental model, fMLP produces a dose dependent stimulation of O_2^- release at concentrations ranging between 1nmol/l and 10 µmol/l. 6-Deoxy Ins(1,4,5)P_3 **27** did not influence this O_2^- generation in spite of its good binding to the isolated receptor. The same was true for the myristoyl derivatives **38** and **39**. However, administration of the 1,4,5-tris(dibutyl)phosphates analogues **30** and **31**, 10 min. prior to the fMLP stimulation resulted in a dose dependent on inhibition of O_2^- generation. The IC_{50} amount to 10 µmol/l for **30** and 3 µmol/l for **31**. In contrast to the stimulation with fMLP none of the compounds is able to block the O_2^- formation initiated by the Ca-ionophore calmycin which raises the intracellular Ca^{2+}-level independently from Ins(1,4,5)P_3. Compound **34** was not active under these conditions.

In addition to the O_2^- generation fMLP enhances the release of β-glucuronidase from these cells 10-20 fold above control. Both compounds **30** and **31** inhibit the fMLP induced release of β-glucuronidase from cytochalasin B treated PMNL without affecting the stimulation by calimycin.

Stimulation of PMNL with fMLP at the same concentrations that used in the superoxide anion and β-glucuronidase experiments caused a rapid increase in the fura-2 dependent fluorescence indicating an increase in intracellular Ca^{2+} within 10 sec. Analogues **30** and **31** show a tendency to slightly increase the basal intracellular Ca^{2+}-level. At the applied highest concentration (10 μmol/l) both tris(dibutyl)phosphate compounds significantly inhibit the rapid enhancement in intracellular Ca^{2+}-level without affecting the velocity of the signal's decline. No effect can be observed when using ionomycin (the fluorescent analogue of calimycin) as agonist. In none of the above mentioned test-systems any significant effect of compound **30** and **31** became obvious, when they were added simultaneously with fMLP. These results can be explained by assuming that a distinct time interval is necessary for the hydrolysis of butylesters.

These results lead to the conclusion that 6-deoxy-Ins(1,4,5)tris(dibutyl)phosphates **30** and **31** cross the cell membrane, are hydrolysed and interfere with the receptor. The specificity of these effects is strengthened by the fact that neither compound interferes with an activation independent of Ins(1,4,5)P_3. Although 3,6-dideoxy-3-fluoro-1,4,5-trisphosphate analogue **45** is comparable to compound **27** in receptor binding assay, the corresponding 3,6-dideoxy 3-fluoro-1,4,5-tris(dibutyl)phosphate **47** was inactive. The same is true for analogues **32** and **33**. Up to now we have not found an explanation for this behaviour.

Additionally, it was found that the intracellular Ca^{2+}-level as well as receptor mediated platelet aggregation is significantly inhibited by analogues **30** and **31**. No effect on the Ins(1,4,5)P_3 independent phorbol-myristyl-acetate (PMA) induced stimulation could be observed.

In the system of smooth muscle cell-proliferation stimulated by fetal calf serum, the 6-deoxy-Ins(1,4,5)tris(dibutyl)phosphates **30** dose-dependent reduced the incorporation of ^{14}C-thymidine. According to the current literature, PLC activation does not play a role in the regulation of this process. Therefore, it is not clear if these effects are due to the claimed specific interaction with the Ins(1,4,5)P_3 pathway.

In none of the above mentioned assays did the tris(dibutyl)phosphate derivative of natural Ins(1,4,5)P_3, synthesized for comparison, elicit agonistic properties. However, in some systems, inhibitory effects occur at the highest concentration (10 μmol/l.). This could be due to its metabolic instability.

These results led to characterize and specify the observed effects in *in vivo* experiments. It was started with an inflammation model. Arachidonic acid applied topically to mice ears induced an inflammation process which was followed by the development of an oedema.

Intravenous administration of 10 mg/kg of 6-deoxy-Ins(1,4,5)tris(dibutyl)phosphates **31** reduced the oedema between 66% after 30 min and 52% after 60 min of the application. These are highly significant biological effects.

Further experiments concerning the inhibition of platelet aggregation *in vivo* and in a shock model are under investigation with promising results.

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EXPERIMENTAL PART

^1H NMR and ^{13}C NMR spectra were recorded on a Bruker spectrometra WP 200, AC 200, AC 250, WM 400 or ARX 400 ; chemical shifts are expressed in parts per million (ppm) referenced to residual chloroform (7.27 ppm). Coupling constants (J) are given in hertz (Hz). Multiplicities are recorded as s (singlet), d (doublet), t (triplet), q (quartet), and m (multiplet or complex). The $[\alpha]_D$ were recorded on Perkin-Elmer 241-MC sodium absorption at 20°C. Mass spectra (m/z (% base peak) were recorded on Atlas CH_4 or AEI MS9 spectrometra. Melting points were determined on a C. REICHERT microscope apparatus and are uncorrected. Elemental analyses were carried out at the "Laboratoire de Microanalyse de l'I.C.S.N." (CNRS, gif/yvette). All solvents were freshly distilled prior to use by standard methods[22]. Flash chromatography was performed on silica-gel Merck 60 230-400 mesh. Thin layer chromatography was performed on precoated plates of silica gel PF_{254} neutralized with sodium bicarbonate. All crystallized compounds were obtained from AcOEt/pentane if not specified.

General procedure for phosphorylation and deprotection steps.

Method A : phosphinylation-oxidation method.

a) Preparation of phosphinylation reagent dialkyloxy(diisopropylamino)phosphine

PCl_3 (4.4 ml) was added under argon to freshly distilled diethyl ether (30 ml). The solution was cooled to -10°C before diisopropylamine (14 mL) was added dropwise. The mixture was stirred over 1 h. at -10°C and then was allowed to warm to 20°C before it was filtered. The filtrate was evaporated to dryness and distillated to give 6.3 g of dichlroro(diisopropylamino)phosphine. This solid intermediate was dissolved in acetonitrile (60 mL) and the solution was maintained under argon. The solution was cooled to -10°C before ethyl diisopropylamine (13.7 ml) was added. To this solution, maintained under argon at -10°C, benzyl alcohol (6.6 mL), butyl alcohol (5.86 ml) or hexyl alcohol (8 ml) dissolved in acetonitrile (40 mL) were added dropwise. The solution was stirred for 1.5 h before it was allowed to warm to 20°C. After 12 h of stirring, the solution was concentrated to dryness and diluted with CH_2Cl_2 (150 mL). Organic layer was washed with saturated Na_2CO_3 and then by water before it was dried ($MgSO_4$) and evaporated under reduced pressure. The yellow oil obtained (8.8 g) is used with no further purification.

b) Phosphinylation reactions.

A mixture of cyclitol and dialkyloxy(diisopropylamino)phosphine (2 eq. per hydroxyl group) was dried for 0.5 h under vacuum (0.05 mm/hg). Sublimated tetrazole (2 eq per phosphinylation reagent), dissolved in dry actetonitrile was added under argon to the mixture. The solution was stirred under argon at r.t. for 1 h.

c) Oxydation P^{III} - P^V

The solution was diluted with CH_2Cl_2 before *t*-BuOOH (2 eq per phosphinylation reagent) were added. The solution was stirred under argon at r.t. for 1 hr.

d) Purification of phosphorylated products.

Aqueous thiosulfate and sodium bicarbonate solution was added for neutralization. Organic layer was extracted with CH_2Cl_2 and concentrated to dryness. The residue was separated by preparative chromatograhy on silica gel or reverse phase.

Method B : Pyrophosphate method

a) Preparation of phosphorylation reagent: tetrabenzylpyrophosphate

DCC (227 mg, 1.1 mmol) was dissolved in dry ethyl ether (1 ml). Dibenzylphosphate (556 mg, 2 mmol) in dry acetonitrile (2ml) and dry ethyl ether (2 ml) were added to the solution. Acetonitrile was added to complete dissolution of the mixture. The solution was stirred for 15 min, filtered through celite and eluted with hexane. The filtrate was concentrated to dryness and crystallized from hexane to give tetrabenzylpyrophosphate (250 mg), m.p. 61-62°C.

b) phosphorylation reactions.

Alcohol was dissolved in the minimun amount of anhydrous THF. The solution was cooled to 0°C before *n*-BuLi (1.1 eq per free hydroxyl group) was added. The solution was stirred for 5 min, then tetrabenzylpyrophosphate (1.3 eq per free hydroxyl group) was added at -40°C. The solution was stirred for 1 h under argon. The mixture was filtered through silica gel and eluted with AcOEt. The filtrate was concentrated to dryness and products were separated by preparative chromatograhy on silica gel, florisil or RP8.

