Synthesis of D-*myo*-inositol 1,4,5-trisphosphate analogues

University of St Andrews

School of Chemistry and
Centre for Biomolecular Sciences
Fife, Scotland

Davide Bello

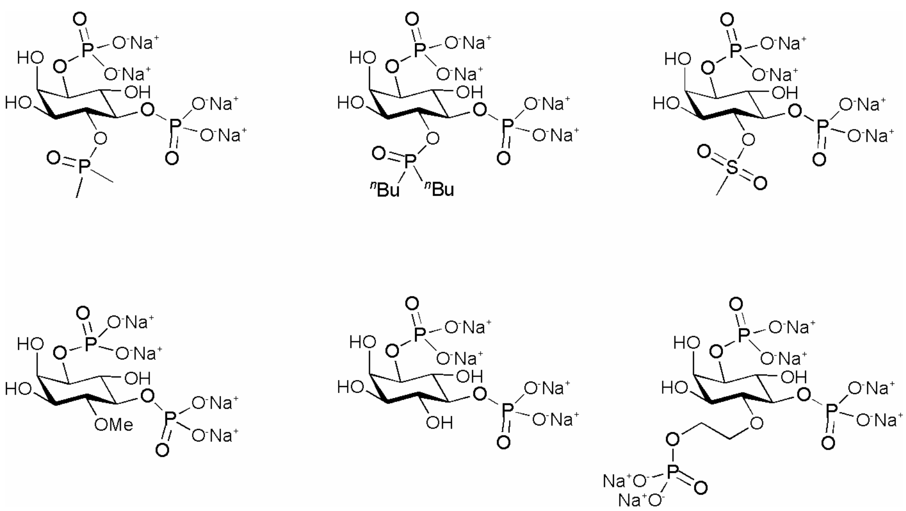
November 2006

Thesis submitted to the University of St Andrews in application for the degree of Doctor of Philosophy

Supervisor: Dr Stuart J. Conway

Abstract

The cytosolic second messenger D-*myo*-inositol 1,4,5-trisphosphate ($InsP_3$), has the ability to mobilise Ca^{2+} from intracellular stores. Ca^{2+} controls a wide range of cellular processes, such as cell division and proliferation, apoptosis, fertilisation, gene transcription and muscle contraction. A number of potent $InsP_3$ receptor agonists are currently known; however, no selective $InsP_3Rs$ antagonists have been reported to date. Using the X-ray crystal structure of the mouse type 1 $InsP_3R$, a range of analogues (below) has been designed with the intention of these compounds acting as competitive $InsP_3Rs$ antagonists. The successful syntheses of these compounds are reported herein.



Declarations

I, Davide Bello, herby certify that this thesis, which is approximately 51000 words in length, has been written by me, that is the record of work carried out by me and that it has not been submitted in any previous application for a higher degree.

Date _______________ Signature of candidate______________________

I was admitted as a research student to the Faculty of Science of the University of St Andrews as a research student in September 2003 and as a candidate for the degree of Doctor of Philosophy in November 2006.

Date _______________ Signature of candidate______________________

I hereby certify that the candidate has fulfilled the conditions of the Resolution and Regulations appropriate for the degree of Doctor of Philosophy in the University of St Andrews and that the candidate is qualified to submit the thesis in application for that degree.

Date _______________ Signature of supervisor___________________

Copyright Declaration

Unrestricted access

In submitting this thesis to the University of St Andrews I understand that I am giving permission for it to be made available for use in accordance with the regulations of the University Library for the time being in force, subject to any copyright vested in the work not being affected thereby. I also understand that the title and abstract will be published, and that a copy of the work may be made and supplied to any *bona fide* library or research worker.

Date _______________ Signed_________________________

Alla mia famiglia

Pe' conto mio la favola più corta
è quella che se chiama Gioventù:
perché... c'era una vorta...
e adesso nun c'è più.

E la più lunga? È quella de la Vita:
la sento raccontà da che sto ar monno,
e un giorno, forse, cascherò dar sonno
prima che sia finita...

Acknowledgements

I would like to thank my supervisor, Dr Stuart Conway, for his invaluable help, advice and encouragement during my PhD work at the University of St Andrews. I would also like to thank the School of Chemistry of St Andrews for funding this work.

I am grateful to Mrs Melanja Smith for her kindness, support and for all the discussions about everything else. I am indebted to her and Dr Tomas Lebl for their patience and help with the NMR analysis. Thanks also to Prof. Alexandra Slawin for her help with solving the crystal structures, to Dr Catherine Botting, Dr Sally Shirran and Mrs Carolyne Horsburgh for helping me out with mass spectrometry and to Mrs Sylvia Williamson for the microanalysis. Thanks also to Dr Gordon Florence and Dr Andrew Smith for answering my endless chemistry questions and proofreading this thesis. Any mistake is solely the responsibility of the author.

Grazie alle persone che amo di più, i miei genitori Mario e Mara e mia sorella Cristina, che ancora mi sopportano nonostante continui ad essere un “protestone”, e grazie ai miei nonni, che sono sempre con me.

I want to thank the people in my group; Joseph, for the unending discussions about too many things; Ross, for introducing me to Scottish culture (Tak awa Aiberdeen an twal mile roun an far ar ye?); Michael for his horrible (though funny) taste in music, Gavin “Gavino”, for being at the same time one of the most brilliant chemists I ever met who taught me many things and a funny half-Welsh lazy dude. Thanks also to the new people in my group; Hajira (you’re da bomb), Tashfeen (one of the sweetest person I ever met), Jess (Éirinn go Brách!), Tom and Neil.

Many thanks to all my friends in St Andrews, Dundee and Edinburgh: Belén, Alejandro, Blerina, Yang Xu, Cristina (la más loca), Dimitris, Nerea, le monsieur Jean-Marie, Mirjana, Bettina, Ana, Henning, Anne Marie. Thanks to my friend Robin, for her sense of humor and her wonderful Indian food. A very special thank to Margaux, who taught me the English language and many other things.

Grazie agli Italiani: Andrea, pe’ esse n’antro romano a Dundee (bella zio), Elena, per le arrampicate; Giulio, per stare fuori come un balcone; Vittoria e Allan, per avermi invitato al loro matrimonio dovo ho magnato come una bestia, Giampiero, Paolo e Francesco per le cene (vasamu li mani). Ringrazio Marcello, che visitai qui anni fa e che in un certo senso mi “introdusse” a St Andrews; compare Antonio e compare Cosimo per le mangiate, le bevute, le chiacchere e l’amicizia.

Grassie a Karen e La Fede, che mi hanno supportato e sopportato in questi anni; a parte le frasi fatte, se ce l’ho fatta è anche merito loro, e per questo hanno tutta la mia stima, pur essendo del Nord (ehehheh).

Grazie ad Armando (aaaaaaoooooo) e Leo (daje ziiii’) sete du’ grossi, spaccate sempre; Clodia per la sua impagabile dolcezza, Dario (alsipscl) e Sole; Andrea (zio cacca), Valentina, Gaia, Piera per sopportare Garro, Gianluca gallicano er cobra pirellone (Garro) per tutte le volte che è venuto a trovarmi qui, e tutte le notti e settimane bukowskiane, e le ceres da soli al pub, i downloads interminabili di materiale altamente educativo, i mioni de cavolate (pe’ usa’ n’eufemismo…) che avemo detto. Te vojo bene. E grazie ovviamente al Dr Alessandro Camangi (e pure a Iggino e l’altro che nun me ricordo come se chiama) per la professionalità con cui ha insegnato a noi tutti ad estimare il solfeggio. Grazie a quer funesto de Andrea (Actarus) e a quell’altro schioppato de Luca (‘mpari); rega’ se vedemo troppo poco, se dovemo inventa’ quarcosa. Grazie anche a Fedes che mi regala momenti esilaranti ad ogni telefonata. E grazie a Franz (aaasd), Betta e Paolo (i + fichi!).
Ciao Micaela, ti penso. Grazie a Marilisa, che non si è mai dimenticata (sei ‘a mejo), a quella scocciata de Rosalba (eh!), a Ele e Andrea per essere sempre lì malgrado tutto.

Eta azkenez, neska ederrengatik ezker, Leticia, geien maite dudana, gaztelainaz itzegiten eta gizon obea izatera irakatzi egin zidana. Nire laztantzu asko maite zaitut.

Table of Contents

List of Abbreviations

°C	degrees Celsius
2-APB	2-aminoethoxydiphenylborate
Å	angstrom
Ac	acetyl
All	allyl
AM	acetoxymethyl
Ar	aryl
ATP	adenosine 5'-trisphosphate
BDCP	tris(2,4,6-tribromophenoxy)dichlorophosphorane
BM	butyryloxymethyl
Bn	benzyl
br s	broad singlet (spectral)
c	concentration
Ca^{2+}	calcium ion
cADPR	cyclic adenosine diphosphate ribose
cAMP	cyclic adenosine 3',5'-monophosphate
CAN	ceric ammonium nitrate
cIMP	inositol 1,2-cyclic phosphate
CNS	central nervous system
C_q	quaternary carbon (spectral)
CSA	camphorsulfonic acid
d	doublet (spectral)
D_6-DMSO	deuterated dimethyl sulfoxide
DAG	diacylglycerol
DDQ	2,3-dichloro-5,6-dicyanobenzoquinone
DIBAL-D	diisobutylaluminium deuteride
DIBAL-H	diisobutylaluminium hydride
D-Ins(1,3,6)PS_3	D-*myo*-inositol 1,3,6-phosphorothioate
D-Ins(1,4,6)PS_3	D-*myo*-Inositol 1,4,6-phosphorothioate
D-InsP_3S_3	D-*myo*-inositol 1,4,5-trisphosphorothioate
DMAP	4-dimethylaminopyridine
DMF	*N,N*-dimethyl formamide
equiv	equivalent

ER	endoplasmic reticulum
Et	ethyl
EtOH	ethanol
FBKP	immunophilin FK506-binding protein
g	grams
GPCRs	G-protein-coupled receptors
GTP	guanosine 5’-trisphosphate
h	hours
Hz	Hertz
^{i}Bu	*iso*-butyl
IC_{50}	inhibitory concentration 50%
Ins(1,3,4)PS_3	DL-*myo*-inositol 1,3,4-phosphorothioate
Ins(1,3,5)PS_3	*myo*-inositol 1,3,5-trisphosphorothioate
Ins(1,4)P_2	*myo*-inositol 1,4-bisphosphate
Ins(1,4)$P_2$5PS	DL-*myo*-Inositol 1,4-bisphosphate-5-phosphorothioate
Ins(1,4,6)PS_3	DL-*myo*-Inositol 1,4,6-phosphorothioate
Ins1PS(4,5)P_2	D-*myo*-inositol 1-phosphorothioate 4,5-bisphosphate
$InsP_3$	D-*myo*-inositol 1,4,5-trisphosphate
$InsP_3$R1	inositol 1,4,5-trisphosphate receptor type 1
$InsP_3$R2	inositol 1,4,5-trisphosphate receptor type 2
$InsP_3$R3	inositol 1,4,5-trisphosphate receptor type 3
$InsP_3$Rs	D-*myo*-inositol 1,4,5-trisphosphate receptors
$InsP_3S_3$	DL-*myo*-inositol 1,4,5-trisphosphorothioate
^{i}Pr	isopropyl
IR	infrared spectroscopy
kDa	kiloDalton
K_i	inhibition constant
L-$InsP_3$	L-*myo*-inositol 1,4,5-trisphosphate
L-$InsP_3S_3$	L-*myo*-inositol 1,4,5-trisphosphorothioate
m	multiplet (spectral); medium (spectral, IR)
M	Molar
m/z (CI)	mass spectrometry, chemical ionisation method
m/z (ES-)	mass spectrometry, negative electrospray method
m/z (ES+)	mass spectrometry, positive electrospray method
*m*CPBA	3-chloroperoxybenzoic acid

Me	methyl
MeCN	acetonitrile
MeOH	methanol
mg	milligrams
MHz	megaHertz
min	minutes
mL	millilitres
mmol	millimoles
mp	melting point
NAADP	nicotinic acid adenine dinucleotide phosphate
^{n}Bu	*n*-butyl
nM	nanoMolar
NMR	nuclear magnetic resonance
NO	nitric oxide
p	pressure
Pg	protecting group
PI-PLC	phosphoinositol-lipid-specific phospholipase C
PKC	protein kinase C
PLC_{β}	phospholipase C type β
PLC_{γ}	phospholipase C type γ
PLC_{δ}	phospholipase C type δ
PLC_{ε}	phospholipase C type ε
PM	propionyloxymethyl
PMA	phosphomolybdic acid
PMB	4-methoxybenzyl
ppm	parts per million
Pr	propyl
Ptd(4,5)InsP_2	phosphatidylinositol 4,5-bisphosphate
PtdIns	phosphatidylinositol
PtdIns(4)P	phosphatidylinositol 4-phosphate
PtdOH	phosphatidic acid
R_f	retention factor
RNA	ribonucleic acid
RT	room temperature
RYR	ryanodine receptor

s	singlet (spectral), strong (spectral, IR); second(s)
S1P	sphingosine 1-phosphate
SERCAs	sarco-endoplasmic reticulum Ca^{2+} ATPases
SOC	store-operated Ca^{2+} channels
sp	septet (spectral)
SR	sarcoplasmic reticulum
t	triplet (spectral)
TBAI	tetra-*n*-butylammonium iodide
TBAS	tetra-*n*-butylammonium sulfate
^{t}BuOH	*tert*-butanol
td	triplet of doublets (spectral)
TEA	triethylamine
Tf	trifluoromethanesulfonyl
THF	tetrahydrofuran
TIPS	triisopropylsilyl
TLC	thin layer chromatography
TMS	tetramethyl silane
TRPV	transient receptor potential vanilloid cation channel
TRPV1	transient receptor potential vanilloid cation channel type 1
TRPV2	transient receptor potential vanilloid cation channel type 2
TRPV3	transient receptor potential vanilloid cation channel type 3
TsOH	4-toluenesulfonic acid
w	weak (spectral, IR)
w/w	weight per unit weight (weight-to-weight ratio)
μL	microlitres
μmol	micromoles

Introduction

1 Introduction

1.1 History

1.1.1. Phospholipids and $InsP_3$

In 1850 Scherer[1] isolated from heart muscle an optically inactive cyclitol possessing an empirical formula of a carbohydrate $[C_n(H_2O)_n]$, which was termed "inosit", after the greek root *inos*, "muscle". The compound name was then translated into the English "inositol", and more recently identified as one of nine possible stereoisomers and named *myo*-inositol (**1**, Figure 1.1).

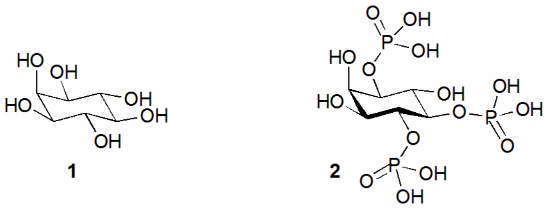


Figure 1.1. The structures of *myo*-inositol (**1**) and $InsP_3$ (**2**).

The existence of inositol phosphates has been known for over eighty years. The first milestone in the discovery of $InsP_3$-signalling was in 1949 when Folch and co-workers[2] isolated a lipid preparation which they called "diphosphoinositide". They assumed that the extract was only one compound; however, the preparation was, in fact, an almost equimolar mixture of phosphatidylinositol (PtdIns), phosphatidylinositol 4-phosphate [PtdIns(4)P] and phosphatidylinositol 4,5-bisphosphate [Ptd(4,5)InsP$_2$], the latter being the phospholipid responsible for the release of $InsP_3$ (**2**, Figure 1.1) by enzymatic hydrolysis, following receptor stimulation (*vide infra*). The metabolic behaviour of diphosphoinositide and other phospholipids was investigated by several groups, but it was not until 1953 that receptor-stimulated lipid turnover was demonstrated by the Hokins.[3]

1.1.2. The "PI" effect

While carrying out studies on the *in vitro* secretion of amylase from respiring pancreas slices stimulated by cholinergic drugs, Lowell and Mabel Hokin found that the addition of acetylcholine stimulated the active secretion of the enzyme, but not its synthesis (as there was no incorporation of ^{32}P into RNA).[3] Analysing the discarded "junk", they found that the lost radioactivity was in the phospholipid fraction and, using a method that allowed the separation and analysis of diacylglycerophospholipids,[4] they showed that the radiolabel was incorporated only in inositol lipids and phosphatidic acid (PtdOH). This became known as the "phosphoinositide" effect ("PI" effect).

In the following 20 years several hypotheses with the intent of explaining the significance of the "PI effect" were developed; this led to some controversies, due to the indirect measurements of the stimulated hydrolysis of inositol lipids.[5] In fact, for many years the PI effect was considered as an event strictly connected with secretion (i.e. of enzymes such as amylase); noticeably, at the same time a number of findings linked the stimulated inositol lipid turnover with some aspects of cell proliferation.[5] It was not until 1964 that Hokin and Hokin[6] deduced that stimulated inositol lipid hydrolysis, with phosphatidylinositol as the presumed substrate, was the initial reaction.

1.1.3. Inositol lipids metabolism is linked to Ca^{2+} homeostasis

Durell and co-workers were first to consider polyphosphoinositol lipids to be involved in receptor-stimulated events.[7] However, detailed studies from Ata Abdel-Latif and Hawthorne[8] on acetylcholine-stimulated phosphodiesteratic cleavage of Ptd(4,5)$InsP_2$, in rabbit iris smooth muscle, apparently showed that there was a requirement for extracellular Ca^{2+} in order to enable the hydrolysis process.[9] This put the phosphoinositol lipids downstream of the Ca^{2+} increase, and therefore remote from the receptors.

In 1975 Michell[10] noticed the coincidence of inositol lipid metabolism with changes in Ca^{2+} homeostasis, and suggested that there was a causal link. Four years later, Berridge and Fain[11] provided the first evidence for Michell's idea; using blowfly salivary glands, organs which are unique in being very permeable to inositol, they were able to prove that the 5-hydroxytryptamine-stimulated breakdown of Ptd(4,5)$InsP_2$ generated inositol phosphates and subsequently mobilised Ca^{2+} from the glands. These inositol phosphates were identified through the measurement of labelled inositol formed by the activity of a dephosphorylating enzyme. Prolonged stimulation resulted in the glands losing their Ca^{2+}, and the response could be restored by supplying inositol to the glands. In the same year Nishizuka and colleagues[12] discovered protein kinase C (PKC) and showed it was a phosphatidylserine-dependent enzyme. In their experiments they found that the huge variability in the efficacy of different batches of phosphatidylserine was due to the presence of various amounts diacylglycerol (DAG) as an impurity. Therefore they proposed that PKC could be regulated *in vivo* by DAG, which was also one of the product of Ptd(4,5)$InsP_2$ hydrolysis.

These findings led Michell *et al.*[13] to put phosphoinositol lipids, and in particular Ptd(4,5)$InsP_2$, upstream of the Ca^{2+} release, as primary substrate for phosphoinositol-lipid-specific phospholipase C (PI-PLC).

1.1.4. The first evidence for $InsP_3$ - Ca^{2+} mobilising capabilities

In 1983 Berridge and co-workers[14] published their findings on inositol phosphates and Ca^{2+} release; their observations provided the missing link between two events, the PI effect (*vide supra*) and Ca^{2+} signalling, which they correctly proposed to be $InsP_3$ acting as a second messenger to mobilise internal Ca^{2+} stores.

Using permeabilised rat pancreatic acinar cells, Berridge and co-workers first demonstrated that $InsP_3$ releases Ca^{2+} only from membrane-bound cellular stores, as $InsP_3$ was unable to release Ca^{2+} from cells that had been pre-treated with a Ca^{2+} ionophore to deplete intracellular Ca^{2+}. In order to identify which intracellular store was sensitive to $InsP_3$, inhibitors of Ca^{2+} uptake were used to reduce the amount of Ca^{2+} available in the store. Cells incubated in the presence of mitochondrial Ca^{2+} inhibitors antimycin A and oligomycin were still sensitive to $InsP_3$, but cells pre-treated with vanadate (which inhibits the Ca^{2+} uptake in the non-mitochondrial pool) did not respond to $InsP_3$. Although this did not clarify which of the non-mitochondrial pools was sensitive to $InsP_3$, it was clear that Ca^{2+} was not released from the mitochondrial store.

Although these experiments were important to prove $InsP_3$ mediated-Ca^{2+} release, the key experiment was the one that proved $InsP_3$ to be a second messenger. Permeabilised cells were treated with carbachol, a compound known to mobilise intracellular Ca^{2+} by binding to external cell-membrane receptors, and $InsP_3$, at different concentrations and in different sequence. The sum of the Ca^{2+} released by carbachol and $InsP_3$ was constant, and in the presence of saturating concentrations of exogenous $InsP_3$ carbachol could no longer release Ca^{2+}, indicating that both the compounds were acting on the same pool of releasable Ca^{2+}. This also indicated that carbachol-induced Ca^{2+} release was mediated by $InsP_3$.

Another important experiment was to study the specificity of the Ca^{2+}-releasing response by testing the effect of *myo*-inositol 1,4-bisphosphate [Ins(1,4)P_2], inositol 1,2-cyclic phosphate (cIMP) and *myo*-inositol; these compounds did not release Ca^{2+}; moreover, when $InsP_3$ was hydrolysed at 100 °C for 30 minutes in the presence of 5 M hydrochloric acid (conditions which randomised the phosphates by bond migration)[15] there was a 50% reduction of Ca^{2+}-release activity, confirming the high specificity in the structure of $InsP_3$.

The evidence of $InsP_3$ being responsible of Ca^{2+} mobilisation increased the interest in Ca^{2+} signalling and inositol chemistry and a flood of subsequent reports extended and consolidated the status of $InsP_3$ as a second messenger.[16]

1.2 Inositols and inositol phosphates - structure, nomenclature and natural occurrence

myo-Inositol **1** represents one of nine possible stereoisomers of hexahydroxy cyclohexane (Figure 1.2).

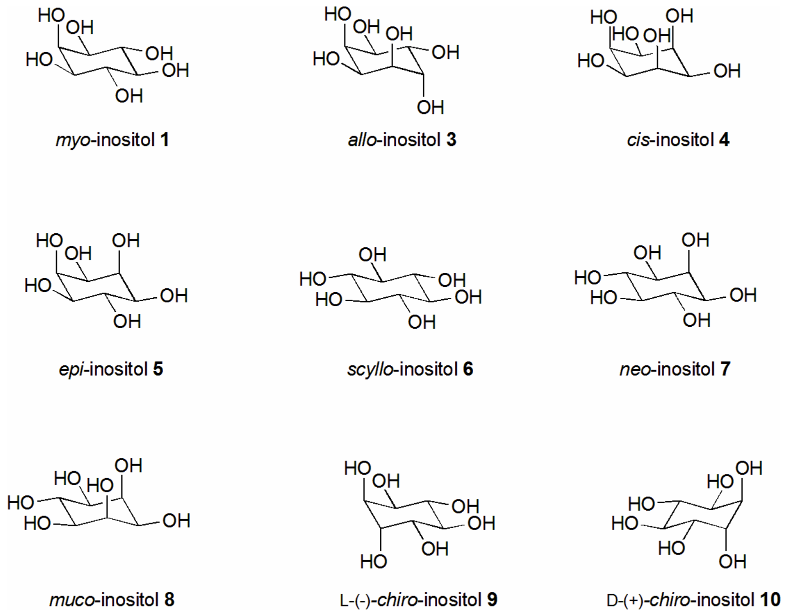


Figure 1.2. The nine isomers of inositol.

The stereoisomers *myo*-inositol **1**, *allo*-inositol **3**, *cis*-inositol **4**, *epi*-inositol **5**, *scyllo*-inositol **6**, *neo*-inositol **7**, *muco*-inositol **8**, (Figure 1.2) contain internal elements of symmetry are therefore optically inactive. The two stereoisomers L-(-)-*chiro*-inositol **9** and D-(+)-*chiro*-inositol **10** are unsymmetrical and form an enantiomeric pair. *myo*-Inositol **1** is a *meso* compound and is the most naturally abundant stereoisomer of the possible nine isomers; for this reason it is generally accepted that the term "inositol" without a prefix refers to *myo*-inositol **1**, whereas the term "inositols" refers to all the nine stereoisomers. The stereoisomer D-(+)-*chiro*-inositol **10** is found in some biological molecules and small quantities of *scyllo*-inositol **6** and *neo*-inositol **7** are present in neuronal tissues.[17,18] Due to the highly symmetric nature of *myo*-inositol **1** and its stereoisomers, there has been much confusion in the scientific literature surrounding inositol phosphates, complicated to the initial strict adherence to the IUPAC rules, in that the addition or removal of a phosphate group would necessitate a swap between the D- and the L- numbering system. In order to circumvent the confusions, Agranoff's turtle[19] has been used (Figure 1.3).

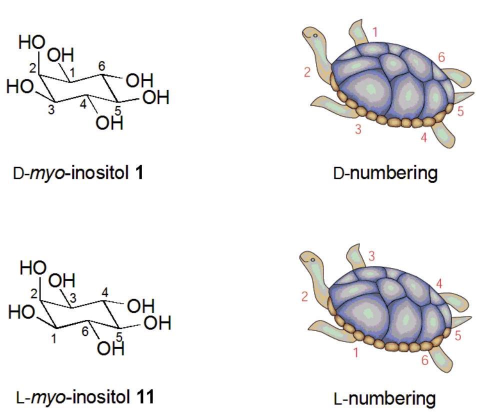


Figure 1.3. Agranoff's turtle rules for numbering inositols (picture taken from Irvine and Schell, 2001).[20]

myo-Inositol **1** is represented in its more thermodynamically stable chair conformation; the head of the turtle resemble the axial hydroxyl group of *myo*-inositol, defined as the 2-position. The D-ring numbering is assigned by using the right front limb of the turtle to define the D-1-position on the *myo*-inositol ring; continuing anticlockwise the left front limb becomes the D-3-position, and so on. In a similar way, the L-ring numbering is assigned by defining as L-1 the left front limb of the turtle, L-2 the head and then proceeding clockwise (Figure 1.3).[21]

1.3 Ca^{2+} signalling and $InsP_3$ intracellular cascade

The first evidence for Ca^{2+} as active compound in the cell goes back to 1883, when Ringer[22] discovered that Ca^{2+} salts were needed in order to allow the contraction of isolated rat hearts. Despite the importance of the discovery, it did not attract particular attention, until the end of the 1950s, when two important discoveries were made: the demonstration by Weber[23] that the binding of Ca^{2+} to myofibrils activated actomyosin; and the finding in the laboratories of Ebashi and Lipmann[24,25] and Hasselbach and Makinose[26] that isolated sarcoplasmic reticulum vesicles accumulated Ca^{2+} by using an ATP-energised system. Thanks to these early discoveries the interest in the signalling role of Ca^{2+} rapidly increased. Today the importance of Ca^{2+} as intracellular messenger is well established.[27,28] Ca^{2+} is responsible for controlling a wide variety of cellular and physiological processes as diverse as cell division and proliferation, apoptosis, fertilisation, gene transcription and muscle contraction. At a very basic level, Ca^{2+} exerts its action when its basal concentration of 100 nM raises to 1000 nM. The versatility arises from the use of an extensive molecular set of components that constitute a so-called Ca^{2+} toolkit. Such a system is structured in order to create Ca^{2+} signals with different spatial and temporal profiles, which activate and regulate many different cellular responses.[28]

The intracellular concentration of Ca^{2+} is elevated in two ways. Either by influx of external Ca^{2+} through transmembrane ion channels, or by release of Ca^{2+} from intracellular stores, subsequent to the activation of ligand gated ion channels. Two components of the Ca^{2+} toolkit, $InsP_3$ and cyclic adenosine diphosphate ribose (cADPR), activate the $InsP_3$ receptors ($InsP_3Rs$)[14] and the ryanodine receptors (RyRs),[29] respectively, releasing Ca^{2+} from the endoplasmic reticulum (ER) or sarcoplasmic reticulum (SR). Other components of the Ca^{2+} toolkit that can release Ca^{2+} from internal stores include nicotinic acid adenine dinucleotide phosphate (NAADP), that may operate by activating a channel on a lysosome-related organelle and sphingosine 1-phosphate (S1P), which is thought to release Ca^{2+} through a pathway that is independent of $InsP_3Rs$ and RYRs.[28]

The intracellular release of Ca^{2+} stimulates a number of Ca^{2+}-dependent events controlled by the variation in the temporal and spatial aspects of the Ca^{2+} signal. The variability of these signals depends on different degrees of excitability of the $InsP_3Rs$ and RYRs, controlled by different levels of the appropriate Ca^{2+}-mobilising messenger. Weak stimulation of $InsP_3Rs$ leads to individual channels opening to give Ca^{2+} blips, where higher levels of stimulation give Ca^{2+} puffs.[28] For the RYRs, a weak stimulation produces Ca^{2+} quarks and higher stimulation gives Ca^{2+} sparks.[27,28] When most of the $InsP_3Rs$ and RYRs are sufficiently sensitive to Ca^{2+}, the Ca^{2+} puffs and sparks can excite neighbouring receptors through Ca^{2+}-induced Ca^{2+} release, leading to an intracellular Ca^{2+} wave. These events can trigger and coordinate different events within the cytosol, such as activation of Ca^{2+}-dependent proteins including calmodulin, alteration of the levels of nitric oxide (NO) and adenosine cyclic 3',5'-monophosphate (cAMP), or can transduce the signal to an adjacent cell through gap junctions.[28] Once Ca^{2+} has completed its signalling functions, a mechanism consisting of pumps and exchangers, brings the intracellular Ca^{2+} levels back to the basal concentration.

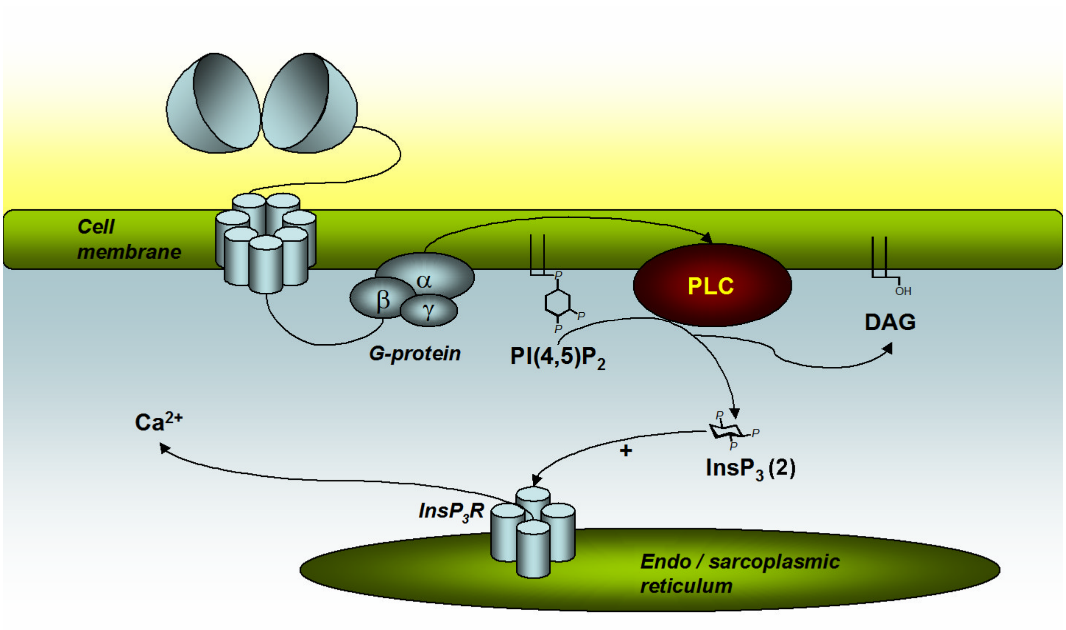


Figure 1.4. Schematic representation of the $InsP_3$ signalling cascade.

$InsP_3$ (**2**, Figure 1.4) is generated by an hydrolytic enzyme, phospholipase C (PLC) from the lipid membrane precursor phosphatidylinositol 4,5-bisphosphate [PI(4,5)P_2]. The several known PLC isoforms are activated by different mechanisms, such as tyrosine kinase-coupled receptors (that activates PLCγ); an increase in Ca^{2+} levels (which activates PLCδ); activation through the RAS gene (PLCε); and G-protein-coupled receptors (GPCR), that activate PLC_β. External signals such as extracellular growth factors, hormones or neurotransmitters arriving at the cell surface engage GPCRs, that are membrane spanning proteins, and activate the G-proteins they are coupled to upon the external agonist binding. The G-proteins are intracellular signal transducers proteins that activate PLC_β through an energy-requiring [guanosine 5'-trisphosphate (GTP) or adenosine 5'-trisphosphate (ATP)] mechanism. PLC_β hydrolyses PI(4,5)P_2 to give DAG and $InsP_3$. The lipophilic DAG remains in the plane of cell membrane and effects signal transduction by activation of PKC. $InsP_3$, which is hydrophilic, diffuses into the cytosol and activates $InsP_3$Rs. The binding of $InsP_3$ to $InsP_3$Rs causes the channel to open releasing Ca^{2+} into the cytosol from a distinct store within the ER.

1.4 $InsP_3$ Receptors

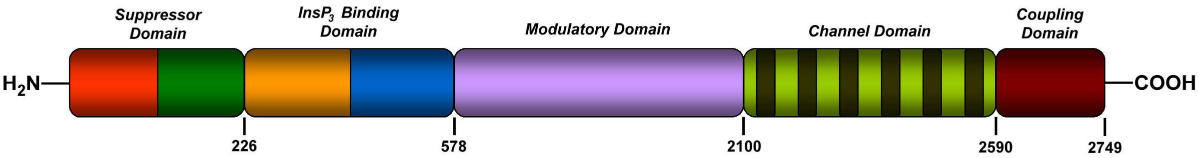


Figure **1.5**. Structure of the $InsP_3R$ type 1 (one of the four subunits in shown). The protein is constituted of 2749 amino acid residues and is divided in five functional subunits (from the *N*-terminal): the suppressor domain; the $InsP_3$ binding domain; the central modulatory region; the channel domain and the coupling domain.[30]

The $InsP_3Rs$ are present in a wide range of organisms including humans, and regulate the level of cytosolic Ca^{2+} (the other major intracellular Ca^{2+} channels are the RYRs). These receptors are situated on the ER (or on the SR in muscle cells) and have been identified in three isoforms.[31] These isoforms possess high sequence homology (60-70% of amino acid residues are conserved in the three receptor subtypes), but differ in their Ca^{2+} dependence, $InsP_3$ affinity and subcellular distributions. The isoforms are also differentially expressed in certain cell types. The $InsP_3R$ type 1 ($InsP_3R1$) is highly expressed in the central nervous system (CNS), especially the cerebellum, with the same cerebellar location in three mammalian species (rat, mouse and hamster).[31] The $InsP_3R$ type 2 ($InsP_3R2$) is present in many tissues with particularly high levels found in the spinal cord and glial cells. The $InsP_3R$ type 3 ($InsP_3R3$) is found in the kidney, brain, gastrointestinal tract and pancreatic islets.[31] The differences in the homology and tissue distribution suggest that each receptor subtype has distinct cellular roles and is possible that interplay between isoforms may be necessary for a cell to control spatial and temporal aspect of Ca^{2+} signalling.[31]

The $InsP_3R1$ is formed of four large subunits; each subunit consists of 2749 amino acid residues (313 kDa) and is divided in five functionally distinct regions (from the *N*-terminal of the polypeptide chain, Figure 1.5): the $InsP_3R$ suppressor domain; the $InsP_3$ binding domain; the central modulatory region; the *C*-terminal channel domain and the coupling regions.[30,32-34] The recent studies of Bosanac[30,32] revealed the molecular architecture of the *N*-terminal region of the $InsP_3R1$, by the elucidation of the crystal structures of both the $InsP_3R$ suppressor domain[32] and the $InsP_3$ binding domain (the latter in complex with $InsP_3$).[30] The $InsP_3R$ suppressor domain is a peptide formed of 223 amino acids (residues 1-223), with a shape resembling a hammer (Figure 1.6). It consists of two subdomains: a head subdomain forming a β-trefoil fold; and an arm subdomain that extrudes away from the β-trefoil structure and features a helix-turn-helix structure (Figure 1.6).[32]

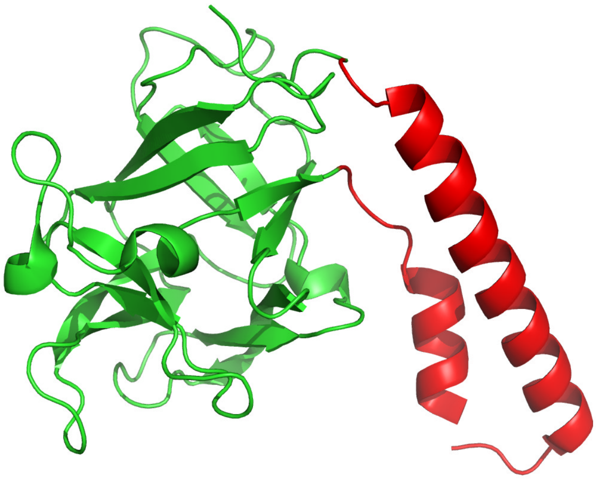


Figure **1.6**. A PyMOL (www.pymol.org) representation of the X-ray crystal structure of the $InsP_3$ suppressor domain of the mouse $InsP_3R1$ (Head-domain in green, Arm-domain in red).[32]

Immediately adjacent to the $InsP_3R$ suppressor domain is the $InsP_3$ binding domain, formed of 381 amino acids (residues 224-604) and consisting of two subdomains forming a cleft in which $InsP_3$ binds, the α-domain containing an "armadillo repeat"-like fold and the β-domain containing the β-trefoil fold (Figure 1.7).[30]

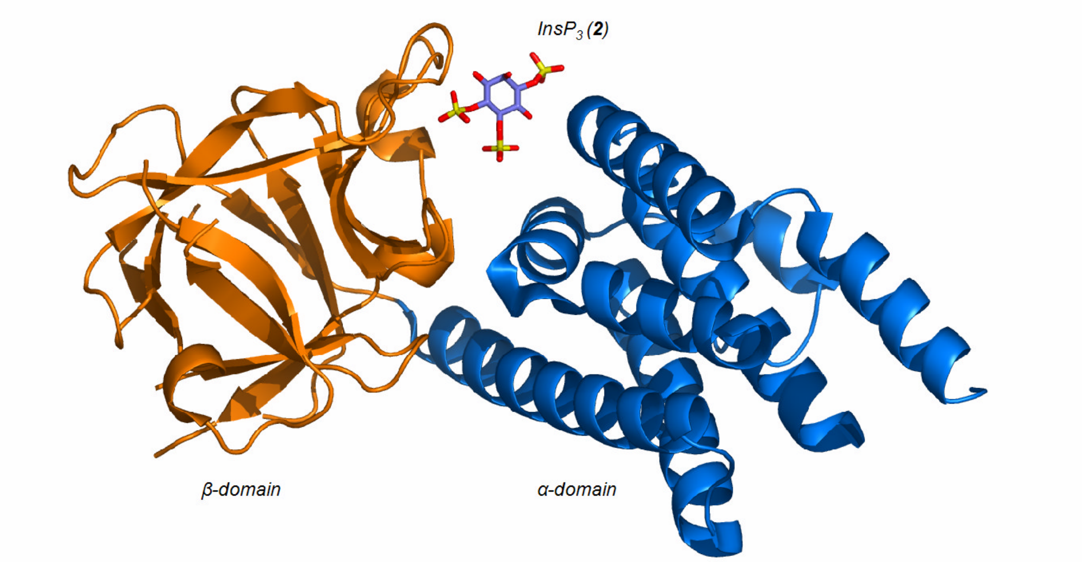


Figure **1.7**. A PyMOL (www.pymol.org) representation of the X-ray crystal structure of the ligand-binding domain of the mouse $InsP_3R1$ with $InsP_3$ (**2**) at the binding site (α-domain in blue, β-domain in orange).[30]

The central modulatory region that separates the channel domain from the $InsP_3$ binding domain is formed of almost 1600 amino acid residues and has been described as the modulatory domain.[31,33] This contains many sites that are thought to regulate the behaviour of the $InsP_3Rs$, including phosphorylation sites (serine amino acid residues) and binding sites for ATP, Ca^{2+} and regulatory proteins [calmodulin, immunophilin FK506-binding protein FBKP)].[31,35] The interactions of these endogenous regulators with the $InsP_3Rs$ govern the pattern of Ca^{2+} release in a manner that allows the fine tuning Ca^{2+} signals in the cellular environment. The channel domain contains the amino acid residues that form the six transmembrane segments channel of the $InsP_3Rs$. The coupling domain is involved in the assembly of the $InsP_3R$ in the tetrameric form and its targeting to the ER.

The elucidation of both the $InsP_3R$ suppressor domain and the $InsP_3$ binding domain (the latter in complex with $InsP_3$),[30,32] together with electron microscopy analysis of isolated $InsP_3Rs$ particles[33] and bio-physiological studies on $InsP_3Rs$,[34] have provided some basis for the understanding of the mechanism by which $InsP_3$ effects the release of Ca^{2+} from the $InsP_3Rs$, although unambiguous evidence is still needed.

The $InsP_3$-induced Ca^{2+} release by $InsP_3$ is positively cooperative,[36,37] suggesting that more than one of the four subunits of the $InsP_3R$ must bind to $InsP_3$ in order to open the channel. There is also evidence that the $InsP_3Rs$ respond to different Ca^{2+} levels,[37,38] suggesting that Ca^{2+} performs as a co-agonist at the $InsP_3Rs$ together with $InsP_3$.[37] The binding of $InsP_3$ seems to inhibit the binding of Ca^{2+} to an inhibitory site and to promote the binding of Ca^{2+} to a stimulatory site, promoting channel opening. Gel filtration experiments on the $InsP_3R1$ showed that a large decrease in the Stoke's radius of the cytosolic portion of the receptor occurs upon the $InsP_3$ binding, suggesting that the activation of the receptor is associated with a large conformational change within the tertiary structure of the protein.[34] Further support to this hypothesis comes from electron cryomicroscopy images[39-41] of the whole $InsP_3R1$ from cerebellum using single-particle analysis. Hamada[39] demonstrated that Ca^{2+} binding induces a conformational change in the tetrameric receptor from the closed state to the open state. $InsP_3$ binds in the cleft formed by the α- and the β-domains in the $InsP_3$ binding domain (Figure 1.7) and in this process it is thought to bring the two domains together. The small modification in the relative positions of the two domains would lead to a much larger conformational change in the $InsP_3R$ with the final effect of opening the channel. The suppressor domain is thought to modulate $InsP_3$ affinity by masking the $InsP_3$ binding site at the binding domain in a manner that $InsP_3$ cannot approach the cleft between the α- and the β-domains. This assumption is supported by site-directed mutagenesis experiments, which identified a number of surface amino acid residues likely to be involved in intramolecular interaction with the $InsP_3$ binding domain and therefore in the $InsP_3$-suppression mechanism.[32] As mentioned above, Ca^{2+} actively participates in receptor activation, but it is not clear where the Ca^{2+} sites are located. It has been recently proposed that the Ca^{2+} binding sites could be positioned on both the $InsP_3$ suppressor domain and the $InsP_3$ binding domain.[32,33,42] It is also known that the $InsP_3$ suppressor domain binds a number of cellular proteins, such as calmodulin, which modulate the activity of the receptor[43] acting like binding partners, therefore these proteins could represent at least part of the Ca^{2+} binding sites.[33] These results indicate that an interplay between the $InsP_3$ suppressor domain and other cellular binding partners could be operating to regulate the $InsP_3$R functions.

1.5 $InsP_3$ receptor agonists

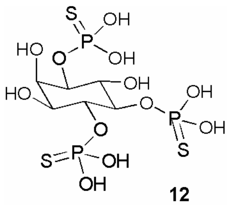
Prior to the discovery of $InsP_3$ acting as a second messenger and mobilising internal Ca^{2+} stores,[14] many inositol phosphates had already been synthesised and there are a number of reviews[44,45] and books[17,46] describing this synthetic work. The findings of Berridge and co-workers[14] considerably increased the interest in the biological investigation of inositol phosphates and many efforts were made towards the synthesis of unnatural $InsP_3$ analogues, in order to establish the key structural requirements for a compound to act as an $InsP_3$R agonist and define a structure-activity relationship profile of $InsP_3$.

In 1986 Ozaki and co-workers[47] reported the first total synthesis of optically pure $InsP_3$. Almost immediately a number of phosphorothioate analogues of $InsP_3$ were synthesised, in which one or more phosphate groups are replaced with the bioisosteric phosphorothioate groups.[48] In 1993 Takahashi and co-workers[49] isolated from *Penicillium brevicompactum* compounds with a chemical structure resembling $InsP_3$, the adenophostins, that showed a Ca^{2+}-mobilising activity higher than $InsP_3$. These compounds were fundamental in the basic understanding of $InsP_3$Rs and related metabolic pathways.