D-4-O-Benzyl-2,3-*O*-cyclohexylidene-6-deoxy-*myo*-inositol-1-(dibenzyl)phosphate 3

The diol **1**[3c] was converted to monophosphate **3** using method B with tetrabenzylpyrophosphate (1.2 eq). Compound **3** was cristallized (65%); m.p. 91°C; $[\alpha]_D$ -0.8° (c 0.95, CH_2Cl_2); ^1H NMR (200MHz, $CDCl_3$): δ: 5.20 (m, 1H, H-1); 4.53 (t, 1H, $J_{1-2}=J_{2-3}=4$, H-2); 4.20 (dd, 1H, $J_{3-2}=4$, $J_{3-4}=8$ Hz, H-3); 3.53 (dd, 1H, $J_{4-3}=8$, $J_{4-5}=9$, H-4); 3.53 (m, 1H, $J_{5-4}=9$, $J_{5-6ax}=8$, $J_{5-6eq}=4$, H-5); 2.13 (m, 1H, $J_{6eq-6ax}=12$, $J_{6eq-5}=4$, $J_{6eq-1}=3$ Hz, H-6eq); 1.73 (m, 1H, $J_{6ax-6eq}=12$, $J_{6ax-5}=8$, $J_{6ax-1}=8$, H-6ax); (Found : C, 66.40; H, 6.54; P, 5.00; $C_{33}H_{39}O_8P$ requires C, 66.65; H, 6.61; P, 5.21).

D-6-Deoxy-*myo*-inositol-1-monophosphate 4

From the (dibenzyl)phosphate **3**: **3** dissolved in the minimun amount of EtOH 95% was hydrogenated for 1 h, under 3 psi, in the presence of the same amount of Pd/C 10%. The catalyst was removed by filtration and washed with water. Tris(hydroxymethyl)aminomethane (TRIS, 2 eq per phosphate) was added before concentration of the filtrate. The aqueous solution was lyophilized and the Ins(1)P **4** was precipitated as bis TRIS-salt; $[\alpha]_D$ -10° (c 1.3, H_2O); ^1H NMR (400MHz, D_2O): δ: 4.07 (m, 1H, H-2); 3.93 (m, 1H, H-1); 3.41 (m, 2H, H-3, H-5); 3.37 (q, 1H, H-4); 2.0 (m, 1H, H-6eq); 1.70 (q, 1H, H-6ax); ^{13}C NMR (63MHz, D_2O); 77.19 (C-3); 75.02 (C-5); 74.43 (C-4); 72.20 (C-2, C-1); 36.26 (C-6) (Found : C, 33.65; H, 7.46; N, 5.38; $C_{14}H_{35}O_{14}N_2P$, H_2O requires C, 33.33; H, 7.39; N, 5.55).

D-2,3;4,5-Di-*O*-cyclohexylidene-6-deoxy-*myo*-inositol 5

Dimethoxycyclohexane (3.5 ml, 5.6 mmol) and camphorsulfonic acid (50 mg) were added to the triol **2**[3c] (459 mg, 1.88 mmol) dissolved in dry *N,N*-dimethylformamide (5 ml). Methanol formed during the course of the reaction was evaporated. The solution was stirred under reduced pressure for 12 h then sodium bicarbonate was added for neutralization. The solution was filtered on silica gel and the solids washed with AcOEt. The filtrate was coevaporated with toluene. The residue was crystallized (n-pentane) to give dicyclohexylidene inositol **5** (99%). m.p. 120-121°C; $[\alpha]_D$ -1° (c 1.2, $CHCl_3$); ^1H NMR (200MHz, $CDCl_3$): δ: 4.26 (m, 2H, $J_{2-1}=4$, $J_{2-3}=5$, $J_{3-4}=7$ H-2, H-3); 4.10 (m, 1H, $J_{1-2}=4$, $J_{1-6ax}=8$, $J_{1-6eq}=6$, H-1); 4.00 (1H, dd, $J_{4-3}=7$, $J_{4-5}=10$, $J_{5-6eq}=4$, H-4); 3.43 (m, 1H, $J_{5-4}=10$, $J_{5-6ax}=10$, $J_{5-6eq}=6$, H-5); 2.33 (m, 1H, $J_{6ax-6eq}=12$, $J_{6eq-1}=J_{6eq-5}=6$, H-6eq); 1.86 (m, 1H, $J_{6ax-6eq}=12$, $J_{6ax-1}=8$, $J_{6ax-5}=10$, H-6ax); (Found : C, 66.86; H, 8.67; $C_{18}H_{28}O_5$ requires C, 66.64; H, 8.77).

D-2,3;4,5-Di-*O*-cyclohexylidene-6-deoxy-*myo*-inositol-1-(dibenzyl)phosphate 6

Myo inositol **5** was phosphorylated using method A, in the presence of dibenzyloxy (diisopropylamino)phosphine reagent, to give the monophosphate **6** (70%); m.p. 76°C ; $[\alpha]_D$ +5° (c 1, CH_2Cl_2); ^1H NMR (200MHz, $CDCl_3$): δ: 4.66 (1H, m, $J_{1-2}=4$, $J_{1-6ax}=J_{1-6eq}=6$, H-1); 4.36 (dd, 1H, $J_{2-1}=4$, $J_{2-3}=6$, H-2); 4.2 (dd, 1H, $J_{3-2}=6$, $J_{3-4}=8$, H-3); 3.76 (dd, 1H, $J_{4-3}=8$, $J_{4-5}=12$, H-4); 3.30 (m, 1H, $J_{5-4}=12$, $J_{5-6ax}=5$, $J_{5-6eq}=12$, H-5); 2.33 (m, 1H, $J_{6ax-6eq}=J_{6eq-5}=12$, $J_{6eq-1}=10$, H-6eq); 2.05 (m, 1H, $J_{6ax-6eq}=12$, $J_{6ax-1}=J_{6ax-5}=5$, H-6ax); (Found : C, 65.44; H, 6,99; P, 5.46; $C_{32}H_{41}O_8P$ requires C, 65.73; H, 7.01; P, 5.29).

L-1,2;5,6-Di-*O*-cyclohexylidene-3-deoxy-*chiro inositol* 8

Dimethoxycyclohexane (1.7 ml, 2.68 mmol) and camphorsulfonic acid (23 mg) were added to triol **7**[3c] (327 mg, 1.34 mmol) in dry *N,N*-dimethylformamide (5 ml). Methanol formed during the course of the reaction was evaporated. The solution was stirred under reduced pressure for 12 h. Sodium bicarbonate was added for neutralization. The solution was filtered and the solids washed with AcOEt. The filtrate was coevaporated with toluene. The residue was purified by chromatograhy on silica gel to give dicyclohexylidene **8** (98%); $[\alpha]_D$ 0° (c 1, CH_2Cl_2); ^1H NMR (200MHz, $CDCl_3$): δ: 4.43 (m, 3H, $J_{6-5}=4$, $J_{6-1}=6$, $J_{1-2}=3$, $J_{1-3ax}=4$, $J_{1-3eq}=6$, H-1, H-2, H-6); 4.23 (m, 1H, $J_{5-4}=7$, $J_{5-6}=4$, H-5); 3.70 (m, 1H, $J_{4-5}=7$, $J_{4-3ax}=6$, $J_{4-3eq}=4$, H-4); 3.00 (m, 1H, $J_{3ax-3eq}=8$, $J_{3eq-4}=4$, $J_{3eq-2}=6$, H-3eq); 1.90 (m, 1H, $J_{3ax-3eq}=8$, $J_{3ax-4}=6$, $J_{3ax-2}=4$, H-3ax); (Found : C, 64.84; H, 26.01; $C_{18}H_{28}O_5$ + 1/2 H_2O requires C, 64.84; H, 26.39).

L-1,2;5,6-Di-*O*-cyclohexylidene-3-deoxy-*chiro*-inositol-4-(dibenzyl)phosphate 9

Dicyclohexylidene **8** was converted into the mono(dibenzyl)phosphate **9** (65%), using phosphorylation method A in the presence of dibenzyloxy(diisopropylamino)phosphine reagent; $[\alpha]_D$ -10° (c 1.02, $CHCl_3$);

^{1}H NMR (200MHz, $CDCl_3$): δ: 4.50 (m, 1H, $J_{6\text{-}5}$=3, $J_{6\text{-}1}$=7, H-6); 4.40 (dd, 1H, $J_{4\text{-}3ax}$=4, $J_{4\text{-}3eq}$=8, H-4); 4.38 (m, 1H, $J_{2\text{-}1}$=6 $J_{2\text{-}3ax}$=3, $J_{2\text{-}3eq}$=6, H-2); 4.35 (d, 1H, $J_{5\text{-}6}$=3, H-5); 4.06 (dd, 1H, $J_{1\text{-}6}$=7, $J_{1\text{-}2}$=6, H-1); 2.26 (m, 1H, $J_{3ax\text{-}3eq}$=12, $J_{3eq\text{-}4}$=8, $J_{3ax\text{-}2}$=7, H-3eq); 1.90 (m, 1H, $J_{3ax\text{-}3eq}$=8, $J_{3ax\text{-}4}$=4, $J_{3ax\text{-}2}$=4, H-3ax); (Found : C, 65.62; H, 6.97; P, 5.16; $C_{32}H_{41}O_8P$ requires C, 65.73; H, 7.01; P, 5.29).