Soon after the first synthesis of $InsP_3$ analogues were completed, it was clear the need of a method for delivering such highly polar compounds into the cell, as the only methods known to test $InsP_3$ and analogues activity was to use detergents to permeabilise the cell membrane or abruptly inject the compounds inside the cell. Following the efforts of some research groups, membrane-permeant analogues of $InsP_3$ were synthesised.[50]

1.5.1. Phosphorothioate analogues of $InsP_3$

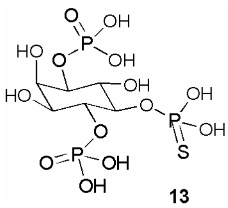
In 1987 Potter and co-workers reported the synthesis of DL-*myo*-inositol 1,4,5-trisphosphorothioate **12** ($InsP_3S_3$) (Figure 1.8).[51] This $InsP_3$ analogue binds with high affinity to the $InsP_3$Rs and is a potent Ca^{2+} mobilising agonist, with a potency approximately 3-4 times less than $InsP_3$.[52,53] $InsP_3S_3$ is not hydrolysed by the 5-phosphatase, displaying in fact increased inhibition of the enzyme with respect to $InsP_3$, with a K_i = 1.7 μM for the D- enantiomer (D-$InsP_3S_3$) and a K_i = 0.50 μM for the L- enantiomer (L-$InsP_3S_3$) *versus* the K_i = 40 μM for $InsP_3$.[54] Remarkably, L-$InsP_3S_3$ has been found to bind to the 3-kinase enzyme, where D-$InsP_3S_3$ is not a substrate for this enzyme.[54] As a result of these properties and despite the fact that L-$InsP_3S_3$ possesses no Ca^{2+}-mobilising activity, $InsP_3S_3$ is able to produce a sustained Ca^{2+} release.[55]



12

Figure 1.8. Structure of DL-$InsP_3S_3$ (**12**).

DL-*myo*-Inositol 1,4-bisphosphate-5-phosphorothioate **13** [Ins(1,4)$P_2$5PS] (Figure 1.9) was synthesised as a racemic mixture in order to investigate whether the substitution of the *C*-5 position phosphate group with a phosphorothioate group would generate a compound as potent as $InsP_3$ but with increased metabolic stability.[56] Despite the fact that Ins(1,4)$P_2$5PS is a full agonist at the $InsP_3$Rs, the affinity for the receptor is 7-fold lower that $InsP_3$ indicating that 4,5-bisphosphate groups of $InsP_3$ are crucial for the affinity. Ins(1,4)$P_2$5PS is a potent inhibitor of the 5-phosphatase and therefore can produce a sustained Ca^{2+} release.[56]

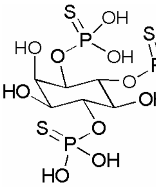


13

Figure 1.9. Structure of DL-Ins(1,4)$P_2$5PS (**13**).

DL-*myo*-Inositol 1,4,6-phosphorothioate **14** [Ins(1,4,6)PS_3] represents a regioisomer of $InsP_3S_3$ and contains the 1,6-bisphosphorothioate groups resembling the 4,5-bisphosphate moieties of $InsP_3$ (Figure 1.10). Ins(1,4,6)PS_3 is a partial agonist at the

$InsP_3Rs$ and shows a low Ca^{2+}-mobilising activity [the D- enantiomer (D-Ins(1,4,6)PS_3) is thought to be the active species in the racemic mixture]. This result suggests that the $InsP_3Rs$ allow a certain degree of tolerance in the distribution of the phosphate groups around the inositol ring, being the receptor able to bind to non-1,4,5-substituted $InsP_3$ analogues.



14

Figure 1.10. Structure of Ins(1,4,6)PS_3 (**14**).

The compound DL-*myo*-inositol 1,3,4-phosphorothioate **15** [Ins(1,3,4)PS_3] (Figure 1.11) displays a Ca^{2+}-mobilising activity similar to Ins(1,4,6)PS_3.[57] The enantiomer L-Ins(1,3,4)PS_3 present in the racemate is thought to be responsible for the activity at the $InsP_3Rs$. This compound can also be called D-*myo*-inositol 1,3,6-phosphorothioate **16** [D-Ins(1,3,6)PS_3] (Figure 1.11) using the D- numbering and is clearly similar to D-Ins(1,4,6)PS_3. The activity of Ins(1,3,4)PS_3 as a partial $InsP_3Rs$ agonist further supports the suggestion that the $InsP_3Rs$ can bind to a variety of $InsP_3$ analogues.[57]

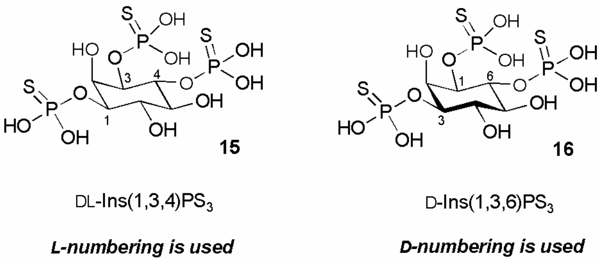
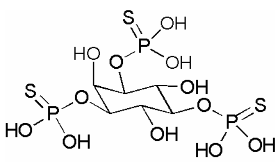


Figure 1.11. Structures of DL-Ins(1,3,4)PS_3 (**15**) and D-Ins(1,3,6)PS_3 (**16**).

myo-Inositol 1,3,5-trisphosphorothioate **17** [Ins(1,3,5)PS_3] is a *meso* compound (Figure 1.12), which inhibits the 5-phosphatase enzyme with a K_i = 0.43 μM and does not release Ca^{2+} from the $InsP_3Rs$.[54] This compound confirms the importance of the 4,5-bisphosphate moiety as a key structural requirement for the activity at the $InsP_3Rs$.



17

Figure 1.12. Structure of Ins(1,3,5)PS_3 (**17**).

D-*myo*-Inositol 1-phosphorothioate 4,5-bisphosphate **18** [Ins1PS(4,5)P_2], synthesised as the optically pure enantiomer (Figure 1.13), is a potent Ca^{2+} mobilising agonist, indicating that the *C*-1 position phosphate group can tolerate conservative substitutions. This compound has been successfully used for the synthesis of a photoaffinity analogue of $InsP_3$ (**19**, Figure 1.13).[58] Such a compound possesses a similar activity as $InsP_3$ to the $InsP_3Rs$, and contains a fluorescent tag connected to the *C*-1 position phosphorothioate group *via* the sulfur atom. Using this compound it has been possible to label the $InsP_3$ binding site of the $InsP_3R$.[58,59] This compound has also been used for the preparation of an affinity matrix, which provides a useful tool for the purification of $InsP_3Rs$.[60,61]

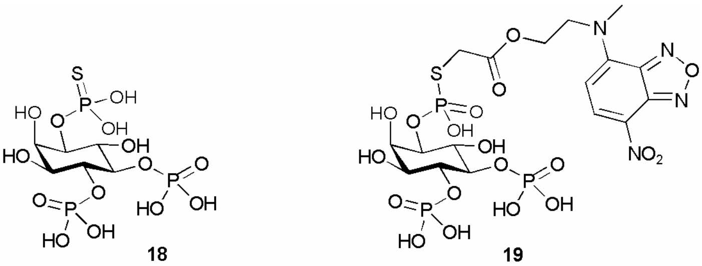


Figure 1.13. Structure of Ins1PS(4,5)P_2 (**18**) and the photoaffinity $InsP_3$ analogue **19**.

1.5.2. The adenophostins

Adenophostins A (**20**) and B (**21**) (Figure 1.14) were isolated from *Penicillium brevicompactum*[49] and have been shown to be full agonists with affinities for $InsP_3Rs$ that are 10-100 fold greater than $InsP_3$.[62-64]

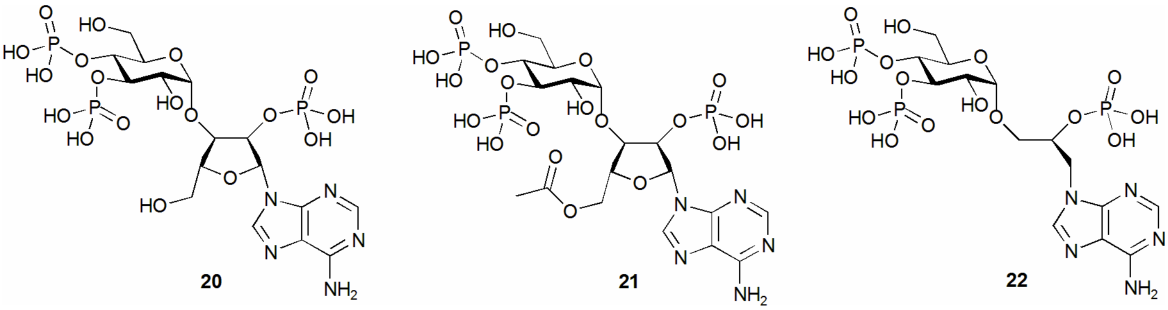


Figure 1.14. Structures of Adenophostin A (**20**), adenophostin B (**21**), acyclophostin (**22**).

The adenophostins resemble $InsP_3$ in that the *trans* diequatorial bisphosphate arrangement flanked by a hydroxyl group, has been identified as a key feature of the adenophostin and contributes to its high affinity for the $InsP_3Rs$. Therefore all synthetic adenophostins analogues, to date, have this arrangement conserved. Attempts to determine which of the remaining structural features of the adenophostins are responsible for their high affinity interactions with $InsP_3Rs$ have resulted in the synthesis and biological evaluation of several related compounds.[63-66]

To date, only the compound acyclophostin (**22**, Figure 1.14)[67] has shown similar activity.[68] These studies showed that the α-D-glucopyranose structure is a good bioisoster of the *myo*-inositol backbone of $InsP_3$ and that the three-dimensional arrangement of the three phosphate groups of adenophostin and its analogues is essential for biological activity. Furthermore, the adenine moiety is able to enhance the activity. Because of the three additional hydrogen-bonding sites on the adenine ring, the high potency of interaction between adenophostin and the $InsP_3R$ that is observed could be explained by the formation of additional hydrogen bonds with respect to $InsP_3$. In order to elucidate the role of the adenine moiety, a number of adenophostin analogues in which the adenine is replaced by different moieties have been synthesised.[69] Since the synthesis of adenophostins and their analogues are more simple than that of optically active $InsP_3$ derivatives, adenophostins provide an alternative approach to develop high-affinity selective ligands for $InsP_3Rs$.

1.5.3. Membrane-permeant analogues of $InsP_3$

The ionic and high polar nature of $InsP_3$ limits its membrane permeability. Disruptive techniques such as microinjection, electroporation and permeabilisation with saponins are required for delivering $InsP_3$ into the cell. Thus, membrane-permeant derivatives of $InsP_3$ would be useful tools for the pharmacological studies of $InsP_3$ and analogues. In order to neutralise the charge present on the phosphate groups of $InsP_3$, the phosphates groups should be protected with moieties that render the whole molecule lipophilic and able to cross the cell membrane. Once the compound has crossed the membrane and is included in the cytosol, the masking groups should be removed by a cytosolic metabolising system in order to restore the phosphate moieties and therefore their biological activity. Various carbonyloxymethyl groups have been investigated by Tsien and co-workers as potential phosphate-masking groups.[50] The rationale behind this choice is that the ester moiety of the masking group could be hydrolysed by non-specific esterase enzymes once in the cytosol, leaving hydroxymethyl phosphate esters that decompose spontaneously to formaldehyde and the free phosphate group (Figure 1.15).

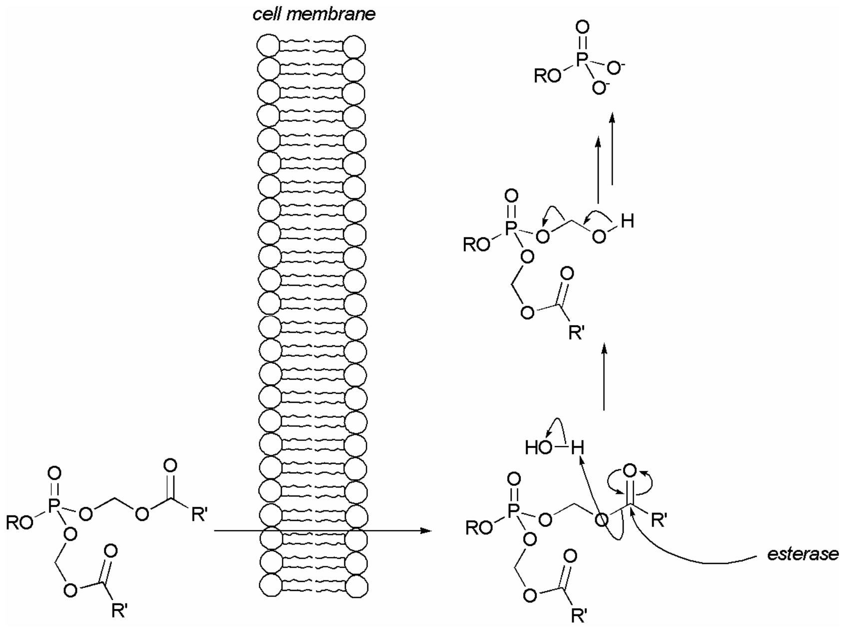


Figure 1.15. Carbonylmethoxy-phosphates are lipophilic and can diffuse across the cell membrane. Cytosolic esterases remove the ester protecting groups, leaving a hydroxymethyl phosphate esters that spontaneously decompose in the free phosphate groups losing formaldehyde.

The methylene linkers are used in order to remove the steric bulk around the ester moiety and therefore allow the access by non-specific esterases. The acetoxymethyl (AM), propionyloxymethyl (PM) and butyryloxymethyl (BM) groups were used to synthesise the corresponding $InsP_3$ derivatives, $InsP_3$/AM (**23**), $InsP_3$/PM (**24**) and $InsP_3$/BM (**25**) (Figure 1.16). These compounds were synthesised as racemic mixtures.[50]

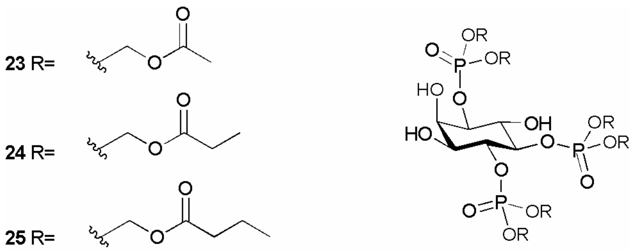


Figure 1.16. Membrane-permeant analogues of $InsP_3$.

The derivative $InsP_3$/AM **23** was not able to mobilise Ca^{2+} when the cells were equilibrated in an extracellular medium containing the compound, whilst microinjections of $InsP_3$/AM directly into the cell caused the release of Ca^{2+}, most likely through regeneration of $InsP_3$ mediated by the esterase enzymes.[50] To explain this experimental outcome it was postulated that the AM groups were not sufficiently lipophilic to allow compound $InsP_3$/AM to cross the cell membrane.

$InsP_3$/PM **24** was found to be active at an extracellular dosing of 20 μM, and $InsP_3$/BM **25** was active at an extracellular dosing of 2 μM. However, the time delay observed between dosing and Ca^{2+} release was 6 minutes for $InsP_3$/BM and between 60 and 100 seconds for $InsP_3$/PM. This result appears to be consistent with the increased steric bulk of the BM esters, which are less accessible to the esterases and therefore cleaved more slowly than the PM esters, less hindered and cleaved more rapidly.

The optically pure D- and L- enantiomers of $InsP_3$/BM have been synthesised by Holmes and co-workers,[70] confirming as expected that the enantiomer D-$InsP_3$/BM is responsible for the Ca^{2+}-mobilising ability of $InsP_3$/BM when applied to the extracellular medium; the enantiomer L-$InsP_3$/BM does show a little Ca^{2+}-mobilising activity, which has been attributed to intracellular migration of the phosphate groups. The racemic $InsP_3$ membrane-permeant derivatives, as well as the optically pure version, have been successfully used to study $InsP_3$Rs-related Ca^{2+} signalling.

1.6 $InsP_3$ antagonists

Despite the relative abundance of $InsP_3$Rs agonists with a binding affinity similar to $InsP_3$, only a few compounds have shown with antagonist activity at the $InsP_3$Rs. These compounds include heparin, xestospongin C, decavanadate, the antimalarial drugs chloroquine, quinine and quinidine, 2-APB and an $InsP_3$ *C*-5 phosphonate analogue.

1.6.1. Heparin

Heparin, a high molecular weight non-membrane-permeant polysulfated polyanion (Figure 1.17) known for its anticoagulant properties, is capable of inhibiting the $InsP_3$-induced Ca^{2+} release.

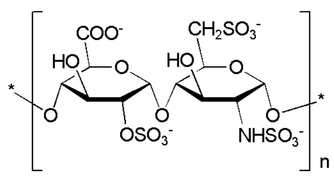
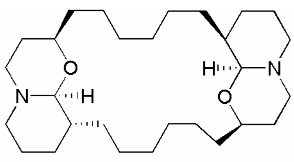


Figure 1.17. Structure of heparin**.**

The potent antagonist activity of heparin has been demonstrated to be competitive and fully reversible, with an affinity of heparin for the binding site of 3 nM.[71] The ability of heparin to bind the $InsP_3$Rs is different for each receptor, being greater for the $InsP_3$R3 than $InsP_3$R2 or $InsP_3$R1.[72] The density of negative charges, contributed by sulfate groups, appears to be important for the effect of heparin and the inhibition decreases dramatically as the size of the heparin chain is reduced below 18-24 monosaccaride units.[73] In addition to its potent competitive inhibition of the $InsP_3$Rs, heparin inhibits the coupling between plasma-membrane receptors and G-proteins,[74] the $InsP_3$ 3-kinase,[75] and stimulates the RYRs.[76] The lack of selectivity of heparin for the $InsP_3$Rs limits the usefulness of the anticoagulant in the study of $InsP_3$-mediated Ca^{2+} signalling in intact cells.

1.6.2. Xestospongin C

Xestospongins are bis-1-oxaquinolizidines isolated from the marine sponge *Xestospongia*.[77] These compounds are potent inhibitors of the $InsP_3$-mediated Ca^{2+} release, with the IC_{50} values ranging from 358 nM to 5.9 μM. As these compounds inhibit the $InsP_3$Rs in a manner that is independent of the concentration of $InsP_3$ and Ca^{2+}, it has not been possible to obtain indications about the nature of the binding site. The most potent compound, xestospongin C (**26**, Figure 1.18), is a membrane-permeant molecule and possess an IC_{50} = 358 nM for the $InsP_3$Rs;[77] it is also able to block the nitric oxide synthase,[78] to release Ca^{2+} from intracellular stores[79,80] and at higher concentrations it inhibits RyRs with a IC_{50} = 10 μM.[77]



26

Figure 1.18. Structure of Xestospongin C (**26**).

1.6.3. Chloroquine, quinine and quinidine

Chloroquine **27**, quinine **28** and quinidine **29** (Figure 1.19) are lipophilic, membrane-permeant antimalarial drugs used against *Plasmodium* parasites that have shown to inhibit the $InsP_3$-mediated Ca^{2+} release from the intracellular Ca^{2+} stores in macrophages.

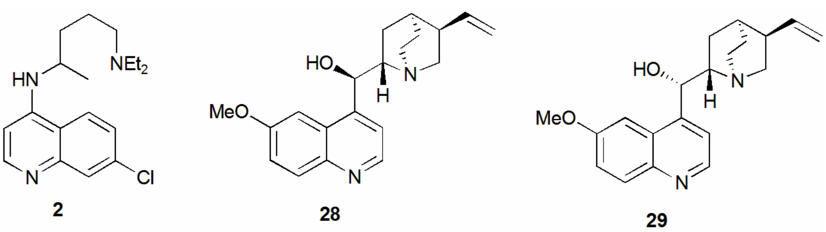


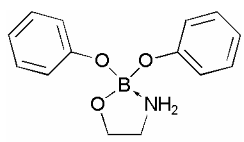
Figure 1.19. Structures of chloroquine (**27**), quinine (**28**) and quinidine (**29**).

Chloroquine blocks the release of Ca^{2+} by preventing the binding of $InsP_3$ to the $InsP_3Rs$, with an $IC_{50} = 10$ μM.[81] It is not clear whether these antimalarial compounds exert their action on the *Plasmodium* organisms by interacting with the Ca^{2+} signalling mechanism; however it has been shown that in permeabilised, isolated *Plasmodium chabaudi* parasites, chloroquine depletes $InsP_3$-sensitive Ca^{2+} stores, suggesting that Ca^{2+} signalling mechanism might be involved in the regulation of growth and differentiation of the parasites.[82] Other properties of these antimalarial compounds include the ability of blocking nicotinic cholinergic receptors at the neuromuscular junctions,[83,84] the alteration of glucose and insulin metabolism by blocking ATP-sensitive K^+ channels,[85,86] inhibition of subclasses of the cytochrome P450.[87] These additional biological properties of the antimalarial drugs chloroquine, quinine and quinidine clearly exclude the application of these compounds as $InsP_3Rs$ selective antagonists.

1.6.4. Decavanadate

Among different vanadium compounds, decavanadate $[(V_{10}O_{26})^{-6}$ at pH 7] inhibits $InsP_3$-mediated Ca^{2+} release by preventing the binding of $InsP_3$ to the $InsP_3Rs$.[88] It has been suggested that the inhibitory activity of decavanadate is due to its ability of bridging the multiple $InsP_3$ binding sites, as oligovanadate and monovanadate, two other vanadium compounds that do not possess this bridging ability, are not $InsP_3Rs$ inhibitors.[36,89] Decavanadate is also able to inhibit the $InsP_3$ 5-phosphatase and the 3-kinase;[90] this low specificity is prevents decavanadate from being a useful tool to investigate $InsP_3$ signalling.

1.6.5. 2-Aminoethoxydiphenylborate



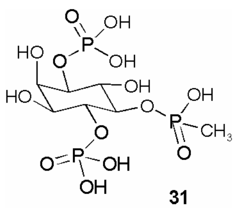
30

Figure 1.20. Structure of 2-aminoethoxydiphenylborate (**30**).

2-Aminoethoxydiphenylborate (2-APB) (**30**, Figure 1.20) is a membrane-permeant compound which inhibits $InsP_3$-mediated Ca^{2+} release with an IC_{50} of 42 μM and with a use-dependent action,[91,92] without affecting the binding of $InsP_3$ to the $InsP_3Rs$.[93] 2-APB also inhibits store-operated Ca^{2+} channels (SOC);[94] this action is not due to the action of 2-APB on the $InsP_3Rs$, as it occurs in cells that do not express the $InsP_3Rs$.[95] It is not clear whether 2-APB interacts directly with the $InsP_3Rs$ as it was originally proposed by Maruyama and co-workers;[91] 2-APB could bind directly to a SOC or a SOC-associated regulatory protein,[95,96] or with a protein promoting or regulating the coupling between the $InsP_3Rs$ and SOC.[97] Furthermore, when applied to the extracellular medium 2-APB is more effective for inhibiting SOC than its intracellular application,[98] suggesting that an extracellular site might be needed in mediating the 2-APB inhibitory action on SOC.[96]

Unlike other $InsP_3Rs$ inhibitors, 2-APB is fairly specific, in the sense that several other Ca^{2+} channels like the RyRs and voltage-operated Ca^{2+} channels are not affected, at least at the concentrations used to inhibit the $InsP_3$-mediated Ca^{2+} release.[91] However, 2-APB is clearly not specific for the $InsP_3Rs$; in some cells types the inhibition of the $InsP_3$-mediated Ca^{2+} release in not observed,[94] and 2-APB has been shown to inhibit sarco-endoplasmic reticulum Ca^{2+} ATPases (SERCAs), leading to gradual Ca^{2+} depletion from the stores.[99] 2-APB also acts as a strong activator of the transient receptor potential vanilloid cation channels (TRPV) type 1 (TRPV1), type 2 (TRPV1), and type 3 (TRPV3).[100]

1.6.6. *C*-5 position methyl phosphonate analogue of $InsP_3$



31

Figure 1.21. Structure the *C*-5 position methyl phosphonate $InsP_3$ analogue (**31**).[101,102]

The compound shown in Figure 1.21, an analogue of $InsP_3$ in which the phosphate group at the *C*-5 position is replaced by a methyl phosphonate, was synthesised as a racemic mixture and displayed a weak activity as inhibitor of the Ca^{2+}.[101,102] This compound could exert its activity by binding to the $InsP_3Rs$ at the same site of $InsP_3$, and the reduced hydrogen-bonding capabilities of the *C*-5 phosphonate moiety could prevent the receptor from undergoing the conformational change thought to be essential for opening the channel and releasing Ca^{2+}. Compound **31** has been only tested towards the inhibition of the release of Ca^{2+}, therefore further studies are necessary to elucidate whether its activity is linked to the inhibition of the $InsP_3Rs$.

1.7 Summary

Since Scherer isolated *myo*-inositol,[1] many efforts have been made towards the understanding of the intimate roles and functions of inositol phosphates in the cellular environment. The discovery by Berridge and co-workers that $InsP_3$ releases Ca^{2+} from intracellular stores increased enormously the interest in the field of Ca^{2+} signalling.[14] The Ca^{2+} signals that $InsP_3$ generates by activating the $InsP_3Rs$ have been shown to be highly organised in spatial and temporal manner, allowing a fine control of the intracellular effects of Ca^{2+}.[28] The action of $InsP_3$ on the $InsP_3Rs$ is modulated by intracellular effectors including ATP, Ca^{2+}, phosphorylating enzymes and regulatory proteins such as calmodulin and FKBP.[31]

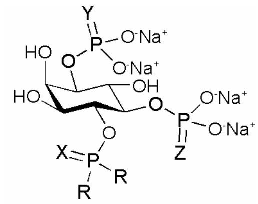
The investigation of $InsP_3$ agonists such as the $InsP_3$ phosphorothioate analogues and the natural products adenophostin A and B allowed researchers to establish the structural requirement for a compound to bind and activate the $InsP_3Rs$.[48] These compounds have found useful applications in the Ca^{2+} signalling field, as well as their membrane permeant analogues, which removed the need of injecting the compound into the cytosol.[50] Although these molecules have provided useful information about structure-activity relationships of $InsP_3$, thus far a compound able to selectively bind and block the $InsP_3Rs$ is still missing. A number of compounds acting as non-specific $InsP_3Rs$ antagonists have been described; the anticoagulant compound heparin, the natural product xestospongin C, the antimalarials chloroquine, quinine and quinidine, the inorganic compound decavanadate and 2-APB have been shown to inhibit the Ca^{2+} release by blocking the $InsP_3Rs$ and also possess many other biological activities. Although 2-APB has found useful applications in a number of studies due to its permeability to the cell membrane and showing no activity at the RyRs and other Ca^{2+} channels, it interacts with other components of the Ca^{2+} toolkit and ion channels, therefore limiting its utility.

In 1991 van Boom and co-workers reported that a compound based on the $InsP_3$ structure was able to inhibit the Ca^{2+} release.[101,102] Although there is no evidence that the molecule interacts with the $InsP_3Rs$, its resemblance to $InsP_3$ suggests that the compound may bind to the $InsP_3$ binding site and disrupt some of the important interactions necessary for the receptor activation.

Result and Discussion (Part One)

2 Results and Discussion (part one)

2.1 Project Aims



32 R = Me; X, Y, Z = O or S
33 R = Alkyl; X, Y, Z = O or S

Figure 2.1. Structures of the proposed $InsP_3Rs$ antagonists.

This project aims to synthesise *C*-4 position-modified $InsP_3$ analogues that may behave as $InsP_3Rs$ antagonists. In Figure 2.1 are shown the general structures designed for such compounds. Analysis of the X-ray crystal structure[30] of $InsP_3R1$ binding domain complexed with $InsP_3$ provides indications of the structural requirements for a compound to bind to this receptor (Figure 2.2). This structure shows that $InsP_3$ (**2**) binds to the receptor in a cleft formed by two domains, named the α- and β- domains (Figure 2.2). In this cleft $InsP_3$ binds to a number of basic amino acid residues; the 1- position (P1) and 5- position (P5) phosphate groups interact predominantly with the α-domain (Figure 2.2, a, b), whereas the 4- position phosphate group (P4) binds mainly to the β-domain (Figure 2.2, c). P1 forms hydrogen-bonds (H-bonds) with residues *R568* and *K569* (cyan) on the α-domain (Figure 2.2, a). P5 forms H-bonds with the residues *R504, K508, R511* and *Y567* (lime), all on the α-domain (Figure 2.2, b). P4 forms H-bonds with the residues *T266, T267* and *G268* (violet) on the β-domain (Figure 2.2, c). In addition, the residues *R265* and *R269* (wheat) form H-bonds with both P4 and P5 (Figure 2.2, d). Gel filtration experiments on the $InsP_3R1$ showed that a large decrease in the Stoke's radius of the cytosolic portion of the receptor occurs upon the $InsP_3$ binding, suggesting that the activation of the receptor is associated with a large conformational change within the tertiary structure of the protein.[33,34] Although the ligand-free crystal structure of the $InsP_3R$ has not been reported and it is therefore not possible to define conclusively which residues move significantly on $InsP_3$ binding, it seems likely that the region that connects the α- and β- domains allows the two domains to move closer on $InsP_3$ binding and this is thought to evoke the conformational change which opens the channel and releases Ca^{2+}.

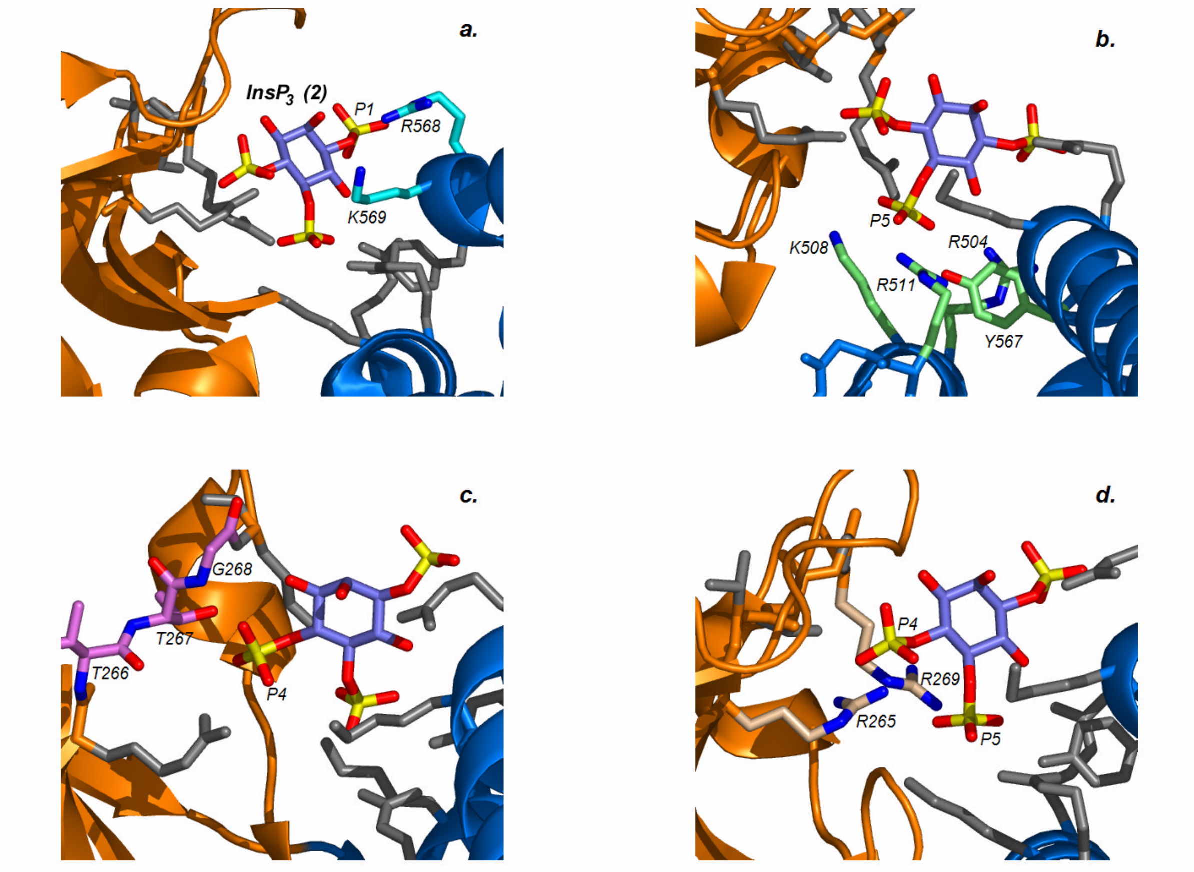


Figure 2.2. A PyMOL (www.pymol.org.) representations of the X-ray crystal structure of the ligand binding domain of the mouse $InsP_3R1$.[30] **a.** P1 forms H-bonds with residues *R568* and *K569* (cyan) on the α-domain (blue). P5 forms H-bonds with the residues *R504, K508, R511* and *Y567* (lime) on the α-domain (blue). P4 forms H-bonds with the residues *T266, T267* and *G268* (violet) on the β-domain (orange). Residues *R265* and *R269* (wheat) form H-bonds with both P4 and P5.

Consequently, any compound that binds to the $InsP_3Rs$ in the same or a similar place to $InsP_3$ but prevents the conformational change will behave as a competitive $InsP_3R$ antagonist.

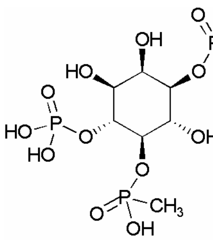
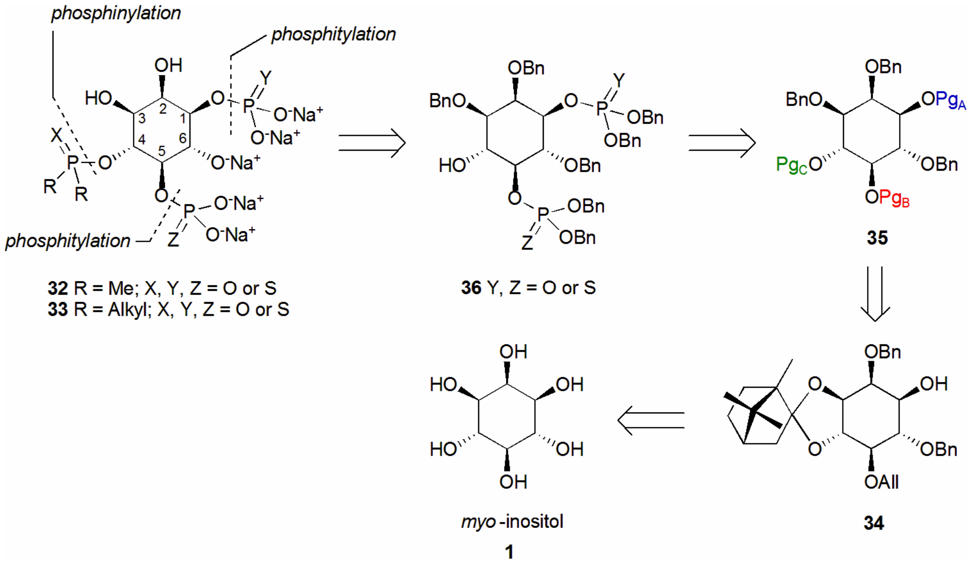

31

Figure 2.3. Structure of the *C*-5 methyl phosphonate $InsP_3$ analogue (**31**).[101,102]

This hypothesis may explain the Ca^{2+} release inhibitory activity of a 5-methyl phosphonate analogue of $InsP_3$ (**31**, Figure 2.3).[101,102] This compound is thought to operate by binding the $InsP_3R$ and partially disrupting the hydrogen-bond network required for activating the receptor because of the presence of the *C*-5 position methyl phosphonate moiety, which possess a different electronic distribution with respect to a phosphate group. If the hypothesis is correct, further modifications of the $InsP_3$ structure may lead to compounds that can selectively block the $InsP_3Rs$. Furthermore, considering that L-$InsP_3$ is not an agonist at the $InsP_3Rs$, the optimum potency for a potential antagonist could be achieved by synthesising the compound in the pure D-ring form.

In order to develop useful, potent and selective $InsP_3Rs$ antagonists a rational design approach based on the above hypothesis and the $InsP_3R1$ crystal structure has been adopted. Replacement of the P4 in the $InsP_3$ structure with non-hydrogen bonding moieties will allow investigations of the structural requirements for $InsP_3R$ antagonist activity. The initial modification in the $InsP_3$ structure will replace the P4 with either a dimethylphosphinyl or a dimethylphosphinothioyl moiety, to give the compounds shown in Figure 2.1. These moieties approximate the tetrahedral geometry of the phosphate group but will not form the same H-bonds as P4 with residues *R265, T266, T267, G268, R269* on the β-domain and *K569* on the α-domain (Figure 2.2, a, b, c, d). This modification will prevent the ability of this analogue to bring the α- and the β-domain together and consequently the receptor will not be activated. P1 and P5 initially will not be modified, in order to leave the hydrogen-bonding interactions with the residues *R568*, *K569*, *R504, K508, R511* and *Y567* (Figure 2.2, a, b) unaltered and maintain the affinity of the compound for the receptor. These alterations to the $InsP_3$ structure will furnish compounds that may be capable of being recognised by the $InsP_3Rs$ and therefore compete with the $InsP_3$ for binding. These compounds would bind to the α-domain but not to the β-domain, thus being unable to effect the conformational change in the $InsP_3Rs$ which is thought to open the channel and release Ca^{2+}.

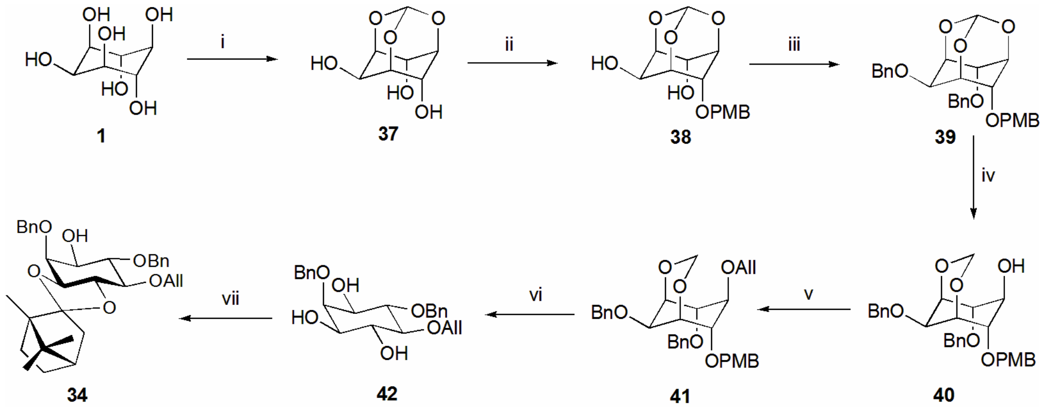
2.2 Retrosynthesis



Scheme 2.1. Proposed retrosynthesis of $InsP_3$ analogues, allowing modifications at the *C*-4.

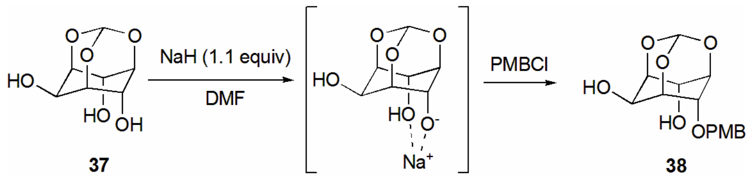
It was proposed that the synthesis of *C*-4 position-modified analogues of $InsP_3$ would be achieved as shown in the retrosynthetic analysis shown in Scheme 2.1. The orthogonally protected inositol intermediate **35** represents a versatile compound, as it could allow the synthesis of at least three classes of $InsP_3$ analogues, modified at the *C*-1, *C*-4 and *C*-5 positions. For the purpose of introducing modifications at the *C*-4 position of $InsP_3$, intermediate **36** was envisaged to be the suitable intermediate to synthesise. The target compound **32** could be prepared by phosphinylation of the alcohol **36** and subsequent hydrogenolysis of the benzyl groups. Removal of protecting groups Pg_A and Pg_B on intermediate **35**, followed by phosphitylation and oxidation of the resulting diol and deprotection of the group Pg_C should furnish alcohol **36**. Compound **35** could be synthesised from the camphor acetal **34** by protecting the *C*-1 hydroxyl group with the protecting group Pg_A, followed by cleavage of the camphor acetal auxiliary, selective benzyl protection of the *C*-3 hydroxyl group over the *C*-4 using the tin-acetal method previously reported by Gigg,[103] and protection of the *C*-4 hydroxyl group with protecting group Pg_C. The camphor acetal **34** could be prepared in seven steps from *myo*-inositol **1** as previously reported.[104,105]

2.3 Synthesis of the enantiopure camphor acetal 34



Scheme 2.2. Synthesis of the camphor acetal **34**. *Reagents and conditions:* **i.** $(EtO)_3CH$ (2.0 equiv), $TsOH{\cdot}H_2O$ (0.3 equiv), DMF, 100 °C, 77% yield. **ii.** NaH (1.1 equiv), PMBCl (1.1 equiv), TBAI (0.05 equiv), DMF, 0 °C to RT, 80% yield. **iii.** NaH (2.5 equiv), BnBr (2.5 equiv), DMF, 0 °C to RT, yield 100%. **iv.** DIBAL-H (2.5 equiv), CH_2Cl_2, 0 °C to RT, 94% yield. **v.** NaH (1.5 equiv), AllBr (1.5 equiv), imidazole (catalytic amount), DMF, 0 °C to RT, 89% yield. **vi.** HCl, MeOH, reflux, 86% yield. **vii. a**. (-)-(*S*)-Camphor dimethyl acetal (3.4 equiv), $TsOH{\cdot}H_2O$ (0.05 equiv), CH_2Cl_2, reflux. **b**. Silica gel column chromatography diastereomeric resolution, 25% yield.

Synthesis of the enantiopure camphor acetal **34** was achieved from *myo*-inositol **1** (Scheme 2.2). Reaction of *myo*-inositol **1** with triethyl orthoformate in the presence of 4-toluenesulfonic acid monohydrate gave the adamantane-like derivative **37**. Treatment of the triol **37** with sodium hydride followed by 4-methoxybenzyl chloride allowed the regioselective protection of one of the two axial hydroxyl groups over the equatorial hydroxyl group, affording the diol **38** as a racemic mixture.



Scheme 2.3. Mechanism of the regioselective protection of the axial hydroxyl group in intermediate **37**.[106]

The regioselective protection of one of the two axial hydroxyl groups is achieved by adding 1.1 equivalents of sodium hydride in small portions to a stirred solution of triol **37** at 0 °C. The high regioselectivity of the reaction is thought to be due to the formation of the sodium chelate complex shown in Scheme 2.3; in this complex, the sodium counter-ion belonging to the alkoxide moiety coordinates to the neighbouring axial hydroxyl group. This stabilises the sodium chelate complex and prevents the formation of the equatorial sodium alkoxide species. Further studies by Billington[106] and co-workers confirmed this experimental outcome, as a loss of regioselectivity is noticed when either the counter-ion or solvent are changed. The subsequent reaction of the sodium chelate with 4-methoxybenzyl chloride affords the 4-methoxybenzyl ether **38** as a mixture of two enantiomers. The X-ray crystal structure of diol **38** (Figure 2.4) demonstrates that only the axial protected compound was obtained.

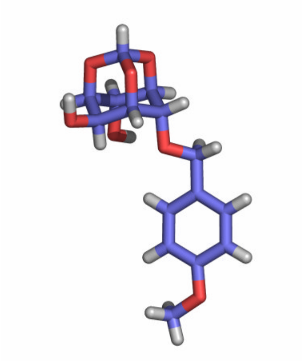
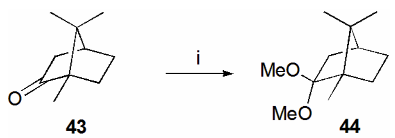


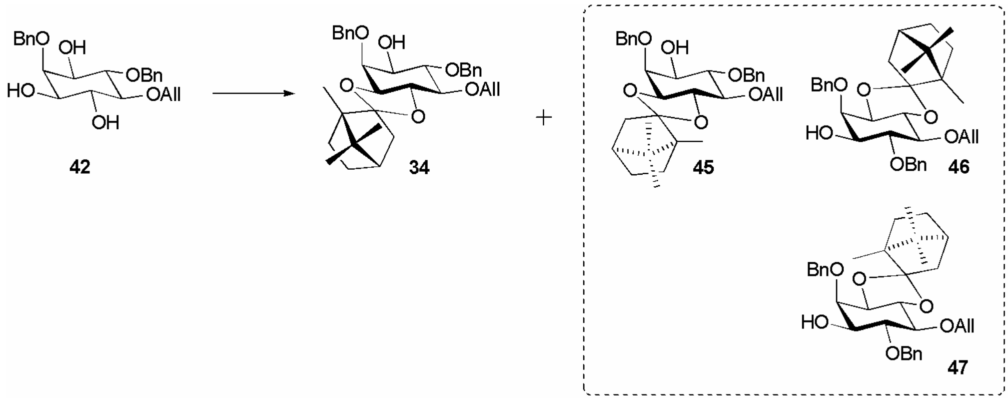
Figure 2.4. A PyMOL (www.pymol.org) representation of the X-ray crystal structure of compound **38** (one of the two enantiomers is shown).

Exhaustive benzylation of diol **38** afforded the fully protected orthoformate **39**, which was then regioselectively reduced to the alcohol **40** by treatment with 2.5 equivalents of diisobutylaluminium hydride (Scheme 2.2).[107,108] The alcohol **40** was then protected by treatment with sodium hydride and allyl bromide in the presence of a catalytic amount of imidazole, to afford compound **41**. Acidic methanolysis effected the removal of the acetal and 4-methoxybenzyl groups to afford the triol **42**. The enantiopure alcohol **34** was prepared by protection of the 3,4-vicinal diol in compound **42** with the chiral auxiliary (1*S*)-(-)-camphor dimethyl acetal **44**.[105] **44** was prepared by stirring at room temperature (1*S*)-(-)-camphor **43** and trimethylorthoformate in the presence of Montmorrilonite® clay K-10 (Scheme 2.4). The reaction afforded a crude mixture containing 75% of the desired product **44** together with a quantity of unreacted starting material **43**. The composition of the crude mixture was calculated by ^{1}H NMR analysis; comparison of the integrations of signals for two of the methyl groups of the acetal **44** [δ_H 0.91 (3H, s) and 0.82 (3H, s)] and the corresponding methyl groups of (1*S*)-(-)-camphor **43** [δ_H 0.92 (3H, s) and 0.84 (3H, s)] indicated a 3:1 ratio in favour of the acetal **44**, corresponding to a yield of 75%.



Scheme 2.4. Synthesis of (1*S*)-(-)-camphor dimethyl acetal **44**. *Reagents and conditions*: $(EtO)_3CH$ (4.0 equiv), K-10 clay, hexane, RT, 75% yield.

The crude mixture containing compound **44** was reacted with triol **42** in the presence of 4-toluenesulfonic acid monohydrate (Schemes 2.2 and 2.5) in dichloromethane under reflux. The reaction proceeds to completeness overnight, to give a mixture of the four diastereomers shown in Scheme 2.5.

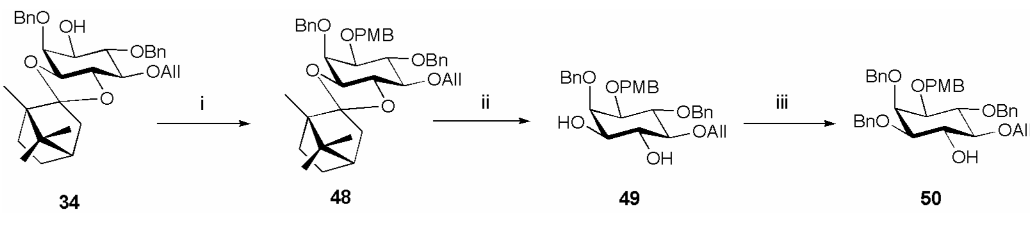


Scheme 2.5. Synthesis of compound **34**. *Reagents and condition:* (-)-(*S*)-Camphor dimethyl acetal (3.4 equiv), TsOH·H_2O (0.05 equiv), CH_2Cl_2, reflux, 25% yield.

Subsequent diastereomeric resolution using silica gel column chromatography allowed the separation of a fraction consisting of the optically pure intermediate **34** obtained in a yield of 25%, from a fraction consisting of an inseparable mixture of the diastereomers **45**, **46** and **47** (Scheme 2.5), obtained in 72% yield. The observed specific rotation of **34** ($[\alpha]_D^{20}$ -11.9) compared well with the literature value ($[\alpha]_D^{22}$ -11.7).[104,105]

2.4 Investigation of the *C*-4 position protecting group

2.4.1. Synthesis of the *myo*-inositol intermediate 50



Scheme 2.6. Synthesis of the intermediate compound **50**. *Reagents and conditions:* **i.** NaH (2.0 equiv), PMBCl (2.0 equiv), THF/DMF, 0 °C to RT, 94% yield. **ii.** AcCl (0.6 equiv), MeOH/CH_2Cl_2 40/60, RT, 79% yield. **iii.** Bu_2SnO (1.1 equiv), TBAI (1 equiv), BnBr (4.8 equiv), 3 Å molecular sieves, MeCN, reflux, 72% yield.[104,105,109]

The secondary alcohol **50**, precursor of the inositol intermediates with the general structure **35** (shown in the retrosynthetic Scheme 2.1) was synthesised in three steps from the enantiopure alcohol **34** (Scheme 2.6) in a manner similar to that reported by Lim and co-workers.[109] The synthesis began with the reaction of alcohol **34** with sodium hydride in dry *N,N*-dimethyl formamide and the subsequent reaction of the sodium alkoxide with 4-methoxybenzyl chloride to give the 4-methoxybenzyl ether **48**. Using this procedure it was not possible to achieve the yield reported in the literature.[109] In Table 1 are summarised the results obtained from a number of experiments carried out to improve the yields and find the optimum experimental conditions for this reaction.

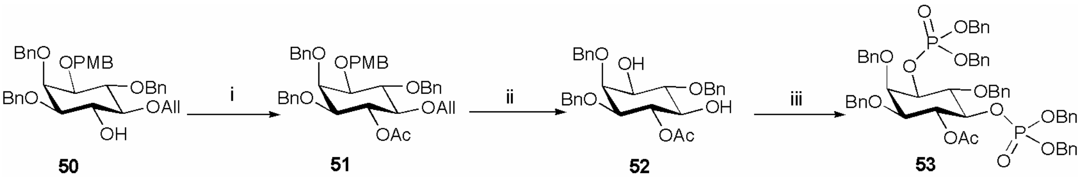
<table>
<tr><th rowspan="2">Experiment</th><th rowspan="2">Add.</th><th colspan="3">Reagents</th><th rowspan="2">Time, temperature and conditions</th><th rowspan="2">Solvent</th><th rowspan="2">Co-solvent</th><th rowspan="2">Yield</th></tr>
<tr><th>NaH</th><th>PMBCl</th><th>Other</th></tr>
<tr><td rowspan="4">1</td><td>a</td><td>1.5 equiv</td><td>1.5 equiv</td><td rowspan="4">imidazole catalytic amount</td><td>Starting material in dry DMF stirred overnight with NaH at RT, then PMBCl added and resulting mixture stirred for 6 h</td><td rowspan="4">dry DMF</td><td rowspan="4"></td><td rowspan="4">17%</td></tr>
<tr><td>b</td><td></td><td>0.5 equiv</td><td>Mixture stirred overnight at RT</td></tr>
<tr><td>c</td><td>0.5 equiv</td><td>0.5 equiv</td><td>Mixture stirred overnight at RT</td></tr>
<tr><td>d</td><td>0.5 equiv</td><td>0.5 equiv</td><td>Mixture stirred 4 h at 40 °C after NaH addition, then overnight at RT after PMBCl addition</td></tr>
<tr><td rowspan="3">2</td><td>a</td><td>1.1 equiv</td><td>1.5 equiv</td><td rowspan="3">TBAI catalytic amount</td><td>Mixture stirred overnight at RT</td><td rowspan="3">dry DMF</td><td rowspan="3"></td><td rowspan="3">84%</td></tr>
<tr><td>b</td><td></td><td>1.0 equiv</td><td>Mixture stirred for 1 h at RT</td></tr>
<tr><td>c</td><td>1.5 equiv</td><td>1.0 equiv</td><td>Mixture stirred for 5 h at RT</td></tr>
<tr><td>3</td><td>a</td><td>1.5 equiv</td><td>1.5 equiv</td><td>TBAI catalytic amount</td><td>Mixture stirred overnight at RT</td><td>dry THF</td><td>dry DMF</td><td>94%</td></tr>
</table>

Table 2.1. Optimisation of the experimental conditions for the synthesis of compound **48**.

In a first attempt, the alcohol **34** was converted in the corresponding sodium alkoxide using 1.5 equivalents of sodium hydride in dry *N,N*-dimethyl formamide and stirring the mixture overnight at room temperature. The subsequent reaction of the sodium alkoxide with 4-methoxybenzyl chloride was not complete after 6 hours (Table 2.1, addition 1-a), as adjudged by the thin layer chromatography analysis. Furthermore, analysis showed the presence of a compound less polar than the starting material and the product, suggesting that a side reaction had occurred. Further amounts of 4-methoxybenzyl chloride and sodium hydride were added to the mixture in order to maximise the yield of the reaction (Table 2.1, additions 1-b, 1-c, 1-d). A catalytic amount of imidazole was added to the mixture as a nucleophilic catalyst (Table 2.1, addition 1-b). The reaction mixture was also warmed to 40 °C for 4 hours in order to enhance the rate of the reaction (Table 2.1, addition 1-d). Thin layer chromatographic analysis after these actions showed the disappearance of the starting material and the presence of the desired compound and a less polar by-product. ^{1}H NMR and ^{13}C NMR analysis of this by-product showed a set of signals similar to those expected for the starting material **34** suggesting that an isomerisation reaction may have occurred; unfortunately mass spectrometry analysis did not lead to an explanation for this experimental observation. Furthermore, the yield of the reaction with respect to the desired compound was only 17% (Table 2.1, reaction 1). In a second attempt to perform the protection of compound **34**, the starting material was stirred for one hour with sodium hydride at room temperature in dry *N,N*-dimethyl formamide and then overnight after the addition of 4-methoxybenzyl chloride and a catalytic amount of tetra-*n*-butylammonium iodide (Table 2.1, addition 2-a). After this time the reaction was not complete and further amounts of sodium hydride and 4-methoxybenzyl chloride were added (Table 2.1, additions 2-b, 2-c). The final thin layer chromatography analysis showed that only the desired compound was formed. The yield of the reaction was 84% (Table 2.1, experiment 2). In further attempts to reduce the required amount of sodium hydride and 4-methoxybenzyl chloride the dry *N,N*-dimethyl formamide solvent was replaced with dry tetrahydrofuran. The alcohol **34** was converted to the sodium alkoxide in dry tetrahydrofuran using sodium hydride. After the subsequent addition of 4-methoxybenzyl chloride and a catalytic amount of tetra-*n*-butylammonium iodide (Table 2.1, experiment 3), the reaction mixture was stirred for 2 hours. The thin layer chromatographic analysis indicated that no reaction had occurred. This result can be explained by the low solubility of the alkoxide in tetrahydrofuran. After adding *N,N*-dimethyl formamide as co-solvent (Table 2.1, reaction 3) the reaction was complete after overnight stirring at RT in a yield of 94%. The experiments performed on the 4-methoxybenzyl protection of compound **34** show that this reaction can be carried out using dry tetrahydrofuran as solvent with dry *N,N*-dimethyl formamide as co-solvent to increase the solubility of the sodium alkoxide. The low yield obtained in the first attempt of this reaction (Table 2.1, experiment 1) can be explained by assuming that the starting material was consumed in a side-reaction due to the prolonged exposure of alcohol **34** to sodium hydride in *N,N*-dimethyl formamide.

The resulting camphor acetal **48** was converted to diol **49** by acidic methanolysis of the chiral auxiliary moiety using acetyl chloride in a methanol/dichloromethane mixture (Scheme 2.6). The resulting compound **49** was regioselectively protected at the *C*-3 hydroxyl group using di-*n*-butyltin oxide, tetra-*n*-butylammonium iodide and benzyl bromide, furnishing the alcohol **50** in a yield of 72% (Scheme 2.6).[103,104]

2.4.2. *C*-4 Position acetyl *myo*-inositol intermediates



Scheme 2.7. Synthesis of the *myo*-inositol derivative **53**. *Reagents and conditions:* **i.** DMAP (0.3 equiv), AcCl (12 equiv), pyridine, RT, 81% yield. **ii. a.** Wilkinson's catalyst, Hunig's base, EtOH, reflux; **b.** AcCl, CH_2Cl_2/MeOH (3:2), RT; **c.** CAN, MeCN/H_2O (4:1), RT; yield over steps **a**, **b** and **c** 72%. **iii. a.** Bis(benzyloxy)-*N,N*-diisopropylamino phosphine (5.0 equiv), 1*H*-tetrazole (5.0 equiv), CH_2Cl_2, RT; **b.** *m*CPBA (5.0 equiv), - 78 °C to RT, 66% yield.

The esterification of alcohol **50** using acetyl chloride and 4-dimethylaminopyridine furnished the intermediate **51** in 81% yield (Scheme 2.7). The structure of this compound was confirmed by X-ray crystallography (Figure 2.5).

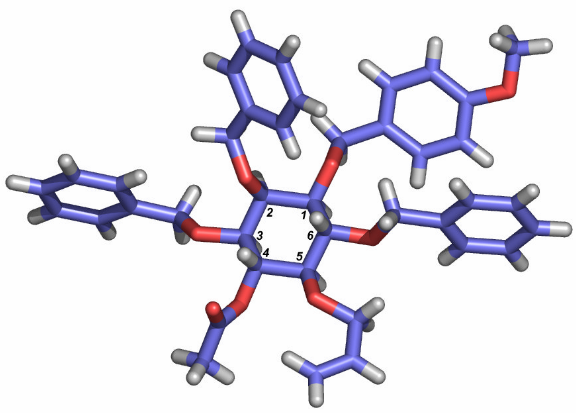
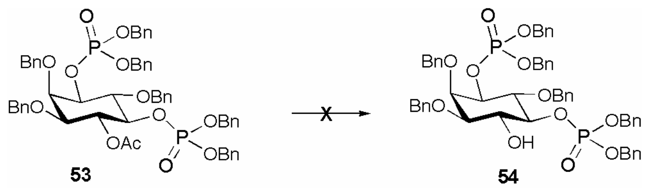


Figure 2.5. A PyMOL (www.pymol.org) representation of the X-ray crystal structure of compound **51.**

The allyl group of compound **51** was selectively removed by isomerisation of the *C*-5 position allyl group using Wilkinson's catalyst to the corresponding vinyl ether intermediate and subsequent alcoholysis of both vinyl and 4-methoxybenzyl groups using 1.0 M hydrochloric acid in ethanol. Using these experimental conditions, the partial hydrolysis of the acetyl group occurred, furnishing the desired compound **52** in 9% yield. A different procedure was developed involving the use of Wilkinson's catalyst. The treatment of the intermediate vinyl ether with acetyl chloride in methanol/dichloromethane and the oxidative cleavage of the 4-methoxybenzyl group using ceric ammonium nitrate in acetonitrile/water (Scheme 2.7), furnishing the desired compound **52** in 72% yield. Using the experimental conditions previously reported by Painter,[104] the diol **52** was phosphitylated and oxidised to afford compound **53** in 66% yield (Scheme 2.7).

In the first attempt to remove the acetyl group in the presence of the phosphate groups in compound **53**, the experimental conditions previously used by Lim were employed,[110] involving the treatment with potassium carbonate (1.1 equivalents) in a 5/3/2 methanol/tetrahydrofuran/water mixture for 5 hours (Scheme 2.8 and Table 2.2, experiment 1). The thin-layer chromatographic analysis indicated that no reaction had occurred and further potassium carbonate (1.0 equivalent) was added. The mixture was analysed after 24 hours (by thin-layer chromatography) and no reaction had occurred. Three more equivalents of potassium carbonate were added, and after 3 hours the thin-layer chromatographic analysis indicated the presence of the starting material and a mixture of more polar compounds, likely to be decomposition products. The potassium carbonate was quenched using a saturated aqueous solution of ammonium chloride. After the aqueous work up, the crude mixture was used as starting material in a further attempt to remove the acetyl group in compound **53** (Scheme 2.8 and Table 2.2, experiment 2). The crude mixture was treated with 1.0 equivalent of sodium hydroxide in methanol for 1.5 hours. The thin-layer chromatographic analysis indicated that no reaction had occurred and further sodium hydroxide (1.0 equivalent) was added to the mixture. Thin-layer chromatographic analysis after 1.5 hours indicated that no reaction had occurred, so the mixture was warmed to 35 ºC for a period of 20 hours. The mixture was analysed by thin-layer chromatography that indicated the complete disappearance of the starting material and the presence of a complex mixture of more polar compounds. Purification by silica gel column chromatography afforded a number of fractions that were analysed by ^{1}H NMR spectrometry, which indicated that the starting material had decomposed.

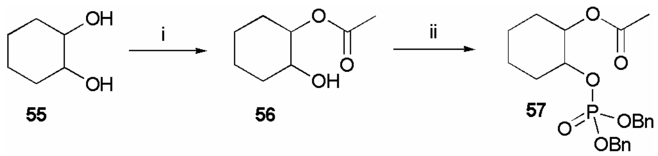


Scheme 2.8. The attempted synthesis of compound **54** through the deprotection of the acetyl group in compound **53**. *Reagent and conditions:* as described in Table 2.2.

Experiment	Reagent	Solvent	Time, Temperature	Yield
1	K_2CO_3 (1.1 equiv) K_2CO_3 (1.0 equiv) K_2CO_3 (3.0 equiv)	MeOH/THF/H_2O 5/3/2	5 h, RT 24 h, RT 3 h, RT	Partial decomposition of starting material
2	NaOH (1.0 equiv) NaOH (1.0 equiv)	MeOH/H_2O 9/1	1.5 h, RT 1.5 h, RT then 20, 35 °C	Decomposition of starting material
3	LiOH (2.1 equiv)	MeOH/H_2O 9/1	12 h, RT	Partial decomposition of starting material
4	Lipase VII	Hexane/wet Et_2O 5/1	7 days, 37.7 °C	No reaction

Table 2.2. Experimental condition used for the removal of acetyl group in compound **53**.

In order to develop the optimal experimental conditions for the removal of the acetyl group in the presence of the phosphate groups in the inositol derivative **53**, the model compound **57** was synthesised in two steps starting from (±)-1,2-*trans*-dihydroxycyclohexane **55** (Scheme 2.9).



Scheme 2.9. Synthesis of the model compound **57**. *Reagents and conditions:* **i**. 4-Dimethylaminopyridine (0.3 equiv), pyridine (1.1 equiv), acetyl chloride (1.1 equiv), CH_2Cl_2, 0 ºC to RT, 60% yield; **ii. a.** Bis(benzyloxy)-*N,N*-diisopropylamino phosphine (2.5 equiv), 1*H*-tetrazole (2.5 equiv), CH_2Cl_2, RT; **b.** *m*CPBA (2.5 equiv), - 78 °C to RT, 87% yield.

Using 4-dimethylaminopyridine as a nucleophilic catalyst, the acetyl protection of one hydroxyl group was achieved by adding a solution of acetyl chloride in dichloromethane to a solution of the starting material dissolved in a large volume of dichloromethane over the period of one hour, in order to reduce the acetylation of both the hydroxyl groups. The resulting mono-acetylated compound **56** was phosphitylated and oxidised to afford the model compound **57** in 87% yield (Scheme 2.9). Table 2.3 shows a number of different reaction conditions that where examined for the removal of the acetyl group in compound **57**. In a first attempt compound **57** was stirred for 2.5 hours in a 9/1 methanol/water solution containing 2.1 equivalents of potassium carbonate (Table 2.3, experiment 1). Using these conditions the alcohol **58** was recovered in 59% yield; however, an undesired trans-esterification side reaction occurred at the phosphate moiety, leading to the by-product **59** in 22% yield (Figure 2.6).

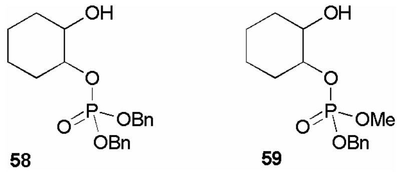


Figure 2.6. Compounds obtained from the deprotection of the acetyl group in model compound **57**.

In order to minimise the unwanted side-reaction, milder carbonates and different solvent systems were then investigated; unfortunately compound **57** was found to be inert towards these reaction conditions (Table 2.3, experiments 2-7). The strong base lithium hydroxide in methanol/water proved to be effective, furnishing the desired compound **58** in a reasonable yield (Table 2.3, experiment 8), together with a small amount of the by-product **59** (6% yield). To overcome the formation of compound **59** it was attempted to carry out the reaction in the presence of lithium hydroxide using benzyl alcohol as solvent, in order to obtain only the desired product **58** from trans-esterification side reaction (Table 2.3, experiment 9). Unfortunately under these experimental conditions no reaction was detected.

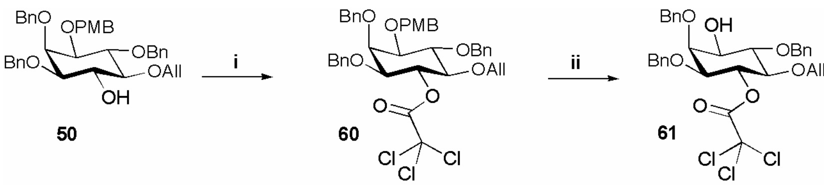
The enzyme Lipase VII from *candida rugosa* (Table 2.3, experiments 10-11) was also investigated. In a first attempt the enzyme was suspended in hexane/water and the mixture shaken for 4 days (Table 2.3, experiment 9). The reaction was found to be incomplete, however the desired compound **58** was obtained in 42% yield. A slight improvement in the final yield was obtained by replacing the solvent with a hexane/wet diethyl ether mixture (Table 2.3, experiment 11).

Experiment	Reagent	Solvent	Time, Temperature	Yield
1	K_2CO_3 (2.1 equiv)	$MeOH/H_2O$ 9/1	2.5 h, RT	59%
2	$BaCO_3$ (2.1 equiv)	$MeOH/H_2O$ 9/1	4 days	No reaction
3	$CaCO_3$ (2.1 equiv)	$MeOH/H_2O$ 9/1	4 days	No reaction
4	Na_2CO_3 (2.1 equiv)	$EtOH/H_2O$ 9/1	5 days	No reaction
5	Na_2CO_3 (2.1 equiv)	THF/H_2O 9/1	5 days	No reaction
6	K_2CO_3 (2.1 equiv)	$EtOH/H_2O$ 9/1	5 days	No reaction
7	K_2CO_3 (2.1 equiv)	THF/H_2O 9/1	5 days	No reaction
8	LiOH (2.1 equiv)	$MeOH/H_2O$ 9/1	30 min, RT	62%
9	LiOH (2.1 equiv)	$BnOH/H_2O$ 9/1	2 days, RT	No reaction
10	Lipase VII	$Hexane/H_2O$ 5/1	4 days, 37.7 ºC	42%
11	Lipase VII	Hexane/wet Et_2O 5/1	3 days, 37.7 ºC	53%

Table 2.3. Experimental condition used in model studies on compound **57**.

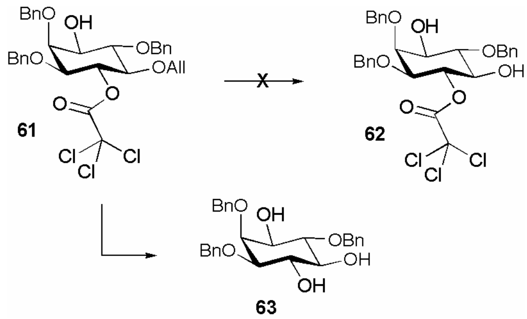
The experimental conditions developed using model compounds **57** were tested on compound **53** (*vide supra*, Table 2.2, experiments 3-4). Compound **53** was dissolved in a methanol/water mixture in the presence of lithium hydroxide (Table 2.2, experiment 3) and the reaction followed by thin-layer chromatography analysis, for a period of 12 hours. Using these conditions the result was the partial decomposition of the starting material. The enzyme Lipase VII was then used to attempt the hydrolysis of the acetyl group in compound **53** (Table 2.2, experiment 4), but after 7 days no conversion had occurred. These last results led to the decision to investigate a different protecting group for the *C*-4 position.

2.4.3. *C*-4 Position trichloroacetyl *myo*-inositol intermediates



Scheme 2.10. Synthesis of compound **61**. *Reagents and conditions:* **i.** Trichloroacetyl chloride (1.5 equiv), pyridine, RT, 30 min, 96% yield; **ii.** DDQ, CH_2Cl_2, RT, 91% yield.

The trichloroacetyl protecting group, due to the inductive effect of the three chlorine atoms vicinal to the carbonyl carbon atom, is much more reactive than the acetyl group towards acidic and basic hydrolysis. Therefore, compound **50** was reacted with trichloroacetyl chloride in pyridine to afford the trichloroacetyl-protected compound **61** in 96% yield (Scheme 2.10). For the removal of the *C*-1 position 4-methoxybenzyl group it was first attempted the reaction with ceric ammonium nitrate in a mixture of acetonitrile/tetrahydrofuran/water. Using these reaction conditions compound **61** was obtained in a yield of 64%. Due to its reactivity, the trichloroacetyl group was adjudged to be too sensitive to the slightly acidic environment generated by the ceric ammonium nitrate. This was confirmed by a second attempt to remove the 4-methoxybenzyl group by using 2,3-dichloro-5,6-dicyanobenzoquinone; this procedure was more successful, furnishing compound **61** in a yield of 91% (Scheme 2.10).

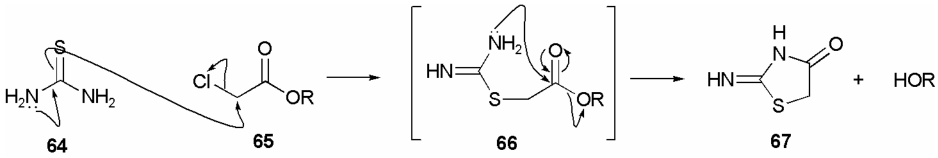


Scheme 2.11. The attempted synthesis of compound **62**. *Reagents and conditions:* **a.** Wilkinson's catalysts (0.6 equiv), Hunig's base (1.0 equiv), EtOH, reflux, 1.5 h. **b.** Acetyl chloride (0.6 equiv), CH_2Cl_2/MeOH, RT, 47% yield.

Compound **61** was reacted with Wilkinson's catalyst in ethanol under reflux in order to isomerise the double bond of the allyl group (Scheme 2.11). After 1.5 hours the ^{1}H NMR analysis indicated that a reaction had occurred at the double bond, but the signals appeared to be inconsistent with the expected signals for the intermediate vinyl ether. However, the crude material was reacted with a catalytic amount of acetyl chloride in methanol/dichloromethane for 2 hours (Scheme 2.11). After purification by silica gel column chromatography, the undesired triol **63** was isolated, indicating that the cleavage of the trichloroacetyl group had occurred under the described reaction conditions. It was thought that the acidic conditions used for the methanolysis of the intermediate vinyl ether were incompatible with the trichloroacetyl group. Therefore, in a further attempt the milder acidic catalyst 4-toluenesulfonic acid was used. After the double bond isomerisation, the crude material was dissolved in methanol/dichloromethane, 4-toluenesulfonic acid added at 0 °C and the mixture stirred for 3 hours at room temperature. TLC analysis indicated the presence of a complex mixture of compounds, likely to be due to decomposition of the starting material, and it was not possible to isolate the desired compound **62**. As a result of the above experimental outcomes, the trichloroacetyl protecting group was judged to be unsuitable for the protection of the *C*-4 position.

2.4.4. *C*-4 Position chloroacetyl *myo*-inositol intermediates

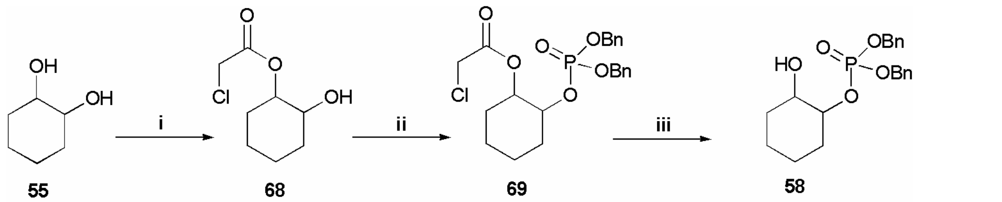
The next protecting group selected was the chloroacetyl group, as this group is more reactive than an acetyl group towards acidic and basic hydrolysis but much less reactive than the trichloroacetyl group. In addition, the chloroacetyl group has a unique deprotection protocol that is based on the reactivity at the carbon atom bearing the chlorine atom and not at the carbonyl centre.[111,112] Furthermore, this deprotection scheme has been previously used by Fraser-Reid to remove the chloroacetyl group in carbohydrate derivatives,[113] therefore it seemed to be a suitable protecting group. The mechanism for this reaction is shown in Scheme 2.12.[112]



Scheme 2.12. Mechanism of the deprotection of the chloroacetyl group using thiourea.[112]

The sulfur atom of thiourea **64** effects the nucleophilic substitution of the chlorine atom in generic compound **65** (Scheme 2.12) leading to the intermediate **66**. This compound undergoes an addition-elimination reaction, releasing the desired alcohol and 2-imino-4-thiazolidinone **67**.

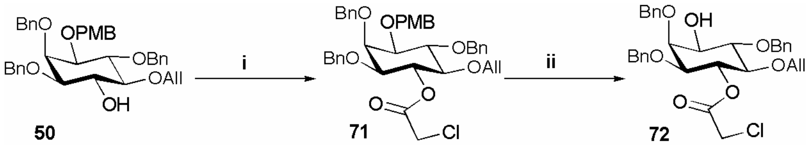
Model studies were carried out in order to test the feasibility of removing the chloroacetyl group in the presence of a neighbouring phosphate group, therefore the model compound **70** was synthesised (Scheme 2.13).



Scheme 2.13. Synthesis of compound **58**. *Reagents and conditions:* **i.** Chloroacetic anhydride (1.2 equiv), DMAP (0.2 equiv), pyridine (1.2 equiv), CH_2Cl_2, RT, 40% yield. **ii. a.** Bis(benzyloxy)-*N,N*-diisopropylamino phosphine (3.0 equiv), 1*H*-tetrazole (7.0 equiv), CH_2Cl_2, RT, 30 min, then H_2O (0.7 equiv). **b.** *m*CPBA (5.0 equiv), - 78 °C to RT, 62% yield. **iii.** Thiourea (10.0 equiv), $NaHCO_3$ (10.0 equiv), MeOH/CH_2Cl_2, 55 °C, 2 h, 61% yield.

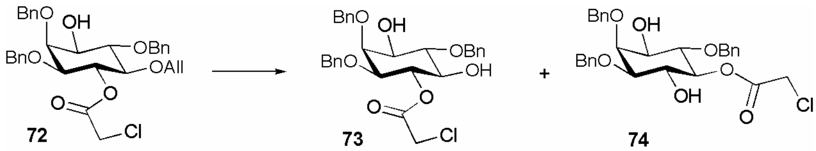
(±)-1,2-*trans*-Dihydroxycyclohexane **55** was converted to compound **68** using chloroacetic anhydride and 4-dimethylaminopyridine in a large volume of dichloromethane to decrease the esterification of both the hydroxyl groups (Scheme 2.13). Compound **68** was then phosphitylated and oxidised using the protocol previously reported by Watanabe;[114] this involves the use of bis(benzyloxy)-*N,N*-diisopropylamino phosphine and 1*H*-tetrazole to phosphitylate compound **68**, followed by treatment with water before oxidising the intermediate phosphite to the corresponding phosphate **69** (Scheme 2.13). Using this procedure compound **69** was synthesised in a yield of 62%. Following the method previously reported by Fraser-Reid,[113] the chloroacetyl group was removed from compound **69** using thiourea to afford compound **98** in 61% yield (Scheme 2.13).

Given the promising results obtained in the model studies on compound **69** (Scheme 2.13), the inositol intermediate **50** was treated with chloroacetic anhydride in pyridine to afford compound **71** in a yield of 90% (Scheme 2.14).



Scheme 2.14. Synthesis of compound **72**. *Reagents and conditions:* **i.** Chloroacetic anhydride (1.5 equiv), pyridine, RT, 90% yield. **ii.** DDQ (2.0 equiv), CH_2Cl_2, RT, 87% yield.

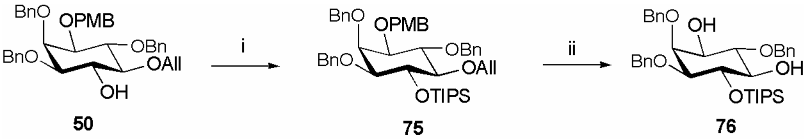
The removal of the *C*-1 position 4-methoxybenzyl group using 2,3-dichloro-5,6-dicyanobenzoquinone in dichloromethane afforded compound **72** in a yield of 87% (Scheme 2.14). The removal of the *C*-5 position allyl group was attempted by using Wilkinson's catalysts to isomerise the double bond and acetyl chloride in methanol as source of hydrochloric acid for the methanolysis of the intermediate vinyl ether. Compound **72** was subjected to these conditions (Scheme 2.15) and the preliminary data collected during the characterisation of the isolated product seemed to provide evidence that compound **73** had been synthesised. Although the thin-layer chromatography indicated the presence of only one spot, further ^{1}H NMR analysis suggested that the *C*-4 position chloroacetyl group had migrated to the *C*-5 position hydroxyl group under the isomerisation-methanolysis reaction conditions, furnishing an inseparable mixture of the two regioisomers **73** and **74** (Scheme 2.15).



Scheme 2.15. The attempted synthesis of compound **73**. *Reagents and conditions:* **a.** Wilkinson's catalysts (0.6 equiv), Hunig's base (1.0 equiv), EtOH, reflux, 1.5 h. **b.** Acetyl chloride (0.6 equiv), CH_2Cl_2/MeOH, RT.

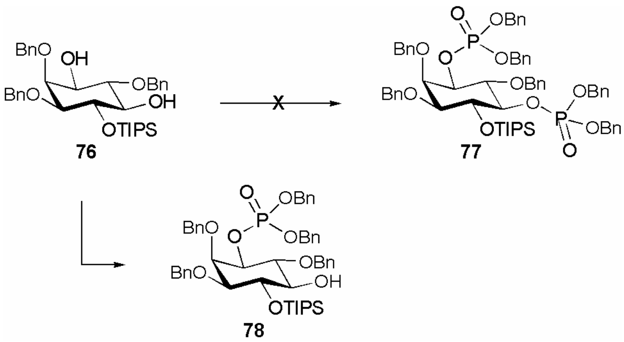
Having assessed that the chloroacetyl protecting group was unsuitable for the protection of the *C*-4 position, it was decided to investigate a different class of protecting groups.

2.4.5. *C*-4 Position triisopropylsilyl *myo*-inositol intermediates



Scheme 2.16. Synthesis of the compound **76**. *Reagents and conditions:* **i.** Triisopropylsilyl triflate (1.5 equiv), 2,6-luditine (4.0 equiv), CH_2Cl_2, 0 ºC to RT, 94% yield. **ii. a.** Wilkinson's catalyst (0.6 equiv), Hunig's base (1.0 equiv), EtOH, reflux, 2.5 h. **b.** Acetyl chloride (0.6 equiv), CH_2Cl_2/MeOH, RT. **c.** DDQ, CH_2Cl_2, RT, yield over 3 steps 62%.

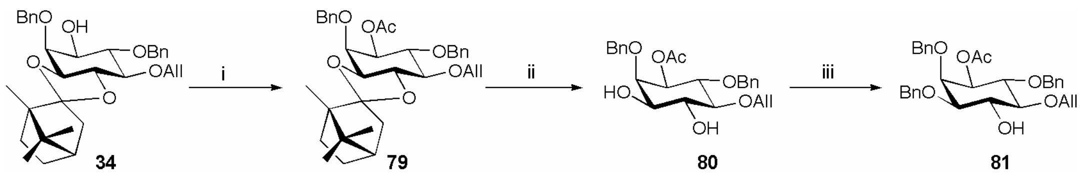
The triisopropylsilyl group was chosen as a potential candidate for the protection of the *C*-4 position of compound **50** because of its relative stability towards the reaction conditions employed to remove the *C*-1 position 4-methoxybenzyl group and the *C*-5 position allyl group. The possibility of selectively removing the triisopropylsilyl moiety using tetra-*n*-butylammonium fluoride, after having installed the phosphate groups, seemed also reasonable. Therefore, alcohol **50** was treated with triisopropylsilyl triflate to afford compound **75** in excellent yield (Scheme 2.16). The *C*-5 position allyl group in compound **75** was removed using Wilkinson's catalyst to isomerise the double bond and acetyl chloride in methanol/dichloromethane to cleave the intermediate vinyl ether (Scheme 2.16). The crude material was then treated with ceric ammonium nitrate, affording the desired diol **76** in a yield of 38%. A better result was obtained by using 2,3-dichloro-5,6-dicyanobenzoquinone as oxidising agent, which allowed the synthesis of compound **76** in a yield of 62% (Scheme 2.16). Compound **76** was then reacted with bis(benzyloxy)-*N,N*-diisopropylamino phosphine and 1*H*-tetrazole in dichloromethane in order to install the phosphate groups at the *C*-1 and *C*-5 positions and synthesise the bisphosphate compound **77** (Scheme 2.17). These reaction conditions did not furnish the desired compound **77**; purification by silica gel column chromatography furnished a compound which was proposed to be the monophosphate **78**, indicating that the phosphitylating reagent reacted only with the *C*-1 position hydroxyl group (Scheme 2.17). It was proposed that the steric hindrance of the *C*-4 position triisopropylsilyl group shields the *C*-5 position hydroxyl group, preventing the latter reacting with the phosphitylating reagent (Scheme 2.17).



Scheme 2.17. The attempted synthesis of compound **77**. *Reagents and conditions:* **a.** Bis(benzyloxy)-*N,N*-diisopropylamino phosphine (5.0 equiv), 1*H*-tetrazole (5.0 equiv), CH_2Cl_2, RT. **b.** *m*CPBA (5.0 equiv), - 78 °C to RT, 13% yield.

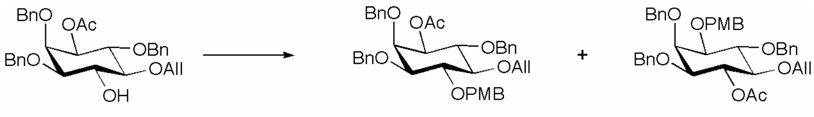
The lack of success in finding a optimal protecting group for the *C*-4 position in the inositol intermediate **50** led to a revision of the protection strategy used thus far; the modifications adopted are described in the next paragraph.

2.5 *C*-1 Position acetic esters: an alternative route to *C*-4 position $InsP_3$ analogues



Scheme 2.18. Synthesis of compound **81**. *Reagents and conditions:* **i.** Acetic anhydride (1.2 equiv), DMAP (0.3 equiv), pyridine, RT, 74% yield. **ii.** Acetyl chloride (0.6 equiv), $MeOH/CH_2Cl_2$, RT, 79% yield. **iii.** Bu_2SnO (1.1 equiv), TBAI (1 equiv), BnBr (4.8 equiv), 3 Å molecular sieves, MeCN, reflux, 56% yield.

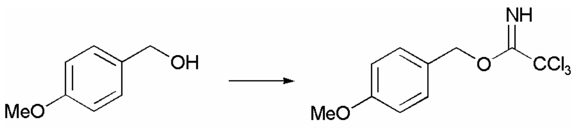
In order to solve the chemical problems related to the *C*-4 position protecting groups, it was decided to synthesise a series of *C*-1 position acetic esters, as shown in Scheme 2.18. The rationale for this new chemical route is that the 4-methoxybenzyl protection of the *C*-4 position hydroxyl group would lead to the fully protected inositol intermediate **82** (Scheme 2.19), which is a regioisomer of compound **51**, the chemical behaviour of which has been previously described in this chapter. The advantage of compound **82** is that the *C*-1 position acetyl group and the *C*-5 position allyl group can be removed using basic hydrolysis and the isomerisation-methanolysis reactions, respectively, without affecting the *C*-4 position 4-methoxybenzyl group. After having installed the two phosphate groups, the *C*-4 position 4-methoxybenzyl group could be removed by using ceric ammonium nitrate without affecting the neighbouring phosphate groups, as previously reported.[109,115] Thus, compound **34** was acetylated at the *C*-1 position using acetic anhydride in pyridine to give intermediate **79** (Scheme 2.18). Removal of the chiral camphor acetal auxiliary by acid-catalised methanolysis furnished the diol **80** in a yield of 79%. Selective benzyl protection of the *C*-3 position hydroxyl group using di-*n*-butyltin oxide chemistry gave the desired alcohol **81** in a yield of 56%.



81 **82** **51**

Scheme 2.19. The attempted synthesis of compound **82**. *Reagents and conditions:* NaH (1.1 equiv), PMBCl (1.1 equiv), DMF, 0 °C to RT, 24 h.

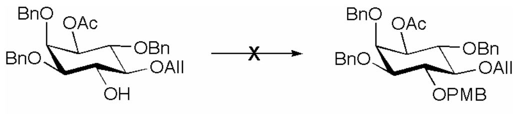
The 4-methoxybenzyl protection of compound **81** was attempted using sodium hydride and 4-methoxybenzyl chloride (Scheme 2.19); the analysis of the resulting product indicated the presence of a mixture of two isomers, which are likely to be compounds **82** and **51** (as judged by ^{1}H NMR and mass spectrometry analysis; *m/z* (ES+) 676 $[M+Na]^{+}$ single peak). It was proposed that the treatment of compound **81** with sodium hydride could set up a series of intermolecular transesterification reactions of the newly formed sodium alkoxide of compound **81** with the acetyl ester at the *C*-1 position in another molecule of compound **81**, leading to the two regioisomers **82** and **51** after the reaction with 4-methoxybenzyl chloride (Scheme 2.19).



83 **84**

Scheme 2.20. Synthesis of 4-methoxybenzyl 2,2,2-trichloroacetimidate **84**. *Reagents and conditions:* 50% aqueous KOH, Cl_3CCN (1.1 equiv), TBAS (0.01 equiv), CH_2Cl_2, - 10 °C to RT, 2 h, 36% yield.

To overcome the problem of the transesterification reaction, the 4-methoxybenzyl protecting group could be installed at the *C*-4 position by using a highly-reactive reagent that would not require the activation of the *C*-4 position hydroxyl group by conversion to the correspondent sodium alkoxide. The reagent 4-methoxybenzyl 2,2,2-trichloroacetimidate **84** has been previously used to install the 4-methoxybenzyl protecting group in compounds sensitive to sodium hydride.[116] This compound was synthesised from 4-methoxybenzyl alcohol **83** using trichloroacetonitrile under phase-transfer catalysis conditions (Scheme 2.20).



81 **82**

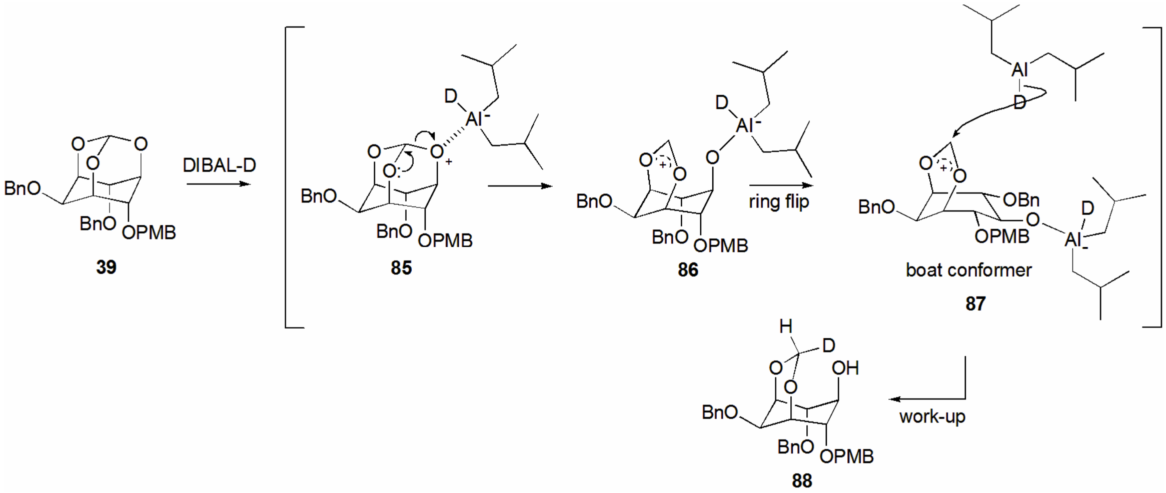
Scheme 2.21. The attempted synthesis of intermediate **82**. Method **A**. *Reagents and conditions:* 4-methoxybenzyl 2,2,2-trichloroacetimidate **84** (2.0 equiv), CSA (catalytic amount), CH_2Cl_2, 0 °C to RT, 15 h. Method **B**. *Reagents and conditions:* 4-methoxybenzyl 2,2,2-trichloroacetimidate **84** (2.0 equiv), TfOH (0.01), Et_2O, RT, 1 day.