L-3-Deoxy-*chiro* -inositol-4-monophosphate 10

The (dibenzyl)phosphate **9** dissolved in the minimun amount of EtOH 95% was hydrogenated for 1 h, under 3 psi, in the presence of the same amount of Pd/C 10%. The catalyst was removed by filtration. Tris(hydroxymethyl)aminomethane (TRIS, 2 eq per phosphate) was added before concentration of water under vacum. The aqueous solution was lyophilized and the *chiro* inositol(4)P **10** was precipitated as bis TRIS-salt; $[\alpha]_D$ -10° (c 1.3, H_2O); ^{1}H NMR (400MHz, D_2O) δ: 4.45-4.23 (m, 4H, H-6, H-5, H-4, H-2), 3.99 (m, 1H, H-1); 2.23 (m, 1H, H-3eq); 1.92 (m, 1H, H-3ax); (Found : C, 33.15; H, 7.70; N, 5.62; $C_{14}H_{35}O_{14}N_2P+H_2O$ requires C, 33.33; H, 7.39; N, 5.55).

L-1,2,4-Tri-*O*-benzyl-5,6-*O*-cyclohexylidene-3-deoxy-*chiro*-inositol 12

NaH (88mg, 3.66 mmol) was added, under argon, to diol **11**[3c] (410 mg, 1.22 mmol) dissolved in *N,N*-dimethylformamide (5 ml). The mixture was stirred for 10 min before benzylbromide (0.44 ml, 3.66 mmol) was added. The solution was stirred for 5 h then methanol was added. The reaction mixture was extracted and the organic layers concentrated to dryness. Flash chromatography on silica gel gave the *chiro* inositol **12** (93%); $[\alpha]_D$ -39° (c 0.27, CH_2Cl_2); ^{1}H NMR (200MHz; $CDCl_3$): δ: 4.37 (dd, 1H, H-6ax); 4.29 (m, 1H, H-5); 3.95 (dd, 1H, H-1); 3.77 (m, 1H, H-2); 3.51 (m, 1H, H-4); 2.00 (m, 2H, H-3ax, H-3eq); ^{13}C NMR (50MHz; $CDCl_3$): δ: 78.9, 77.2, 76.3, 76.0; 74.6 (C-1, C-2, C-4, C-5, C-6); 29.9 (C-3); 37.5, 35.1, 24.9, 23.9, 23.6 (C_6H_{10}); (Found : C, 76.74; H, 7.66; $C_{33}H_{38}O_5$ requires C, 77.01; H, 7.44).

L-1,2,4-Tri-*O*-benzyl-3-deoxy-*chiro*-inositol 13

Acetic acid (12 ml) and water (6 ml) were added to the *chiro* inositol **12** (581 mg, 1.13 mmol). The solution was stirred at 60°C for 2 h. then cooled to r.t. and coevaporated with toluene under reduced pressure. The residue was purified by flash chromatography on silica gel. The diol **13** was isolated by cristallization (95%); m.p. 79-80°C; ^{1}H NMR (200MHz; $CDCl_3$): δ: 4.37 (dd, 1H, H-6); 4.29 (m, 1H, H-5); 3.95 (dd, 1H, H-1); 3.77 (m ,1H, H-2); 3.51 (m, 1H, H-4); 2 (m, 2H, H-3ax, H-3eq); ^{13}C NMR (50MHz; $CDCl_3$): δ: 76.5 (C-5, C-6); 74.7, 73.2, 70.3 (C-1, C-2, C-4); 29.3 (C-3); (Found : C, 74.18; H, 6.82; $C_{27}H_{30}O_5$ requires C, 74.63; H, 6.96).

L-1,2,4,5-Tretra-*O*-benzyl-3-deoxy-*chiro*-inositol 14

Sodium carbonate (107 mg, 0.78 mmol) was added to the diol **13** (340 mg, 0.78 mmol) dissolved in CH_2Cl_2 (8 ml). KOH (123 mg, 2.2 mmol), Aliquot 336 (40 mg) and benzylbromide (0.09 ml, 0.78 mmol) were added to the solution which was stirred for 5h. The reaction mixture was extracted and the organic layers concentrated to dryness. Flash chromatography on silica gel gave the tetrabenzylether **14** (80%); $[\alpha]_D$ -11° (c 1, CH_2Cl_2); ^{1}H NMR (400MHz; $CDCl_3$): δ: 3.96 (dd, 1H, $J_{4\text{-}5}$=9, $J_{5\text{-}6}$=3, H-5); 3.88 (dd, 1H, $J_{1\text{-}6}$= $J_{5\text{-}6}$=3, H-6); 3.81 (bs, 1H, H-1); 3.75 (ddd, 1H, $J_{2\text{-}3ax}$= 12, $J_{2\text{-}3eq}$=3, $J_{1\text{-}2}$=2, H-2); 3.6 (ddd, 1H, $J_{3ax\text{-}4}$=11, $J_{4\text{-}5}$=9, $J_{3ax\text{-}4}$=4.5, H-4); 2.24 (m, 1H, H-3eq); 1.9 (ddd, 1H, $J_{2\text{-}3ax}$=12, $J_{3ax\text{-}4}$=11, $J_{3eq\text{-}3ax}$=12, H-3ax); ^{13}C NMR (63MHz; $CDCl_3$): δ: 73.8, 72.6, 71, 70.6 (C-1, C-2, C-4, C-5, C-6); 26.8 (C-3); (Found : C, 77.74; H, 6.89; $C_{34}H_{36}O_5$ requires C, 77.83; H, 6.92).

L-1,2,4,5-Tetra-*O*-benzyl-3-deoxy-*chiro*-6-inosose 15

N-oxy-4-methylmorpholine (214 mg, 1.82 mmol), tetra-n-butylammonium per-ruthenate (43 mg, 0.12 mmol) and molecular sieves (600 mg) were added to a solution of **14** (639 mg, 1.22 mmol) in anhydrous CH_2Cl_2 (10 ml). The mixture was stirred for 45 min before addition of isopropanol (10ml). The stirring was maintained for another 30 min before concentration and filtration through florisil (eluent : AcOEt). The filtrate was concentrated under reduce pressure and used with no further purification (98 %); IR : 1738 cm^{-1}.

D-1,2,4,5-Tetra-*O*-benzyl-6-deoxy-*myo*-inositol 16

$NaBH_4$ (12 mg, 0.31 mmol) was added at 0°C to a solution of the ketone **15** (104 mg, 0.21 mmol) in EtOH (4ml). The solution was stirred for 30 min before addition of aqueous solution of NaCl. The mixture was stirred overnight and then coevaporated twice with isopropanol and filtered through celite. The filtrate was concentrated and chromatographed on silica gel to give the *myo* inositol **16** (75%); $[\alpha]_D$ +21° (c 1.0, CH_2Cl_2); ^1H NMR (400MHz; $CDCl_3$): δ: 4.00 (m, 1H, H-4); 3.87 (dd, 1H, $J_{2-3}=J_{3-4}=4.5$, H-3); 3.82 (m,1H, H-2); 3.74 (ddd, 1H, $J_{1-6ax}=11$, $J_{1-6eq}=4$ 5, $J_{1-2}=4$, H-1); 3.62 (m, 1H, H-5); 2.20 (m, 1H, H-6eq); 2.00 (ddd, 1H, $J_{5-6ax}=12$, $J_{1-6ax}=11$, $J_{6eq-6ax}=12.5$, H-6ax); (Found : C, 77.96; H, 7.04; $C_{34}H_{36}O_5$ requires C, 77.83; H, 6.92).