In a first attempt the compound **81** was stirred in dichloromethane in the presence of 4-methoxybenzyl trichloroacetimidate **84** and camphorsulfonic acid for 15 h (Scheme 2.21, method A). TLC analysis indicated the presence of a complex mixture of compounds which could not be purified by column chromatography. The reaction was repeated using triflic acid as catalyst and diethyl ether as solvent (Scheme 2.21, method B). TLC analysis indicated the presence of an inseparable mixture of compounds.

As a result of this experimental outcome, the *C*-1 position acetic esters were judged to be not suitable for the synthesis of *C*-4 position-modified $InsP_3$ analogues.

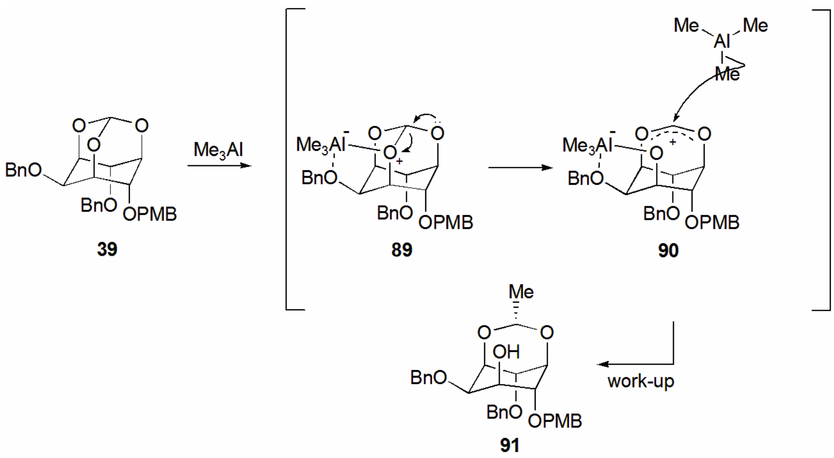
2.6 Selected Reaction Mechanisms

2.6.1. Diisobutylaluminium hydride-mediated cleavage



Scheme 2.22. Mechanism of the diisobutylaluminium deuteride-mediated cleavage of orthoformate **39**.[107,108]

The diisobutylaluminium deuteride (DIBAL-D) mediated-cleavage of orthoformate **39** has been previously investigated by Holmes[107,108] and the proposed mechanism is shown in Scheme 2.22. DIBAL-D can behave as a Lewis acid since has an empty *3p* orbital on the aluminium atom. This orbital coordinates to the *C*-5 position oxygen atom over the *C*-1 position and the *C*-3 position oxygen atoms. The *C*-5 position oxygen atom is thought to be more accessible than the other two oxygen atoms of the orthoformate moiety due to the presence of the *C*-2 position benzyl group, which is free to rotate around the C-O bond, generating a hindered environment proximal to the *C*-1 position and the *C*-3 position oxygen atoms. Therefore DIBAL-D can coordinate only to the *C*-5 position oxygen atom to give the intermediate **85**. This rearranges to the oxacarbenium species **86**, which is thermodynamically unstable due to the unfavourable 1-3 diaxial interactions between the transient *C*-5 position aluminium moiety and the acetal ring and thus undergoes a ring flip, leading to the more stable boat conformer **87**. This intermediate reacts with the second equivalent of diisobutylaluminium deuteride which donates a deuteride atom exclusively from the less hindered face of the acetal moiety. The reaction with the deuteride reagent affords nearly 100% yield of the alcohol **88**.[107,108]

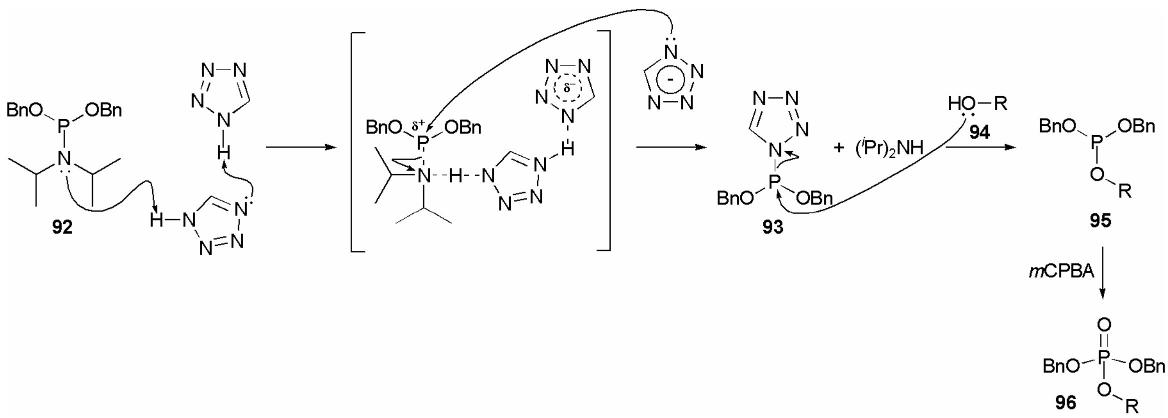


Scheme 2.23. Mechanism of trimethylaluminium-mediated cleavage of orthoformate **39**.[107,108]

The reaction of orthoformate **39** with trimethylaluminium has also been investigated by Holmes.[107,108] This reaction leads to compound **91**, as shown in Scheme 2.23. Trimethylaluminium is a Lewis acid, much less hindered than diisobutylaluminium hydride, and reacts with **39** forming a chelate complex with the *C*-2 position oxygen atom and either the *C*-1 position or the *C*-3 position oxygen atoms, to give the intermediate **89**. This rearranges to the oxacarbenium species **90**, which reacts with the methyl carbanion donated from the other equivalent of trimethylaluminium, affording compound **91**.

The use of a bulky reagent as diisobutylaluminium hydride or a regent with reduced steric hindrance as trimethylaluminium allows to modify the reaction outcome and achieve a different selectivity in the cleavage of the orthoformate moiety in compound **39**, allowing the development of different synthetic strategies.

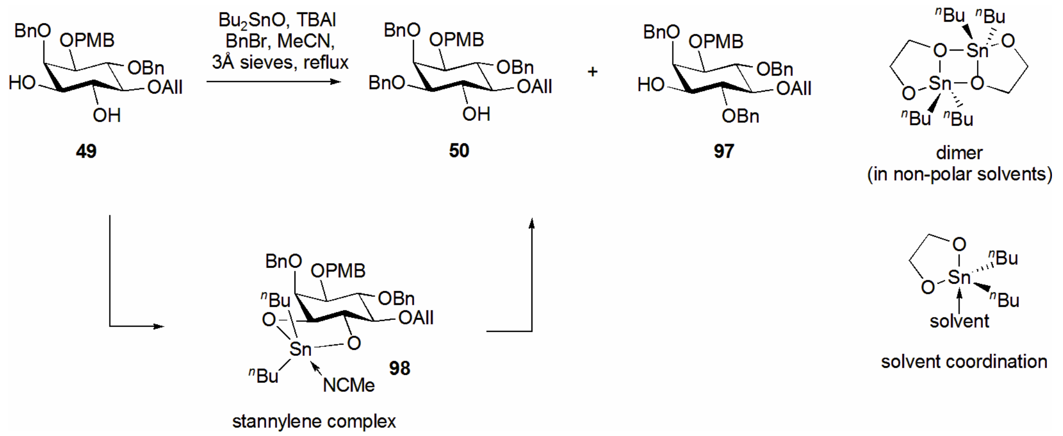
2.6.2. Phosphitylation and oxidation of alcohols to phosphates



Scheme 2.24. Mechanism of the 1*H*-tetrazole catalysed phosphitylation of alcohols.[117,118]

The mechanism of the phosphitylation-oxidation procedure in shown in Scheme 2.24. The most used catalyst in phosphoramidite chemistry is 1*H*-tetrazole, because of its behaviour as both acidic and nucleophilic catalyst. As established by kinetic studies on phosphitylation of alcohols,[117,118] the phosphoramidite **92** is first protonated by 1*H*-tetrazole, then a second, anionic, 1*H*-tetrazole reacts with the partially positive-charged phosphorus atom to give the tetrazolide intermediate **93**, which is the reactive species that effects the phosphitylation of the alcohol **94**, yielding the phosphite **95**.[117,118] This is not usually isolated, but oxidised directly to the corresponding phosphate **96** by treatment with an oxidising agent such as 3-chloroperoxybenzoic acid (*m*CPBA in Scheme 2.24).

2.6.3. Selective benzylation of the *C*-3 position with di-*n*-butyltin oxide



Scheme 2.25. Mechanism of the selective protection of diol **49**.[103,104]

The highly regioselective protection procedure was previously reported by Gigg and co-workers.[103] This method involves the use of di-*n*-butyltin oxide in acetonitrile under reflux (Scheme 2.25) to form the stannane acetal **98** *in situ* (in order to assist the stannane acetal formation a Soxhlet extractor filled with activated 3 Å molecular sieves was used to remove the formed water from the reaction mixture).[103] Although the reaction mechanism has not been unambiguously proven, studies of stannane derivatives using ^{119}Sn NMR spectroscopy suggest that the ^{119}Sn atom is penta- or hexa- coordinated.[119] While in the solid state it is known that penta-coordinated stannane compounds exist as dimers (Scheme 2.25), in solution and in the presence of a polar solvent such as acetonitrile the stannane acetal could exist as a penta-coordinated complex (**98**, Scheme 2.25). In this complex the two oxygen atoms at the *C*-3 and *C*-4 positions are differentiated; the *C*-3 position oxygen atom lies on the apical position of the complex, the *C*-4 position oxygen atom occupies the equatorial position. In this configuration, the apical bond of the complex is longer than the equatorial bond. In the presence of benzyl bromide, the *C*-3 position apical oxygen atom reacts preferentially over the *C*-4 position oxygen atom, and this can be explained by assuming that the *C*-3 oxygen atom is more accessible to a bulky alkylating reagent such as benzyl bromide than the *C*-4 oxygen atom, and also more reactive being the apical, which has a longer oxygen-tin bond than the equatorial one. The reaction proceeds quantitatively to furnish a mixture of the *C*-3 position (**50**) and *C*-4 position (**97**) benzyl-protected compounds (Scheme 2.25).

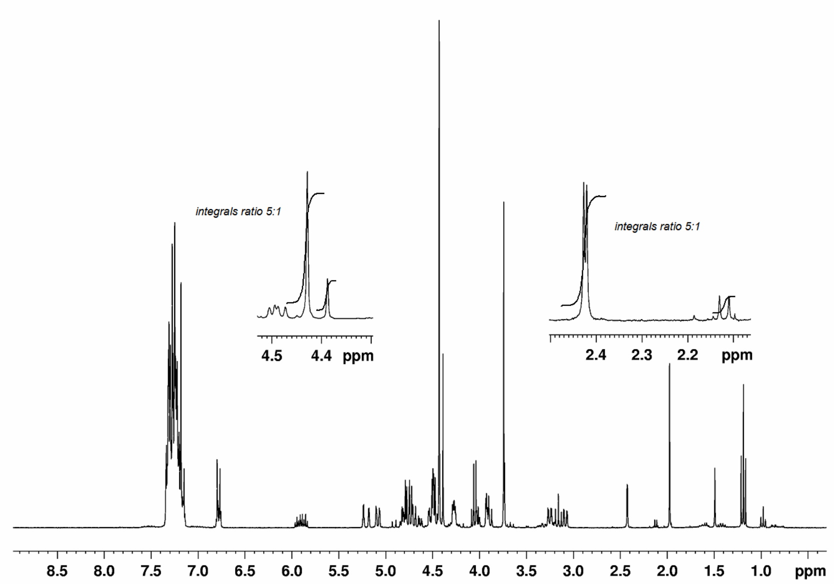


Figure 2.7. ^{1}H NMR spectrum of a crude mixture of compounds **50** and **97** after the benzyl protection using the tin acetal method.

^{1}H NMR analysis of the crude mixture indicated a 5:1 ratio mixture of the two compounds, in favour of the desired regioisomer **50**. This ratio was assessed by comparing the integrations of the two signals for the 4-methoxybenzyl group of the *C*-3 position **50** and *C*-4 position **97** benzyl-protected compounds [δ_H 4.43 (OC*H*$_3$, compound **50**) and 4.39 (OC*H*$_3$, compound **97**)] as shown in Figure 2.7. The same result was obtained by comparison of the integrations of the signal for the hydroxyl group in the two isomers **50** and **97** [δ_H 2.42 (O*H*, compound **50**) and 2.12 (O*H*, compound **97**)] (Figure 2.7).

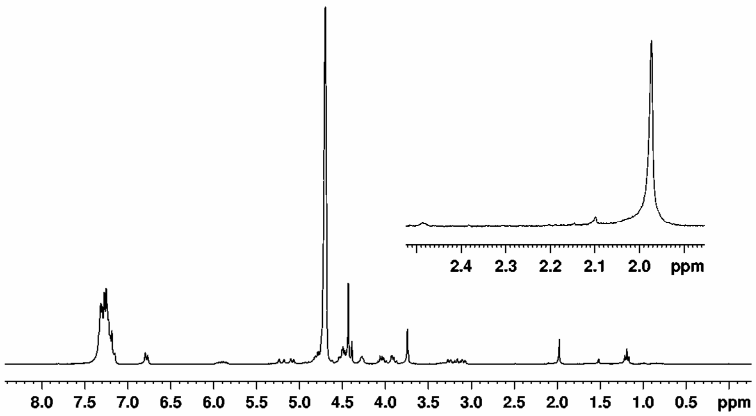


Figure 2.8. ^{1}H NMR spectrum of a crude mixture of compounds **50** and **97** after treatment with D_2O.

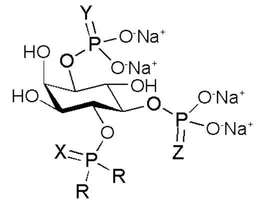
Assignment of the signals at δ_H 2.42 and δ_H 2.12 to the hydroxyl groups of the corresponding compounds was performed by ^{1}H NMR analysis of a sample from the crude of the reaction, after treatment with deuterium oxide. The two hydroxyl groups signals disappeared as result of the exchange of the hydrogen/deuterium atoms (Figure 2.8).

2.7 Summary

The analysis of the crystal structure of the $InsP_3R1$ binding domain provided essential information about the structural requirements for a compound to behave as an $InsP_3R$ antagonist. *C*-4 position-modified $InsP_3$ analogues with the general structure **32** and **33** (Figure 2.9), prepared as pure D-enantiomers, are proposed to be $InsP_3Rs$ antagonists. In order to synthesise such compounds, a chemical route starting from *myo*-inositol has been designed; this route makes use of a previously reported method for the separating the D-inositol enantiomers from the L-enantiomers.[104,105]

The protecting groups examined for masking the *C*-4 position in inositol intermediates were all found to be not suitable for synthesising *C*-4 position-modified $InsP_3$ analogues. A different approach, involving the use of an acetyl group to mask the *C*-1 position of the inositol ring was found to be incompatible with the reaction conditions used through the synthetic steps.

The next chapter describes the modifications adopted to complete the synthesis of *C*-4 position-modified $InsP_3$ analogues.



32 R = Me; X, Y, Z = O
33 R = Alkyl; X, Y, Z = O or S

Figure 2.9. Structures of the proposed $InsP_3Rs$ antagonists.

Result and Discussion (Part Two)

3 Results and Discussion (part two)

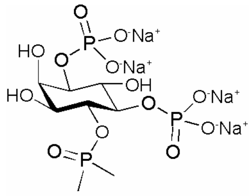
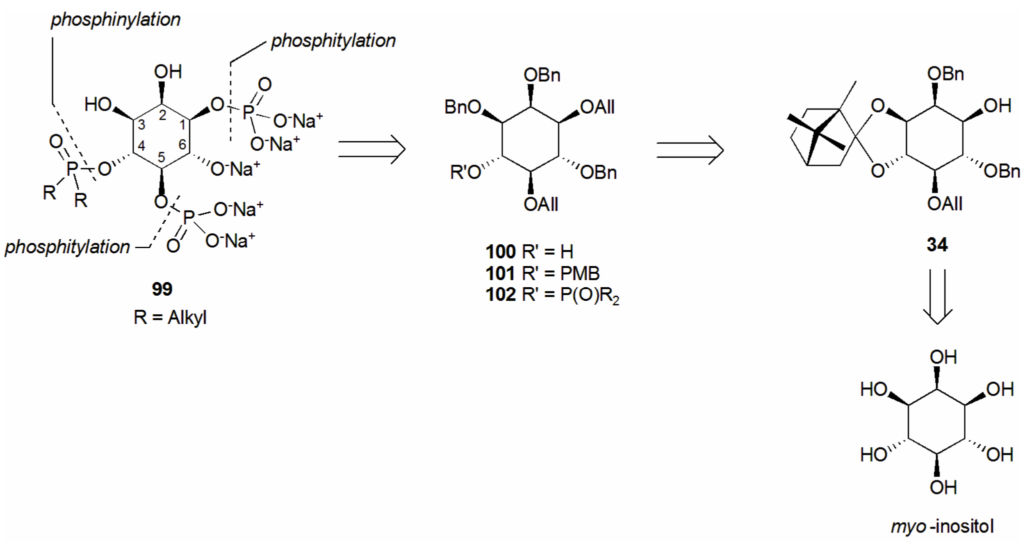


Figure 3.1. Structure of the C-4 position-modified $InsP_3$ analogue **32**.

The *C*-4 position-modified $InsP_3$ analogue **32** shown in Figure 3.1 has been proposed as a competitive antagonist of the $InsP_3R$ (*vide supra*). As described in chapter 2, it was not possible to achieve the synthesis of such compound using the proposed route, due to problems encountered during the later stages in the synthetic procedure. The strategy used thus far was therefore revised and a new plan for the synthesis developed. The retrosynthetic analysis in Scheme 3.1 describes the new proposed synthesis of *C*-4 position-modified $InsP_3$ analogues starting from *myo*-inositol.

3.1 Retrosynthesis

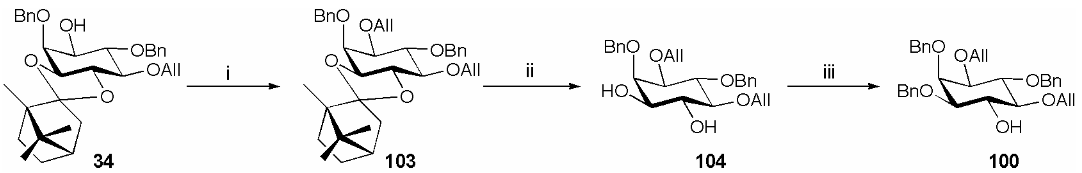


Scheme 3.1. Proposed retrosynthesis of $InsP_3$ analogues, allowing modifications at the *C*-4.

It was proposed that compounds with the structure **99** could be prepared in five steps from intermediate **101** by deprotection of the allyl groups, phosphitylation and oxidation of the resulting *C*-1 and *C*-5 hydroxyl groups, deprotection of the 4-methoxybenzyl group, phosphinylation of the resulting *C*-4 position hydroxyl group and final hydrogenolysis of the benzyl groups (Scheme 3.1). The use of two allyl protecting groups at the *C*-1 and the *C*-5 positions would allow the installation of the required phosphate groups in one synthetic step and would also allow the use of the 4-methoxybenzyl group for protecting the *C*-4 position hydroxyl group (intermediate **101**). In chapter 2, the 4-methoxybenzyl group was shown to be stable to the reaction conditions used to remove allyl groups; furthermore, it has been previously reported that the 4-methoxybenzyl can be removed in the presence of phosphate groups using oxidising agents, such as ceric ammonium nitrate.[115] Therefore, compound **101** could be prepared by 4-methoxybenzyl protection of the *C*-4 position hydroxyl group in compound **100**, which in turn could be synthesised from the camphor acetal **34** by allyl protection of the *C*-1 hydroxyl group, removal of the camphor acetal auxiliary and selective benzyl protection of the *C*-3 hydroxyl group using the di-*n*-butyltin oxide method.[103]

It was envisaged that compound **100** could be a useful intermediate, as it would allow the synthesis of compound **99** in four steps. The synthesis could be achieved by phosphinylation of the *C*-4 position hydroxyl group to give compound **102**, followed by removal of the allyl groups, phosphitylation and oxidation of the resulting diol and final hydrogenolysis of the benzyl groups (Scheme 3.1). This procedure would also shorten the synthetic route by avoiding the use of a protecting group for the *C*-4 position hydroxyl group in compound **100**. The camphor acetal **34** required for the proposed synthetic route could be prepared in seven steps from *myo*-inositol **1** as previously described in chapter 2.[104,105]

3.2 Synthesis of the bis-allyl *myo*-inositol derivative 100

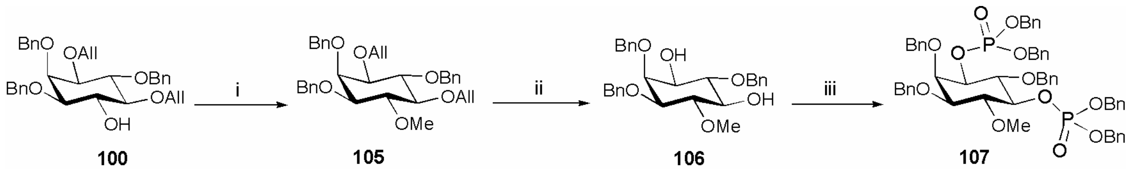


Scheme 3.2. Synthesis of compound **100**. *Reagents and conditions:* **i.** Allyl bromide (1.2 equiv), sodium hydride (1.2 equiv) imidazole (catalytic amount), TBAI (catalytic amount), THF/DMF, 0 °C to RT, 91% yield. **ii.** Acetyl chloride (0.6 equiv), $MeOH/CH_2Cl_2$, RT, 88% yield. **iii.** Bu_2SnO (1.1 equiv), TBAI (1.0 equiv), BnBr (4.8 equiv), 3 Å molecular sieves, MeCN, reflux, 71% yield.

Compound **100** was synthesised in three steps from the enantiopure compound **34** (Scheme 3.2). Allyl protection of the *C*-1 position hydroxyl group of intermediate **34** afforded compound **103** in high yield. The removal of the camphor acetal auxiliary using acetyl chloride in dichloromethane/methanol as a hydrochloric acid source furnished the diol **104** in 88% yield; this compound was selectively benzylated at the

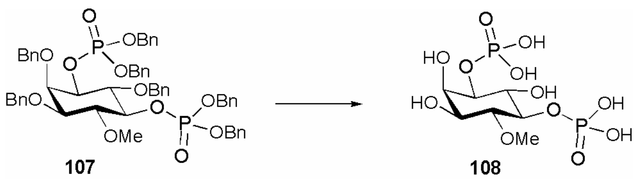
C-3 position using di-*n*-butyltin oxide and benzyl bromide. ^{1}H NMR analysis of the crude reaction mixture indicated the presence of two compounds; estimation of the relative ratio of the compounds, and therefore of the selectivity, was not possible, due to the signals for the two compounds not being fully resolved. Hovever, purification of the crude mixture afforded intermediate **100** in 71% yield.

3.3 Synthesis of (-)-1D-4-*O*-methyl-*myo*-inositol 1,5-bisphosphate (sodium salt) 109



Scheme 3.3. Synthesis of compound **107**. *Reagents and conditions:* **i.** MeI (1.1 equiv), NaH (1.1 equiv), THF, 0 °C to RT, 91% yield. **ii. a.** Wilkinson's catalyst, Hunig's base, EtOH, reflux. **b.** AcCl, CH_2Cl_2/MeOH (3:2), RT, 79% yield. **iii. a.** Bis(benzyloxy)-*N,N*-diisopropylamino phosphine (5.0 equiv), 1*H*-tetrazole (5.0 equiv), CH_2Cl_2, RT. **b.** *m*CPBA (5.0 equiv), - 78 °C to RT, 66% yield.

The *C*-4 position-modified $InsP_3$ analogue (-)-1D-4-*O*-methyl-*myo*-inositol 1,5-bisphosphate (sodium salt) **109** was synthesised in order to both obtain preliminary information about the biological activity at the $InsP_3$Rs and test the experimental conditions to be used for the final hydrogenolysis of the benzyl protecting groups.
Compound **105** was synthesised from intermediate **100** using sodium hydride and methyl iodide in tetrahydrofuran (Scheme 3.3). Wilkinson's catalyst was used to isomerise the allyl groups to the corresponding vinyl ethers, followed by acidic methanolysis to furnish compound **106** in good yield. Phosphitylation and oxidation of diol **106** gave the perbenzylated compound **107** in 66% yield.



Scheme 3.4. Synthesis of compound **108**. *Reagents and conditions:* H_2, Pd/C (10%) (0.4 equiv), EtOH, RT, 10 h. These reaction conditions may have caused the transesterification of the free phosphate groups to the neighbouring hydroxyl groups.

The final hydrogenolysis of the benzyl groups was first attempted by using palladium on activated carbon as a catalyst (Scheme 3.4) under an atmosphere of hydrogen in ethanol. This procedure should furnish the final compound **108** with the two phosphate groups in the free phosphoric acid form. The reaction yielded a material possessing the same molecular mass as compound **108** [*m/z* (ES+) 377 $(M+Na)^+$; (ES-) 353 $(M-H)^-$].

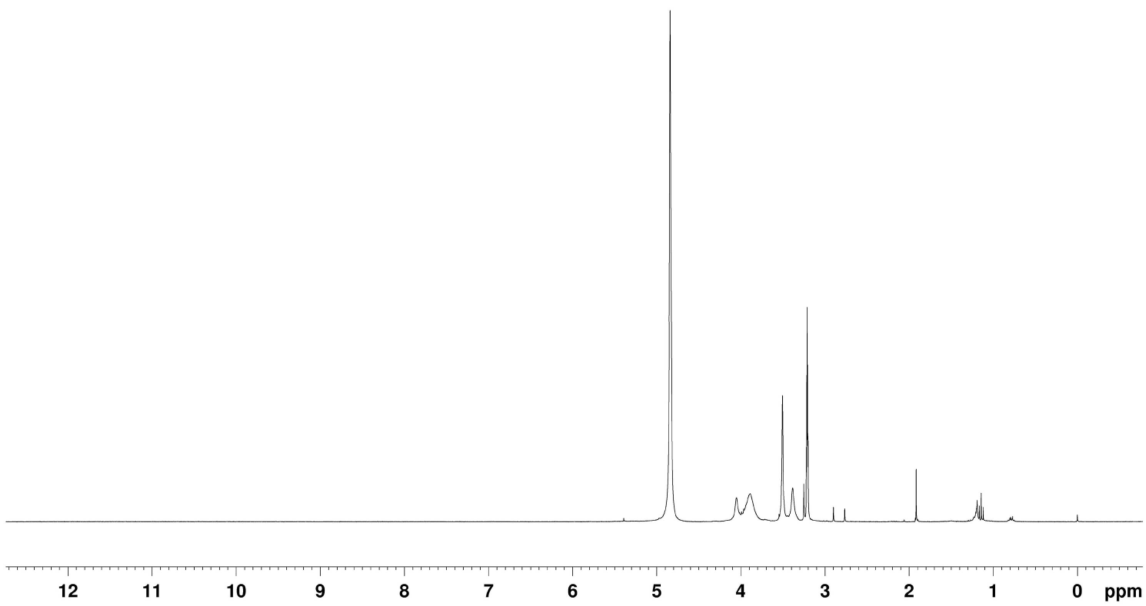


Figure 3.2. ^{1}H NMR spectrum of the material obtained from catalytic hydrogenolysis of compound **107** as described in Scheme 3.4.

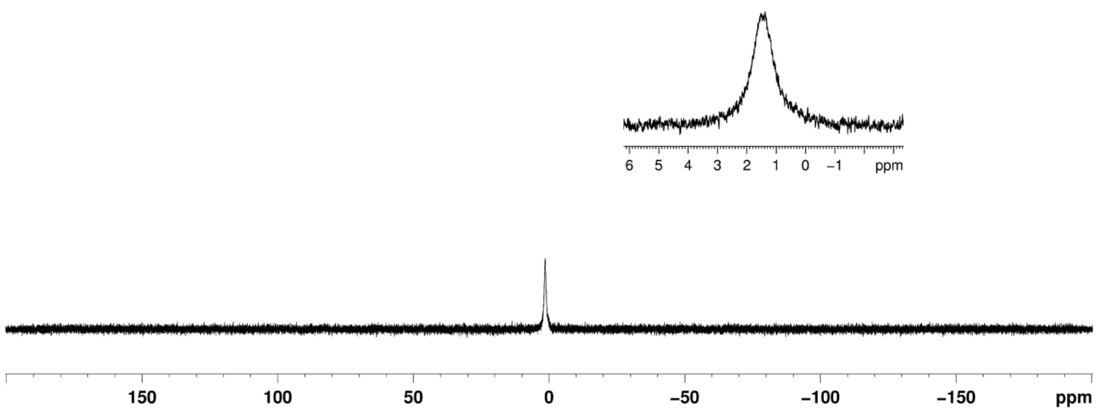
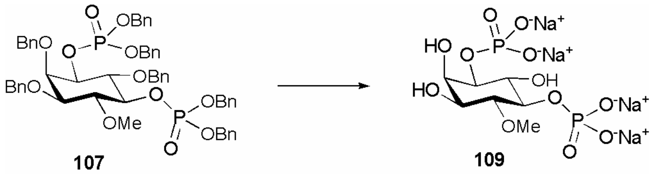


Figure 3.3. ^{31}P NMR spectrum of the material obtained from catalytic hydrogenolysis of compound **107** as described in Scheme 3.4.

^{1}H NMR analysis indicated the presence of broad inositol proton signals (Figure 3.2), and ^{31}P NMR analysis revealed a very broad signal centred around the phosphate signals region (Figure 3.3). The line broadening in both the ^{1}H NMR and the ^{31}P NMR spectra was attributed to the presence of the two free phosphoric acid groups in compound **108**; however, the signal broadening hampered the correct assignment of the NMR signals to the structure of compound **108**. Any inhomogeneity in the composition of the final compound **108**, resulting from phosphate group migration, would be reflected in the biological activity assessments, leading to flawed results. Therefore, an accurate and unambiguous assignment of the ^{1}H NMR and ^{31}P NMR signals is essential.

A previously reported method[109] for the hydrogenolysis of benzyl groups in inositol phosphate intermediates involves the use of palladium black in *tert*-butanol/water in the presence of sodium hydrogen carbonate. This method would furnish the final compounds as sodium salts; the function of the sodium hydrogen carbonate is to convert the newly formed phosphoric acid groups in sodium phosphates and therefore minimise the undesired transesterification reaction. The phosphates have been shown to give sharp ^{1}H and ^{31}P NMR signals.[104] In addition the sodium salts of phosphates can often be lyophilised to give solid products.



107 **109**

Scheme 3.5. Synthesis of compound **109**. *Reagents and conditions:* H_2, Pd black (20.0 equiv), $NaHCO_3$ (4.0 equiv), $^{t}BuOH/H_2O$ 6/1, RT, 4 h, yield 82% yield.

The hydrogenolysis reaction was attempted using compound **107** (Scheme 3.5), furnishing the desired final compound *(-)-1D-4-O-methyl-myo-inositol 1,5-bisphosphate (sodium salt)* **109** in 82% yield. ^{1}H NMR analysis confirmed the presence of the expected signals for the inositol ring (Figure 3.4). The ^{31}P NMR spectrum showed two sharp signals at δ_P 3.6 and δ_P 3.0, indicating that the two phosphate groups had not migrated (Figure 3.5).

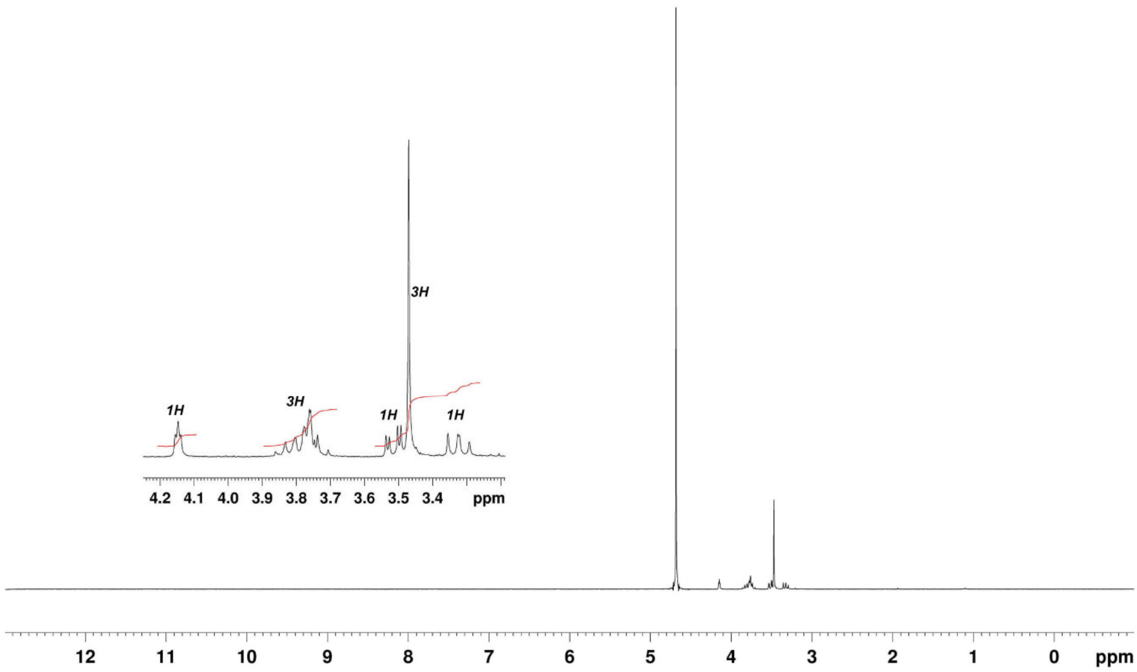


Figure 3.4. ^{1}H NMR spectrum of (-)-1D-4-*O*-methyl-*myo*-inositol 1,5-bisphosphate (sodium salt) **109.**

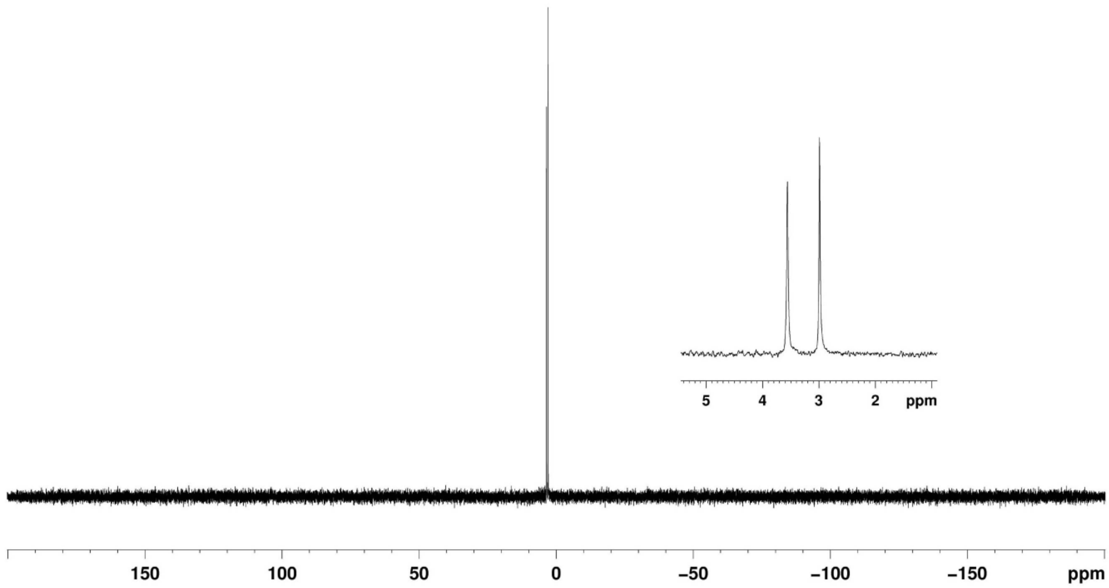
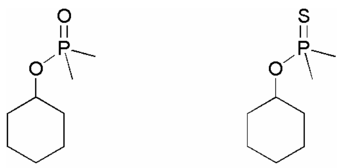


Figure 3.5. ^{31}P NMR spectrum of (-)-1D-4-O-methyl-myo-inositol 1,5-bisphosphate (sodium salt) **109**.

3.4 Development of a phosphinylation method for the synthesis of *myo*-inositol derivatives

Having developed the synthesis of a *C*-4 position-modified inositol analogue, it was necessary to develop conditions for the installation of the dimethylphosphinyl moiety on compound **100**.

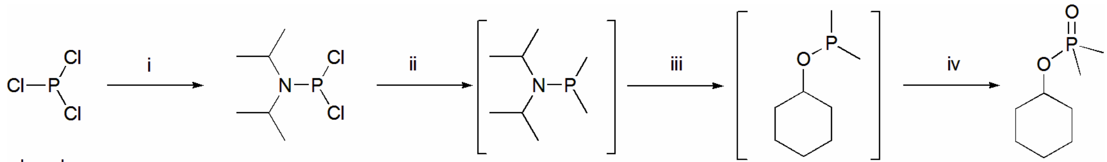
3.4.1. *In situ* generation of the phosphinylating reagent



110 **111**

Figure 3.6. The dimethylphosphinate **110** and dimethylphosphinothioate **111** model compounds.

The cyclohexyl dimethylphosphinate **110** and the cyclohexyl dimethylphosphinothioate **111** (Figure 3.6) were synthesised as model compounds to develop the conditions required for the phosphinylation of *myo*-inositol intermediates.

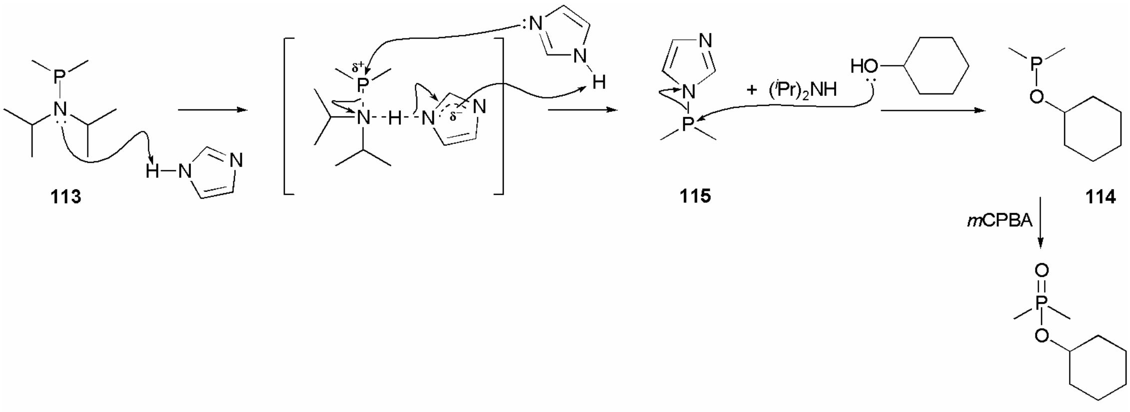


phosphorus trichloride **112** **113** **114** **110**

Scheme 3.6. Synthesis of cyclohexyl dimethylphosphinate **110**. *Reagents and conditions:* **i.** *N,N*-Diisopropylamine (2.0 equiv), Et_2O, - 10°C to RT, 73% yield. **ii.** MeLi (3.1 equiv), Et_2O, - 78 °C to RT. **iii.** Cyclohexanol (0.5 equiv), imidazole (2.0 equiv), CH_2Cl_2, - 78 °C to RT. **iv.** *m*CPBA (2.0 equiv), CH_2Cl_2, - 78 °C to RT. Yield over steps **ii**, **iii** and **iv** 63%.

The synthesis of compound **110** was achieved from phosphorus trichloride (Scheme 3.6). Treatment with *N,N*-diisopropylamine in diethyl ether afforded, after Kugelrohr distillation, compound **112** in a yield of 73%. Dialkylation with methyl lithium yielded the presumed intermediate **113**, as judged by ^{31}P NMR (δ_P 8.7), which was converted *in situ* to the presumed intermediate phosphinite **114** (δ_P 112.0), by addition to cyclohexanol and imidazole in dichloromethane and then oxidised to the desired product **110**.

The established phosphoramidite chemistry has been considered in order to rationalise the mechanism of the phosphinylation reaction of cyclohexanol (Scheme 3.7).[117] In Scheme 3.6 imidazole is used as the catalyst in place of 1*H*-tetrazole. Since two equivalents of imidazole are added, a possible reaction mechanism is proposed shown in Scheme 3.7. By analogy with the phosphitylation mechanism, the rate-limiting step is likely to be the protonation of the nitrogen atom of the *N,N*-diisopropylamine moiety, as the second equivalent of imidazole can easily trap the developing phosphorus cation to give the intermediate imidazolide **115** (Scheme 3.7). This species then reacts with cyclohexanol to give the phosphinite **114**. This hypothesised mechanism seems to be reasonable if compared with the nucleophilic catalysis in phosphoramidite alcoholysis previously discussed (Scheme 2.24).[117,118] Intermediate **114** is oxidised to the phosphinate **110** by treatment *in situ* with two equivalents of 3-chloroperoxybenzoic acid (Scheme 3.7).



Scheme 3.7. Proposed mechanism for the phosphinylation of cyclohexanol.

Compound **110** displayed analytical and spectroscopic data consistent with the assigned structure. As expected, in the ^{1}H NMR spectrum (Figure 3.7) the signal of the six hydrogen atoms on the two methyl groups was split into a doublet as a result of the coupling with the phosphorus atom ($J_{P\text{-}H}$ 13.8).

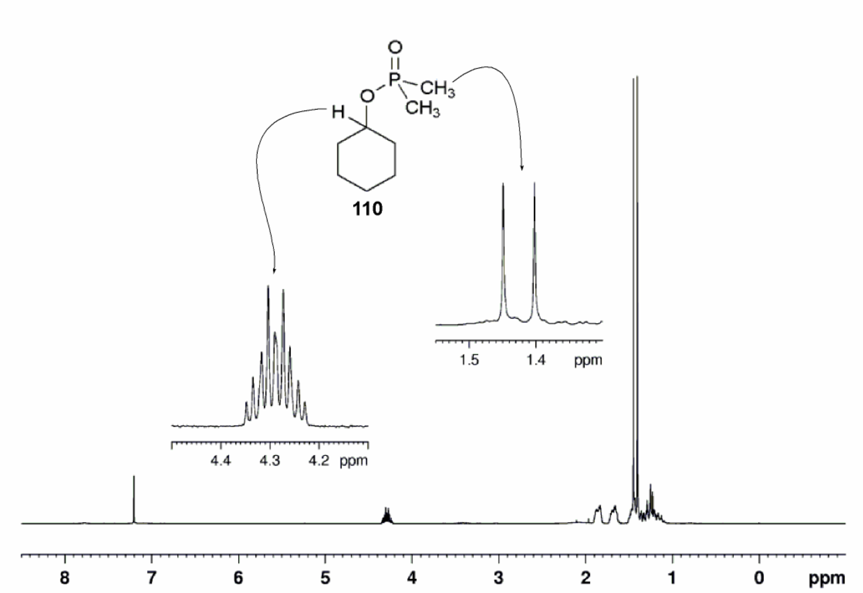


Figure 3.7. ^{1}H NMR spectrum of cyclohexyl dimethylphosphinate **110**.

The analysis of the ^{13}C NMR spectrum (Figure 3.8) showed the expected couplings of the carbon atoms with the phosphorus atom: the *C*-1 position carbon atom is coupled (^{2}J constant) with the phosphorus atom; the *C*-2 and *C*-6 position carbon atoms are coupled (^{3}J constant) with the phosphorus atom; the large ^{1}J constant confirms the two methyl groups bonded to the phosphorus atom. The ^{31}P NMR spectrum shows one signal (δ_P 52.0), which correlates well with data for a similar compound.[120]

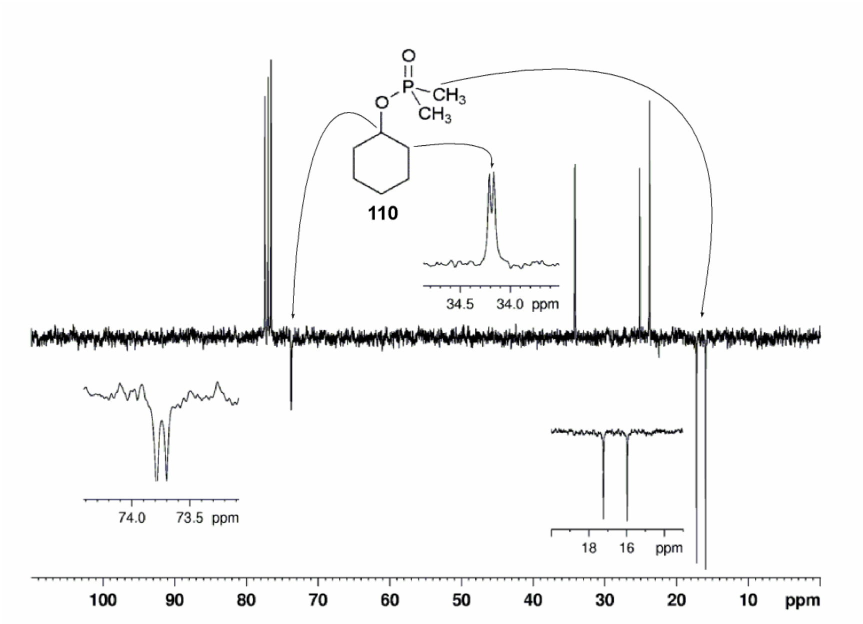
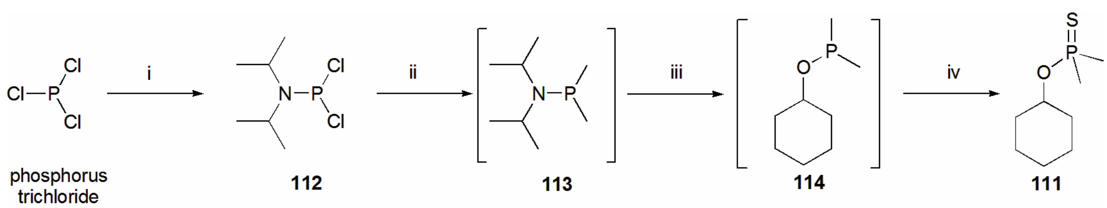


Figure 3.8. ^{13}C NMR spectrum of cyclohexyl dimethylphosphinate **110**.