D-1,2,4,5-Tretra-*O*-benzyl-6-deoxy-*myo*-inositol-3-(dibenzyl)phosphate 17

Alcohol **16** was phosphorylated to the 3-monophosphate **17** using method A in the presence of dibenzyloxy(diisopropylamino)phosphine reagent (71%); $[\alpha]_D$ +8° (c 1,6 CH_2Cl_2); ^1H NMR (400MHz; $CDCl_3$): δ: 4.72 (m, 1H, H-3); 4.21 (m, 1H, H-4); 3.81 (m, 1H, H-1); 3.71 (m, 2H, H-2, H-5); 2.37 (ddd, 1H, $J_{1-6ax}=J_{5-6ax}=11$, $J_{6eq-6ax}=12$, H-6ax); 1.85 (m, 1H, H-6eq); ^{13}C NMR (50MHz; $CDCl_3$): δ: 74.1, 73.55, 73.5 (C-1, C-2, C-4, C-6); 29.0 (C-3); (Found : C, 73.46; H, 6.47; P, 4.03; $C_{48}H_{49}O_8P$ requires C, 73.46; H, 6.28; P, 3.94).

D-6-Deoxy-*myo*-inositol-3-monophosphate 18

(Dibenzyl)phosphate **17** dissolved in the minimun amount of EtOH 95% was hydrogenated for 1 h, under 3 psi, in the presence of the same amount of Pd/C 10%. The catalyst was removed by filtration. Tris(hydroxymethyl)aminomethane (TRIS, 2 eq per phosphate) was added before concentration *in vacuo*. The aqueous solution was lyophilized and the 6-deoxy *myo* inositol(3)P **18** was precipitated as bis TRIS-salt; $[\alpha]_D$ 0° (c 1, H_2O); ^1H NMR (400MHz, D_2O): δ: 4.28 (m, 1H, H-1); 4.07 (m, 2H, H-2, H-5); 4.02 (m, 1H, H-3); 3.52 (m, 1H, H-4); 2.17 (m ,1H, H-6ax); 1.79 (m, 1H, H-6eq); ^{13}C NMR (50MHz; $CDCl_3$): δ: 79.4 (C-3); 75.1 (C-4); 71.2 (C-5); 70.9 (C-2); 70.6 (C-1); 32.7 (C-6); (Found : C, 34.61; H, 7.37; N, 5.27; P, 6.09; $C_{14}H_{34}O_{14}N_2P$ requires C, 34.68; H, 7.05; N, 5.77; P, 6.37).

D-4-O-Benzyl-2,3-*O*-cyclohexylidene-6-deoxy-*myo*-inositol-1,5-bis(dibenzyl)phosphate 19

Treatment of diol **1** by method A in the presence of dibenzyloxy(diisopropylamino)phosphine gave the bis(dibenzyl)phosphates **19** as a yellow oil (78%); $[\alpha]_D$ -13° (c 1, CH_2Cl_2); ^1H NMR (200MHz; $CDCl_3$): δ: 5.00 (d, 1H, $J_{1-2}=4$, H-1); 4.56 (dd, 1H, $J_{5-4}=8$, $J_{5-6ax}=12$, H-5); 4.33 (dd, 1H, $J_{2-1}=4$, $J_{2-3}=5$, H-2); 4.20 (dd, 1H, $J_{3-2}=5$, $J_{3-4}=7$, H-3); 4.06 (dd, 1H, $J_{4-3}=7$, $J_{4-5}=8$, H-4); 1.96 (m, 2H, H-6ax, H-6eq); (Found C, 66.19; H, 6,35; P, 7.18; $C_{47}H_{52}O_{11}P_2$ requires C, 66.03; H, 6.13; P, 7.24).

D-6-Deoxy-*myo*-inositol-1,5-bisphosphate 20

Bis(dibenzyl)posphate **19** dissolved in the minimun amount of EtOH 95% was hydrogenated for 1 h, under 4-5 psi, in the presence of the same amount of Pd/C 10%. The catalyst was removed by filtration. Tris(hydroxymethyl)aminomethane (TRIS, 2 eq. per phosphate) was added before concentration *in vacuo*. The aqueous solution was lyophilized and the *myo* inositol(1,5)P_2 **20** was precipitated as tetra-TRIS-salt; $[\alpha]_D$ -3° (c 1.3, H_2O); ^1H NMR (400MHz, D_2O): δ: 4.41-3.64 (m, 4H, H-1, H-3, H-4, H-5); 2.08 (m, 1H, H-6eq); 1.74 (m, 1H, H-6ax); ^{13}C NMR (63MHz; D_2O): δ: 76.8 (C-3); 75.1, 74.7 (C-1, C-2, C-4); 29.7, 27.2 (C-5, C-6); ^{31}P NMR (81MHz; D_2O): δ:+6.15-6.22 (P1, P5); (Found C, 32.63; H, 7.42; N, 6.84; P, 7.24; $C_{22}H_{58}O_{23}N_4P_2$ requires C, 32.67; H, 7.23; N, 6.93; P, 7.66).

D-2,3-*O*-Cyclohexylidene-5,6-dideoxy-*myo*-inositol-1,4-bis(dibenzyl)phosphate 22

Treatment of the 5,6-dideoxy *myo* inositol **21** by method A in the presence of dibenzyloxy (diisopropylamino)phosphine reagent gave the bis(dibenzyl)posphates **22** as a yellow oil (72%); $[\alpha]_D$ +20° (c 1.8, $CHCl_3$); ^1H NMR (250MHz; $CDCl_3$) δ: 5.00 (m 8H, $C\underline{H}_2Ph$); 4.51 (m, 1H, H-4); 4.33 (m, 2H, H-1, H-3); 3.93 (t, 1H, $J_{1-2}=J_{2-3}=4$, H-2); 2.00-1.20 (m, 4H, H-5ax, H-5eq, H-6ax, H-6eq); ^{13}C NMR (63MHz; $CDCl_3$): δ: 111.0 (O-C-O); 78.1 (C-3); 74.7, 74.1, 74.0 (C-1, C-2, C-4); 69.3 ($4\underline{C}H_2Ph$); 25.7, 25.0

(C-5, C-6); ^{31}P NMR (81MHz; $CDCl_3$): δ: -1.06 (P_1, P_4); (Found: C, 63.34; H, 6.26; P, 7.41; $C_{40}H_{46}O_{10}P_2,1/2H_2O$ requires C, 63.40; H, 6.25; P, 8.17).

D-5,6-Dideoxy-*myo*-inositol-1,4-bisphosphate 23

Bis(dibenzyl)phosphates **22** dissolved in the minimun amount of EtOH 95% was hydrogenated for 1 h, under 4-5 psi, in the presence of the same amount of Pd/C 10%. The catalyst was removed by filtration. Tris(hydroxymethyl)aminomethane (TRIS, 2 eq. per phosphate) was added before concentration *in vacuo*. The aqueous solution was lyophilized and the *myo* inositol(1,4)P_2 **23** was precipitated as tetra-TRIS-salt; $[\alpha]_D$ -3° (c 1.0, H_2O); ^1H NMR (400MHz, D_2O): δ: 4.80 (s, 1H, OH); 4.20-3.90 (m, 3H, H-1, H-3, H-4); 2.00 (m, 1H, H-6eq); 1.80 (m, 2H, H-6ax, H-5eq); 1.30 (m, 1H, H-5ax); ^{13}C NMR (63MHz; D_2O): δ: 76.8 (C-3); 75.1, 74.7 (C-1, C-2, C-4); 29.7, 27.2 (C-5, C-6); ^{31}P NMR (81MHz; D_2O): δ:+6.26 (P1, P4); (Found C, 31.71; H, 7.81; N, 6.64; $C_{22}H_{58}N_4O_{23}P_2$, $2H_2O$ requires C, 31.88; H, 7.54; N, 6.76).

L-1-*O*-Benzyl-5,6-*O*-cyclohexylidene-3-deoxy-*chiro*-inositol-2,4-bis(dibenzyl)phosphate 24

Cyclohexylidene diol **11** was converted into the bis(dibenzyl)phosphate **24** (68%), using phosphorylation method A in the presence of dibenzyloxy(diisopropylamino)phosphine reagent; $[\alpha]_D$ -7° (c 1, CH_2Cl_2), ^1H NMR (200MHz, $CDCl_3$): δ: 5.00 (m, 1H, J_{4-3}=8, J_{4-3ax}=4 , J_{4-3eq}=8, H-4); 4.80 (m, 2H, CH_2Ph); 4.70 (m, 1H, J_{2-1}=4, J_{2-3ax}=3, J_{2-3eq}=6, H-2); 4.26 (dd, 1H, J_{6-5}=5, J_{6-1}=6, H-6); 4.22 (dd, 1H, J_{1-2}=4, J_{1-6}=6, H-1); 4.00 (dd, 1H, J_{5-4}=8, J_{5-6}=5 H-5); 2.23 (m, 2H, $J_{3ax-3eq}$=12, J_{3ax-4}=4, J_{3ax-2}=3, J_{3eq-4}=8, J_{3eq-2}=6, H-3ax, H-3eq); (Found C, 66.26; H, 6.39; P, 7.49; $C_{47}H_{52}O_{11}P_2$ requires C, 66.03; H, 6.13; P, 7.24).

L-3-Deoxy-*chiro*-inositol-2,4-bisphosphate 25

The (dibenzyl)phosphate **9** dissolved in the minimun amount of EtOH 95% was hydrogenated for 1 h, under 3 psi, in the presence of the same amount of Pd/C 10%. The catalyst was removed by filtration. Tris(hydroxymethyl)aminomethane (TRIS, 2 eq per phosphate) was added before concentration *in vacuo*. The aqueous solution was lyophilized and the *chiro* inositol(2,4)P_2 **25** was precipitated as tetraTRIS-salt. $[\alpha]_D$ -3° (c 1.2, H_2O); (Found C, 32.70; H, 7.32; N, 7.00; P, 7.43; $C_{22}H_{58}O_{23}N_4P_2$ requires C, 32.67; H, 7.23; N, 6.93; P, 7.66).

D-2,3-*O*-Cyclohexylidene-6-deoxy-*myo*-inositol-1,4,5-tris(dibenzyl)phosphate 26

Trisphosphate **26** was prepared by phosphorylation method A from triol **2** in the presence of dibenzyloxy(diisopropylamino)phosphine reagent. Compound **26** was crystallized (75%); m.p. 76°C; $[\alpha]_D$ +4° (c 0.8, $CHCl_3$); ^1H NMR (200MHz; $CHCl_3$): δ: 4.80 (dd, 1H, J_{3-4}=7, J_{4-5}=9, H-4); 4.50 (m, 1H, J_{1-2}=3, J_{1-6ax}=12, J_{1-6eq}=4, H-1); 4.40 (dd, 1H, J_{2-1}=3, J_{2-3}=5, H-2); 4.20 (m, 1H, J_{5-4}=9, J_{5-6ax}=12, J_{5-6eq}=4, H-5); 4.00 (dd, 1H, J_{3-2}=5, J_{3-4}=7, H-3); 2.45 (m, 1H, $J_{6eq-6ax}$=13, J_{6eq-1}=4, J_{6eq-5}=12, H-5); 2.22 (m, 1H, $J_{6eq-6ax}$=13, J_{6ax-1}=12, J_{6ax-5}=4, H-5); (Found C, 63.18; H, 6.10; P, 9.09; $C_{54}H_{59}O_{14}P_3$ requires C, 63.27; H, 5.80; P, 9.07).

D-6-Deoxy-*myo*-inositol-1,4,5-trisphosphate 27

Tris(dibenzyl)phosphate **26** dissolved in the minimun amount of EtOH 95% was hydrogenated for 1 h, under 3 psi, in the presence of the same amount of Pd/C 10%. The catalyst was removed by filtration. Tris(hydroxymethyl)aminomethane (TRIS, 2 eq. per phosphate) was added before concentration *in vacuo*. The aqueous solution was lyophilized and the Ins(1,4,5)P_3 **27** was precipitated as hexa TRIS-salt; $[\alpha]_D$ -3° (c 1.08, H_2O); ^1H NMR (400MHz; D_2O): δ: 4.10 (m, 1H, H-2); 4.06 (m, 1H, H-5); 3.90 (m, 1H, H-1); 3.60 (bs, 1H, H-3); 3.59 (dd, 1H, H-4); 2.20 (m, 1H, H6-eq); 1.90 (m, 1H, H6-ax); ^{13}C NMR (50MHz; $CDCl_3$): δ: 79.5 (C-3); 74.4 (C-1); 73.8 (C-4, C-5); 71.4 (C-2); 34.9 (C-6); (Found C, 30.37; H, 7.39; N, 6.83; $C_{30}H_{81}O_{32}N_6P_3$+ 3 H_2O requires C, 30.40; H, 7.40; N, 7.09).

D-6-Deoxy-*myo*-inositol-1,2-cyclic-4,5-trisphosphate 28

Tri(dibenzyl)phosphate **26** dissolved in the minimun amount of EtOH 95% was hydrogenated for 1 h under 2 psi in the presence of the same amount of Pd/C 10%. The catalyst was removed by filtration and water

was removed *in vacuo* then the residue was lyophilized to give the cyclic phoshate **28** quantitatively; $[\alpha]_D$ -0.5° (c 0.92, H_2O); ^{31}P NMR (81MHz; D_2O): δ: - 5.85 (P1,2), + 6.34 (P4, P5); (Found C, 18.64; H, 3.17; P, 24.30; $C_6H_{13}O_{13}P_3$ requires C, 18.66; H, 3.39; P, 24.07).

D-2,3-*O*-Cyclohexylidene-6-deoxy-*myo*-inositol-1,4,5-tris(dibutyl)phosphate 30

Treatement of triol **2** by phosphorylation method A in the presence of dibutyloxy (diisopropylamino)phosphine reagent gave trisphosphate **30** (60%); $[\alpha]_D$ +4° (c 3, $CHCl_3$); ^1H NMR (200MHz; $CDCl_3$): δ: 4.65 (m, 1H, H-1); 4.50 (m, 2H, H-2, H-3); 4.25 (m, 1H, H-5); 4.10 (m, 13H, OCH_2, H-4); 2.55 (dt, 1H, $J_{6eq-6ax}$=12; J_{6eq-1}=J_{6eq-5}=4, H-6eq); 2.3 (q, 1H, $J_{6ax-6eq}$ =J_{6ax-1}=J_{6ax-5}=12, H-6ax); 1.60 (m, 34H, $OCH_2C\underline{H}_2$, C_6H_{10}); 0.90 (t, 18H, CH_3); ^{13}C NMR (50MHz; $CDCl_3$): δ: 111.4 (O-C-O); 80.8 (C-4); 77.4 (C-3); 74.8 (C-2); 73.0 (C-5); 70.6 (C-1); 67.7 (6$\underline{C}H_2O$); 32.4 (6$\underline{C}H_2CH_2O$); 31.8 (C-6); 18.7 (6$\underline{C}H_2CH_3$); 13.6 (6$\underline{C}H_3$). ^{31}P NMR (81MHz; $CDCl_3$): δ: -1.35, -1.10, -0.75 (P_1, P_4, P_5); (Found C, 52.61; H, 8;94; P, 10.92; $C_{36}H_{71}O_{14}P_3$ requires C, 52.67; H, 8;72; P, 11.32).

D-6-Deoxy-*myo*-inositol-1,4,5-tris(dibutyl)phosphate 31

To a solution of thrisphosphate **30** (200mg, 0.24 mmol) dissolved in dry acetonitrile (10ml), $BF_3.Et_2O$ (0.5 ml) was added and stirred 12 h at r.t.. After evaporation to dryness, the residue was diluted with dioxane (10ml) and aq. HCl 37% (1ml) was added. The mixture was stirred over 3 h before neutralization with sodium bicarbonate and evaporation of organic layer. The residue was chromatographed on silica gel to give **31** (88%); m.p. 139-141°C; $[\alpha]_D$ -17° (c 1.1, $CHCl_3$); ^1H NMR (250MHz; $CDCl_3$): δ: 4.40 (q, 1H, J_{4-3} =J_{4-5}=9, H-4); 4.30 (m, 1H, H-1); 4.20 (m, 2H, H-2, H-5); 4.0 (m, 12H, C$\underline{H}_2$O); 3.5 (dd, 1H, J_{3-4}=9; J_{3-2}=3, H-3); 2.40 (m, 1H, H-6eq); 2.30 (q, 1H; $J_{6ax-6eq}$=J_{6ax-1}=J_{6ax-5}=12, H-6ax); 1.60 (m, 12H, C$\underline{H}_2CH_2O$); 1.35 (m, 12H, C$\underline{H}_2CH_3$); 0.90 (t, 18H, CH_3). ^{13}C NMR (63MHz; $CDCl_3$): δ: 80.51 (C-4); 73.57 (C-5); 72.29 (C-1); 71.30 (C-2, C-3); 68.48, 67.87 (6$\underline{C}H_2O$); 32.31 (6$\underline{C}H_2CH_2O$); 31.54 (C-6); 18.70 (6$\underline{C}H_2CH_3$); 13.61 (6$\underline{C}H_3$); ^{31}P NMR (81MHz; $CDCl_3$): δ: -1.23, +0.80 (P_1, P_4, P_5); (Found C, 47.90; H, 8.86; P, 12.09; $C_{30}H_{63}O_{14}P_3$+1/2 H_2O requires C, 48.05; H, 8;61; P, 12.39).