Scheme 3.8. Synthesis of cyclohexyl dimethylphosphinothioate **111**. *Reagents and conditions:* **i.** *N,N*-Diisopropylamine (2.0 equiv), Et_2O, - 10°C to RT, 73% yield. **ii.** MeLi (3.1 equiv), Et_2O, - 78 °C to RT. **iii.** Cyclohexanol (0.5 equiv), imidazole (2.0 equiv), CH_2Cl_2, - 78 °C to RT. **iv.** Molecular sulfur (2.0 equiv), CH_2Cl_2, RT. Yield over **ii**, **iii** and **iv** steps 53%.

The cyclohexyl dimethylphosphinothioate **111** was synthesised following the same synthetic route used for the synthesis of compound **110**. Starting from phosphorus trichloride, the presumed cyclohexyl dimethylphosphinite **114** was prepared and oxidised *in situ* using two equivalents of molecular sulfur (Scheme 3.8). In the ^{1}H NMR spectrum (Figure 3.9) the six methyl group hydrogen atoms were coupled with the phosphorus atom ($^{2}J_{P-H}$ 13.3).

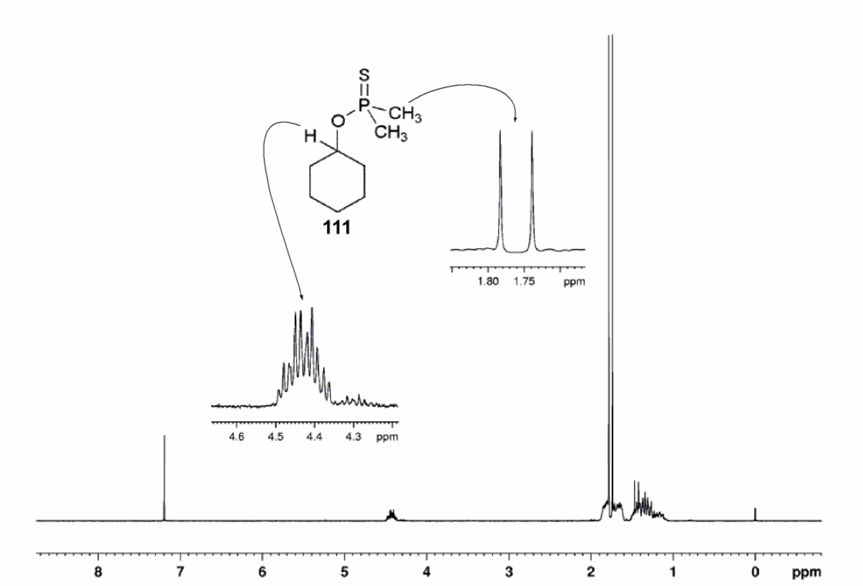


Figure 3.9. ^{1}H NMR spectrum of cyclohexyl dimethylphosphinothioate **111**.

The ^{13}C NMR spectrum showed the following couplings (Figure 3.9). The *C*-1 position (^{2}J constant), *C*-2 position and *C*-6 position (^{3}J constant) carbon atoms were coupled with the phosphorus atom, and the two methyl groups carbon atoms were coupled with a ^{1}J value of 75.0 Hz. The ^{31}P NMR spectrum shows one signal (δ_P 91.0). These data are in good agreement with the literature values.[121,122]

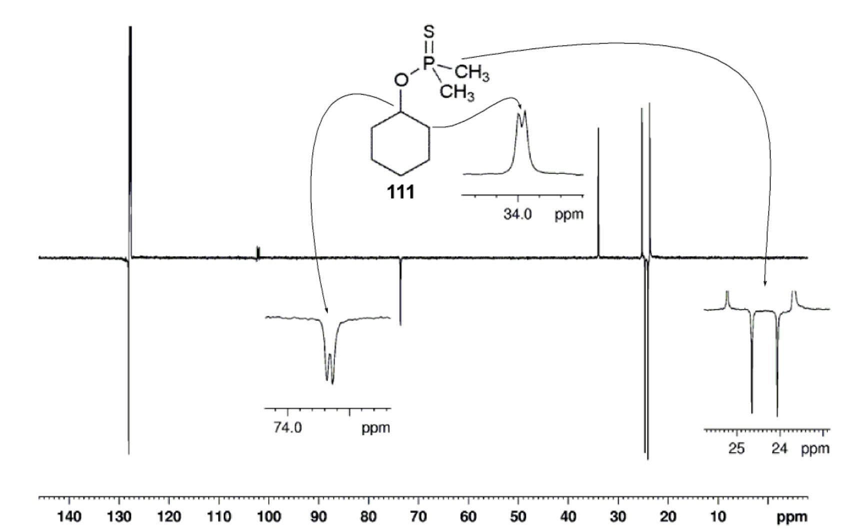
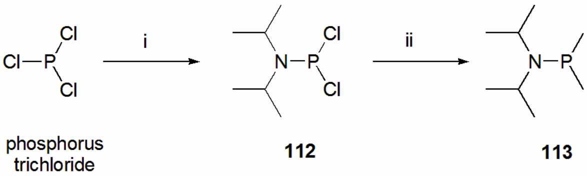


Figure 3.10. ^{13}C NMR spectrum of cyclohexyl dimethylphosphinothioate **111**. The signal at δ_C 128.0 was assigned to of $C_6D_XH_Y$, present as contaminant of the locking solvent C_6D_6.

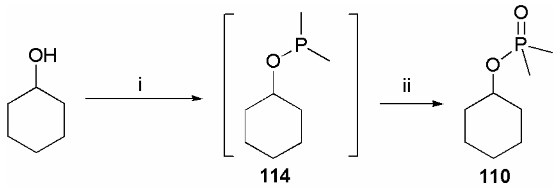
3.4.2. Phosphinylation using a pre-synthesised phosphinylating reagent

The above method for the installation of the dimethylphosphinyl functional group proved to be efficient on simple alcohols such as cyclohexanol; however, it was envisaged that the use of an excess of methyl lithium could limit the application of the method to inositol intermediates containing functional groups sensitive to such strong bases. Therefore, a milder and more general procedure for the phosphinylation of alcohols was developed. This involved the synthesis and purification of compound **113** following a literature procedure (Scheme 3.9).[123]



Scheme 3.9. Synthesis of Diisopropylamino dimethylphosphine **113**. *Reagents and conditions:* **i.** *N,N*-Diisopropylamine (2.0 equiv), Et_2O, - 10 °C to RT, 73% yield. **ii.** Methyl magnesium bromide (3.0 equiv), Et_2O, - 78 to RT, 1h, 58% yield.

Compound **113** was prepared as described above by treating phosphorus trichloride with *N,N*-diisopropylamine. Dialkylation of **112** with methyl magnesium bromide in diethyl ether afforded, after Kugelrohr distillation under inert atmosphere, pure diisopropylamino dimethylphosphine **113** (δ_P 8.3).



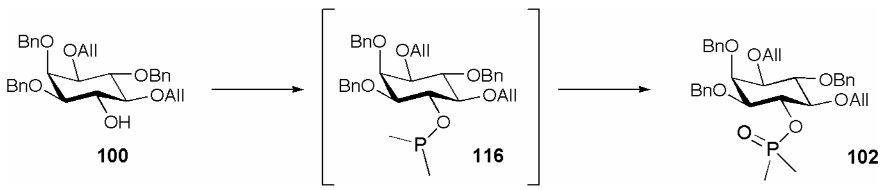
Scheme 3.10. Synthesis of cyclohexyl dimethylphosphinate **110**. *Reagents and conditions:* **i.** Diisopropylamino dimethylphosphine **113** (2.5 equiv), 1*H*-tetrazole (2.5 equiv), CH_2Cl_2, - 78 °C to RT, 1.5 h. **ii.** *m*CPBA (2.5 equiv), CH_2Cl_2, - 78 °C to RT, 90% yield.

The freshly synthesised compound **113** was used as phosphinylating reagent (Scheme 3.10). Cyclohexanol was added to a solution of the phosphine **113** and 1*H*-tetrazole in dry dichloromethane at - 78 °C. After stirring the resulting mixture at room temperature for 1.5 hours, the presumed intermediate phosphinite **114** was shown to be present in the mixture by ^{31}P NMR analysis (δ_P 112.3). Oxidation of the phosphinite **114** with 3-chloroperoxybenzoic acid gave the phosphinate **110** in 90% yield. Using this procedure it was possible to improve the yield of the phosphinylation reaction.

3.5 Towards the synthesis of *C*-4 position-modified $InsP_3$ analogues

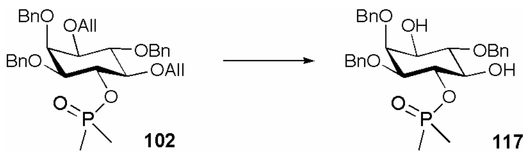
The phosphinylation method described above was used for the installation of the dimethylphosphinyl moiety at the *C*-4 position in the inositol intermediate **100**.

3.5.1. Synthesis of the intermediate *C*-4 position dimethylphosphinyl *myo*-inositol derivative 102



Scheme 3.11. Synthesis of dimethylphosphinate **102**. *Reagents and conditions:* **a.** Diisopropylamino dimethylphosphine (2.5 equiv), 1*H*-tetrazole (2.5 equiv), CH_2Cl_2, RT. **b.** *m*CPBA, CH_2Cl_2, 0 °C to RT, 94% yield.

The phosphinylation procedure described above was used to synthesise compound **102** (Scheme 3.11). Alcohol **100** was added to a solution of diisopropylamino dimethylphosphine **113** and 1*H*-tetrazole in dry dichloromethane at - 78 °C. The reaction was monitored using ^{31}P NMR, which indicated the presence of the presumed intermediate **116** in the reaction mixture (δ_P 130.0). Oxidation with 3-chloroperoxybenzoic acid gave the dimethylphosphinate **102** in high yield.



Scheme 3.12. Synthesis of compound **117**. *Reagents and conditions:* as shown in Table 3.1.

In a first attempt to remove the allyl groups and synthesise the diol **117** (Scheme 3.12), Wilkinson's catalyst was used to isomerise the allyl groups to the correspondent vinyl ether groups (Table 3.1, experiment 1). After heating compound **102** under reflux in the presence of the Wilkinson's catalyst, ^{1}H NMR analysis indicated that a change had occurred in the set of signals for the allyl protons; however, it was not possible to establish whether the allyl groups had been converted to the vinyl ether groups. The crude material obtained after removing the solvent was treated with acetyl chloride in methanol/dichloromethane. TLC analysis indicated the presence of a mixture of compounds more polar than the starting material; the attempted purification by column chromatography failed, and ^{1}H NMR and ^{31}P NMR analysis of the crude mixture indicated that decomposition of the starting material had occurred (lack of the expected signals for the dimethylphosphinyl group). It was proposed that the dimethylphosphinyl moiety may interact with the rhodium atom in the catalyst, leading to undesired side reactions. Consequently, a series of experimental conditions were investigated in order to remove the two allyl groups on compound **102** and synthesise compound **117** (Scheme 3.12 and Table 3.1).

Experiment	Reagents	Solvents	Time, Temperature	Yield
1	**i.** Wilkinson's catalyst, Hunig's base **ii.** Acetyl chloride	**i.** EtOH **ii.** $MeOH/CH_2Cl_2$	**i.** 4 h, reflux **ii.** 3 h, RT	Decomposition of the starting material
2	Pd/C (10%), $TsOH{\cdot}H_2O$	$MeOH/H_2O$ 8/3	20 h, reflux	Allyl removed, product isomerised
3	SmI_2, TEA, H_2O	THF	2 days, RT	No reaction
4	SmI_2, $^{i}PrNH_2$, H_2O	THF	2 days, RT	No reaction
5	Pd/C (10%), $TsOH{\cdot}H_2O$	$MeOH/H_2O$ 8/3	8 h, reflux	No reaction
6	Pd/C (10%), $TsOH{\cdot}H_2O$	$MeOH/H_2O$ 8/3	24 h, 60 °C	21% yield

Table 3.1. Experimental condition investigated for the removal of allyl groups in compound **102**.

Following the procedure recently reported by Chen,[124] compound **102** was dissolved in a mixture of methanol/water and heated under reflux in the presence of palladium on activated carbon and 4-toluenesulfonic acid monohydrate (Table 3.1, experiment 2). According to this procedure, the palladium catalyst would effect the isomerisation of the allyl groups, that would then be cleaved by the solvent under acidic catalysis conditions promoted by the 4-toluenesulfonic acid. After 20 hours the TLC analysis indicated the complete disappearance of the starting material and the presence of a number of more polar compounds. Purification by column chromatography furnished a material that was characterised by ^{1}H NMR and ^{31}P NMR analysis; it was proposed that this material consisted of the two regioisomeric compounds **117** and **118** shown in figure 3.11.

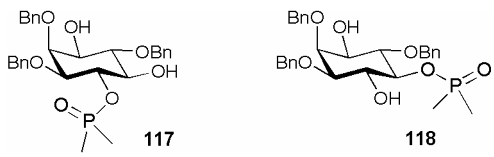


Figure 3.11. Structures of the two presumed regioisomeric compounds **117** and **118**.

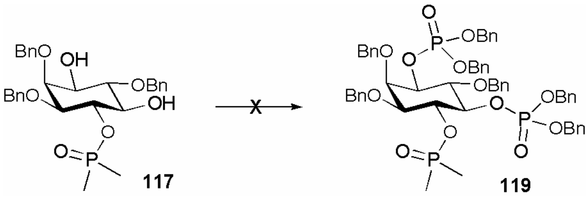
This result was explained by assuming that the prolonged heating in methanol in the presence of the acidic catalyst 4-toluenesulfonic acid monohydrate promoted the isomerisation of compound **117** to compound **118** by intramolecular transesterification of the *C*-4 position dimethylphosphinyl group to the newly formed *C*-5 position hydroxyl group.

Samarium iodide has recently been shown to effect the selective reductive cleavage of unsubstituted allyl protecting groups in carbohydrates.[125] The method seemed to be a mild and effective approach to achieve the synthesis of compound **117**. Compound **102** and dry triethylamine (20 equiv) were dissolved in a 0.1 M solution of samarium iodide (5 equiv) in dry tetrahydrofuran and water (15 equiv) was added in order to initiate the reaction (Table 3.1, experiment 3). After stirring the mixture for two days TLC analysis indicated that no reaction had occurred. The reaction was repeated using the same procedure and conditions but using dry isopropylamine as a base which, according to the literature procedure,[125] should have increased the reaction rate (Table 3.1, experiment 4). After two days the starting material was found to be unreacted by TLC analysis. It was proposed that the reactivity of the samarium iodide reagent could be decreased by interactions with the *C*-4 position dimethylphosphinyl group.

The removal of the allyl groups using the palladium on activated carbon in the presence of 4-toluenesulfonic acid could be the method of choice if it was possible to control and avoid the undesired transesterification of the *C*-4 position dimethylphosphinyl group. It was therefore attempted to carry out the reaction by heating under reflux compound **102** in methanol/water for a period of 8 h (Table 3.1, experiment 5). TLC analysis indicated that no reaction had occurred, suggesting that a prolonged reaction time was needed. The reaction was repeated by heating the methanol/water mixture to 60 °C for a period of 24 hours (Table 3.1, experiment 6). TLC analysis indicated that the starting material had been completely consumed and that a number of more polar compounds were present. Purification by column chromatography afforded the crude diol **117** in 21% yield.

Although the above described method furnished compound **117** in low yield (Table 3.1, experiment 6), it was decided to attempt the following step consisting in the phosphitylation and oxidation of diol **117** to compound **119** (Scheme 3.13).

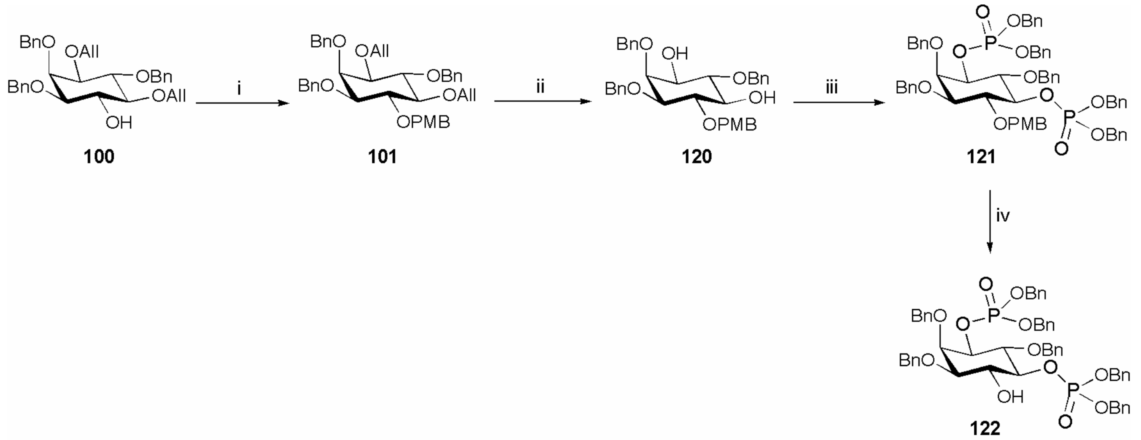
3.5.2. Towards the synthesis of the *C*-4 position *myo*-inositol intermediate 119 - Method A



Scheme 3.13. Attempted phosphitylation and oxidation of compound **117**. *Reagents and conditions:* **a.** Bis(benzyloxy)-*N,N*-diisopropylamino phosphine (5.0 equiv), 1*H*-tetrazole (5.0 equiv), CH_2Cl_2, RT. **b.** *m*CPBA (5.0 equiv), - 78 °C to RT. A complex mixture of compound was obtained instead of the desired compound **119**.

In order to install the two phosphate groups on intermediate **117** the well established phosphoramidite chemistry was employed. Compound **117** dissolved in dichloromethane was added to a mixture of the phosphitylating reagent bis(benzyloxy)-*N,N*-diisopropylamino phosphine and 1*H*-tetrazole (Scheme 3.13). After oxidation with 3-chloroperoxybenzoic acid, TLC analysis of the reaction mixture indicated the presence of a number of compounds. Purification by column chromatography furnished a compound that was analysed by ^{1}H NMR and ^{31}P NMR. The analysis indicated the obtained material was constituted of a mixture of at least two compounds; these compounds could be isomers of either the starting material **117** or the desired product **119**, or compounds deriving from the partial phosphitylation and oxidation of compound **117**. One explanation for the described experimental outcome was given by considering that acidic catalyst 1*H*-tetrazole used in the reaction could promote the transesterification of the *C*-4 position dimethylphosphinyl moiety with the neighbouring hydroxyl groups.

3.6 Synthesis of the key intermediate (-)-1D-2,3,6-tris-*O*-benzyl-*myo*-inositol 1,5-bis(dibenzylphosphate) 122



Scheme 3.14 Synthesis of compound **122.** *Reagents and conditions:* **i.** NaH (1.1 equiv), PMBCl (1.1 equiv), TBAI (0.05 equiv), DMF, 0 °C to RT, 95% yield. **ii. a.** Wilkinson's catalyst (0.4 equiv), BuLi (1.6 equiv), THF, reflux, 7 h. **b.** AcCl (0.6 equiv), CH_2Cl_2/MeOH (3/2), RT, 89% yield. **iii. a.** Bis(benzyloxy)-*N,N*-diisopropylamino phosphine (5.0 equiv), 1*H*-tetrazole (5.0 equiv), CH_2Cl_2, RT. **b.** *m*CPBA (5.0 equiv), - 78 °C to RT, 75% yield. **iv.** CAN (6.0 equiv), MeCN/H_2O (4/1), RT, 2 h, 73% yield.

As described above, the installation of the dimethylphosphinyl moiety at the *C*-4 position in intermediate **100** introduced a series of problems related to the stability of this group towards the experimental conditions to be used in the following synthetic steps. It was necessary, therefore, to make use of a protecting group at the *C*-4 position as described in the retrosynthetic Scheme 3.1 (*vide supra*). The synthesis of the key intermediate **122** was achieved from compound **100** in four steps (Scheme 3.14). Protection of the *C*-4 position hydroxyl group in compound **100** with 4-methoxybenzyl chloride afforded intermediate **101** in high yield. The following removal of the two allyl groups in compound **101** using the Wilkinson's catalyst method furnished the diol **120** in moderate yield (Table 3.2, experiment 1). In order to find the optimal reaction conditions for the removal of the two allyl groups in intermediate **101** and improve the reaction yields, a number of different methods were investigated (Table 3.2).

Experiment	Reagents	Solvents	Time, Temperature	Yield
1	**i.** Wilkinson's catalyst, Hunig's base **ii.** Acetyl chloride	**i.** EtOH **ii.** $MeOH/CH_2Cl_2$	**i.** 3 h, reflux **ii.** 3 h, RT	44%
2	Pd/C (10%), $TsOH{\cdot}H_2O$	$MeOH/H_2O$ 4/1	3 h, reflux	32%
3	Pd/C (10%)	$MeOH/H_2O$ 4/1	15 h, reflux	Decomposition of the starting material
4	**i.** KO^tBu **ii.** Acetyl chloride	**i.** Dry DMSO **ii.** $MeOHl/CH_2Cl_2$	**i.** 3.5 h, reflux **ii.** 3 h, RT	20%
5	**i.** Wilkinson's catalyst, BuLi **ii.** Acetyl chloride	**i.** THF **ii.** $Methanol/CH_2Cl_2$	**i.** 6 h, reflux **ii.** 3 h, RT	89%

Table 3.2. Investigation of different experimental condition for the removal of the allyl groups in compound **101**

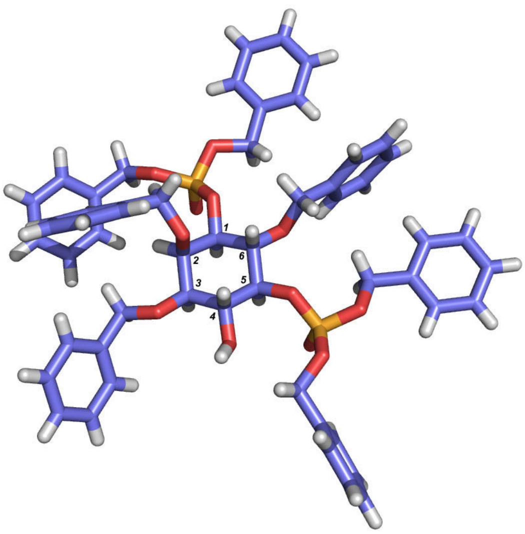
The method described by Chen[124] using palladium on activated carbon in the presence of 4-toluenesulfonic acid was developed to remove allyl groups in inositol intermediates containing one or more 4-methoxybenzyl groups, therefore seemed to be ideal for the removal of the two allyl groups in compound **101**. This procedure furnished the desired compound **120** in 32% yield (Table 3.2, experiment 2). The reaction was complete in three hours, and taking into consideration that the 4-methoxybenzyl protecting group is known to be unstable in acidic environments, the low yield could be due to the decomposition of either the starting material or the reaction product.

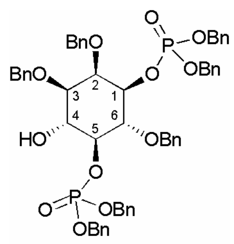
The above method[124] was modified by removing the 4-toluenesulfonic acid from the reaction mixture (Table 3.2, experiment 3), the rationale being that the palladium catalyst would isomerise the allyl groups to the vinyl ether group, which could then be removed by using milder reaction conditions. The reaction was monitored by TLC analysis and reached completion after 15 hours. ^1H NMR analysis of the resulting material revealed loss of the signals for the allyl protons, as well as those for the aromatic protons, indicating that complete decomposition of the starting material had occurred. This result was explained assuming that the palladium catalyst in the presence of the protic solvents methanol and water had effected the reductive cleavage of the protecting groups on the inositol ring.

Gigg[126] reported the isomerisation of allyl groups by using a strong hindered base, such as potassium *tert*-butoxide. The reaction was attempted by heating compound **101** to reflux in the presence of potassium *tert*-butoxide (Table 3.2, experiment 4). ^1H NMR indicated that the allyl groups had isomerised, and the resulting material was treated with acetyl chloride in methanol/dichlorometane to effect the methanolysis of the vinyl ether groups, furnishing compound **120** in 20% yield. These reaction conditions were judged to be too harsh, therefore this procedure was abandoned.

The isomerisation of the allyl groups using Wilkinson's catalyst provided the best results, although the yields were moderate (Table 3.2, experiment 1). One known drawback of Wilkinson's catalyst promoted isomerisation of allyl groups is that the allyl ethers are partially reduced to the propyl ethers, which are unreactive towards the acidic methanolysis necessary to unveil the hydroxyl groups. This could explain the moderate yield obtained in the experiment 1 shown in Table 3.2. According to the procedure previously described by Boons,[127] treatment of the Wilkinson's catalyst with *n*-butyl lithium furnishes a catalyst that effects the isomerisation of allyl groups to the corresponding vinyl ether groups without any detectable trace of the reduced propyl ether by-products. The Wilkinson's catalyst was therefore pre-treated with *n*-butyl lithium and then used to isomerise the allyl groups in compound **101**. ^{1}H NMR analysis indicated complete isomerisation of the allyl groups, and the following removal of the intermediate vinyl ethers furnished the desired diol **120** in 89% yield.

Having found a high-yielding procedure for the synthesis of compound **120**, it was phosphitylated and oxidised to furnish intermediate **121** in good yield, which was in turn treated with ceric ammonium nitrate in acetonitrile/water to give the desired key intermediate **122** in 73% yield (Scheme 3.14). The structure and absolute stereochemistry of compound **122** was confirmed by X-ray crystallography (Figure 3.11).





122

Figure 3.11. A PyMOL (www.pymol.org) representation of the X-ray crystal structure of compound **122.**

3.7 Synthesis of *C*-4 position-modified $InsP_3$ analogues

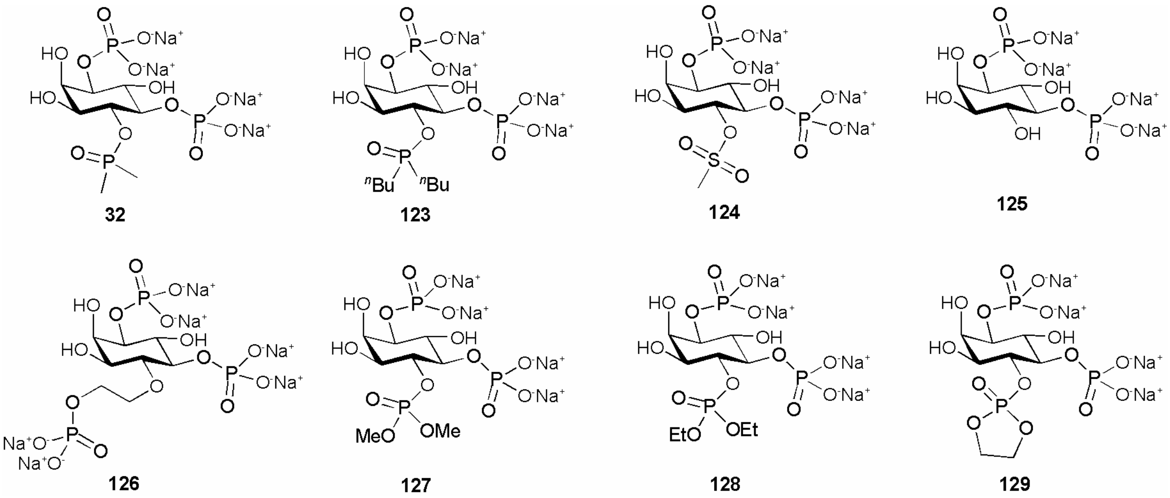
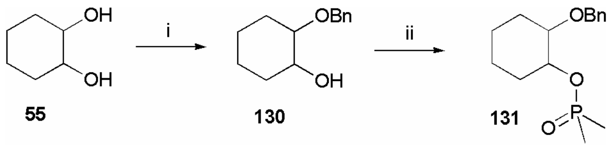


Figure 3.13. Structures of the *C*-4 position-modified $InsP_3$ analogues to be synthesised.

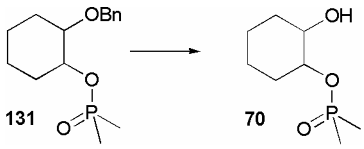
The key intermediate **122** represents a versatile compound, as it allows the synthesis of a series of *C*-4 position-modified $InsP_3$ analogues. Analysis of the $InsP_3R1$ binding domain crystal structure indicates that the introduction of a moiety approximating the geometry of a phosphate group but with reduced hydrogen-bonding capabilities may lead to compounds that are able to antagonise the $InsP_3Rs$. Figure 3.13 shows the target compounds to be synthesised in order to assess the structural requirements for the optimum antagonist activity at the $InsP_3Rs$. The dimethylphosphinyl compound **32**, the di-*n*-butylphosphinyl compound **123**, the three phosphoryl compounds **127**, **128** and **129** and the mesyl compound **124** approximate the geometry of the *C*-4 position phosphate group of $InsP_3$, but possess different electronic distribution and steric bulkiness and will therefore provide information about the structural requirements needed for achieving a inhibitory activity at the $InsP_3Rs$. Compound **125** represents the simplest *C*-4 position-modified $InsP_3$ analogue and will provide basic information on the effect of removing most of the hydrogen bonding interactions at the *C*-4 position. Compound **126** will be synthesised in order to investigate whether a phosphate group positioned away from the inositol ring can be used to lock the α- and β-domains at the $InsP_3$ binding site in the opened position.

3.7.1. Model studies on the stability of the dimethylphosphinyl group towards the hydrogenolysis reaction



Scheme 3.15. Synthesis of the model compound **131**. *Reagents and conditions:* **i.** NaH (1.1 equiv), BnBr (1.1 equiv), THF, 0 ºC to RT, 40% yield; **ii. a.** Diisopropylamino dimethylphosphine (2.5 equiv), 1*H*-tetrazole (2.5 equiv), CH_2Cl_2, RT. **b.** *m*CPBA, 0 °C to RT, 89% yield.

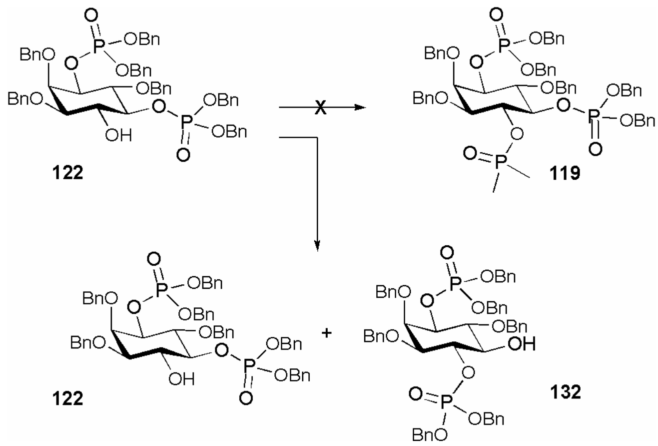
In view of the forthcoming synthesis of compound **32**, it was decided to assess the stability of the dimethylphosphinyl moiety towards the experimental conditions previously used for the hydrogenolysis of benzyl groups. Model compound **131** was synthesised in two steps starting from (±)-1,2-*trans*-dihydroxycyclohexane **55** (Scheme 3.15). The benzyl protection of one of the two hydroxyl groups furnished the alcohol **130** that was phosphinylated by using diisopropylamino dimethylphosphine and oxidised to give compound **131** in high yield.



Scheme 3.16. The hydrogenolysis of the benzyl group in model compound **131**. *Reagents and conditions:* H_2, Pd black (20.0 equiv), $NaHCO_3$ (4.0 equiv), ^{t}BuOH/H_2O (6/1), RT, 7 h, 92% yield.

The hydrogenolysis of the benzyl group in model compound **131** proceeded smoothly furnishing compound **70** in 92% yield (Scheme 3.16); the sodium hydrogen carbonate present in the mixture had no effect on the dimethylphosphinyl moiety, confirming the efficacy of the method.

3.7.2. Towards the synthesis of the *C*-4 position *myo*-inositol intermediate 119 - Method B

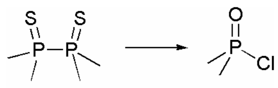


Scheme 3.17. Attempted synthesis of compound **119**. *Reagents and conditions:* **a.** Diisopropylamino dimethylphosphine **113** (2.5 equiv), 1*H*-tetrazole (2.5 equiv), CH_2Cl_2, RT. **b.** *m*CPBA, CH_2Cl_2, 0 °C to RT. It is thought that the reaction led to the partial isomerisation of starting material to regioisomer **132**.

The previously developed phosphinylation method was used to install the dimethylphosphinyl group at the *C*-4 position in compound **122**. Intermediate **122** was added to a mixture of diisopropylamino dimethylphosphine **113** and 1*H*-tetrazole in dichloromethane (Scheme 3.17). After 15 h the ^{31}P NMR analysis indicated that the signal for the intermediate phosphinite (expected to be in the region of δ_P 130-100, as seen in the similar intermediate **116**, Scheme 3.11) was not present. TLC analysis revealed the presence of a small amount of starting material and a less polar compound. The mixture was treated with 3-chloroperoxybenzoic acid and purification by column chromatography afforded a 17% of the starting material and a 33% of the less polar compound. ^{1}H NMR and ^{31}P NMR analysis indicated that this material could have the structure of compound **132** (Scheme 3.17). Mass spectrometry analysis was also consistent with the proposed structure [*m/z* (ES+) 993 $(M+Na)^+$]. The absence of reaction could be explained by assuming that the bulky phosphinylating reagent could not react with the *C*-4 position hydroxyl group because of the steric hindrance of the *C*-5 position phosphate group. The isomerisation of compound **122** to compound **132** could be ascribed to an acidic catalysed transesterification reaction catalysed by the 1*H*-tetrazole, although it is possible that the phosphinylating species could be responsible of promoting the isomerisation reaction.

3.7.3. Synthesis of (+)-1D-4-*O*-dimethylphosphinyl-*myo*-inositol 1,5-bisphosphate (sodium salt) 32

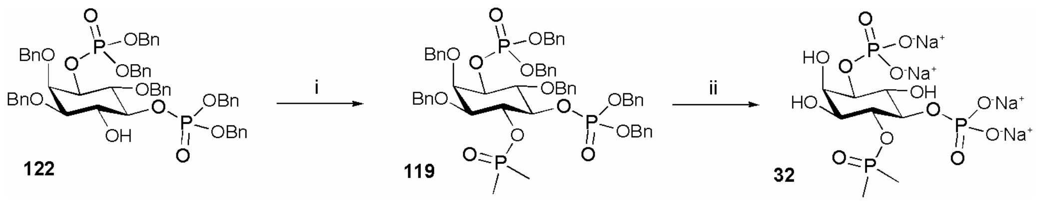
The developed phosphinylation method using the reagent diisopropylamino dimethylphosphine **113** failed when applied to compound **122** (Scheme 3.17). Ramage[128] reported the use of dialkyl phosphinates as protecting groups in peptide synthesis. The procedure used to install such protecting groups involved the synthesis of a highly reactive dialkyl phosphinic chloride and its reaction with the compound to be protected in the presence of a base.



133 **134**

Scheme 3.18. Synthesis of dimethylphosphinic chloride **134**. *Reagents and conditions:* Thionyl chloride (4.8 equiv), toluene, 0 °C to RT, then reflux, 1.5 h, 59% yield.

Following the procedure described by Ramage,[128] tetramethyl diphosphine disulfide **133** was treated with thionyl chloride in toluene to give, after purification by Kugelrohr distillation under inert atmosphere, the desired dimethylphosphinic chloride **134** (Scheme 3.18).



Scheme 3.19. Synthesis of (+)-1D-4-*O*-dimethylphosphinyl-*myo*-inositol 1,5-bisphosphate (sodium salt) **32**. *Reagents and conditions:* **i.** Dimethylphosphinic chloride (4.0 equiv), 2,6-lutidine (5.0 equiv), DMF, - 42 °C to RT, 22 h, 76% yield. **ii.** H_2, Pd black (20.0 equiv), $NaHCO_3$ (4.0 equiv), ^{t}BuOH/H_2O (6/1), RT, 7 h, 93% yield.

The dimethylphosphinic chloride reagent **134** was reacted with compound **122** in the presence of 2,6-lutidine to afford compound **119** in good yield (Scheme 3.19). The final hydrogenolysis of the benzyl groups was achieved by using palladium black in the presence of sodium hydrogen carbonate as previously described. The reaction afforded the final compound *(+)-1D-4-O-dimethylphosphinyl-myo-inositol 1,5-bisphosphate (sodium salt)* **32** in excellent yield.

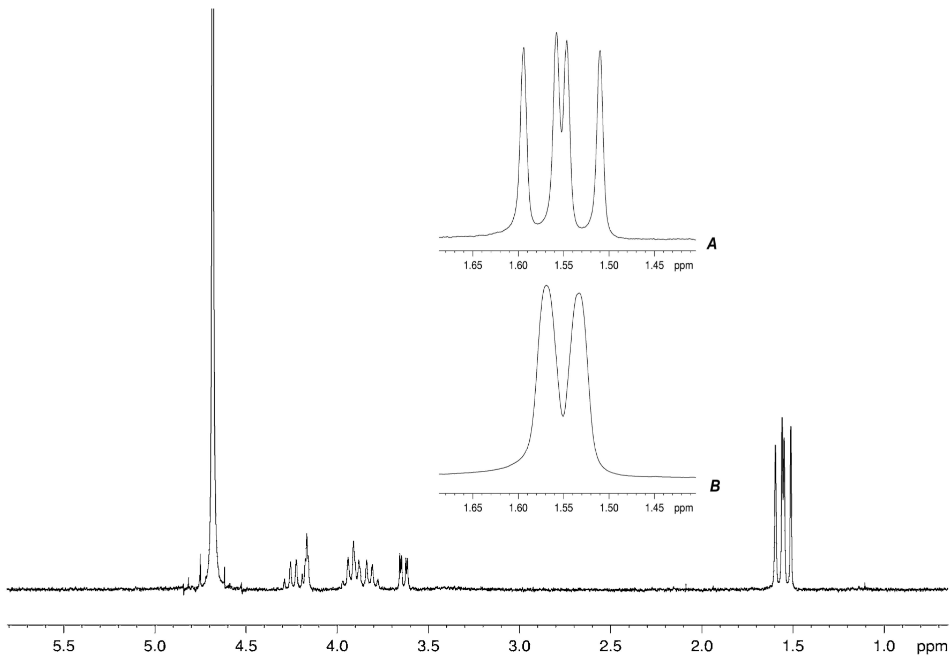


Figure 3.14. ^{1}H NMR spectrum of compound **32**. ***A*** - Signals for the two methyl groups of the *C*-4 position dimethylphosphonate moiety. Each methyl group signal is split in a doublet by the neighbouring phosphorus atom. ***B*** - ^{31}P-decoupled ^{1}H NMR spectrum of compound **32**, showing the signals for the two methyl groups of the *C*-4 position dimethylphosphonate moiety. The coupling with the neighbouring phosphorus atom has been removed by the decoupling sequence.

Figure 3.14 is shown the ^{1}H NMR spectrum of compound **32**. The expansion ***A*** shows the two doublets for the two diastereotopic methyl groups of the *C*-4 position dimethylphosphonate moiety. The expansion ***B*** shows the signals for the two methyl groups as they appear in the ^{31}P-decoupled ^{1}H NMR spectrum of compound **32**. The couplings of the ^{1}H nuclei with the neighbouring ^{31}P nucleus have been removed by the decoupling sequence.

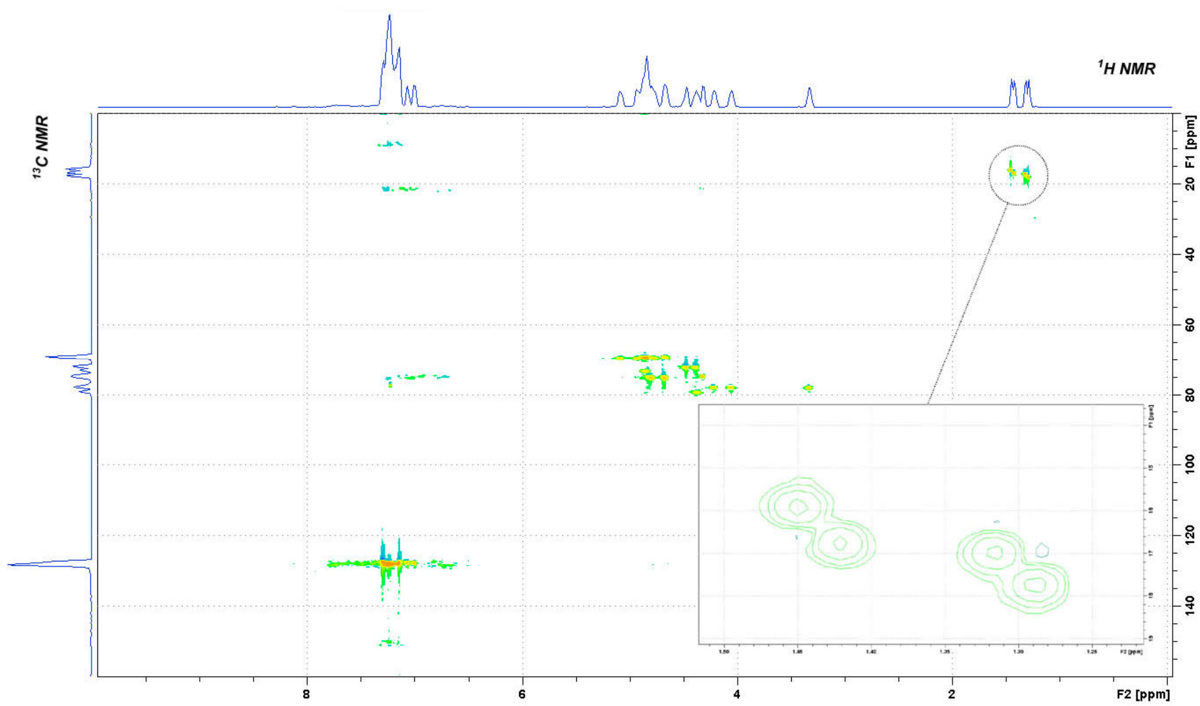
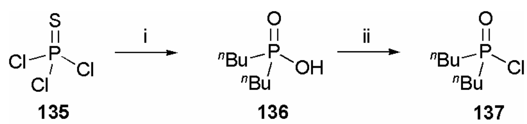


Figure 3.15. HSQC 2D-spectrum of compound **32**. The expansion shows the signals for the two methyl groups of the *C*-4 position dimethylphosphonate moiety. In the magnified area are shown the correlation signals between ^{1}H and ^{13}C nuclei of the two methyl groups.

Figure 3.15 shows the heteronuclear single quantum correlation (HSQC) spectrum of compound **32**. This technique allowed the assignment of the $^{1}J_{CP}$ constants for the *C*-4 position dimethylphosphinyl moiety by transferring the known ^{1}H nuclei assignments onto the ^{13}C nuclei.

3.7.4. Synthesis of (-)-1D-4-*O*-di-*n*-butylphosphinyl-*myo*-inositol 1,5-bisphosphate (sodium salt) 123

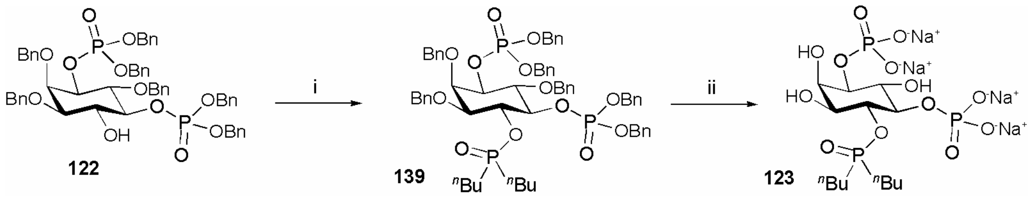
The bulky *C*-4 position di-*n*-butylphosphinyl $InsP_3$ analogue was synthesised in two steps from intermediate **122** using the di-*n*-butylphosphinic chloride reagent **137** (Scheme 3.21).



Scheme 3.20. Synthesis of di-*n*-butylphosphinic choride **137**. *Reagents and conditions:* **i. a.** *n*-Butylmagnesium bromide (4.0 equiv), Et_2O, 0 °C to RT, then reflux, 1 h. **b.** HNO_3 (30%), 0 °C to RT, then 70 °C, 1 h, 31% yield. **ii.** Thionyl chloride, toluene, 0 °C to RT, then reflux, 30 min, 84% yield.

Reagent **137** was synthesised in two steps from thiophosphoryl chloride **135** (Scheme 3.20).[128,129] The starting material was reacted with the freshly prepared

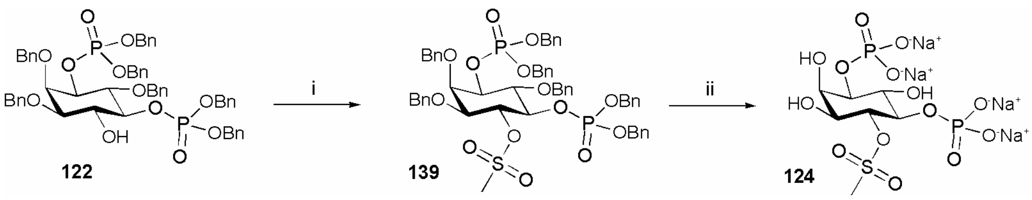
Grignard reagent *n*-butylmagnesium bromide to furnish a mixture of compounds. This material could be directly treated with thionyl chloride and converted to compound **137**;[129] however, this procedure was not used as the by-products that could be present in the mixture could lead, over the treatment with thionyl chloride, to the undesired compound di-*n*-butylphosphinothioyl chloride, which would be difficult to separate from the desired compound **137**.[128] Therefore, the mixture obtained from the reaction of compound **135** with the Grignard reagent was oxidised using nitric acid. During the oxidation step the by-products are converted into the di-*n*-butylphosphinic acid **136**. Chlorination of the pure compound **136** with thionyl chloride affords, after vacuum distillation, the desired reagent **137** in 84% yield.



Scheme 3.21. Synthesis of (+)-1D-4-*O*-di-*n*-butylphosphinyl-*myo*-inositol 1,5-bisphosphate (sodium salt) **123**. *Reagents and conditions:* **i.** Di-*n*-butylphosphinic chloride (4.0 equiv), TEA (5.0 equiv), DMAP (catalytic amount), DMF, - 42 °C to RT, 15 h, 65% yield. **ii.** H_2, Pd black (20.0 equiv), $NaHCO_3$ (4.0 equiv), $^tBuOH/H_2O$ (10/1), RT, 8 h, 95% yield.

Compound **123** was synthesised in two steps from the intermediate **122** (Scheme 3.21). The freshly synthesised di-*n*-butylphosphinic chloride **137** was reacted with compound **122** in the presence of triethylamine and 4-dimethylaminopyridine to give the intermediate **139** in 65% yield; hydrogenolysis in the presence of sodium hydrogen carbonate furnished the final compound *(+)-1D-4-O-di-n-butylphosphinyl-myo-inositol 1,5-bisphosphate (sodium salt)* **123** in high yield.

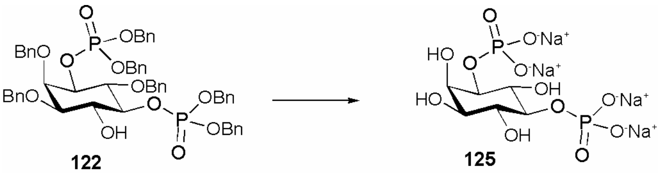
3.7.5. Synthesis of (+)-1D-4-*O*-methylsulfonyl-*myo*-inositol 1,5-bisphosphate (sodium salt) 124



Scheme 3.22. Synthesis of (+)-1D-4-*O*-methylsulfonyl-*myo*-inositol 1,5-bisphosphate (sodium salt) **124**. *Reagents and conditions:* **i.** Methanesulfonyl chloride (4.0 equiv), TEA (5.0 equiv), DMAP (catalytic amount), CH_2Cl_2, 0 °C to RT, 2 days, 56% yield. **ii.** H_2, Pd black (20.0 equiv), $NaHCO_3$ (4.0 equiv), $^tBuOH/H_2O$ (10/1), RT, 8 h, 91% yield.

The synthesis of compound **124** was achieved in two steps from the intermediate **122** (Scheme 3.22). Methanesulfonyl chloride was reacted with compound **122** in the presence of triethylamine and 4-dimethylaminopyridine, furnishing the desired compound **139** in 56% yield. The hydrogenolysis of the benzyl protecting groups in the presence of sodium hydrogen carbonate gave the final compound *(+)-1D-4-O-methylsulfonyl-myo-inositol 1,5-bisphosphate (sodium salt)* **124** in 91% yield.

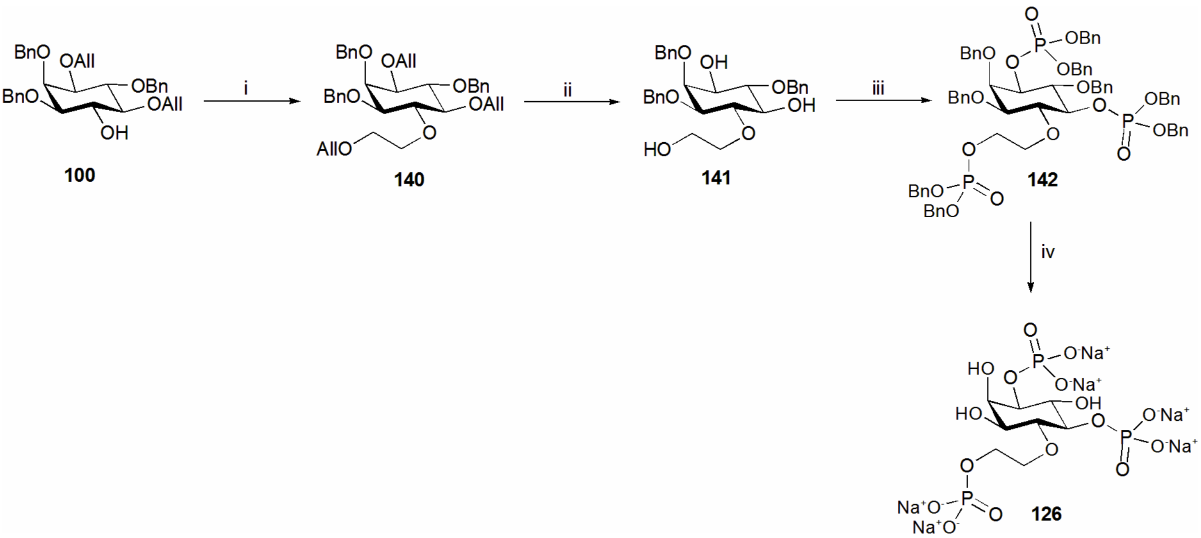
3.7.6. Synthesis of (+)-1D-*myo*-inositol 1,5-bisphosphate (sodium salt) 125



Scheme 3.23. Synthesis of (+)-1D-*myo*-inositol 1,5-bisphosphate (sodium salt) **125**. *Reagents and conditions:* H_2, Pd black (20.0 equiv), $NaHCO_3$ (4.0 equiv), ^{t}BuOH/H_2O (5/1), RT, 8 h, 92% yield.

The synthesis of compound **125** was achieved from intermediate **122**. The hydrogenolysis of the benzyl protecting groups by hydrogenolysis in the presence of sodium hydrogen carbonate furnished (+)-1D-*myo*-inositol 1,5-bisphosphate (sodium salt) **125** in 92% yield (Scheme 3.23).

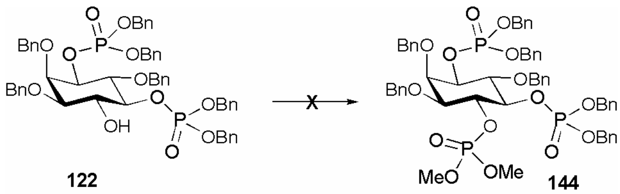
3.7.7. Synthesis of (-)-1D-4-*O*-(2-phosphoryloxy)ethyl-*myo*-inositol 1,5-bisphosphate (sodium salt) 126



Scheme 3.24. Synthesis of (-)-1D-4-*O*-(2-phosphoryloxy)ethyl-*myo*-inositol 1,5-bisphosphate (sodium salt) **126**. *Reagents and conditions:* **i.** NaH (1.2 equiv), 2-allyloxyethyl bromide **143** (1.2 equiv), TBAI (catalytic amount), DMF, 0 °C to RT, 15 h, 80% yield. **ii. a.** Wilkinson's catalyst (0.1 equiv), BuLi (0.4 equiv), THF, reflux, 6 h. **b.** AcCl (0.6 equiv), CH_2Cl_2/MeOH (3/2), RT, 80% yield. **iii. a.** Bis(benzyloxy)-*N,N*-diisopropylamino phosphine (7.5 equiv), 1*H*-tetrazole (7.5 equiv), CH_2Cl_2, RT. **b.** *m*CPBA (7.5 equiv), - 78 °C to RT, 46% yield. **iv.** H_2, Pd black (20.0 equiv), $NaHCO_3$ (6.0 equiv), ^{t}BuOH/H_2O (5/1), RT, 8 h, 89% yield.

Compound **126** was synthesised in four steps from intermediate **100** (Scheme 3.24). Alcohol **100** was treated with sodium hydride and then reacted with freshly synthesised 2-allyloxyethyl bromide **143** to give compound **140** in good yield. Removal of the three allyl protecting groups using Wilkinson's catalyst pre-treated with *n*-butyl lithium furnished the triol **141** in 80% yield. This was phosphitylated using the standard phosphoramidite method and oxidised to intermediate **142**, which was hydrogenolysed in the presence of sodium hydrogen carbonate to give the final compound *(-)-1D-4-O-(2-phosphoryloxy)ethyl-myo-inositol 1,5-bisphosphate (sodium salt)* **126** in 89% yield (Scheme 3.24).

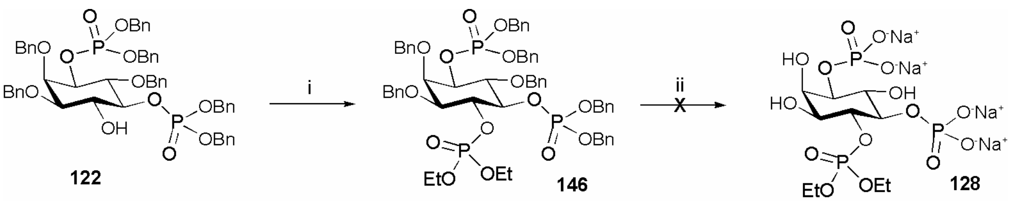
3.7.8. Towards the synthesis of the *C*-4 position dimethylphosphoryl *myo*-inositol derivative 144



Scheme 3.25. The attempted synthesis of compound **144**. Method ***A***: *Reagents and conditions:* Dimethyl chlorophosphate (4.0 equiv), 2,6-lutidine (5.0 equiv), DMAP (catalytic amount), DMF, - 42 °C to RT, 2 days. Method ***B***. *Reagents and conditions:* **a.** Dimethyl chlorophosphite **145** (10.0 equiv), Hunig's base (20.0 equiv), DMF, - 42 °C to RT, 15 h. **b.** *m*CPBA (10.0 equiv), DMF, - 42 °C to RT, 30 min.

The synthesis of the *C*-4 position dimethylphosphoryl compound **144** was first attempted using dimethyl chlorophosphate in the presence of 2,6-lutidine (Scheme 3.25, method A). After 2 days the starting material was found to be unreacted. It is thought that the low reactivity of the phosphorylating reagent dimethyl chlorophosphate prevented the formation of compound **144**. It was therefore decided to use the more reactive reagent dimethyl chlorophosphite **145**. This compound was freshly synthesised and used for the phosphitylation of compound **122** in the presence of Hunig's base (Scheme 3.25, method B). The reaction afforded a mixture of compounds which could not be purified by column chromatography. ^{1}H NMR analysis indicated the presence of non-inositol related impurities, and the ^{31}P NMR spectrum showed both signals not related with those expected for the product, and signals that could correspond to phosphate groups and therefore to the desired product. Since some of the phosphorus-containing impurities were present as contaminants in the ^{31}P NMR spectrum of the dimethyl chlorophosphite reagent **145**, this compound was synthesised again and more carefully purified by vacuum distillation and the preparation of compound **144** further attempted. This second experiment yielded a mixture of compounds displaying ^{1}H NMR and ^{31}P NMR signals similar to those for the mixture obtained from the previous experiment, indicating that the impurities present in chlorophosphite reagent **145** could not be removed by vacuum distillation. It is thought that these impurities could affect the outcome of the phosphinylation reaction of compound **122**.

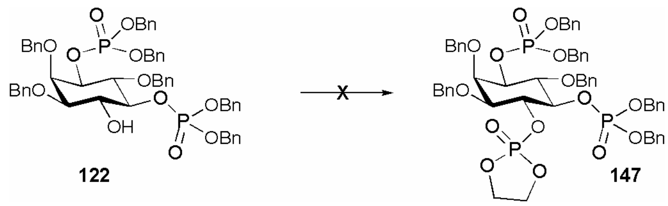
3.7.9. Towards the synthesis of the *C*-4 position diethylphosphoryl $InsP_3$ analogue 128



Scheme 3.26. Attempted synthesis of compound **128**. *Reagents and conditions:* **i. a.** Diethyl chlorophosphite (3.0 equiv), TEA (4.0 equiv), CH_2Cl_2, - 78 °C to RT, 4 h. **b.** *m*CPBA (3.0 equiv), CH_2Cl_2, - 78 °C to RT, 30 min, 52% yield. **ii.** H_2, Pd black (20.0 equiv), $NaHCO_3$ (4.0 equiv), $^{t}BuOH/H_2O$ (8/1), RT, 7 h.

The intermediate **146** was synthesised by phosphitylation and oxidation using diethyl chlorophosphite as phosphitylating reagent. The intermediate **122** was reacted with diethyl chlorophosphite in the presence of triethylamine (Scheme 3.26). TLC analysis after four hours indicated the complete consumption of the starting material and the presence of a less polar compound, which is thought to be the intermediate phosphite. The reaction mixture was treated with 3-chloroperoxybenzoic acid to furnish compound **146** in 52% yield. The removal of the benzyl protecting groups was attempted by using the hydrogenolysis procedure previously described. Treatment of **146** with palladium black in the presence of sodium hydrogen carbonate yielded a material whose ^{1}H NMR spectrum displayed very broad signals; moreover, ^{31}P NMR analysis indicated the presence of a number of signals in the region of the phosphate groups, suggesting that the *C*-4 position diethylphosphate group could have undergone an intramolecular transesterification reaction with the neighbouring hydroxyl group. Although the intermolecular transesterification is less likely to occur because of the steric hindrance of the phosphate groups, it could have also contributed to yielding a mixture of compounds.

3.7.10. Towards the synthesis of the *C*-4 position ethylenephosphoryl *myo*-inositol derivative 147



122 **147**

Scheme 3.27. Attempted synthesis of compound **147**. Method ***A***. *Reagents and conditions:* **i. a.** 2-Chloro-1,3,2-dioxaphospholane (6.0 equiv), TEA (8.0 equiv), CH_2Cl_2, - 78 °C to RT, 15 h. **b.** *m*CPBA (6.0 equiv), CH_2Cl_2, - 78 °C to RT, 30 min. Method ***B***. *Reagents and conditions:* **i. a.** 2-Chloro-1,3,2-dioxaphospholane (15.0 equiv), pyridine, - 42 °C to RT, 15 h. **b.** *m*CPBA (6.0 equiv), CH_2Cl_2, - 78 °C to RT, 30 min.

The synthesis of the *C*-4 position diethylphosphoryl compound **147** was attempted by reacting intermediate **122** with 2-chloro-1,3,2-dioxaphospholane in the presence of triethylamine (Scheme 3.27, method A). After 15 hours TLC analysis could not establish whether the reaction had occurred, and the reaction mixture was treated with 3-choroperoxybenzoic acid. Purification by column chromatography afforded the unreacted starting material. The reaction was attempted again using the 2-chloro-1,3,2-dioxaphospholane and pyridine as both the base and the solvent (Scheme 3.27, method B). The pyridine was removed under reduced pressure after 15 hours and keeping the residue under an inert atmosphere, this was dissolved in dichloromethane and treated with 3-choroperoxybenzoic acid. Purification by column chromatography furnished the unreacted starting material.

3.8 Future work

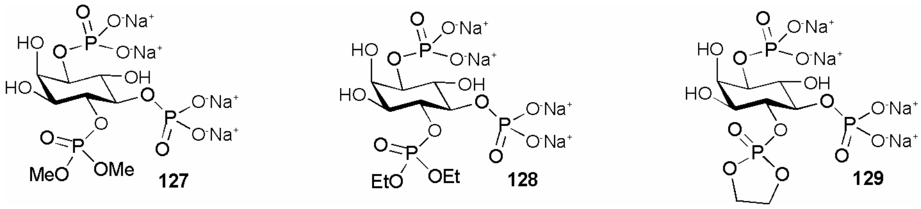
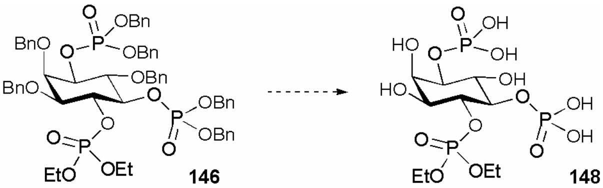


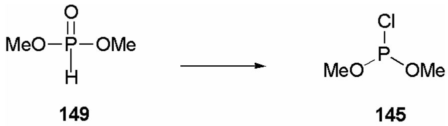
Figure 3.16. Structure of the C-4 position-modified $InsP_3$ analogues compounds to be synthesised.

In order to assess the activity of the *C*-4 position phosphoryl $InsP_3$ analogues at the $InsP_3Rs$, it is intended to complete the synthesis of compounds **127**, **128** and **129** (Figure 3.16).



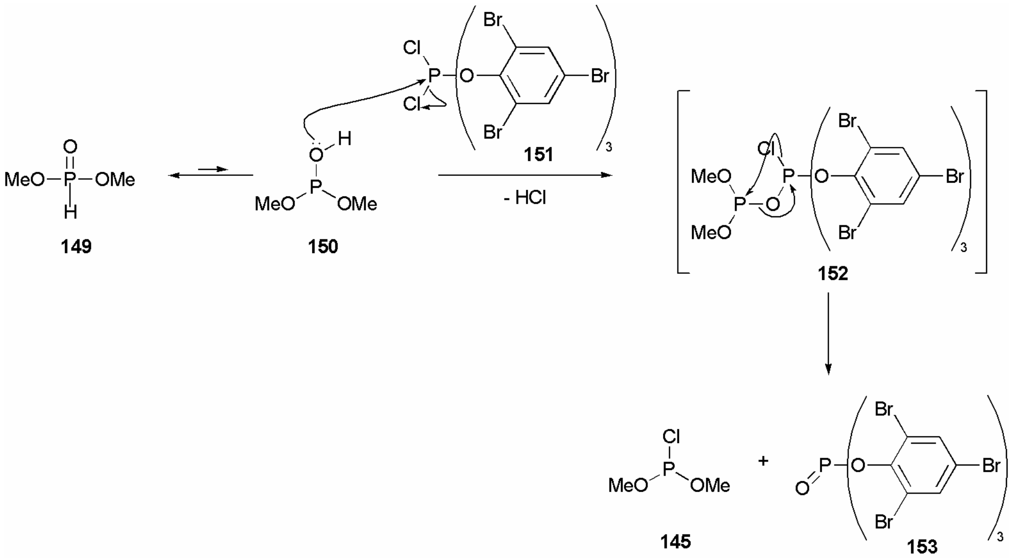
Scheme 3.28. Proposed synthesis of compound **148**. *Reagents and conditions:* H_2, $Pd(OAc)_2$, $Pd(O_2CCF_3)_2$, AcOH, 18 °C.

As previously described, the removal of the benzyl protecting groups from compound **146** using palladium black in the presence of sodium hydrogen carbonate failed to furnish the desired compound **128**; it is thought that a transesterification reaction occurred, shifting the diethylphosphate group around the inositol ring. Tsien[50] reported a method for the removal of benzyl groups in inositol intermediates where the phosphate groups were masked in order to achieve membrane-permeant properties; this method involves the use of palladium acetate and palladium trifluoroacetate as catalysts in glacial acetic as solvent. Performing the reaction at 18 °C it was possible to efficiently remove the benzyl protecting groups from the inositol ring and preserve intact the masked phosphate groups. These reaction condition will be tested on compound **146** to synthesise compound **148** which will be obtained with the phosphate groups in the free-acids form (Scheme 3.28).



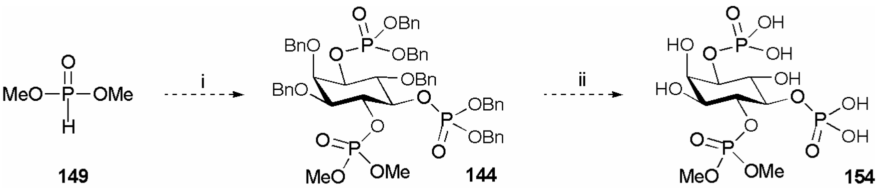
Scheme 3.29. Synthesis of compound **145** (as reported by Hata).[130] *Reagents and conditions:* BDCP **151**, pyridine, RT.

Hata[130] has previously reported a procedure for the preparation of dimethyl chlorophosphite **145** by non-oxidative chlorination of dimethyl hydrogen phosphonate **149** using the reagent tris(2,4,6-tribromophenoxy)dichlorophosphorane (BDCP) (**151**, Scheme 3.29). The method allows the conversion of compound **149** to the chlorophosphite **145** in high yield and avoids the formation of by-products. Scheme 3.30 shows the reaction mechanism proposed by Hata.[130]



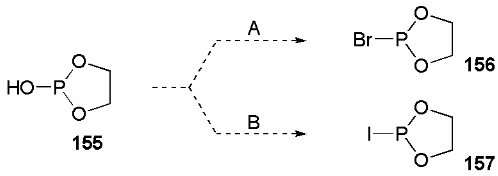
Scheme 3.30. Mechanism of the non-oxidative chlorination of dimethyl hydrogen phosphonate **149** to dimethyl chlorophosphite **145** as reported by Hata.[130]

Dimethyl hydrogen phosphonate exists as a mixture of the two tautomeric form **149** and **150** (Scheme 3.30). Compound **150** reacts with BDCP **151** to yield the intermediate species **152** which collapses to the dimethyl chlorophosphite **145** and the inert compound tris(2,4,6-tribromophenoxy) phosphate **153**.[130]



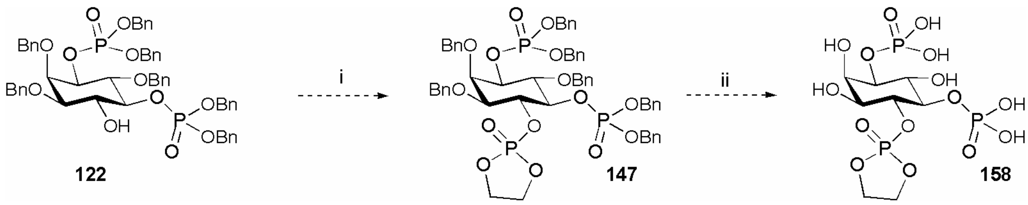
Scheme 3.31. Proposed synthesis of compound **154**. *Reagents and conditions:* **i. a.** BDCP, pyridine, RT. **b. 122**, pyridine, - 42 °C to RT. **c.** *m*CPBA, CH_2Cl_2, - 78 °C to RT. **ii.** H_2, $Pd(OAc)_2$, $Pd(O_2CCF_3)_2$, AcOH, 18 °C.

The method will be used to attempt the synthesis of dimethyl chlorophosphite **145** from dimethyl hydrogen phosphonate; reagent **145** would be generated *in situ*, thus avoiding to introduce impurities in the phosphinylation step of compound **122** (Scheme 3.31); in such a way compound **144** would be purified and rigorously characterised; final hydrogenolysis using palladium acetate and palladium trifluoroacetate in glacial acetic acid at 18 °C would afford compound **154**, with the phosphate groups in the free-acids form (Scheme 3.31).



Scheme 3.32. Synthesis of reagents **156** and **157**. *Reagents and conditions: **A*** - PBr_3, toluene, RT. ***B*** - TMS-I, toluene, RT.

As previously described the phosphinylation of compound **122** using the reagent 2-chloro-1,3,2-dioxaphospholane failed. To overcome this problem a more reactive reagent will be used, that is, 2-bromo-1,3,2-dioxaphospholane **156** or 2-iodo-1,3,2-dioxaphospholane **157** (Scheme 3.32). These compounds could be prepared from ethylene hydrogen phosphite **155** by bromination with phosphorus tribromide or by iodination with trimethylsilyl iodide (Scheme 3.32).[131,132]



Scheme 3.33. Proposed synthesis of compound **158**. *Reagents and conditions:* **i. a.** 2-bromo-1,3,2-dioxaphospholane **156** or 2-iodo-1,3,2-dioxaphospholane **157**, TEA, CH_2Cl_2, - 78 °C to RT. **b.** *m*CPBA, CH_2Cl_2, - 78 °C to RT. **ii.** H_2, $Pd(OAc)_2$, $Pd(O_2CCF_3)_2$, AcOH, 18 °C.

Compound **122** would be phosphitylated and oxidised to the intermediate **147** (Scheme 3.33). The removal of the benzyl protecting groups by hydrogenolysis using palladium acetate and palladium trifluoroacetate in glacial acetic acid at 18 °C would afford compound **158**, with the phosphate groups in the free-acids form (Scheme 3.33).

3.9 Summary and conclusions

The aim of this project of synthesising a series of *C*-4 position-modified $InsP_3$ analogues as pure enantiomers has been achieved.

As a result of the studies towards the synthesis of such compounds, a robust synthetic route starting from *myo*-inositol has been developed. This route has allowed the synthesis of the key intermediate (-)-1D-2,3,6-tris-*O*-benzyl-*myo*-inositol 1,5-bis(dibenzylphosphate) **122** as pure enantiomer. Using this intermediate, the *C*-4 position-modified $InsP_3$ analogues **32**, **109**, **123**, **124**, **125** and **126** shown in Figure 3.17 were synthesised in high yields. These compounds are predicted to act as $InsP_3Rs$ competitive antagonists.

The intermediate **122** will allow the synthesis of a wider range of *C*-4 position-modified $InsP_3$ analogues, thus helping the process of both achieving the optimal biological activity and acquiring more information about the behaviour of $InsP_3Rs$.

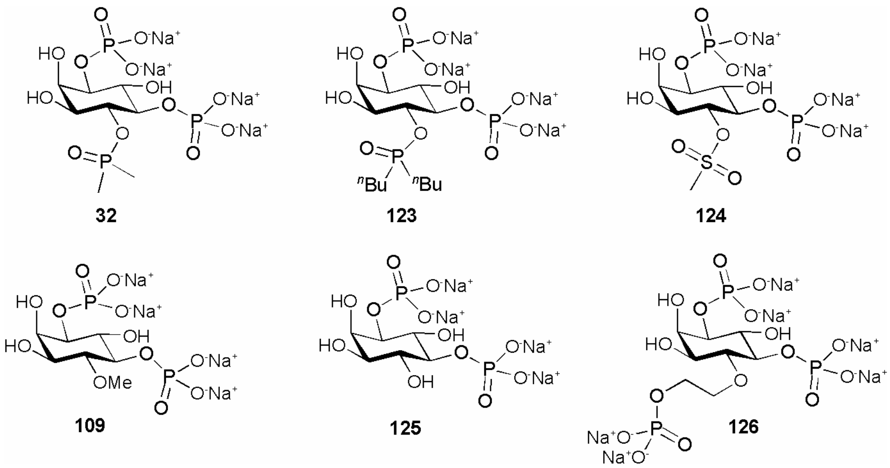


Figure 3.17. Structures of the *C*-4 position-modified $InsP_3$ analogues synthesised.

Experimental section

4 Experimental Section

4.1 General

^{1}H NMR spectra were recorded on a Bruker Avance 300 (300.1 MHz) instrument, Bruker Avance 500 (499.9 MHz) instrument or a Varian Gemini 2000 (300.0 MHz) instrument, using deuteriochloroform (or other indicated solvent) as reference and internal deuterium lock. The chemical shift data for each signal are given as δ in units of parts per million (ppm) relative to tetramethylsilane (TMS) where δ_{TMS} = 0.00 ppm. The multiplicity of each signal is indicated by: s (singlet); br s (broad singlet); d (doublet); t (triplet); td (triplet of doublets); dd (doublet of doublets); ddd (doublet of doublet of doublets); dddd (doublet of doublet of doublet of doublets); ddt (doublet of doublet of triplets); sp (septet) or m (multiplet). The number of protons (n) for a given resonance is indicated by nH. Aryl protons are indicated by Ar*H*. Coupling constants (*J*) are quoted in Hz and are recorded to the nearest 0.1 Hz.

^{13}C NMR spectra were recorded on a Bruker Avance 300 (75.5 MHz) instrument using the PENDANT sequence and internal deuterium lock or on a Varian Gemini 2000 (75.5 MHz) instrument using proton decoupling and internal deuterium lock. The chemical shift data for each signal are given as δ in units of ppm relative to TMS where δ_{TMS} = 0.00 ppm. Aryl carbons are indicated by Ar*C*H and Ar*C*; quaternary carbons are indicated by C_q. Where appropriate, coupling constants (*J*) are quoted in Hz and are recorded to the nearest 0.1 Hz.

^{31}P NMR spectra were recorded on Bruker Avance 300 (121.5 MHz), or Varian Gemini 2000 (121.4 MHz) instruments using proton decoupling and internal deuterium lock. The chemical shift data for each signal are given as δ in units of ppm relative to an external standard of 85% H_3PO_4.

IR spectra were recorded on a Perkin-Elmer Paragon series 1000 FTIR spectrometer as thin films between potassium bromide discs or nujol mull or as potassium bromide disks as indicated. Absorption maxima are reported in wavenumbers (cm^{-1}). Intensities of the maxima are quoted as strong (s), medium (m), weak (w).

Melting points were determined using a Gallenkamp MF-370 or an Electrothermal 9100 melting point apparatus and are uncorrected.

Optical rotations were measured using an Optical Activity AA-1000 automatic polarimeter or a Bellingham+Stanley Ltd ADP220 instrument, in cells with a path length of 2 dm or 1 dm. The concentration (*c*) is expressed in g/100 mL (equivalent to g/0.1 dm^3). Specific rotations are denoted $[\alpha]_D^T$ and are given in units of 10^{-1} deg cm^2 g^{-1} (T= ambient temperature in °C).

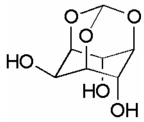
Analytical thin layer chromatography (TLC) was carried out on pre-coated 0.25 mm ICN Biomedicals GmbH 60 F_{254} silica gel plates. Visualisation was by absorption of UV light, or thermal development after dipping in either an ethanolic solution of phosphomolybdic acid (PMA) or an aqueous solution of potassium permanganate, potassium carbonate and sodium hydroxide.

Flash Column chromatography was carried out on silica gel (Apollo Scientific Ltd 40-63 micron) or on activated aluminium oxide (Acros, 50-200 micron, neutral) as indicated, under a positive pressure of compressed air.

Kugelrohr bulb-to-bulb distillations were carried out using a Büchi GKR-51 machine. Boiling points are the actual oven temperatures.

Dichloromethane was distilled from calcium hydride in a recycling still. Diethyl ether was distilled from sodium in a recycling still using benzophenone ketyl as an indicator. Anhydrous *N,N*-dimethyl formamide was purchased from Aldrich UK and dried by distillation from 4 Å molecular sieves onto 4 Å molecular sieves under an atmosphere of nitrogen. Chemicals were purchase from Acros UK, Aldrich UK, Avocado UK, Fisher UK or Fluka UK. All solvents and reagents were purified and dried, where necessary, by standard techniques.[133] Where appropriate and if not stated otherwise, all non aqueous reactions were performed under an inert atmosphere of nitrogen or argon, using a vacuum manifold with nitrogen passed through 4 Å molecular sieves and self-indicating silica gel. *In vacuo* refers to the use of a rotary evaporator attached to a diaphragm pump. Hexane refers to *n*-hexane and petroleum ether to the fraction boiling between 40-60 ^oC. Room temperature (RT) refers to the temperature of 25 °C.

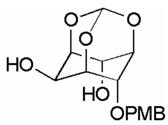
4.1.1. 2,4,10-Trioxatricyclo[3.3.1.1^{3,7}]decane-6,8,9-triol 37



37

myo-Inositol **1** (10 g, 55.5 mmol, 1.0 equiv) was dissolved in dry *N,N*-dimethyl formamide (160 mL) under an atmosphere of nitrogen. Triethylorthoformate (18.5 mL, 16.5 g, 111.0 mmol, 2.0 equiv) and 4-toluenesulfonic acid monohydrate (2.7 g, 14.4 mmol, 0.3 equiv) were added with stirring. The reaction mixture was heated to 100 °C and stirred for 16 h. The mixture was then cooled to room temperature and the 4-toluenesulfonic acid quenched with a saturated aqueous solution of sodium hydrogen carbonate (10 mL). The resulting solid was removed by filtration and the mother liquor concentrated under reduced pressure. Most of the sodium 4-toluenesulfonate was removed by crystallisation from methanol, and the resulting mixture was concentrated under reduced pressure to give a yellow residue. Purification by silica gel column chromatography, eluting with methanol and chloroform (10/90), yielded 2,4,10-trioxatricyclo-[3.3.1.1^{3,7}]decane-6,8,9-triol **37** (16.2 g yield, 77%) as a colourless solid. R_f 0.52 (ethyl acetate/acetonitrile 80/20); mp 220 °C dec. (*from methanol/chloroform*, Lit.[106] 300-302 °C); δ_H (500 MHz; D_6-DMSO) 5.47 (1H, br s, equatorial OH), 5.45 (2H, d, *J* 1.2, 2 × axial OH), 5.31 (1H, d, *J* 6.4, O_3CH) 4.30-4.22 (2H, m, inositol ring), 4.08-4.03 (1H, m, inositol ring), 4.02-3.96 (1H, m, inositol ring), 3.96-3.92 (2H, m, inositol ring). These data are in good agreement with the literature values.[106,134]

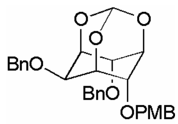
4.1.2. 6-[(4'-Methoxy)benzyloxy]-2,4,10-trioxatricyclo[3.3.1.1^{3,7}]decane-8,9-diol 38



38

2,4,10-Trioxatricyclo-[3.3.1.1^{3,7}]decane-6,8,9-triol **37** (15.0 g, 79.0 mmol, 1.0 equiv) was dissolved in dry *N,N*-dimethyl formamide (250 mL) under an atmosphere of nitrogen. The resulting mixture was cooled to 0 °C and sodium hydride (3.5 g, 60% dispersion in mineral oil, 87.0 mmol, 1.1 equiv) was added portionwise with vigorous stirring. The suspension was allowed to warm to RT and stirred for 2 h. The mixture was re-cooled to 0 °C and tetra-*n*-butylammonium iodide (2 mg, 4 μmol, 0.05 equiv) and 4-methoxybenzyl chloride (12.2 mL, 13.6 g, 86.8 mmol, 1.1 equiv) were added. The resulting slurry was allowed to warm to RT and stirred overnight. The sodium hydride was quenched by addition of water (20 mL) and the resulting mixture was concentrated under reduced pressure. The resulting oil was reconstituted in ethyl acetate (80 mL) and water (80 mL), the layers separated and the aqueous layer extracted with ethyl acetate (3 × 50 mL). The combined organic layers were washed with brine (50 mL), dried (magnesium sulfate), filtered and concentrated under reduced pressure. The resulting solid was purified by silica gel column chromatography, eluting with ethyl acetate and hexane (20/80, then 25/75, then 30/70, then 40/60), to yield 6-[(4'-methoxy)benzyloxy]-2,4,10-trioxatricyclo-[3.3.1.1^{3,7}]decane-8,9 diol **38** (19.5 g yield, 80%) as a colourless solid. R_f 0.32 (ethyl acetate/hexane 50/50); mp 100-102 °C (*from ethyl acetate/hexane*, Lit.[110] 100-101 °C); δ_H (300 MHz; $CDCl_3$) 7.25 (2H, d, *J* 8.7, Ar*H*), 6.91 (2H, d, *J* 8.7, Ar*H*), 5.44 (1H, d, *J* 1.2, O_3CH) 4.63 (1H, d, J_{AB} 11.5, OCH_AH_B), 4.58 (1H, d, J_{AB} 11.5, OCH_AH_B), 4.40-4.39 (2H, m, inositol ring), 4.27-4.19 (3H, m, inositol ring), 4.10 (1H, m, inositol ring), 3.83 (3H, s, OCH_3), 3.79 (1H, d, *J* 10.5, OH), 3.13 (1H, d, *J* 11.7, OH). These data are in good agreement with the literature values.[110,135]

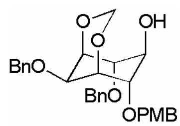
4.1.3. 8,9-Bis(benzyloxy)-6-[(4'-methoxy)benzyloxy]-2,4,10-trioxatricyclo[3.3.1.1^{3,7}]decane 39



39

6-[(4'-Methoxy)benzyloxy]-2,4,10-trioxatricyclo-[3.3.1.1^{3,7}]decane-8,9 diol **38** (19.8 g, 63.7 mmol, 1.0 equiv) was dissolved in dry *N,N*-dimethyl formamide (200 mL) under an atmosphere of nitrogen. The mixture was cooled to 0 °C and sodium hydride (6.4 g, 60% dispersion in mineral oil, 159.4 mmol, 2.5 equiv) was added portionwise. The mixture was allowed to warm to RT and stirred for 2 h, then re-cooled to 0 °C and benzyl bromide (27.2 g, 18.9 mL, 159.4 mmol, 2.5 equiv) was added dropwise, keeping the temperature at 0 °C. The mixture was allowed to warm to RT and stirred overnight. The sodium hydride was quenched by addition of water (20 mL). The solvent was removed under reduced pressure and the resulting oil was reconstituted in ethyl acetate (50 mL) and water (50 mL). The layers were separated and the aqueous layer was extracted with ethyl acetate (3 × 50 mL). The combined organic layers were washed with brine (30 mL), dried (magnesium sulfate), filtered and concentrated under reduced pressure. The resulting yellow oil was purified by silica gel column chromatography, eluting with ethyl acetate and petroleum ether (20/80, then 40/60), to yield 6-[(4'-methoxy)benzyloxy]-2,4,10-trioxatricyclo[3.3.1.1^{3,7}]decane **39** as a colourless oil (31.2 g yield, 100%); R_f 0.7 (ethyl acetate/petroleum ether 40/60); δ_H (300 MHz; $CDCl_3$) 7.40-7.20 (10 H, m, 2 × Ar*H*), 7.13 (2H, d, *J* 8.8, $OCH_2C_6H_4OCH_3$), 6.81 (2H, d, *J* 8.8, $OCH_2C_6H_4OCH_3$), 5.53 (1H, d, *J* 1.3, O_3CH), 4.65 (2H, s, OCH_2Ph), 4.62 (1H, d, J_{AB} 11.8, OCH_AH_BPh), 4.55 (1H, d, $J_{A'B'}$ 11.3, $OCH_{A'}H_{B'}$-$C_6H_4OCH_3$), 4.47 (1H, d, J_{AB} 11.8, OCH_AH_BPh), 4.44-4.40 (2H, m,1 × $OCH_{A'}H_{B'}$-$C_6H_4OCH_3$ and 1 × inositol ring), 4.35-4.26 (4H, m, inositol ring), 4.05-4.03 (1H, m, inositol ring), 3.81 (3H, s, OCH_3). These data are in good agreement with the literature values.[135]

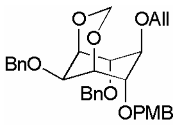
4.1.4. 8,9-Bis(benzyloxy)-6-[(4'-methoxy)benzyloxy]-2,4-dioxatricyclo[3.3.1.]nonan-7-ol 40



40

8,9-Bis(benzyloxy)-6-[(4'-methoxy)benzyloxy]-2,4,10-trioxatricyclo[3.3.1.1^{3,7}]decane **39** (17.0 g, 34.7 mmol, 1.0 equiv) was dissolved in dry dichloromethane (150 mL) under an atmosphere of nitrogen. The resulting mixture was cooled to 0 °C and a 1.0 M solution of diisobutylaluminium hydride in hexanes (86.9 mL, 86.9 mmol, 2.5 equiv) was added dropwise, keeping the temperature at 0 °C. The mixture was allowed to reach the RT and then stirred for 4 h. The reaction mixture was cannulated onto a vigorously stirred 1.0 M aqueous solution of sodium potassium tartrate (100 mL) and saturated aqueous solution of ammonium chloride (100 mL). The resulting mixture was stirred overnight to destroy the aluminium salts. The combined organic layers were washed with brine (50 mL), dried (magnesium sulfate), filtered and concentrated under reduced pressure to yield 8,9-bis(benzyloxy)-6-[(4'-methoxy)benzyloxy]-2,4-dioxatricyclo-[3.3.1.]nonan-7-ol **40** (16.1 g yield, 94%) as a colourless oil. R_f 0.29 (ethyl acetate/hexane 40/60); δ_H (300 MHz; $CDCl_3$) 7.31-7.17 (10 H, m, 2 × Ar*H*), 7.12 (2H, d, *J* 8.7, $OCH_2C_6H_4OCH_3$), 6.76 (2H, d, *J* 8.7, $OCH_2C_6H_4OCH_3$), 5.48 (1H, d, *J* 5.1, O_3CHH), 4.60 (1H, d, J_{AB} 12.0, OCH_AH_BPh), 4.59 (1H, d, *J* 5.1, O_3CHH), 4.54 (2H, s, OCH_2Ph), 4.53 (1H, d, $J_{A'B'}$ 11.5, $OCH_{A'}H_{B'}$-$C_6H_4OCH_3$), 4.50 (1H, d, J_{AB} 12.0, OCH_AH_BPh), 4.43 (1H, d, $J_{A'B'}$ 11.5, $OCH_{A'}H_{B'}$-$C_6H_4OCH_3$), 4.38-4.32 (2H, m, inositol ring), 4.22-4.20 (1H, m, inositol ring), 3.96-3.90 (2H, m, inositol ring), 3.88 (1H, d, *J* 10.2, inositol ring), 3.73 (3H, s, OCH_3), 2.90 (1H, d, *J* 10.2, OH). These data are in good agreement with the literature values.[135]

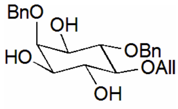
4.1.5. 8,9-Bis(benzyloxy)-6-[(4'-methoxy)benzyloxy]-7-(allyl)-2,4-dioxatricyclo-[3.3.1.]nonane 41