D-2,3-*O*-Cyclohexylidene-6-deoxy-*myo*-inositol-1,4,5-tris(dihexyl)phosphate 32

Trisphosphate **32** was prepared by phosphorylation method A from triol **2** in the presence of dihexyloxy(diisopropylamino)phosphine reagent (60%); $[\alpha]_D$ +3° (c 0.6, $CHCl_3$); ^1H NMR (400MHz, $CDCl_3$): δ: 4.65 (m, 1H, H-1); 4.45 (dd, 2H, $J_{3,4}$=$J_{4,5}$=10, H-2, H-4); 4.15 (m, 1H, H-5); 4.05 (m, 13H, H-3, OCH_2); 2.50 (dt, 1H, $J_{6eq-6ax}$=12, J_{6eq-1}=J_{6eq-5}=4, H-6eq); 2.20 (q, 1H, $J_{6ax-6eq}$ =J_{6ax-1}=J_{6ax-5}=12, H-6ax); 1.60 (m, O$\underline{C}H_2CH_2$, C_6H_{10}); 1.30 (m, C$\underline{H}_2CH_3$, C_6H_{10}); 0.85 (t, 1H, CH_3); ^{13}C NMR (63MHz; $CDCl_3$): δ: 111.8 (O-C-O); 80.9 (C-4); 77.5 (C-3); 74.8 (C-2); 72.9 (C-5); 70.6 (C-1); 68.2 ($\underline{C}H_2O$); 31.9 (C-6); 22.6 (6$\underline{C}H_2CH_3$); 14.0 [6($\underline{C}H_3CH_2)_5$]; ^{31}P NMR (81MHz; $CDCl_3$): δ: -1.33, -1.08, -0.84 (P_1, P_4, P_5); (Found C, 57.95; H, 9.52; P, 9.75; $C_{48}H_{95}O_{14}P_3$ requires C, 58.28; H, 9.68; P, 9.40).

D-6-Deoxy-*myo*-inositol-1,4,5-tris(hexyl)phosphate 33

To a solution of trisphosphate **32** (99 mg, 0.1 mmol) dissolved in dry acetonitrile (5 ml), $BF_3.Et_2O$ (0.25 ml) was added and stirred 12 h at r.t.. After evaporation to dryness, the residue was diluted with dioxane (5 ml) and aq HCl 37% (0.5 ml) was added. The mixture was stirred over 3 h before neutralization with sodium bicarbonate and evaporation of organic layer. The residue was chromatographed on silica gel to give **33** (80%); $[\alpha]_D$ -15° (c 0.4, $CHCl_3$); S.M. (FAB): 909 $[MH]^+$; ^1H NMR (400MHz, $CDCl_3$): δ: 5.20 (bs, 1H, OH); 4.40 (q, 1H, H-4); 4.30 (m, 1H, H-1); 4.25 (bs, 1H, H-2); 4.20 (m, 1H, H-5); 4.05 (m, 12H, OCH_2); 3.60 (d, 1H, H-3); 3.05 (bs, 1H, OH); 2.40 (m, 1H, H-6eq); 2.35 (q, 1H, $J_{6ax-6eq}$ =J_{6ax-1}=J_{6ax-5}=12, H-6ax); 1.70 (m, 12H, $OCH_2C\underline{H}_2$); 1.30 (m, 12H, C$\underline{H}_2CH_3$); 0.90 (t, 18H, CH_3); ^{13}C NMR (63MHz; $CDCl_3$): δ: 80.61 (C-4); 73.6 (C-5); 72.4 (C-1); 71.3 (C-2. C-3); 69.0-68.0 (6$\underline{C}H_2O$); 31.7 (C-6); 22.6 (6$\underline{C}H_2CH_3$); 14.1 (6($\underline{C}H_3CH_2)_5$); ^{31}P NMR (81MHz; $CDCl_3$): δ: -1.33, -0.99, +1.73 (P_1. P_4. P_5); (Found C, 55.23; H, 9.61; P, 9.97; $C_{42}H_{87}O_{14}P_3$ requires C, 55.49; H, 9.65; P, 10.22).

D-6-Deoxy-2,3-di-*O*-palmitoyl-*myo*-inositol-1,4,5-tris(dibutyl)phosphate 34

To a solution of diol **31** (74 mg, 0.1 mmol), dissolved in anhydrous CH_2Cl_2 (10 ml) were added palmitic acid (77 mg, 0.3 mmol), dicyclohexyl carbodiimide (62 mg, 0.3 mmol.) and DMAP (20 mg). The solution was stirred 6 h at r.t., filtered on celite and the filtrate was concentrated under reduced pressure. The residue was flash chromatographed on silica gel to give the crystalline diester **34** (78%); m.p. 37-39°C; $[\alpha]_D$ +8° (c 1.8, $CHCl_3$); ^1H NMR (250MHz, $CDCl_3$): δ: 5.60 (bs, 1H, H-2); 4.90 (dd, 1H, J_{3-2}=2.5, J_{3-4}=10, H-3); 4.70 (q, 1H, J_{4-3}=J_{4-5}=10, H-4); 4.50 (m, 1H, H-1); 4.30 (m, 1H, H-5); 4.05 (m, 12H, OCH_2); 2.65 (m, 1H, $J_{6eq-6ax}$=12, J_{6eq-1}=J_{6eq-5}=4, H-6eq); 2.35 (m, 4H, CH_2CO); 2.20 (q, 1H, J_{6ax-1}=J_{6ax-5}=$J_{6ax-6eq}$=12, H-6ax); 1.70 (m, 16H, $C\underline{H}_2CH_2CO$, $C\underline{H}_2CH_2O$); 1.40 (m, 60H, $(C\underline{H}_2)_{12}$, $C\underline{H}_2CH_3$); 0.9 (m, 24H, CH_3); ^{13}C NMR (63MHz; $CDCl_3$): δ: 172.6, 172.4 (2C=O), 76.9, 73.3 (C-1, C-4, C-5); 69.5 (C-2; C-3); 68.0 ($6\underline{C}H_2O$); 32.3 ($6\underline{C}H_2CH_2O$); 32.0 (C-6); 18.7 ($\underline{C}H_2CH_3$); 14.2, 13.7 ($\underline{C}H_3(CH_2)_3$, $\underline{C}H_3(CH_2)_{14}$); ^{31}P NMR (81MHz; $CDCl_3$): δ: -2.58, -2.30 (P_1, P_4, P_5); (Found C, 61.55; H, 10.24; P, 7.49; $C_{62}H_{123}O_{16}P_3$ requires C, 61.16; H, 10.18; P, 7.63).

D-6-Deoxy-*myo*-inositol-1,4,5-tris(dibenzyl)phosphate 35

Trisphosphate **26** (163 mg, 0.15 mmol) was treated with a HCl (1N)/ methanol solution for 2 h at r.t.. The mixture was evaporated to dryness and the residue was purified by flash chormatography on silica gel. Crystallization gave the tris(dibenzyl)phosphate **35** (76%); m.p. 122°C; $[\alpha]_D$ -1° (c 1, CH_2Cl_2); ^1H NMR (200MHz, $CDCl_3$): δ: 4.50 (t, 1H, J_{4-3}=J_{4-5}=10, H-4); 4.20 (m, 3H, J_{2-3}=2, H-1, H-2, H-5); 3.40 (m, 1H, H-3); 2.30 (m, 2H, H-6ax, H-6eq); (Found C, 64.05; H, 5.27; P, 9.63; $C_{48}H_{51}O_{14}P_3$ requires C, 63.99; H, 5.47; P, 9.56).

D-6-Deoxy-3-*O*-(β-benzyloxy)myristoyl-*myo*-inositol-1,4,5-tris(dibenzyl)phosphate 36

To the diol **35** (95 mg, 0.1 mmol) dissolved in dry CH_2Cl_2 (10 ml) were added DCC (41 mg, 0.12 mmol), DMAP (10 mg) and β-benzyloxymyristic acid (40 mg, 0.12 mmol). After 4h of stirring at r.t., the mixture was filtered on celite and the filtrate was concentrated. The residue was chromatographed on silica gel to give the crystalline monoester **36** (60%); m.p. 88-90°C; $[\alpha]_D$ 0° (c 1, CH_2Cl_2); ^1H NMR (200MHz; $CDCl_3$): δ: 4.85 (m, 13H, H-3 et $C\underline{H}_2Ph$); 4.70 (q, 1H, J_{4-3}=J_{4-5}=9, H-4); 4.45 (bs, 2H,$C\underline{H}_2Ph$); 4.10 (m, 1H, H-1); 4.00 (bs, 1H, H-2); 3.65 (m, 1H, H-5); 3.40 (m, 1H, $C\underline{H}OBn$); 2.50-2.00 (m, 4H, H-6eq, H6ax, $C\underline{H}_2C=O$); 0.80 (t, 3H, CH_3); ^{13}C NMR (50MHz; $CDCl_3$): δ: 172.3 (C=O); 79.1 (C-4); 77.1, 76.4, 75.7 (C-1, C-5, C-2); 71.0 (C-3); 33.0 (C-6); (Found C, 65.68; H, 6.73; P, 7.47; $C_{69}H_{83}O_{16}P_3$ requires C, 65.70; H, 6.73; P, 7.37).