41

8,9-Bis(benzyloxy)-6-[(4'-methoxy)benzyloxy]-2,4-dioxatricyclo-[3.3.1.]nonan-7-ol **40** (27.0 g, 54.6 mmol, 1.0 equiv) was dissolved in dry *N,N*-dimethyl formamide (250 mL) under an atmosphere of nitrogen. The resulting mixture was cooled to 0 °C and sodium hydride (3.3 g, 60% dispersion in mineral oil, 82.3 mmol, 1.5 equiv) was added portionwise with stirring. The resulting mixture was allowed to warm to RT and stirred for 2 h, then it was re-cooled to 0 °C and imidazole (catalytic amount) and allyl bromide (9.9 g, 7.1 mL, 82.3 mmol, 1.5 equiv) were added. The resulting mixture was allowed to warm to RT and stirred overnight. The sodium hydride was quenched by addition of water (30 mL). The solvent was removed under reduced pressure and the residue reconstituted in ethyl acetate (100 mL) and water (100 mL). The layers were separated and the aqueous layer extracted with ethyl acetate (3 × 100 mL). The combined organic layers were washed with brine (50 mL), dried (magnesium sulfate), filtered and concentrated under reduced pressure. The resulting yellow oil was purified by silica gel column chromatography, eluting with ethyl acetate and hexane (30/70) to yield 8,9-bis(benzyloxy)-6-[(4'-methoxy)benzyloxy]-7-(allyl)-2,4-dioxatricyclo-[3.3.1.]nonane **41** as a colourless oil (26.1 g yield, 89%). R_f 0.43 (ethyl acetate/hexane 40/60); δ_H (300 MHz; $CDCl_3$) 7.42-7.28 (10 H, m, 2 × Ar*H*), 7.26 (2H, d, *J* 8.7, $OCH_2C_6H_4OCH_3$), 6.89 (2H, d, *J* 8.7, $OCH_2C_6H_4OCH_3$), 5.90 (1H, ddt, *J* 17.2, 10.3, 5.6, C*H*=CH_2), 5.25 (1H, ddt, *J* 17.2, 1.8, 1.5, CH=C*H*H), 5.20 (1H, d, *J* 5.4, O_3C*H*H), 5.18 (1H, ddt, *J* 10.3, 1.8, 1.3, CH=CH*H*), 4.84 (1H, d, *J* 5.4, O_3CH*H*), 4.68 (1H, d, J_{AB} 11.8, OCH_AH_BPh), 4.66 (2H, s, OCH_2Ph), 4.61 (1H, d, J_{AB} 11.8, OCH_AH_BPh), 4.61 (1H, d, $J_{A'B'}$ 11.5, $OCH_{A'}H_{B'}C_6H_4OCH_3$), 4.54 (1H, d, $J_{A'B'}$ 11.5, $OCH_{A'}H_{B'}C_6H_4OCH_3$), 4.28-4.24 (2H, m, inositol ring), 4.15 (2H, ddd, *J*, 5.6, 1.5, 1.3, $OCH_2CH=CH_2$), 3.84 (1H, t, *J* 2.0 inositol ring), 3.82 (3H, s, OCH_3), 3.54 (1H, t, *J* 5.6, inositol ring). These data are in good agreement with the literature values.[135]

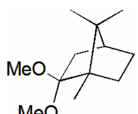
4.1.6. (±)-5-*O*-Allyl-2,6-*O*-dibenzyl-*myo*-inositol 42



42

8,9-Bis(benzyloxy)-6-[(4'-methoxy)benzyloxy]-7-(allyl)-2,4-dioxatricyclo-[3.3.1.] nonane **41** (27.8 g, 52.1 mmol, 1.0 equiv) was dissolved in methanol (400 mL) and concentrated hydrochloric acid (48 mL) was added. The mixture was heated under reflux for 6 h, then cooled to 0 °C. The hydrochloric acid was quenched by cautious addition of sodium hydrogen carbonate (50 g). The formed solid was removed by filtration and the solvent evaporated under reduced pressure. Purification by silica gel column chromatography, eluting with ethyl acetate and hexane (30/70, then 40/60) and then ethyl acetate furnished (±)-5-*O*-allyl-2,6-*O*-dibenzyl-*myo*-inositol **42** as a colourless solid (18.0 g yield, 86%). R_f 0.41 (ethyl acetate); mp 118-120 °C (*from ethyl acetate/hexane*, Lit.[135] 111-112 °C); δ_H (300 MHz; $CDCl_3$) 7.35-7.20 (10 H, m, 2 × Ar*H*), 5.90 (1H, ddt, *J* 17.2, 10.3, 5.6, C*H*=CH_2), 5.23 (1H, ddt, *J* 17.2, 1.8, 1.5, CH=C*H*H), 5.12 (1H, ddt, *J* 10.3, 1.8, 1.3, CH=CH*H*), 4.85 (1H, d, J_{AB} 11.5, OC$H_A$$H_B$Ph), 4.81 (1H, d, $J_{A'B'}$ 11.3, OC$H_{A'}$$H_{B'}$Ph), 4.70 (1H, d, $J_{A'B'}$ 11.3, OC$H_{A'}$$H_{B'}$Ph), 4.67 (1H, d, J_{AB} 11.5, OC$H_A$$H_B$Ph), 4.35-4.20 (2H, m, OCH_2CH=CH_2), 3.93 (1H, t, *J* 2.8, 2-H), 3.77-3.62 (2H, m, inositol ring), 3.53-3.46 (1H, m, inositol ring), 3.43-3.35 (1H, m, inositol ring), 3.14 (1H, t, *J* 9.0 inositol ring), 2.56 (1H, br s, OH) 2.34 (1H, d, *J* 6.9, OH), 2.28 (1H, d, *J* 4.9, OH). These data are in good agreement with the literature values.[135]

4.1.7. (1*S*)-(-)-Camphor dimethyl acetal 44

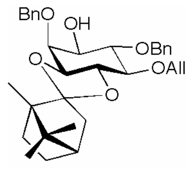


44

(1*S*)-(-)-Camphor (25.0 g, 164.2 mmol, 1.0 equiv), trimethylorthoformate (69.7 g, 71.9 mL, 656.9 mmol, 4.0 equiv) and Montmorillonite K-10 clay (45.0 g) were stirred in hexane (200 mL) under an atmosphere of nitrogen for 24 h. The clay was removed by filtration and washed with hexane (2 × 50 mL). The combined organic extracts were concentrated under reduced pressure to yield a colourless oil (32.4 g), which was used without any further purification in the next step. The oil is estimated to contain 75 % (1*S*)-(-)-camphor dimethyl acetal **44** and 25 % of (1*S*)-(-)-camphor, using NMR analysis. R_f 0.66 (diethyl ether/hexane 30/70); δ_H (300 MHz; $CDCl_3$) 3.22

(3H, s, OCH_3,), 3.16 (3H, s, OCH_3), 2.23-2.15 (1H, m, camphor ring), 1.80-1.62 (3H, m, camphor ring), 1.41-1.18 (2H, m, camphor ring), 1.75-1.10 (1H, d, *J* 12.8, 4-H), 0.96 (3H, s, 1-CH_3), 0.91 (3H, s, 7-CH_3), 0.82 (3H, s, 7-CH_3). These data are in good agreement with the literature values.[136]

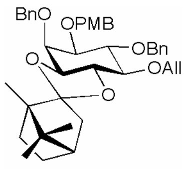
4.1.8. (-)-1D-5-*O*-Allyl-2,6-bis-*O*-benzyl-3-*O*-*endo*-4-*O*-*exo*-(L-1',7',7'-trimethylbicyclo[2.2.1]hept-2'-ylidene)-*myo*-inositol 34



34

(±)-5-*O*-Allyl-2,6-*O*-dibenzyl-*myo*-inositol **42** (17.1 g, 42.6 mmol, 1.0 equiv), crude (1*S*)-(-)-camphor dimethyl acetal **44** (28.3 g, 75% w/w, 127.8 mmol, 3 equiv) and 4-toluenesulfonic acid monohydrate (405.1 mg, 2.1 mmol, 0.05 equiv) were dissolved in dry dichloromethane (200 mL) and heated under reflux under an atmosphere of nitrogen. After 8 h the reaction was adjudged to be incomplete by TLC analysis and a further amount of crude (1*S*)-(-)-camphor dimethyl acetal **44** was added (4.0 g, 75% w/w, 18.0 mmol, 0.4 equiv) and the resulting mixture was stirred overnight. The solvent was removed under reduced pressure and the crude mixture was stored in the fridge. The crude mixture was divided in three batches and purified by silica gel column chromatography eluting with the following solvent system: ethyl acetate and petroleum ether 5/95 (6000 mL), 6/94 (2000 mL), 7/93 (2000 mL), 8/92 (2000 mL), 9/91 (2000 mL), 10/90 (8000 mL), 20/80 (5000 mL) (the undesired diastereoisomers were collected with the solvent system 10/90 ethyl acetate/petroleum ether) to afford the required diastereoisomer (-)-1D-5-*O*-allyl-2,6-bis-*O*-benzyl-3-*O*-*endo*-4-*O*-*exo*-(L-1',7',7'-trimethylbicyclo[2.2.1]hept-2'-ylidene)-*myo*-inositol **34** as a colourless oil (5.6 g yield, 25%); R_f 0.29 (ethyl acetate/hexane 20/80); $[\alpha]_D^{20}$ -11.9 (*c* 0.2 in $CHCl_3$; Lit.[135] $[\alpha]_D^{22}$ -11.7, *c* 1.3 in $CHCl_3$); δ_H (300 MHz; $CDCl_3$) 7.35-7.17 (10H, m, 2 × Ar*H*), 5.88 (1H, ddt, *J* 17.4, 10.5, 5.6 C*H*=CH_2), 5.24 (1H, ddt, *J* 17.4, 1.8, 1.5, CH=C*H*H), 5.09 (1H, ddt, *J* 10.5, 1.8, 1.3, CH=CH*H*), 4.93 (1H, d, J_{AB} 11.5, OC$H_A$$H_B$Ph), 4.85 (1H, d, $J_{A'B'}$ 11.1, OC$H_{A'}$$H_{B'}$Ph), 4.70 (1H, d, $J_{A'B'}$ 11.1, OC$H_{A'}$$H_{B'}$Ph), 4.60 (1H, d, J_{AB} 11.5, OC$H_A$$H_B$Ph), 4.32 (1H, dddd, *J* 12.8, 5.6, 1.5, 1.3, CH*H*CH=CH_2), 4.16-4.03 (3H, m, 1 × C*H*HCH=CH_2 + 2 × inositol ring) 3.90 (1H, t, *J* 9.7, inositol ring), 3.65-3.54 (2H, m, 2 × inositol ring).3.22 (1H, dd, *J* 9.7, 1.8, inositol ring), 2.38 (1H, d, *J* 7.7, O*H*), 2.07 (1H, dt, *J* 13.6, 4.0, camphor ring), 1.88-1.77 (1H, m, camphor ring), 1.70-1.58 (2H, m, 2 × camphor ring), 1.38 (1H, d, *J* 13.5, camphor ring), 1.21-1.04 (2H, m, 2 × camphor ring), 0.95 (3H, s, CH_3), 1.22-1.06 (3H, m, 3 × camphor ring), 0.79 (3H, s, CH_3), 0.78 (3H, s, CH_3). These data are in good agreement with the literature values.[135]

4.1.9. (-)-1D-5-*O*-Allyl-2,6-bis-*O*-benzyl-1-*O*-(4'-methoxybenzyl)-3-*O*-*endo*-4-*O*-*exo*-(L-1',7',7'-trimethylbicyclo[2.2.1]hept-2'-ylidene)-*myo*-inositol 48

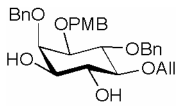


48

Sodium hydride (112 mg, 60 % dispersion in mineral oil, 2.8 mmol, 1.5 equiv) was suspended in dry tetrahydrofuran (30 mL) under an atmosphere of nitrogen and the resulting suspension was cooled to 0 °C. A solution of (-)-1D-5-*O*-allyl-2,6-bis-*O*-benzyl-3-*O*-*endo*-4-*O*-*exo*-(L-1',7',7'-trimethylbicyclo[2.2.1]hept-2'-ylidene)-*myo*-inositol **34** (1.0 g, 1.9 mmol, 1.0 equiv) in dry tetrahydrofuran (20 mL) was added by cannula. The resulting mixture was allowed to warm to RT and stirred for 1 h. The mixture was re-cooled to 0 °C and 4-methoxybenzyl chloride (668 mg, 380 µL, 2.8 mmol, 1.5 equiv), tetra-*n*-butylammonium iodide (catalytic amount) and dry *N,N*-dimethyl formamide (20 mL) were added. The resulting mixture was allowed to warm to RT and stirred overnight. The sodium hydride was quenched with water (10 mL), the solvent removed under reduced pressure and the resulting yellow residue reconstituted in ethyl acetate (20 mL) and water (20 mL). The layers were separated and the aqueous layer was extracted with ethyl acetate (3 × 20 mL). The combined organic layers were dried (magnesium sulfate), filtered and concentrated under reduced pressure to yield a yellow oil. Purification by silica gel column chromatography, eluting with ethyl acetate/hexane (10/90) afforded (-)-1D-5-*O*-allyl-2,6-bis-*O*-benzyl-1-*O*-(4'-methoxybenzyl)-3-*O*-*endo*-4-*O*-*exo*-(L-1',7',7'-trimethyl bicyclo[2.2.1]hept-2'-ylidene)-*myo*-inositol **48** (1.2 g yield, 94%) as a colourless oil; R_f 0.45 (ethyl acetate/hexane 20/80); $[\alpha]_D^{22}$ -18.4 (*c* 0.5 in $CHCl_3$; Lit.[135] $[\alpha]_D^{22}$ -20.1, *c* 2.6 in $CHCl_3$); δ_H (300 MHz; $CDCl_3$) 7.48-7.28 (10H, m, 2 × Ar*H*), 7.21 (2H, d, *J* 8.8, $OCH_2C_6H_4OCH_3$), 6.82 (2H, d, *J* 8.8, $OCH_2C_6H_4OCH_3$), 5.98 (1H, ddt, *J* 17.4, 10.2, 5.6, C*H*=CH_2), 5.33 (1H, ddt, *J* 17.4, 1.8, 1.5, CH=C*H*H), 5.17 (1H, ddt, *J* 10.2, 1.8, 1.3, CH=CH*H*), 4.91 (1H, d, J_{AB} 12.3, OCH_AH_BPh), 4.90 (1H, d, $J_{A'B'}$ 10.8, $OCH_{A'}H_{B'}Ph$), 4.84 (1H, d, $J_{A'B'}$ 10.8, $OCH_{A'}H_{B'}Ph$), 4.81 (1H, d, J_{AB} 12.3,

OCH_AH_BPh), 4.54 (1H, d, $J_{A''B''}$ 12.2, $OCH_{A''}H_{B''}C_6H_4OCH_3$), 4.50 (1H, d, $J_{A''B''}$ 12.2, $OCH_{A''}H_{B''}C_6H_4OCH_3$) 4.40 (1H, dddd, *J* 13.1, 5.6, 1.5, 1.3, C*H*HCH=CH_2), 4.21 (1H, dddd, *J* 13.1, 5.6, 1.5, 1.3, CH*H*CH=CH_2), 4.15-4.10 (1H, m, inositol ring), 4.03 (1H, t, *J* 9.7, inositol ring), 3.87 (1H, t, *J* 9.0, inositol ring), 3.72 (3H, s, OCH_3), 3.43-3.34 (2H, m, inositol ring), 3.08 (1H, dd, *J* 9.7, 1.8, inositol ring), 2.14 (1H, dt, *J* 13.3, 3.3, camphor ring), 2.00-1.90 (1H, m, camphor ring),1.77-1.68 (2H, m, camphor ring), 1.43 (1H, d, *J* 13.6, camphor ring), 1.24-1.15 (3H, m, camphor ring),1.03 (3H, s, CH_3), 0.88 (3H, s, CH_3), 0.87 (3H, s, CH_3). These data are in good agreement with the literature values.[135]

4.1.10. (-)-1D-5-*O*-Allyl-2,6-bis-*O*-benzyl-1-*O*-(4-methoxybenzyl)-*myo*-inositol 49

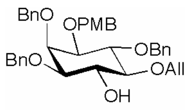


49

(-)-1D-5-*O*-Allyl-2,6-bis-*O*-benzyl-1-*O*-(4'-methoxybenzyl)-3-*O*-*endo*-4-*O*-*exo*-(L-1',7',7'-trimethylbicyclo[2.2.1]hept-2'-ylidene)-*myo*-inositol **48** (386 mg, 585 µmol, 1.0 equiv) was dissolved in methanol (8 mL) and dichloromethane (12 mL) under an atmosphere of nitrogen and acetyl chloride (28 mg, 25 µL, 70.0 µmol, 0.6 equiv) was added. The resulting mixture was stirred for 4h at RT, then the generated hydrochloric acid was quenched by the addition of triethylamine (1 mL) and the solvent was removed under reduced pressure. Purification by silica gel column chromatography, eluting with ethyl acetate/hexane (30/70, then 50/50) and then ethyl acetate afforded (-)-1D-5-*O*-allyl-2,6-bis-*O*-benzyl-1-*O*-(4-methoxybenzyl)-*myo*-inositol **49** as a colourless solid (270 mg, 88% yield); R_f 0.6 (ethyl acetate / hexane 20/80); mp 123-125 °C (*from ethyl acetate/hexane*, Lit.[109] 125-126 °C); $[\alpha]_D^{22}$ -26.5 (*c* 0.4 in $CHCl_3$; Lit.[109] $[\alpha]_D^{22}$ -26.4, *c* 1.2 in $CHCl_3$); δ_H (300 MHz; $CDCl_3$) 7.35-7.12 (12H, m, Ar*H* and 2 × $OCH_2C_6H_4OCH_3$), 6.79 (2H, d, *J* 9.0, $OCH_2C_6H_4OCH_3$), 5.89 (1H, ddt *J* 17.2, 10.2, 5.6 C*H*=CH_2), 5.21 (1H, ddt, *J* 17.2, 1.8, 1.5, CH=C*H*H), 5.10 (1H, ddt, *J* 10.2, 1.8, 1.3, CH=CH*H*), 4.98 (1H, d, J_{AB} 11.5, OCH_AH_BPh), 4.83 (1H, d, $J_{A'B'}$ 10.8, $OCH_{A'}H_{B'}Ph$), 4.73 (1H, d, $J_{A'B'}$ 10.8, $OCH_{A'}H_{B'}Ph$), 4.60 (1H, d, $J_{A''B''}$ 11.3, $OCH_{A''}H_{B''}C_6H_4OCH_3$), 4.59 (1H, d, J_{AB} 11.5, OCH_AH_BPh), 4.54 (1H, d, $J_{A''B''}$ 11.3, $OCH_{A''}H_{B''}C_6H_4OCH_3$), 4.34 (1H, dddd, *J* 12.4, 5.6, 1.5, 1.3, C*H*HCH=CH_2), 4.20 (1H, dddd, *J* 12.4, 5.6, 1.5, 1.3, CH*H*CH=CH_2), 3.90 (1H, t, *J* 2.6, inositol ring), 3.85 (1H, t, *J* 9.5, inositol ring), 3.74 (3H, s, OCH_3), 3.72 (1H, t, *J* 9.5, inositol ring), 3.36

(1H, dd, *J* 9.7, 2.6, inositol ring), 3.30 (1H, dd, *J* 9.7, 2.6, inositol ring), 3.12 (1H, t, *J* 9.3, inositol ring), 2.3 (1H, br s, O*H*), 1.50 (1H, br s, O*H*). These data are in good agreement with the literature values.[109]

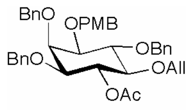
4.1.11. (-)-1D-5-*O*-Allyl-1-*O*-(4-methoxybenzyl)-2,3,6-tris-*O*-benzyl-*myo*-inositol 50



50

(-)-1D-5-*O*-Allyl-2,6-bis-*O*-benzyl-1-*O*-(4-methoxybenzyl)-*myo*-inositol **49** (200 mg, 384 µmol, 1.0 equiv), di-*n*-butyltin oxide (105 mg, 423 µmol, 1.1 equiv), tetra-*n*-butylammonium iodide (142 mg, 384 µmol, 1.0 equiv) and benzyl bromide (315 mg, 220 µL, 1.8 mmol, 4.8 equiv) were dissolved in acetonitrile under an atmosphere of nitrogen. The mixture was heated under reflux for 24 h using soxhlet apparatus filled with 3 Å molecular sieves to remove water generated in the reaction. The reaction mixture was cooled to RT and the solvent was removed under reduced pressure. The residue was suspended in water (10 mL) and extracted with ethyl acetate (3 × 20 mL). The combined organic layers were washed with a saturated aqueous solution of sodium hydrogen carbonate (10 mL) and the formed solid was removed by filtration through Celite®. The filtrate was washed with brine, dried (magnesium sulfate), filtered and concentrated under reduced pressure. Purification by silica gel column chromatography (twice), eluting with diethyl ether/petroleum ether (20/80) yielded (-)-1D-5-*O*-allyl-1-*O*-(4-methoxybenzyl)-2,3,6-tris-*O*-benzyl-*myo*-inositol **50** as a colourless solid (170 mg yield, 72%). R_f 0.43 (diethyl ether / petroleum ether 60/40); mp 60-61 °C (*from diethyl ether / petroleum ether*, Lit.[109] 60-61 °C); $[\alpha]_D^{20}$ - 0.9 (*c* 0.4 in $CHCl_3$; Lit.[109] $[\alpha]_D^{20}$ - 0.6, *c* 0.4 in $CHCl_3$); δ_H (300 MHz; $CDCl_3$, sodium hydrogen carbonate in the NMR tube) 7.34-7.13 (17H, m, Ar*H* and 2 × $OCH_2C_6H_4OCH_3$), 6.78 (2H, d, *J* 8.7, $OCH_2C_6H_4OCH_3$), 5.90 (1H, ddt *J* 17.2, 10.2, 5.6 C*H*=CH_2), 5.21 (1H, ddt, *J* 17.2, 1.8, 1.5, CH=C*H*H), 5.09 (1H, ddt, *J* 10.2, 1.8, 1.3, CH=CH*H*), 4.81 (1H, d, J_{AB} 10.8, OCH_AH_B), 4.80 (1H, d, $J_{A'B'}$ 12.0, $OCH_{A'}H_{B'}$), 4.73 (1H, d, J_{AB} 10.8, OCH_AH_B), 4.70 (1H, d, $J_{A'B'}$ 12.0, $OCH_{A'}H_{B'}$), 4.53 (1H, d, $J_{A''B''}$ 11.5, $OCH_{A''}H_{B''}$), 4.52 (1H, d, $J_{A'''B'''}$ 12.0 $OCH_{A'''}H_{B'''}$), 4.47 (1H, d, $J_{A''B''}$ 11.5, $OCH_{A''}H_{B''}$), 4.46 (1H, d, $J_{A'''B'''}$ 12.0, $OCH_{A'''}H_{B'''}$), 4.34-4.21 (2H, m, C*H*HCH=CH_2), 4.03 (1H, t, *J* 9.7, inositol ring), 3.92 (1H, t, *J* 2.3, inositol ring), 3.89 (1H, d, *J* 9.5, inositol ring), 3.75 (3H, s, OCH_3), 3.25 (1H, dd, *J* 9.5, 2.3, inositol ring), 3.16 (1H, t, *J* 9.3, inositol ring), 3.08 (1H, dd, *J* 9.5, 2.3, inositol ring), 2.43 (1H, br s, O*H*). These data are in good agreement with the literature values.[109]

4.1.12. (+)-1D-4-*O*-Acetyl-5-*O*-allyl-1-*O*-4-methoxybenzyl-2,3,6-tris-*O*-benzyl-*myo*-inositol 51

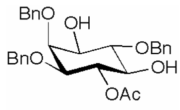


51

(-)-1D-5-*O*-Allyl-1-*O*-(4-methoxybenzyl)-2,3,6-tris-*O*-benzyl-*myo*-inositol **50** (800 mg, 1.3 mmol, 1.0 equiv) was dissolved in dry pyridine (30 mL) under an atmosphere of nitrogen. 4-Dimethylaminopyridine (48 mg, 39 μmol, 0.3 equiv) was added, followed by acetyl chloride (308 mg, 280 μL, 3.9 mmol, 3.0 equiv) and the resulting mixture was stirred for 6h. The pH of the mixture was adjusted to pH 7 using a 10 % aqueous solution of ammonium chloride. The solvent was removed under reduced pressure and the residue reconstituted in ethyl acetate (20 mL) and water (20 mL). The layers were separated and the aqueous layer extracted with ethyl acetate (3 × 10 mL). The combined organic layers were dried (magnesium sulfate), filtered and concentrated under reduced pressure. Purification by silica gel column chromatography, eluting with diethyl ether/hexane (20/80) yielded *(+)-1D-4-O-acetyl-5-O-allyl-1-O-4-methoxybenzyl-2,3,6-tris-O-benzyl-myo-inositol* **51** (62 mg yield, 81%) as a colourless solid (Found: C, 73.3; H, 6.85. $C_{40}H_{44}O_8$ requires C, 73.6; H, 6.8); R_f 0.45 (ethyl acetate/hexane 30/70); mp 96-98 °C (*from ethyl acetate/hexane)*; $[\alpha]_D^{20}$ + 4.2 (*c* 0.54 in $CHCl_3$); ν_{max} (nujol)/cm^{-1} 3036.7 (w), 2926.6 (s), 2856.6 (s), 1732.9 (s, C=O), 1612.7 (w), 1512.8 (m), 1452.7 (m), 1367.7 (m), 1302.6 (w), 1237.6 (s), 1137.5 (m), 1097.5 (m), 1047.5 (m), 1012.5 (w), 927.4 (m), 832.4 (w), 727.3 (s), 692.3 (m); δ_H (300 MHz; $CDCl_3$) 7.33-7.13 (17H, m, Ar*H* and 2 × $OCH_2C_6H_4OCH_3$), 6.77 (2H, d, *J* 8.7, $OCH_2C_6\mathit{H}_4OCH_3$), 5.78 (1H, ddt *J* 17.2, 10.5, 5.6 C*H*=CH_2), 5.51 (1H, t, *J* 10.0, axial 4-H), 5.13 (1H, ddt, *J* 17.2, 1.8, 1.5, CH=C*H*H), 5.03 (1H, ddt, *J* 10.5, 1.8, 1.3, CH=CH*H*), 4.80 (1H, d, J_{AB} 10.8, $OC\mathit{H}_AH_B$), 4.79 (1H, d, $J_{A'B'}$ 12.3, $OC\mathit{H}_{A'}H_{B'}$), 4.77-4.73 (1H, m, $OCH_A\mathit{H}_B$), 4.70 (1H, d, $J_{A'B'}$ 12.3, $OCH_{A'}\mathit{H}_{B'}$), 4.48 (1H, d, $J_{A''B''}$ 11.3 $OC\mathit{H}_{A''}H_{B''}$), 4.45 (1H, d, $J_{A'''B'''}$ 12.0, $OC\mathit{H}_{A'''}H_{B'''}$), 4.43 (1H, d, $J_{A''B''}$ 11.3, $OCH_{A''}\mathit{H}_{B''}$), 4.34 (1H, d, $J_{A'''B'''}$ 12.0, $OCH_{A'''}\mathit{H}_{B'''}$), 4.21 (1H, dddd, *J* 12.5, 5.6, 1.5, 1.3, C*H*HCH=CH_2), 4.03-3.95 (2H, m, 1 × CH*H*CH=CH_2 and 1 × inositol ring), 3.90 (1H, t, *J* 2.3, inositol ring), 3.89 (1H, d, *J* 9.5, inositol ring), 3.73 (3H, s, $OC\mathit{H}_3$), 3.25-3.15 (3H, m, inositol ring); δ_C (75 MHz;

$CDCl_3$) 170.3 (*C*=O), 159.6 (Ar*C*OCH$_3$), 139.2 (Ar*C*), 139.1 (Ar*C*), 138.4 (Ar*C*), 135.4 (*C*H=CH$_2$), 130.8 (Ar*C*), 129.7 (Ar*C*H), 128.8 (Ar*C*H), 128.7 (Ar*C*H), 128.6 (Ar*C*H), 128.5 (Ar*C*H), 128.3 (Ar*C*H), 128.1 (Ar*C*H), 128.0 (Ar*C*H), 127.8 (Ar*C*H), 127.7 (Ar*C*H), 117.0 (CH=*C*H$_2$), 114.2 (Ar*C*H), 81.8 (inositol ring), 81.7 (inositol ring), 80.6 (inositol ring), 78.6 (inositol ring), 76.2 (*C*H$_2$), 74.5 (*C*H$_2$), 74.3 (*C*H$_2$), 73.7 (inositol ring), 73.6 (inositol ring), 72.8 (*C*H$_2$), 72.5 (*C*H$_2$), 55.7 (O*C*H$_3$), 35.7 [C(O)*C*H$_3$]; *m/z* (ES+) [Found: (M+Na)$^+$ 675.2914. $C_{40}H_{44}O_8Na$ requires M^+, 675.2934], *m/z* (ES+) 675 ([M+Na]$^+$, 100%), 413 (10).

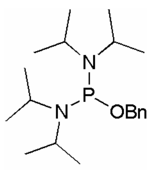
4.1.13. 1-D-*O*-Acetyl-2,3,6-tris-*O*-benzyl-*myo*-inositol 52



52

(+)-1D-4-*O*-Acetyl-5-*O*-allyl-1-*O*-4-methoxybenzyl-2,3,6-tris-*O*-benzyl-*myo*-inositol **51** (100 mg, 153 μmol, 1.0 equiv) was dissolved in ethanol (8 mL) under an atmosphere of nitrogen and Wilkinson’s catalyst (43 mg, 46 μmol, 0.3 equiv) and Hunig’s base (20 mg, 27 μL, 153 μmol, 1.0 equiv) were added. The resulting suspension was heated under reflux for 1.5 h. The mixture was cooled to RT and an aliquot was removed for ^{1}H NMR analysis, which indicated that the double bond had isomerised. The reaction mixture was filtered through Celite® and concentrated under reduced pressure to yield a dark oil. This material was dissolved in methanol/dichloromethane (2/3, 8 mL) under an atmosphere of nitrogen and acetyl chloride (7 mg, 6 μL, 92 μmol, 0.6 equiv) was added. The resulting mixture was stirred for 2 h at RT. The generated hydrochloric acid was quenched with triethylamine (20 μL) and the solvent removed under reduced pressure. The residue was reconstituted in ethyl acetate (10 mL) and water (10 mL) and the aqueous layer extracted with ethyl acetate (3 × 10 mL). The combined organic layers were dried (magnesium sulfate), filtered and concentrated under reduced pressure to yield a yellow residue. This material was dissolved in acetonitrile/water (8/2, 10 mL) and ceric ammonium nitrate (504 mg, 919 μmol, 6.0 equiv) was added. The resulting orange solution was stirred for 2h and then concentrated under reduced pressure. The residue was reconstituted in ethyl acetate (10 mL) and water (10 mL) and the aqueous layer extracted with ethyl acetate (3 × 10 mL). The combined organic layers were washed with a saturated aqueous solution of sodium hydrogen carbonate (10 mL), brine (10 mL), then dried (magnesium sulfate), filtered and concentrated under reduced pressure. Purification by silica gel column chromatography, eluting with ethyl acetate/petroleum ether (30/70) afforded *1D-4-O-acetyl-2,3,5-tris-O-benzyl-myo-inositol* **52** as a colourless waxy solid (54 mg yield, 72%); R_f 0.42 (ethyl acetate/petroleum ether); $[\alpha]_D^{20}$ + 17.0 (*c* 0.35 in $CHCl_3$); v_{max} (*KBr disc*)/cm^{-1} 3445.8 (s), 3031.2 (m), 2878.3 (s), 1747.8 (s, C=O), 1496.3 (m), 1455.6 (m),1372.2 (m), 1237.0 (s), 1025.8 (s), 933.4 (m), 820.1 (w), 735.0 (s) and 697.1 (s); δ_H (300 MHz; $CDCl_3$) 7.28-7.18 (15H, m, Ar*H*), 5.37 (1H, t, *J* 9.7, axial 4-H), 4.93 (1H, d, J_{AB} 11.5, OCH_AH_B), 4.79 (1H, d, $J_{A'B'}$ 11.3, $OCH_{A'}H_{B'}$), 4.73 (1H, d, $J_{A'B'}$ 11.3, $OCH_{A'}H_{B'}$), 4.59 (1H, d, J_{AB} 11.5, OCH_AH_B), 4.57 (1H, d, $J_{A''B''}$ 12.1 $OCH_{A''}H_{B''}$), 4.48 (1H, d, $J_{A''B''}$ 12.1 $OCH_{A''}H_{B''}$), 3.96 (1H, br s, inositol ring), 3.62 (1H, t, *J* 9.2, inositol ring), 3.46-3.40 (2H, m, inositol ring), 3.35 (1H,dd, *J* 10.0, 1.8, inositol ring), 2.40 (1H, br s, O*H*), 2.25 (1H, br s, O*H*), 2.01 (3H, s, OCH_3); δ_C (75 MHz; $CDCl_3$) 171.6 (*C*=O), 138.8 (Ar*C*), 138.7 (Ar*C*), 138.2 (Ar*C*), 129.0 (Ar*C*H), 128.9 (Ar*C*H), 128.88 (Ar*C*H), 128.5 (Ar*C*H), 128.46 (Ar*C*H), 128.35 (Ar*C*H), 128.3 (Ar*C*H), 128.2 (Ar*C*H), 127.9 (Ar*C*H), 82.8 (inositol ring), 78.7 (inositol ring), 77.1 (inositol ring), 75.7 (CH_2), 75.3 (CH_2), 74.6 (inositol ring), 74.0 (inositol ring), 73.0 (CH_2), 72.5 (inositol ring), 21.5 [C(O)CH_3]; *m/z* (ES+) [Found: $(M+Na)^+$ 515.2054. $C_{29}H_{32}O_7Na$ requires M^+, 515.2046], *m/z* (ES+) 515 ($[M+Na]^+$, 100%).

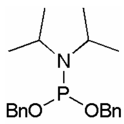
4.1.14. Benzyloxy bis(*N,N*-diisopropylamino)phosphine 159



159

Phosphorus trichloride (18 mL, 28.3 g, 206.3 mmol, 1.0 equiv) was dissolved in dry diethyl ether (200 mL) under an atmosphere of nitrogen and dry pyridine (16.3 g, 16.7 mL, 206.3 mmol, 1.0 equiv) was added. The resulting mixture was cooled to - 78 °C and a solution of benzyl alcohol (22.3 g, 21.3 mL, 206.3 mmol, 1.0 equiv) in dry diethyl ether (150 mL) was added dropwise over 1 h. The mixture was allowed to warm to RT and stirred for 1.5 h. The resulting white precipitate was removed by Schlenk filtration and the remaining solid was washed with dry diethyl ether (40 mL). The filtrate was placed under an atmosphere of nitrogen and cooled to -10 °C. Dry *N,N*-diisopropylamine (85.5 g, 110.7 mL, 845.9 mmol, 4.1 equiv) was added dropwise over 15 min. The mixture was allowed to warm to RT and stirred overnight. The resulting white precipitate was removed by Schlenk filtration and the filtrate was concentrated under reduced pressure to give the title compound **159** as an oil (51.5 g, 74% yield); δ_H (300 MHz; $CDCl_3$) 7.31-7.13 (5H, m, Ar*H*), 4.60 (2H, d, *J* 7.2, OCH_2Ph), 3.56-3.44 [4H, m, $NCH(CH_3)_2$], 1.11 [24H, dd, *J* 6.7, 3.6 $NCH(CH_3)_2$]; δ_P (121 MHz, $CDCl_3$) 124.8. These data are in good agreement with the literature values.[135]

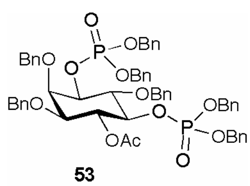
4.1.15. Bis(benzyloxy)-*N,N*-diisopropylamino phosphine 92



92

Benzyloxy bis(*N,N*-diisopropylamino)phosphine **159** (3.0 g, 8.7 mmol, 1.0 equiv) was dissolved in dry dichloromethane (15 mL) under an atmosphere of nitrogen and 1*H*-tetrazole (0.43 M solution in acetonitrile, 8.2 mL, 3.6 mmol, 0.4 equiv) was added. Dry benzyl alcohol (957 mg, 916 µL, 8.7 mmol, 1.0 equiv) was slowly added using a syringe pump over 30 min. The resulting mixture was stirred for 2 h. The solvent was removed under reduced pressure to give a colourless residue. Purification by silica gel column chromatography, eluting with triethylamine/ethyl acetate/petroleum ether (5/15/80) gave the bis(benzyloxy)-*N,N*-diisopropylamino phosphine **92** as a colourless oil (2.4 g yield, 78%); δ_H (300 MHz; $CDCl_3$) 7.31-7.18 (10H, m, Ar*H*), 4.71 (2H, dd, J_{AB} 12.8, J_{HP} 8.4, 1 × OCH_AH_B and 1 × $OCH_{A'}H_{B'}$), 4.63 (2H, dd, J_{AB} 12.8, J_{HP} 8.4, 1 × OCH_AH_B and 1 × $OCH_{A'}H_{B'}$), 3.69-3.57 [2H, m, $NCH(CH_3)_2$], 1.14 [12H, d, *J* 6.9, $NCH(CH_3)_2$]; δ_P (121 MHz, $CDCl_3$) 148.8. These data are in good agreement with the literature values.[104]

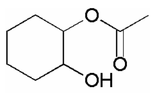
4.1.16. 1-D-*O*-Acetyl-2,3,6-tris-*O*-benzyl-*myo*-inositol 1,5-bis(dibenzylphosphate) 53



53

Bis(benzyloxy)-*N,N*-diisoproplyamino phosphine **92** (357 mg, 1.0 mmol, 5.0 equiv) was stirred with 1*H*-tetrazole (0.43 M solution in acetonitrile, 2.4 mL, 1.0 mmol, 5.0 equiv) under an atmosphere of nitrogen for 30 min. (+)-1-D-*O*-Acetyl-2,3,6-tris-*O*-benzyl-*myo*-inositol **52** (102 mg, 207 µmol, 1.0 equiv) dissolved in dry dichloromethane (5 mL) was added and the resulting mixture stirred overnight. The mixture was cooled to - 78 °C and 3-chloroperoxybenzoic acid (60% w/w, 179 mg, 1.0 mmol, 5.0 equiv) was added. The mixture was allowed to warm to RT and stirred for 30 min. The 3-chloroperoxybenzoic acid was quenched with a 10% aqueous solution of sodium hydrogen sulfite (5 mL). The resulting mixture was stirred for 10 min, then the layers were separated and the aqueous layer extracted with dichloromethane (3 × 5 mL). The combined organic layers were washed with a 10% aqueous solution of sodium hydrogen carbonate (5 mL), brine (5 mL), dried (magnesium sulfate), filtered and concentrated under reduced pressure. Purification by silica gel column chromatography, eluting with ethyl acetate/petroleum ether (30/70) yielded *1-D-O-acetyl-2,3,6-tris-O-benzyl-myo-inositol 1,5-bis (dibenzyl phosphate)* **53** as a colourless oil (139 mg yield, 66%); R_f 0.47 (ethyl acetate/petroleum ether 50/50); δ_H (300 MHz; $CDCl_3$) 7.30-6.95 (35H, m, Ar*H*), 5.59 (1H, t, *J* 9.8, axial 4-H), 4.85-4.60 (14H, m, OCH_2Ph), 4.46-4.26 (2H, m, inositol ring), 4.20-4.13 (1H, m, inositol ring), 4.09-4.01 (1H, m, inositol ring), 3.28 (1H, dd, *J* 10.2, 1.7, inositol ring), 1.79 (3H, s, CH_3); δ_P (121 MHz, $CDCl_3$) -0.39, -0.56; *m/z* (ES+) 1035 ($[M+Na]^+$, (100%).

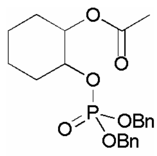
4.1.17. (±)-1-*O*-Acetyl-1,2-*trans*-dihydroxycyclohexane 56



56

(±)-1,2-*trans*-Dihydroxycyclohexane **55** (5.0 g, 43.04 mmol, 1.0 equiv) was dissolved in dry dichloromethane (400 mL) under an atmosphere of nitrogen. 4-Dimethylaminopyridine (1.6 g, 12.9 mmol, 0.3 equiv) and dry pyridine (3.7 g, 3.8 mL, 47.3 mmol, 1.1 equiv) were added and the resulting mixture was cooled to 0 ºC. Acetyl chloride (3.7 g, 3.4 mL, 47.3 mmol, 1.1 equiv) dissolved in dry dichloromethane (100 mL) was added dropwise over 1 h. The mixture was warmed to RT and stirred overnight. The solvent was removed under reduced pressure and the residue reconstituted in ethyl acetate (50 mL) and water (50 mL). The layers were separated and the organic layer extracted with ethyl acetate (3 × 50 mL). The combined organic layers were washed with brine, dried (magnesium sulfate), filtered and concentrated under reduced pressure. Purification by silica gel column chromatography, eluting with ethyl acetate/petroleum ether (30/70, then 50/50) gave the less polar diacetyl derivative (±)-1,2-*O*-diacetyl-1,2-*trans*-dihydroxycyclohexane as a colourless oil (2.4 g yield, 28%). Further elution with ethyl acetate/petroleum ether (70/30) yielded (±)-1-*O*-acetyl-1,2-*trans*-dihydroxycyclohexane **56** as a colourless solid (4.1 g yield, 60%); mp 37-39 ºC (*from ethyl acetate/petroleum ether*, Lit.[137] 39-40 ºC), δ_H (300 MHz; $CDCl_3$) 4.54-4.46 (1H, m, C*H*OC(O)CH_3), 3.52-3.44 (1H, m, C*H*OH), 2.30 (1H, s, O*H*), 2.02 (3H, s, CH_3), 2.01-1.93 (2H, m, CH_2CHOC(O)CH_3), 1.67-1.62 (2H, m, CH_2CHOH), 1.30-1.19 (4H, m, CH_2CH_2). These data are in good agreement with the literature values.[137]

4.1.18. (±)-1-*O*-Acetyl-1,2-*trans*-dihydroxycyclohexane 2-(dibenzylphosphate) 57

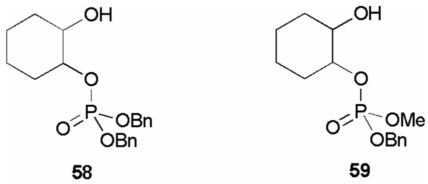


57

Bis(benzyloxy)-*N,N*-diisopropylamino phosphine **92** (2.7 g, 7.9 mmol, 2.5 equiv) was stirred with 1*H*-tetrazole (0.43 M in acetonitrile, 18.4 mL, 7.9 mmol, 2.5 equiv) under an atmosphere of nitrogen for 30 min. (±)-1-*O*-Acetyl-1,2-*trans*-dihydroxycyclohexane **56** (0.5 g, 3.2 mmol, 1.0 equiv) dissolved in dry dichloromethane (20 mL) was added and the resulting mixture stirred overnight. TLC analysis indicated the reaction to be incomplete and further bis(benzyloxy)-*N,N*-diisopropylamino phosphine (0.6 g, 1.6 mmol, 0.5 equiv) was added. The mixture was stirred for 2 h, then cooled to - 78 ºC and 3-chloroperoxybenzoic acid was added. The mixture was warmed to RT and stirred for 30 min. The reaction was quenched with a 10% aqueous solution of sodium hydrogen sulfite (10 mL) and the resulting mixture stirred for 30 min. The layers were separated and the aqueous layer extracted with dichloromethane (3 × 20 mL). The combined organic layers were washed with a 10% aqueous solution of sodium hydrogen carbonate (20 mL), brine (20 mL), then dried (magnesium sulfate), filtered and concentrated under reduced pressure. Purification by silica gel column chromatography, eluting with ethyl acetate/petroleum ether (30/70) gave *(±)-1-O-acetyl-1,2-trans-dihydroxycyclohexane 2-(dibenzylphosphate)* **57** as a colourless oil (1.1 g yield, 87%); Rf 0.66 (ethyl acetate); δ_H (300 MHz; $CDCl_3$) 7.38-7.32 (10H, m, Ar*H*), 5.04-5.00 (4H, m, OCH_2Ph), 4.85-4.78 [1H, m, C*H*OP(O)$(OBn)_2$], 4.40-4.30 [1H, m, C*H*OC(O)CH_3], 2.18-2.00 [2H, m, CH_2CHOP(O)$(OBn)_2$], 1.90 (3H, s, CH_3), 1.73-1.68 [2H, m, CH_2CHOC(O)CH_3], 1.58-1.22 (4H, m, CH_2CH_2); δ_P (121 MHz, $CDCl_3$) -0.67; *m/z* (ES+) [Found: $[M+H]^+$ 419.1617. $C_{22}H_{28}O_6P$ requires *$[M+H]^+$*, 419.1624];

m/z (ES+) 419 ($[M+H]^+$, (5%), 221 $[C_8H_{14}O_5P]^+$ (20), 179 $[C_8H_{12}O_4P]^+$ (10), 141 $[C_8H_{13}O_2]^+$ (100), 91 $[Bn]^+$ (10).

4.1.19. (±)-1,2-*trans*-Dihydroxycyclohexane 1-(dibenzylphosphate) 58



Method 1.