D-6-Deoxy-2,3-Di-*O*-(β-benzyloxy)myristoyl-*myo*-inositol-1,4,5-tris(dibenzyl)phosphate 37

To the diol **35** (95 mg, 0.1 mmol) dissolved in dry CH_2Cl_2 (10 ml) were added DCC (82 mg, 0.24 mmol), DMAP (20 mg) and β-benzyloxymyristic acid (80 mg, 0.24 mmol). After 4h of stirring at r.t., the mixture was filtered on celite and the filtrate was concentrated. The residue was chromatographed on silica gel to give the diester **37** (84%); $[\alpha]_D$ 0° (c 1, CH_2Cl_2); ^1H NMR (200MHz; $CDCl_3$): δ: 5.65 (bs, 1H, H-2); 5.00 (m, 14H, H-3, H-4, $C\underline{H}_2Ph$); 4.40 (m, 6H, H-1, H-5, $C\underline{H}_2Ph$); 3.80 (m, 1H, $C\underline{H}OBn$); 3.65 (m, 1H, $C\underline{H}OBn$); 2.50 (m, 5H, H-6eq, $2C\underline{H}_2C=O$); 2.20 (m, 1H, H-6ax); 0.85 (t, 6H, $2CH_3$); ^{13}C NMR (63MHz; $CDCl_3$): δ: 170.8, 170.2 ($2\underline{C}=O$); 77.1 (C-4); 75.7, 75 .2 (C-1, C-5); 73.4 (C-2); 69.3 (C-3); 32.5 (C-6); (Found C, 68.32; H, 7.06; P, 5.88; $C_{90}H_{115}O_{18}P_3$ requires C, 68.51; H, 7.35; P, 5.89).

D-6-Deoxy-3-*O*-(β-hydroxy)myristoyl-*myo*-inositol-1,4,5-trisphosphate 38

Tris(dibenzyl)phosphate **36** dissolved in the minimun amount of EtOH 95% was hydrogenated for 2 h under 4-5 psi in the presence of the same amount of Pd/C 10%. The catalyst was removed by filtration on Whatman paper. Tris(hydroxymethyl)aminomethane (TRIS, 2 eq. per phosphate) was added and the aqueous solution was concentrated *in vacuo*. After lyophilization, the trisphosphate **38** was precipitated as hexa TRIS-salt.;$[\alpha]_D$ 0° (c 0.92, H_2O); ^1H NMR (400MHz; D_2O): δ: 4.80 (m, 38H, $O\underline{H}$, $N\underline{H}_2$); 4.40 (m, 1H, OH); 4.10 (m, 2H, H-2, H-5); 3.80 (m, 1H, H-1); 3.50 (dd, 1H, H-4); 2.10 (m, 1H, H-6eq); 2.00 (m, 1H, H-6ax); 1.45

(m, 2H, CH_2CO); 1.20 (m, 20H, $(CH_2)_{10}$); 0.80 (t, 3H, CH_3); (Found C, 38.75; H, 7.72; N, 7.42; P, 7.02; $C_{47}H_{107}O_{34}N_6P_3$ requires C, 38.93; H, 7.95; N, 6.10; P, 6.85).

D-6-Deoxy-2,3-di-*O*-(β-hydroxy)myristoyl-*myo*-inositol-1,4,5-trisphosphate 39

Tris(dibenzyl)phosphate **37** dissolved in the minimun amount of EtOH 95% was hydrogenated for 2 h under 4-5 psi in the presence of the same amount of Pd/C 10%. The catalyst was removed by filtration on Whatman paper. Tris(hydroxymethyl)aminomethane (TRIS, 2 eq. per phosphate) was added and the aqueous solution was concentrated *in vacuo*. After lyophilization, the trisphosphate **39** was precipitated as hexa TRIS-salt; $[\alpha]_D$ 0° (c 1, H_2O); ^1H NMR (250MHz; D_2O): δ: 4.80 (m, 38H, O<u>H</u>, N<u>H</u>$_2$); 4.40-3.00 (m, 43H, H-1, H-2, H-3, H-4, H-5, C<u>H</u>$(CH_2)_{10}$, 6 $(CH_2OH)_3$); 2.80-2.00 (m, 6H, H-6eq, H-6ax, 2C<u>H</u>$_2$C=O); 1.60-1.00 (m, 40H, 2(C<u>H</u>$_2)_{10}$); 0.80 (t, 6H, $2CH_3$); (Found C, 44.01; H, 8.52; N, 5.27; P, 6.00; $C_{58}H_{133}O_{36}N_6P_3$ requires C, 43.98; H, 8.46; N, 5.31, P, 5.87)

D-2-*O*-Benzyl-3,6-dideoxy-*myo*-inositol-1,4,5-tris(dibenzyl)phosphate 41

Triol **40** was converted to the tris(dibenzyl)phosphate **41** using method A in the presence of dibenzyloxy(diisopropylamino)phosphine reagent (55%); ^1H NMR (250MHz; $CDCl_3$): δ: 4.56 (bs, 1H, H-4); 4.3 (m, 2H, H-1, H-5); 3.73 (m, 1H, H-2); 2.43 (m, 1H, H-3ax); 2.33 (m, 2H, H-6ax, H-6eq); 1.33 (m, 1H, H-3eq); ^{13}C NMR (63MHz; $CDCl_3$): δ: 75.1, 74.4, 73.7 (C-1, C-2, C-4, C-5); 32.3, 31.1 (C-3, C-6); (Found C, 64.19; H, 5.67; O, 21.30; P, 9.12; $C_{55}H_{57}O_{13}P_3$,1/2 H_2O requires C, 64.27; H, 5.68; O, 21.00; P, 9.04).

D-3,6-dideoxy-*myo*-inositol-1,4,5-trisphosphate 42

Tris(dibenzyl)phosphate **41** dissolved in the minimun amount of EtOH 95% was hydrogenated for 2 h, under 4-5 psi, in the presence of the same amount of Pd/C 10%. The catalyst was removed by filtration on Whatman paper. Tris(hydroxymethyl)aminomethane (TRIS, 2 eq. per phosphate) was added and the aqueous solution was concentrated *in vacuo*. After lyophilization, the trisphosphate **42** was precipitated as hexa TRIS-salt; ^1H NMR (250MHz; $CDCl_3$): δ: 3.70 (m, 4H, H-1, H-2, H-4, H-5); 2.29 (m, 4H, H-3ax, H-3eq, H-6ax H-6eq); ^{13}C NMR (63MHz; $CDCl_3$): δ: 75.6, 73.6, 73.0 (C-1, C-4, C-5); 69.7 (C-2); 35.4, 34.5 (C-3, C-6); ^{31}P NMR (81MHz; D_2O): δ: 6.47 (P_1, P_4, P_5); (Found : C, 33.13; H, 7.41; N, 7.04; P, 7.04; $C_{30}H_{81}O_{31}N_6P_3$, EtOH requires C, 33.11; H, 7.53; N, 7.23; P, 8.00).

D-2-*O*-Benzyl-3,6-dideoxy-3-fluoro-*myo*-inositol-1,4,5-tris(dibenzyl)phosphate 44

3-Fluoro triol **43** was converted to the tris(dibenzyl)phosphate **44** using phosphorylation method A in the presence of dibenzyloxy(diisopropylamino)phosphine reagent (50%); $[\alpha]_D$ -10° (c 0.4, $CHCl_3$); ^1H NMR (250MHz; $CDCl_3$): δ: 5.00 (m, 12H1/2, 1/2H-3, $6PhCH_2OP$); 4.85 (d, 1/2H, J_{3-4}=8, 1/2H-3); 4.75 (m, 1H, H-1); 4.60 (m, 2H, H-4, H-5); 4.55 (dd, 2H, C_2-O-CH_2Ph); 3.95 (bs, 1H, H-2); 2.35 (m, 1H, H-6eq); 2.20 (q, 1H, $J_{6ax-6eq}=J_{6ax-1}=J_{6ax-5}$=11, H-6ax); ^{13}C NMR (63MHz; $CDCl_3$): δ: 89.7, 86.7 (C-3; J_{C3-F}=185); 76.0 (C-2); 75.0, 74.7 (C-4, J_{C4-F}= 21); 74.0 (C_2-O-CH_2Ph); 72.3 (C-1, C-5); 69.7 (6<u>C</u>H_2Ph); 31.86 (C-6); ^{31}P NMR (81MHz; $CDCl_3$): δ: -1.55, -1.33, -1.09 (P_1, P_4, P_5); (Found C, 62.68; H, 5.57; P, 8.82; $C_{55}H_{56}O_{13}FP_3$, 1/2 H_2O requires C, 62.62; H, 5.54; P, 8.81).

D-3,6-Dideoxy-3-fluoro-*myo*-inositol-1,4,5-trisphosphate 45

Tris(dibenzyl)phosphate **44** dissolved in the minimun amount of EtOH 95% was hydrogenated for 2 h, under 4-5 psi, in the presence of the same amount of Pd/C 10%. The catalyst was removed by filtration on Whatman paper. Tris(hydroxymethyl)aminomethane (TRIS, 2 eq. per phosphate) was added and the aqueous solution was concentrated *in vacuo*. After lyophilization, the bisphosphate **45** was precipitated as hexa TRIS-salt; $[\alpha]_D$ 0° (c 0.6, H_2O); ^1H NMR (400MHz, D_2O): δ: 5.1(bd, 1H, J_{H3-F}=48, H-3); 4.75 (bs, 27H, OH); 4.25 (m, 4H, H-1, H-2, H-4, H-5); 2.25 (bs, 1H, H-6eq); 1.90 (bs, 1H, H-6ax); ^{13}C NMR (63MHz; D_2O): δ: 92.8, 90.0 (C-3; J_{C3-F}=175); 73.6, 73.4 (C-2; J_{C2-F}=15); 70.4, 69.6, 69.1, 68.7 (C-1, C-4, C-5); 32.4 (C-6). ^{31}P NMR (81MHz; D_2O): δ: 6.50 (P_1, P_4, P_5); (Found : C, 31.72; H, 7.40; N, 7.43; $C_{30}H_{80}O_{31}N_6FP_3$ requires C, 31.80; H, 7.12; N, 7.42).

D-2-*O*-Benzyl-3,6-dideoxy-3-fluoro-*myo*-inositol-1,4,5-tris(dibutyl)phosphate 46

3-Fluoro triol **43** was converted to the tris(dibenzyl)phosphate **44** using phosphorylation method A in the presence of dibutyloxy(diisopropylamino)phosphine reagent (65%); $[\alpha]_D$ -12° (c 0.8, $CHCl_3$); ^1H NMR (250MHz; $CDCl_3$): δ: 5.05 (dd, 1H, J_{H3-F}=48; J_{3-4}=8, H-3); 4.75 (dd, 2H, $C\underline{H}_2Ph$); 4.65 (m, 1H, H-1); 4.55 (m, 1H, H-5); 4.05 (m, 14H, H-2, H-4, $6C\underline{H}_2O$); 2.50 (m, 1H, H-6eq); 2.30 (q, 1H, $J_{6ax-6eq}=J_{6ax-1}=J_{6ax-5}$=12, H-6ax); 1.71 (m, 12H, $C\underline{H}_2CH_2O$); 1.40 (m, 12H, $C\underline{H}_2CH_3$); 0.90 (t, 18H, $C\underline{H}_3$); ^{13}C NMR (50MHz; $CDCl_3$): δ: 90.3, 86.6 (C-3; J_{C3-F}=183); 75.8, 75.7, 75.4, 74.9 (C-2, C-4); 74.2 ($\underline{C}H_2Ph$); 72.1 (C-1, C-5); 67.9 ($6\underline{C}H_2O$); 32.5 ($6\underline{C}H_2CH_2O$); 32.1 (C-6); 18.8 ($6\underline{C}H_2CH_3$); 13.6 ($6(\underline{C}H_3)_3$); ^{31}P NMR (81MHz; $CDCl_3$): δ: -1.29, -1.14, -0.93 (P_1, P_4, P_5); (Found C, 53.22; H, 8.14; P, 10.81; $C_{37}H_{68}O_{13}FP_3$ requires C, 53.36; H, 8.23; P, 11.16).

D-3,6-dideoxy-3-fluoro-*myo*-inositol-1,4,5-tris(dibutyl)phosphate 47

Tris(dibenzyl)phosphate **46** dissolved in the minimun amount of AcOEt was hydrogenated for 2 h, under 4-5 psi, in the presence of the same amount of Pd/C 10%. The catalyst was removed by filtration on Whatman paper. The organic layer was removed *in vacuo* to give the dideoxy tris(dibutyl)phosphate **47** quantitatively; $[\alpha]_D$ -20° (c 0.7, $CHCl_3$); S.M. (FAB): 744 [MH^+]; ^1H NMR (250MHz;$CDCl_3$): δ: 5.10 (bd, 1H, J_{H3-F}=48, H-3); 4.65 (m, 1H, H-4); 4.55 (m, 2H, H-1, H-5); 4.35 (bs, 1H, H-2); 4.10 (m, 12H, $OC\underline{H}_2$); 2.45 (m, 1H, H-6eq); 2.25 (q, 1H, $J_{6ax-6eq}=J_{6ax-1}=J_{6ax-5}$=12, H-6ax); 1.70 (m, 12H, OCH_2CH_2); 1.40 (m, 12H, $C\underline{H}_2CH_3$); 0.90 (t, 18H, CH_3); ^{13}C NMR (63MHz; $CDCl_3$): δ: 91.2, 88.2 (C-3; J_{C3-F}=180); 75.5 (C-4); 73.2, 72.2 (C-1, C-5); 68.5 (C-2); 68.1 ($6\underline{C}H_2O$); 32.4 ($6\underline{C}H_2CH_2O$); 31.5 (C-6); 18.7 ($6\underline{C}H_2CH_3$); 13.6 ($6\underline{C}H_3$); ^{31}P NMR (81MHz; $CDCl_3$): δ: -1.49, -1.25, -1.15 (P_1, P_4, P_5); (Found : C, 48.80; H, 8.12; P, 12.37; $C_{30}H_{62}O_{13}FP_3$ requires C, 48.51; H, 8.41; P, 12.51).

L-5,6-*O*-cyclohexylidene-3-deoxy-*chiro*-inositol-1,2,4-tris(dibenzyl)phosphate 48

Cyclohexylidene diol **7** was converted into the tris(dibenzyl)phosphate **48** (75%), using phosphorylation method A in the presence of dibenzyloxy(diisopropylamino)phosphine reagent; $[\alpha]_D$ -14° (c 0.63, $CHCl_3$); ^1H NMR (200MHz, $CDCl_3$): δ: 5.03 (m, 1H, J_{4-3}=8, J_{4-3ax}=4 , J_{4-3eq}=8, H-4); 4.70 (m, 1H, J_{2-1}=4, J_{2-3ax}=3, J_{2-3eq}=6, H-2); 4.52 (dd, 1H, J_{1-2}=4, J_{1-6}=6, H-1); 4.26 (dd, 1H, J_{6-5}=5, J_{6-1}=6, H-6); 4.00 (dd, 1H, J_{5-4}=8, J_{5-6}=5, H-5); 2.23 (m, 2H, $J_{3ax-3eq}$=12, J_{3ax-4}=4, J_{3ax-2}=3, J_{3eq-4}=8, J_{3eq-2}=6, H-3ax, H-3eq); (Found C, 63.07; H, 6.05 P, 8.84; $C_{54}H_{59}O_{14}P_3$ requires C, 63.27; H, 5.80; P, 9.07).

L-3-deoxy-*chiro*-inositol-1,2,4-trisphosphate 49

Tris(dibenzyl)phosphate **48** dissolved in the minimun amount of EtOH 95% was hydrogenated for 1 h, under 3 psi, in the presence of the same amount of Pd/C 10%. The catalyst was removed by filtration. The filtrate was diluted with water and tris(hydroxymethyl)aminomethane (TRIS, 2 eq per phosphate) was added before concentration *in vacuo*. The aqueous solution was lyophilized and the Ins(1,4,5)P_3 **27** was precipitated as hexa TRIS-salt. ^1H NMR (400MHz, D_2O): δ: 4.70 (m, 1H, H-4); 4.46 (m, 1H, H-2); 4.23 (m, 2H, H-6, H-5); 3.92 (m, 1H, H-1); 2.20 (m, 2H, H-3eq, H-3ax); ^{31}P NMR (81MHz; D_2O): δ: 6.42-6.33 (P_1, P_2, P_4); (Found C, 30.39; H, 7.35; N, 6.85; $C_{30}H_{81}O_{32}N_6P_3$+3 H_2O requires C, 30.40; H, 7.40; N, 7.09).

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