(±)-1-*O*-Acetyl-1,2-*trans*-dihydroxycyclohexane 2-(dibenzylphosphate) **57** (50 mg, 119 μmol, 1.0 equiv) was dissolved in methanol/water (9/1, 2 mL) and potassium carbonate (35 mg, 251 μmol, 2.1 equiv) was added. The resulting mixture stirred at RT for 2.5 h. TLC analysis indicated the reaction to be mostly complete and the presence of two compounds more polar than the starting material of R_f 0.53 and 0.45 (ethyl acetate). The potassium carbonate was quenched with a saturated aqueous solution of ammonium chloride to pH 7. The solvent was removed under reduced pressure and the residue reconstituted in ethyl acetate (2 mL) and water (2 mL). The layers were separated and the aqueous layer extracted with ethyl acetate (3 × 5 mL). The combined organic layers were washed with brine (5 mL), dried (magnesium sulfate), filtered and concentrated under reduced pressure. Purification by silica gel column chromatography, eluting with ethyl acetate/petroleum ether (70/30) yielded *(±)-1,2-trans-dihydroxycyclohexane 1-(dibenzylphosphate)* **58** as a colourless solid (26 mg yield, 59%); mp 81-83 °C (*from ethyl acetate/petroleum ether*); δ_H (300 MHz; $CDCl_3$) 7.36-7.33 (10H, m, Ar*H*), 5.13-5.00 (4H, m, OCH_2Ph), 4.10-4.00 [1H, m, C*H*OP(O)$(OBn)_2$], 3.57-3.49 (1H, m, C*H*OH), 3.12 (1H, br s, O*H*) 2.06-1.99 (2H, m, CH_2CHOP(O)$(OBn)_2$], 1.69-1.66 (2H, m, CH_2CHOH), 1.42-1.16 (4H, m, CH_2CH_2); δ_P (121 MHz, $CDCl_3$) 0.92; *m/z* (CI+) [Found: $[M+H]^+$ 377.1509 $C_{20}H_{26}O_5P$ requires *$[M+H]^+$*, 377.1518]; *m/z* (CI+) 377 $[M+H]^+$ (50%), 285 $[M - Bn]^+$ (50), 279 $[C_{14}H_{16}O_4P]^+$ (70), 189 $[C_8H_{14}O_3P]^+$ (10), 181 $[C_6H_{14}O_4P]^+$ (80), 179 $[C_6H_{12}O_4P]^+$ (50), 171 $[C_8H_{12}O_2P]^+$ (20), 91 $[Bn]^+$ (100). Further elution with ethyl acetate/petroleum ether (70/30) gave the more polar compound yielded *(±)-1,2-trans-dihydroxycyclohexane 1-(benzyl methyl phosphate)* **59** as a colourless oil (8 mg yield, 22%); δ_H (300 MHz; $CDCl_3$) 7.42-7.35 (5H, m, Ar*H*), 5.14-5.10 (2H, m, OCH_2Ph), 4.14-4.03 [1H, m, C*H*OP(O)(OBn)(OMe)], 3.74 (3H, d, *J* 11.3, OCH_3), 3.57-3.50 (1H, m, C*H*OH), 2.13-2.00 (2H, m, CH_2CHOP(O)(OBn)(OMe)], 1.75-1.65 (2H, m, CH_2CHOH), 1.46-1.20 (4H, m, CH_2CH_2); δ_P (121 MHz, CDCl$_3$) 2.07, 1.96; *m/z* (CI+) [Found: $[M+H]^+$ 301.1199 $C_{14}H_{22}O_5P$ requires *$[M+H]^+$*, 301.1205]; *m/z* (CI+) $[M+H]^+$ 301 (30%), 300 $[M]^+$ (15), 209 $[M - Bn]^+$ (10), 203 $[C_8H_{12}O_4P]^+$ (100), 202, $[C_8H_{11}O_4P]^+$ (50), 189 $[C_8H_{14}O_3P]^+$ (10), 171 $[C_8H_{12}O_2P]^+$ (20), 113 $[CH_6O_4P]^+$ (40), 91 $[Bn]^+$ (90).

Method 2.

(±)-1-*O*-Acetyl-1,2-*trans*-dihydroxycyclohexane 2-(dibenzylphosphate) **57** (50 mg, 119 µmol, 1.0 equiv) was dissolved in methanol/water (9/1, 2 mL) and lithium hydroxyde (11 mg, 251 µmol, 2.1 equiv) was added. The resulting mixture stirred at RT for 30 min. TLC analysis indicated the reaction to be mostly complete and the presence of two compounds more polar than the starting material of R_f 0.50 and 0.44 (ethyl acetate). The reaction was quenched with a saturated aqueous solution of ammonium chloride to pH 7. The solvent was removed under reduced pressure and the residue reconstituted in ethyl acetate (2 mL) and water (2 mL). The layers were separated and the aqueous layer extracted with ethyl acetate (3 × 5 mL). The combined organic layers were washed with brine (5 mL), dried (magnesium sulfate), filtered and concentrated under reduced pressure. Purification by silica gel column chromatography, eluting with ethyl acetate/petroleum ether (70/30) yielded *(±)-1,2-trans-dihydroxycyclohexane 1-(dibenzylphosphate)* **58** as a colourless solid (28 mg yield, 62%); mp 79-82 °C (*from ethyl acetate/petroleum ether*).

Method 3.

(±)-1-*O*-Acetyl-1,2-*trans*-dihydroxycyclohexane 2-(dibenzylphosphate) **57** (50 mg, 119 µmol, 1.0 equiv) was dissolved in hexane (10 mL). Lipase VII (from *candida rugosa*, 1.0 g, 1140 units) and water (1 mL) were added. The resulting mixture was shaken at 37.7 ºC for 3 days. TLC analysis indicated the reaction to be incomplete, and a further amount of Lipase VII (from *candida rugosa*, 0.5 g, 570 units) and water (1 mL) were added and the mixture shaken for 1 day at 37.7 ºC. The solvent was removed under reduced pressure and the resulting residue crushed using a mortar and pestle. The resulting powder was washed with ethyl acetate (4 × 10 mL). The combined organic layers were dried (magnesium sulfate), filtered and concentrated under reduced pressure to give a yellow solid. Purification by silica gel column chromatography, eluting with ethyl acetate/petroleum ether (70/30), yielded the title compound **58** as a colourless solid (19 mg yield, 42%); mp 80-81 °C (*from ethyl acetate/petroleum ether*).

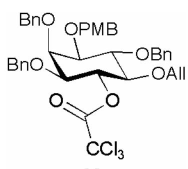
Method 4.

(±)-1-*O*-Acetyl-1,2-*trans*-dihydroxycyclohexane 2-(dibenzylphosphate) **57** (50 mg, 119 μmol, 1.0 equiv) was dissolved in hexane (10 mL). Lipase VII (from *candida rugosa*, 1.0 g, 1140 units) and wet diethyl ether (2 mL) were added. The resulting mixture was shaken at 37.7 ºC for 3 days. TLC analysis indicated the reaction to be incomplete. The solvent was removed under reduced pressure and the resulting dry residue crushed using a mortar and pestle. The resulting powder was washed with ethyl acetate (4 × 10 mL). The combined organic layers were dried (magnesium sulfate), filtered and concentrated under reduced pressure to give a yellow solid. Purification by silica gel column chromatography, eluting with ethyl acetate/petroleum ether (70/30), yielded the title compound **58** as a colourless solid (24 mg yield, 53%); R_f 0.53 (ethyl acetate); mp 81-82 °C (*from ethyl acetate/petroleum ether*).

Method 5.

(±)-1-*O*-Chloroacetyl-1,2-*trans*-dihydroxycyclohexane 2-(dibenzylphosphate) **69** (40 mg, 88 μmol, 1.0 equiv) was dissolved in a methanol/dichloromethane mixture (50/50, 4 mL) under an atmosphere of nitrogen. Thiourea (67 mg, 880 μmol, 10.0 equiv) was added and the resulting mixture was stirred at 55 °C for 2 h. The mixture was cooled to RT, diluted with dichloromethane (10 mL) and washed with a saturated acqueous solution of sodium hydrogen carbonate (5 mL). The layers were separated and the acqueous layer extracted with dichloromethane (3 × 5 mL). The combined organic layers were dried (magnesium sulfate), filtered and concentrated under reduced pressure. Purification by silica gel column chromatography, eluting with ethyl acetate/petroleum ether (70/30) furnished (±)-1,2-trans-dihydroxycyclohexane 1-(dibenzylphosphate) **58** as a colourless solid (20 mg yield, 61%); R_f 0.5 (ethyl acetate); mp 79-81 °C (*from ethyl acetate/petroleum ether*).

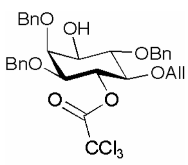
4.1.20. 1-D-4-*O*-Trichloroacetyl-5-*O*-allyl-1-*O*-(4-methoxybenzyl)-2,3,6-tris-*O*-benzyl-*myo*-inositol 60



60

(-)-1D-5-*O*-Allyl-1-*O*-(4-methoxybenzyl)-2,3,6-tris-*O*-benzyl-*myo*-inositol **50** (100 mg, 164 μmol, 1.0 equiv) was dissolved in dry pyridine (2 mL) under an atmosphere of argon. Trichloroacetyl chloride (45 mg, 28 µL, 246 µmol, 1.5 equiv) was added and the resulting mixture stirred for 30 min. The trichloroacetyl chloride was quenched with water (2 mL) and the solvent removed under reduced pressure. The resulting residue was reconstituted in ethyl acetate (5 mL) and water (5 mL), the layers separated and the aqueous layer extracted with ethyl acetate (3 × 5 mL). The combined organic layers were washed with a saturated aqueous solution of sodium hydrogen carbonate (5 mL), then dried (magnesium sulfate), filtered and concentrated under reduced pressure. Purification by silica gel column chromatography, eluting with ethyl acetate/petroleum ether (10/90) yielded *1-D-4-O-trichloroacetyl-5-O-allyl-1-O-(4-methoxybenzyl)-2,3,6-tris-O-benzyl-myo-inositol* **60** as a colourless solid (119 mg yield, 96%); Rf 0.62 (ethyl acetate/petroleum ether 40/60); mp 140-142 °C (*from ethyl acetate/petroleum ether*) δ_H (300 MHz; $CDCl_3$) 7.32-7.17 (15H, m, Ar*H*), 7.13 (1H, d, *J* 8.7, $OCH_2C_6H_4OCH_3$), 6.86 (2H, d, *J* 8.7, $OCH_2C_6H_4OCH_3$), 5.78 (1H, ddt *J* 17.1, 10.5, 5.6 C*H*=CH_2), 5.54 (1H, t, *J* 9.7, inositol ring), 5.12 (1H, ddt, *J* 17.1, 1.6, 1.5, CH=C*H*H), 5.04 (1H, ddt, *J* 10.5, 1.6, 1.5 CH=CH*H*), 4.83 (1H, d, J_{AB} 10.7, OCH_AH_B), 4.74 (2H, s, $OCH_{A'}H_{B'}$), 4.71 (1H, d, J_{AB} 10.7, OCH_AH_B), 4.48 (1H, d, $J_{A''B''}$ 11.5, $OCH_{A''}H_{B''}$), 4.45-4.42 (3H, m, 1 × $OCH_{A''}H_{B''}$ and 2 × $OCH_{A'''}H_{B'''}$), 4.25 (1H, ddt, *J* 12.0, 5.6, 1.6, C*H*HCH=CH_2), 4.10-3.98 (2H, m, 1 × CH*H*CH=CH_2 and 1 × inositol ring), 3.88 (1H, t, *J* 2.1, inositol ring), 3.74 (3H, s, OCH_3), 3.40-3.30 (2H, m, inositol ring), 3.24 (1H, dd, *J* 9.7, 2.3, inositol ring); δ_C (75 MHz; $CDCl_3$) 161.0 (*C*=O), 159.3 (Ar*C*OCH_3), 138.6 (Ar*C*), 138.4 (Ar*C*), 137.3 (Ar*C*), 134.4 (*C*H=CH_2), 130.1 (Ar*C*), 129.3 (Ar*C*H), 128.35 (Ar*C*H), 128.3 (Ar*C*H), 128.1 (Ar*C*H), 127.9 (Ar*C*H), 127.8 (Ar*C*H), 127.7 (Ar*C*H), 127.53 (Ar*C*H), 127.5 (Ar*C*H), 127.4 (Ar*C*H), 116.91 (CH=CH_2), 113.8 (Ar*C*H), 90.2 [C(O)*C*Cl_3], 81.4 (inositol ring), 80.4 (inositol ring), 79.9 (inositol ring), 79.0 (inositol ring), 77.9 (inositol ring), 75.7 (*C*H_2), 74.3 (*C*H_2), 74.1 (*C*H_2), 73.2 (inositol ring), 72.4 (*C*H_2), 72.3 (*C*H_2), 55.2 (O*C*H_3); *m/z* (ES+) 777 ($[M+Na]^+$, 100%), 779 (95).

4.1.21. 1-D-4-*O*-Trichloroacetyl-5-*O*-allyl-2,3,6-tris-*O*-benzyl-*myo*-inositol 61

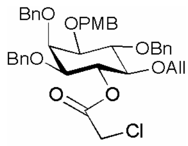


61

1-D-4-*O*-Trichloroacetyl-5-*O*-allyl-1-*O*-(4-methoxybenzyl)-2,3,6-tris-*O*-benzyl-*myo*-inositol **60** (49 mg, 64 µmol, 1.0 equiv) was dissolved in dichloromethane (3 mL).

2,3-Dichloro-5,6-dicyanobenzoquinone (29 mg, 129 μmol, 2.0 equiv) was added and the mixture stirred for 2 h. The mixture was diluted with dichloromethane (5 mL) and washed with a saturated aqueous solution of sodium hydrogen carbonate (5 mL). The layers were separated and the aqueous layer extracted with dichloromethane (3 × 5 mL). The combined organic layers were dried (magnesium sulfate), filtered and concentrated under reduced pressure. Purification by silica gel column chromatography, eluting with ethyl acetate/petroleum ether (20/80) gave 1-D-4-*O-trichloroacetyl-5-O-allyl-2,3,6-tris-O-benzyl-myo-inositol* **61** as a colourless gum (37 mg yield, 91%); R_f 0.5 (ethyl acetate/petroleum ether 40/60); δ_H (300 MHz; $CDCl_3$) 7.45-7.26 (15H, m, Ar*H*), 5.90 (1H, ddt *J* 17.1, 10.5, 5.6 C*H*=CH_2), 5.62 (1H, t, *J* 10.0, inositol ring), 5.24 (1H, ddt, *J* 17.1, 1.8, 1.5, CH=C*H*H), 5.15 (1H, ddt, *J* 10.5, 1.5, 1.3 CH=CH*H*), 4.97 (1H, d, J_{AB} 11.5, OC$H_A$$H_B$), 4.87 (1H, d, $J_{A'B'}$ 11.1, OC$H_{A'}$$H_{B'}$) 4.78 (1H, d, $J_{A'B'}$ 11.1, OC$H_{A'}$$H_{B'}$), 4.65 (1H, d, J_{AB} 10.7, OC$H_A$$H_B$), 4.62 (2H, s, OC$H_{A''}$$H_{B''}$), 4.34 (1H, dd, *J* 11.8, 5.6, C*H*HCH=CH_2), 4.20 (1H, d, *J* 11.8, 5.6, CH*H*CH=CH_2), 4.07 (1H, t, *J* 2.6, inositol ring), 3.86 (1H, t, *J* 9.2, inositol ring), 3.58-3.44 (3H, m, inositol ring); *m/z* (ES+) 657 ($[M+Na]^+$, 100%), 659 (95).

4.1.22. 1-D-4-*O*-Chloroacetyl-5-*O*-allyl-1-*O*-(4-methoxybenzyl)-2,3,6-tris-*O*-benzyl-*myo*-inositol 71

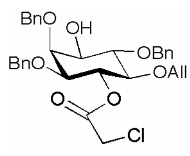


71

(-)-1D-5-*O*-Allyl-1-*O*-(4-methoxybenzyl)-2,3,6-tris-*O*-benzyl-*myo*-inositol **50** (50 mg, 82 μmol, 1.0 equiv) was dissolved in dry pyridine (1 mL) under an atmosphere of nitrogen. Chloroacetic anhydride (21 mg, 123 μmol, 1.5 equiv) was added and the mixture stirred for 3 h. The chloroacetic anhydride was quenched with water (200 μL) and the solvent removed under reduced pressure. The resulting residue was reconstituted in dichloromethane (3 mL) and water (3 mL), the layers separated and the aqueous layer extracted with dichloromethane (3 × 5 mL). The combined organic layers were dried (magnesium sulfate), filtered and concentrated under reduced pressure. Purification by silica gel column chromatography, eluting with ethyl acetate/petroleum ether (10/90) gave *1-D-4-O-chloroacetyl-5-O-allyl-1-O-(4-methoxybenzyl)-2,3,6-tris-O-benzyl-myo-inositol* **71** (51 mg yield, 90%) as a colourless solid (Found: C, 69.9; H, 6.65. $C_{40}H_{43}ClO_8$ requires C, 69.9; H, 6.3); R_f

0.6 (ethyl acetate/petroleum ether 40/60); $[\alpha]_D^{22}$ + 8.06 (*c* 0.5 in $CHCl_3$); mp 90-91 °C (*from ethyl acetate/petroleum ether*); ν_{max} (*KBr disc*)/cm^{-1} 3033.4 (w), 2916.8 (w), 2969.3 (w), 1759.8 (s), 1512.7 (m), 1453.3 (m), 1363.9 (m), 1306.2 (m), 1246.0 (m), 1196.2 (m), 1138.8 (s), 1094.7 (s), 1011.5 (m), 926.3 (w), 833.3 (s), 726.4 (s), 696.0 (m); δ_H (300 MHz; $CDCl_3$) 7.35-7.12 (17H, m, 15 × Ar*H* and 2 × $OCH_2C_6H_4OCH_3$), 6.77 (2H, d, *J* 8.7, $OCH_2C_6\mathit{H}_4OCH_3$), 5.75 (1H, ddt *J* 17.2, 10.5, 5.6 C*H*=CH_2), 5.52 (1H, t, *J* 9.7, inositol ring), 5.10 (1H, ddt, *J* 17.2, 1.6, 1.5, CH=C*H*H), 5.04 (1H, ddt, *J* 10.5, 1.6, 1.5 CH=CH*H*), 4.82 (1H, d, J_{AB} 10.5, $OC\mathit{H}_AH_B$), 4.76 (2H, s, $OC\mathit{H}_{A'}\mathit{H}_{B'}$), 4.69 (1H, d, J_{AB} 10.5, $OCH_A\mathit{H}_B$), 4.50-4.41 (3H, m, 2 × $OC\mathit{H}_{A''}\mathit{H}_{B''}$ and 1 × $OCH_{A'''}\mathit{H}_{B'''}$), 4.30 (1H, d, *J* 12.3, $OCH_{A'''}\mathit{H}_{B'''}$) 4.19 (1H, ddt, *J* 12.5, 5.6, 1.6, C*H*HCH=CH_2), 4.07-3.92 (2H, m, 1 × CH*H*CH=CH_2 and 1 × inositol ring), 3.90-3.87 (3H, m, 2 × $COC\mathit{H}_2Cl$ and 1 × inositol ring), 3.75 (3H, s, $OC\mathit{H}_3$), 3.30-3.23 (2H, m, inositol ring), 3.21 (1H, t, *J* 2.6, inositol ring); δ_C (75 MHz; $CDCl_3$) 166.3 (*C*=O), 159.3 (Ar*C*OCH_3), 138.7 (Ar*C*), 138.6 (Ar*C*), 137.8 (Ar*C*), 134.8 (*C*H=CH_2), 130.3 (Ar*C*), 129.3 (Ar*C*H), 128.5 (Ar*C*H), 128.3 (Ar*C*H), 128.2 (Ar*C*H), 128.1 (Ar*C*H), 128.0 (Ar*C*H), 127.8 (Ar*C*H), 127.6 (Ar*C*H), 127.4 (Ar*C*H), 116.8 (CH=*C*H_2), 113.8 (Ar*C*H), 81.4 (inositol ring), 80.9 (inositol ring), 80.1 (inositol ring), 78.0 (inositol ring), 75.8 (*C*H_2), 75.3 (inositol ring), 74.1 (*C*H_2), 74.0 (*C*H_2), 73.3 (inositol ring), 72.5 (*C*H_2), 72.1 (*C*H_2), 55.3 (O*C*H_3), 40.9 [C(O)*C*H_2Cl]; *m/z* (ES+) [Found: $(M+Na)^+$ 709.2531 $C_{40}H_{43}O_8NaCl$ requires M^+, 709.2544]; *m/z* (ES+) 709 ($[M+Na]^+$, 100%).

4.1.23. 1-D-4-*O*-Chloroacetyl-5-*O*-allyl-2,3,6-tris-*O*-benzyl-*myo*-inositol 72

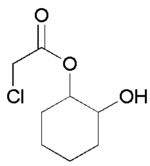


72

1-D-4-*O*-Chloroacetyl-5-*O*-allyl-1-*O*-(4-methoxybenzyl)-2,3,6-tris-*O*-benzyl-*myo*-inositol **71** (95 mg, 138 µmol, 1.0 equiv) was dissolved in dichloromethane (6 mL) and 2,3-dichloro-5,6-dicyanobenzoquinone (63 mg, 276 µmol, 2.0 equiv) was added. The resulting mixture stirred for 2 h, then diluted with dichloromethane (5 mL) and washed with a saturated aqueous solution of sodium hydrogen carbonate (5 mL). The layers were separated and the aqueous layer extracted with dichloromethane (3 × 5 mL). The combined organic layers were dried (magnesium sulfate), filtered and concentrated under reduced pressure. Purification by silica gel column chromatography, eluting with ethyl acetate/petroleum ether (20/80) yielded *1-D-4-O-*

chloroacetyl-5-O-allyl-2,3,6-tris-O-benzyl-myo-inositol **72** as a colourless gum (68 mg yield, 87%); R_f 0.48 (ethyl acetate/petroleum ether 40/60); $[\alpha]_D^{22}$ + 9.73 (*c* 0.5 in $CHCl_3$); v_{max} (*thin film*)/cm^{-1} 3548.0 (br s), 3031.2 (m), 2874.5 (s), 1751.9 (s), 1497.4 (m), 1454.4 (s), 1407.6 (m), 1363.9 (m), 1282.0 (s), 1129.8 (s), 1071.1 (s), 927.5 (m), 797.4 (w), 736.1 (s), 698.0 (s); δ_H (300 MHz; $CDCl_3$) 7.37-7.29 (15H, m, Ar*H*), 5.87 (1H, ddt *J* 17.2, 10.5, 5.6 C*H*=CH_2), 5.61 (1H, t, *J* 9.7, inositol ring), 5.23 (1H, ddt, *J* 17.2, 1.5, 1.3, CH=C*H*H), 5.15 (1H, dd, *J* 10.5, 1.3, CH=CH*H*), 5.00 (1H, d, J_{AB} 11.5, OC$H_A$$H_B$), 4.86 (1H, d, $J_{A'B'}$ 11.3, OC$H_{A'}$$H_{B'}$), 4.77 (1H, d, $J_{A'B'}$ 11.3, OC$H_{A'}$$H_{B'}$), 4.67 (1H, d ,*J* 11.5, OC$H_A$$H_B$), 4.66 (1H, d, *J* 12.3, OC$H_{A''}$$H_{B''}$), 4.50 (1H, d, *J* 12.3, OC$H_{A''}$$H_{B''}$), 4.30 (1H, ddt, *J* 12.5, 5.6, 1.5, C*H*HCH=CH_2), 4.11 (1H, ddt, *J* 12.5, 5.6, 1.5, C*H*HCH=CH_2), 4.06 (1H, t, *J* 2.6, inositol ring), 4.01 (1H, d, *J* 14.6, COC*H*HCl), 3.96 (1H, d, *J* 14.6, COCH*H*Cl), 3.81 (1H, t, *J* 9.5, inositol ring), 3.48 (1H, dd, *J* 9.7, 2.6, inositol ring), 3.42 (1H, dd, *J* 9.7, 2.3, inositol ring), 3.36 (1H, t, *J* 9.5, inositol ring); δ_C (75 MHz; $CDCl_3$) 166.3 (1C, *C*=O), 138.4 (Ar*C*), 138.3 (Ar*C*), 137.7 (Ar*C*), 134.6 (*C*H=CH_2), 128.5 (Ar*C*H), 128.4 (Ar*C*H), 128.1 (Ar*C*H), 128.0 (Ar*C*H), 127.94 (Ar*C*H), 127.9 (Ar*C*H), 127.8 (Ar*C*H), 127.5 (Ar*C*H), 117.0 (CH=*C*H_2), 81.9 (inositol ring), 80.8 (inositol ring), 78.3 (inositol ring), 76.2 (inositol ring), 75.6 (*C*H_2), 75.3 (inositol ring), 74.8 (*C*H_2), 74.2 (*C*H_2), 72.4 (*C*H_2), 72.2 (inositol ring), 40.9 [C(O)*C*H_2Cl]; *m/z* (ES+) [Found: $(M+Na)^+$ 589.1967. $C_{32}H_{35}O_7Na$ requires M^+, 589.1969]; *m/z* (ES+) 589 ($[M+Na]^+$, 100%).

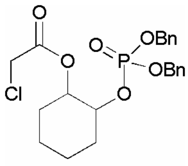
4.1.24. (±)-1-*O*-Chloroacetyl-1,2-*trans*-dihydroxycyclohexane 68



68

(±)-1,2-*trans*-Dihydroxycyclohexane **55** (1.0 g, 8.6 mmol, 1.0 equiv) was dissolved in dry dichloromethane (100 mL) under an atmosphere of nitrogen. Dry pyridine (815 mg, 0.8 mL, 10.3 mmol, 1.2 equiv) and 4-dimethylaminopyridine (210 mg, 1.7 mmol, 0.2 equiv) were added, followed by and chloroacetic anhydride (1.8 g, 10.3 mmol, 1.2 equiv), the resulting mixture stirred for 6 h. The chloroacetic anhydride was quenched with water (10 mL), the layers were separated and the aqueous layer extracted with dichloromethane (3 × 10 mL). The combined organic layers were washed with brine (10 mL), dried (magnesium sulfate), filtered and concentrated under reduced pressure. Purification by silica gel column chromatography, eluting with ethyl acetate/petroleum ether (40/60) gave *(±)-1-O-chloroacetyl-1,2-trans-dihydroxycyclohexane* **68** as a colourless solid (639 mg yield, 40%); R_f 0.68 (ethyl acetate); mp 79-81 °C (*from ethyl acetate/petroleum ether*); δ_H (300 MHz; $CDCl_3$) 4.64-4.56 [1H, m, C*H*OC(O)CH_2Cl], 4.06 [1H, d, *J* 14.4, C(O)C*H*HCl], 4.00 [1H, d, *J* 14.4, C(O)CH*H*Cl], 3.58-3.05 (1H, m, C*H*OH), 2.02-1.98 (2H, m, CH_2), 1.68-1.65 (2H, m, CH_2), 1.33-1.18 (4H, m, CH_2CH_2).

4.1.25. (±)-1-*O*-Chloroacetyl-1,2-*trans*-dihydroxycyclohexane 2-(dibenzylphosphate) 69

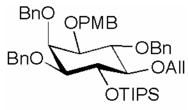


69

Bis(benzyloxy)-*N,N*-diisoproplyamino phosphine **92** (538 mg, 1.6 mmol, 3.0 equiv) and 1*H*-tetrazole (253 mg, 3.6 mmol, 7.0 equiv) were dissolved in dry dichloromethane (5 mL) under an atmosphere of nitrogen. (±)-1-*O*-Chloroacetyl-1,2-*trans*-dihydroxycyclohexane **68** (100 mg, 519 μmol, 1.0 equiv) dissolved in dry dichloromethane (2 mL) was added by cannulation and the resulting mixture stirred for 30 min. Water (21 μL) was added and the resulting mixture stirred for 15 min. The mixture was then cooled to - 78 °C and 3-chloroperoxybenzoic acid (75% w/w, 598 mg, 2.6 mmol, 5.0 equiv) was added. The resulting mixture allowed to warm to RT and stirred for 30 min. The 3-chloroperoxybenzoic acid was quenched with a 10% aqueous solution of sodium hydrogen sulfite (10 mL), the layers were separated and the aqueous layer extracted with ethyl acetate (3 × 5 mL). The combined organic layers were washed with a saturated aqueous solution of sodium hydrogen carbonate (5 mL), brine (5 mL), then dried (magnesium sulfate), filtered and concentrated under reduced pressure. Purification by silica gel column chromatography, eluting with ethyl acetate/petroleum ether (20/80) yielded *(±)-1-O-chloroacetyl-1,2-trans-dihydroxycyclohexane 2-(dibenzylphosphate)* **69** as a colourless solid (145 mg yield, 62%); R_f 0.3 (ethyl acetate/petroleum ether 40/60); mp 64-67 °C (*from ethyl acetate/petroleum ether*); δ_H (300 MHz; $CDCl_3$) 7.36-7.28 (10H, m, Ar*H*), 4.98-4.85 (4H, m, CH_2OPh), 4.83-4.75 (1H, m, C*H*OP), 4.32-4.18 [1H, m, C*H*OC(O)CH_2Cl], 3.80 [1H, d, *J* 14.4, C(O)C*H*HCl], 3.77 [1H, d, *J* 14.4, C(O)CH*H*Cl], 2.15-1.90 (2H, m, CH_2), 1.70-1.58 (2H, m, CH_2), 1.45-1.20 (4H, m,

CH_2CH_2); δ_P (121 MHz, $CDCl_3$) - 0.62; *m/z* (ES+) 474 ($[M+Na]^+$, 100%), 301 (50) ($C_{14}H_{15}NaOP_4$).

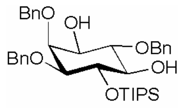
4.1.26. 1-D-4-*O*-Triisopropylsilyl-5-*O*-allyl-1-*O*-4-methoxybenzyl-2,3,6-tris-*O*-benzyl-*myo*-inositol 75



75

(-)-1D-5-*O*-Allyl-1-*O*-(4-methoxybenzyl)-2,3,6-tris-*O*-benzyl-*myo*-inositol **50** (50 mg, 82 µmol, 1.0 equiv) was dissolved in dry dichloromethane (1 mL) under an atmosphere of argon. The mixture was cooled to 0 °C and 2,6-luditine (35 mg, 38 µL, 327 µmol, 4.0 equiv) and triisopropylsilyl triflate (38 mg, 33 µL, 123 µmol, 1.5 equiv) were added. The mixture was allowed warm to RT and stirred overnight. The triisopropylsilyl triflate was quenched with water (2 mL) and the mixture was diluted with dichloromethane (5 mL), the layers separated and the aqueous layer extracted with dichloromethane (3 × 2 mL). The combined organic layers were dried (magnesium sulfate), filtered and concentrated under reduced pressure. Purification by silica gel column chromatography, eluting with ethyl acetate/petroleum ether (10/90) furnished *1D-4-O-triisopropylsilyl-5-O-allyl-1-O-4-methoxybenzyl-2,3,6-tris-O-benzyl-myo-inositol* **75** as a deliquescent colourless solid (59 mg yield, 94%); R_f 0.6 (ethyl acetate/petroleum ether 20/80); δ_H (300 MHz; $CDCl_3$) 7.40-7.23 (17H, m, 15 × Ar*H* and 2 × $OCH_2C_6H_4OCH_3$), 6.86 (2H, d, *J* 8.7, $OCH_2C_6H_4OCH_3$), 5.96 (1H, ddt *J* 17.4, 10.5, 5.4 C*H*=CH_2), 5.25 (1H, ddt, *J* 17.4, 1.8, 1.5, CH=C*H*H), 5.13 (1H, dd, *J* 10.5, 1.8, CH=CH*H*), 4.90 (1H, d, J_{AB} 10.5, OCH_AH_B), 4.86 (1H, d, $J_{A'B'}$ 12.0, $OCH_{A'}H_{B'}$), 4.77 (1H, d, J_{AB} 10.5, OCH_AH_B), 4.68 (1H, d, $J_{A'B'}$ 12.0, $OCH_{A'}H_{B'}$), 4.60 (1H, d, $J_{A''B''}$ 11.3, $OCH_{A''}H_{B''}$), 4.58 (1H, d, $J_{A''B''}$ 11.5 $OCH_{A'''}H_{B'''}$), 4.54 (1H, d, $J_{A''B''}$ 11.3, $OCH_{A''}H_{B''}$), 4.48 (1H, d, $J_{A'''B'''}$ 11.5, $OCH_{A'}H_{B''}$), 4.47-4.40 (1H, m, C*H*HCH=CH_2), 4.34-4.25 (2H, m, 1 × CH*H*CH=CH_2 and 1 × inositol ring), 4.05-3.93 (2H, m, inositol ring), 3.85 (3H, s, OCH_3), 3.35 (1H, dd, *J* 9.7, 2.3, inositol ring), 3.20 (1H, t, *J* 9.0, inositol ring), 3.13 (1H, dd, *J* 9.5, 2.0, inositol ring), 1.15-1.32 [(21H, m, 3 × C*H*(CH_3)$_2$].

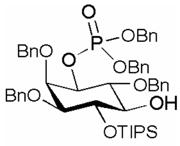
4.1.27. 1D-4-*O*-Triisopropylsilyl-2,3,6-tris-*O*-benzyl-*myo*-inositol 76



76

1D-4-*O*-Triisopropylsilyl-5-*O*-allyl-1-*O*-4-methoxybenzyl-2,3,6-tris-*O*-benzyl-*myo*-inositol **75** (56 mg, 73 µmol, 1.0 equiv) was dissolved in ethanol (5 mL) under an atmosphere of nitrogen. Wilkinson's catalysts (21 mg, 22 µmol, 0.3 equiv) and Hunig's base (9 mg, 13 µL, 73 µmol, 1.0 equiv) were added and the resulting mixture heated under reflux for 2.5 h. The mixture was cooled to RT and an aliquot was removed for ^{1}H NMR analysis, which indicated complete isomerisation of the allyl group. The mixture was filtered through Celite® and the filtrate concentrated under reduced pressure. The resulting residue was dissolved in methanol/dichloromethane (2/3, 5 mL) under an atmosphere of nitrogen and acetyl chloride (9 mg, 8 µL, 117 µmol, 1.6 equiv) was added. The resulting mixture was stirred for 2 h, then the generated hydrochloric acid was quenched with triethylamine (20 µL) and the solvent removed under reduced pressure. The resulting solid was dissolved in dichloromethane (1.5 mL), 2,3-dichloro-5,6-dicyanobenzoquinone (35 mg, 146 µmol, 2.0 equiv) was added and the resulting mixture stirred at RT for 3 h. The reaction mixture was diluted with dichloromethane (5 mL), washed with a saturated solution of sodium hydrogen carbonate (5 mL), the layers separated and the aqueous layer extracted with dichloromethane (3 × 2 mL). The combined organic layers were washed with brine (5 mL), dried (magnesium sulfate), filtered and concentrated under reduced pressure. Purification by silica gel column chromatography, eluting with ethyl acetate/petroleum ether (10/90, then 15/85) yielded *1D-4-O-triisopropylsilyl-2,3,6-tris-O-benzyl-myo-inositol* **76** as a colourless oil (27 mg yield, 62%); Rf 0.32 (ethyl acetate/petroleum ether 20/80); δ_H (300 MHz; $CDCl_3$) 7.21-7.22 (15H, m, Ar*H*), 4.88 (1H, d, J_{AB} 11.3, OC$H_A$$H_B$), 4.86 (1H, d, $J_{A'B'}$ 11.5, OC$H_{A'}$$H_{B'}$), 4.82 (1H, d, J_{AB} 11.3, OC$H_A$$H_B$), 4.70-4.68 (2H, m, OC$H_{A''}$$H_{B''}$), 4.58 (1H, d, $J_{A'B'}$ 11.5, OC$H_{A'}$$H_{B'}$), 4.24 (1H, t, *J* 9.0, inositol ring), 4.05 (1H, t, *J* 2.3, inositol ring), 3.73 (1H, t, *J* 9.2, inositol ring), 3.61-3.49 (1H, m, inositol ring), 3.51 (1H, t, *J* 8.2, inositol ring), 3.33 (1H, d, *J* 9.2, inositol ring), 1.14-1.10 [(21H, m, 3 × C*H*(CH_3)$_2$]; *m/z* (ES+) 629 [M+Na]$^+$.

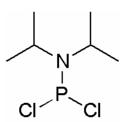
4.1.28. 1-D-4-*O*-Triisopropylsilyl-2,3,6-tris-*O*-benzyl-*myo*-inositol 1-(dibenzyl)phosphate 78



78

Bis(benzyloxy)-*N,N*-diisopropylamino phosphine **92** (125 mg, 360 µmol, 5.0 equiv) was stirred with 1*H*-tetrazole (0.43 M in acetonitrile, 837 µL, 360 µmol, 5.0 equiv) under an atmosphere of for 30 min. 1D-4-*O*-Triisopropylsilyl-2,3,6-tris-*O*-benzyl-*myo*-inositol **76** (44 mg, 73 µmol, 1.0 equiv) dissolved in dry dichloromethane (5 mL) was added *via* cannulation and the resulting mixture stirred overnight. The mixture was cooled to - 78 °C and 3-chloroperoxybenzoic acid (75% w/w, 104 mg, 360 µmol, 5.0 equiv) was added. The resulting mixture was allowed to warm to RT and stirred for 30 min. The reaction was quenched with a 10% aqueous solution of sodium hydrogen sulfite (5 mL) and the resulting mixture stirred for 30 min. The layers were separated and the aqueous layer extracted with dichloromethane (3 × 5 mL). The combined organic layers were washed with a 10% aqueous solution of sodium hydrogen carbonate (5 mL), brine (5 mL), then dried (magnesium sulfate), filtered and concentrated under reduced pressure to yield a colourless oil. Purification by silica gel column chromatography, eluting with ethyl acetate/petroleum ether (20/80) yielded *1-D-4-O-triisopropylsilyl-2,3,6-tris-O-benzyl-myo-inositol 1-(dibenzyl) phosphate* **78** as a colourless oil (8 mg, yield 13%). Rf 0.70 (ethyl acetate/petroleum ether 40/60); δ_H (300 MHz; $CDCl_3$) 7.39-7.19 (25H, m, Ar*H*), 5.08 (1H, d, J_{AB} 12.0, OCH_AH$_B$), 4.06 (1H, d, $J_{A'B'}$ 12.0, OC$H_{A'}$H$_{B'}$), 5.03-4.91 (4H, m, 1 × OCH$_A$$H_B$, 1 × OCH$_{A'}$$H_{B'}$, and 2 × OC$H_{A''}$$H_{B''}$), 4.84 (1H, d, *J* 11.0, OC$H_{A'''}$H$_{B'''}$), 4.79 (1H, d, *J* 11.0, OCH$_{A'''}$$H_{B'''}$), 4.76 (1H, d, *J* 11.5, OC$H_{A''''}$H$_{B''''}$), 4.71 (1H, d, *J* 11.5, OCH$_{A''''}$$H_{B''''}$), 4.05 (1H, t, *J* 2.3, inositol ring), 3.73 (1H, t, *J* 9.2, inositol ring), 3.61-3.49 (1H, m, inositol ring), 3.51 (1H, t, *J* 8.2, inositol ring), 3.33 (1H, d, *J* 9.2, inositol ring), 1.14-1.10 [(21H, m, 3 × C*H*(CH_3)$_2$]; δ_P (121 MHz, $CDCl_3$) - 0.70.

4.1.29. Diisopropylphosphoramidous dichloride 112

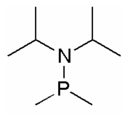


112

Phosphorus trichloride (34.6 g, 22.0 mL, 252.2 mmol, 1.0 equiv) was dissolved in dry diethyl ether (150 mL) under an atmosphere of nitrogen and cooled to -10 °C.

Dry *N,N*-diisopropylamine (51.0 g, 70.7 mL, 504.3 mmol, 2.0 equiv) in dry diethyl ether (100 mL) was added by cannulation over 1.5 h, keeping the temperature below 0 °C. The resulting mixture was stirred at 0 °C for 2.5 h, then warmed to RT and stirred for 1 h. The solvent was removed under reduced pressure and the remaining and the resulting oil purified by Kugelrohr distillation to afford diisopropylphosphoramidous dichloride **112** as a colourless oil (37.2 g yield, 73%); bp 70 °C (5 mbar); δ_H (300 MHz; $CDCl_3$) 3.93 [2H, sp, *J* 6.9, 2 × C*H*(CH_3)$_2$], 1.28 [12H, d, *J* 6.9, 2 × CH_2(C*H*$_3$)$_2$]; δ_P (121 MHz, $CDCl_3$) 170.8 These data are in good agreement with the literature values.[138]

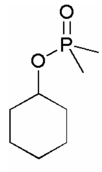
4.1.30. Diisopropylamino dimethylphosphine 113



113

Diisopropylphosphoramidous dichloride **112** (5.0 g, 4.6 mL, 24.7 mmol, 1.0 equiv) was dissolved in dry diethyl ether (50 mL) under an atmosphere of nitrogen. The mixture was cooled to - 78 °C and methyl magnesium bromide (3.0 M solution in diethyl ether, 19.0 mL, 56.9 mmol, 2.3 equiv) was added dropwise over 20 min. The mixture was allowed to warm to RT and stirred for 1 h. The reaction was adjudged to be complete by ^{31}P NMR analysis, and the resulting white precipitate removed by Schlenk filtration. The filtrate was concentrated under reduced pressure and the resulting oil was purified by Kugelrohr distillation, furnishing diisopropylamino dimethylphosphine **113** (2.3 g yield, 58%) as a colourless oil; bp 30 °C (13 mbar); δ_H (300 MHz; $CDCl_3$) 3.15 [2H, sp, *J* 6.1, 2 × C*H*(CH_3)$_2$], 0.95 [18H, m, 2 × CH_2(C*H*$_3$)$_2$ and 2 × C*H*$_3$]; δ_P (121 MHz, $CDCl_3$) 8.3. These data are in good agreement with the literature values.[123]

4.1.31. Cyclohexyl dimethylphosphinate 110



110

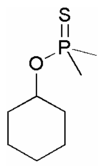
Method 1.

Diisopropylphosphoramidous dichloride **112** (474 mg, 433 µL, 2.3 mmol, 2.0 equiv) was dissolved in dry diethyl ether (20 mL) under an atmosphere of nitrogen. The resulting mixture was cooled to - 78 °C and methyl lithium (1.6 M in hexane, 3.0 mL, 4.9 mmol, 4.2 equiv) was added dropwise over 30 min. The resulting mixture was stirred for 1 h at - 78 °C then warmed to RT when the reaction was adjudged to be incomplete by ^{31}P NMR analysis. The mixture was re-cooled to - 78 °C and methyl lithium (1.6 M in hexane, 1.4 mL, 2.3 mmol, 2.0 equiv) was added dropwise over 5 min. The mixture was stirred for 30 min at - 78 °C and then warmed to RT. The reaction was adjudged to be complete by ^{31}P NMR analysis (δ_P 8.7) and the reaction mixture was cannulated onto a stirred solution of cyclohexanol (116 mg, 121 µL, 1.2 mmol, 1.0 equiv) and imidazole (158 mg, 2.3 mmol, 2.0 equiv) in dry dichloromethane (10 mL) under an atmosphere of nitrogen at - 78 °C. The resulting mixture was warmed to RT and stirred overnight. The reaction was adjudged to be complete by ^{31}P NMR analysis (δ_P 112.0) and cooled to - 78 °C and 3-chloroperoxybenzoic acid (75% w/w, 400 mg, 2.3 mmol, 2.0 equiv) was added. The resulting mixture was stirred for 10 min at - 78 °C then was warmed to RT and stirred for 30 min. The 3-chloroperoxybenzoic acid was quenched with a 10% aqueous solution of sodium hydrogen sulfite (10 mL). The layers were separated and the aqueous layer extracted with dichloromethane (3 × 10 mL). The combined organic layers were washed with a saturated solution of sodium hydrogen carbonate (10 mL), brine (10 mL), dried (magnesium sulfate), filtered and concentrated under reduced pressure. Purification by silica gel column chromatography, eluting with triethylamine/ethyl acetate (1/99) then triethylamine/methanol/ethyl acetate (1/4/95) gave *cyclohexyl dimethylphosphinate* **110** as a deliquescent solid [175 mg yield, 83% (with respect to cyclohexanol)]; R_f 0.47 (methanol/ethyl acetate 30/70); ν_{max} (*KBr disc*)/cm^{-1} 2932.9 (s), 2853.1 (m), 1718.3 (s), 1654.2 (w), 1508.3 (w), 1457.8 (s), 1376.4 (w), 1259.4 (m), 1217.4 (m), 1079.1 (s), 1020.2 (s), 865.0 (w), 801.7 (m), 771.3 (m) and 697.8 (w); δ_H (300 MHz; $CDCl_3$) 4.35-4.22 (1H, m, OC*H*), 1.91-1.80 (2H, m, cyclohexane ring), 1.91-1.73 (2H, m, cyclohexane ring), 1.43 (6H, d, $J_{P\text{-}H}$ 13.8, 2 × C*H*$_3$), 1.38-1.08 (6H, m, cyclohexane ring); δ_C (75 MHz; $CDCl_3$) 73.8 [d, $J_{P\text{-}C}$ 6.6, P(O)O*C*H], 34.2 (d, $J_{P\text{-}C}$ 3.3, *C*-2 position *C*H_2 and *C*-6 position *C*H_2), 23.8 (*C*-4 position *C*H_2), 22.5 (*C*-3 position *C*H_2 and *C*-5 position *C*H_2) 16.6 (d, $J_{P\text{-}C}$ 95.0, 2 × *C*H_3); δ_P (121 MHz; $CDCl_3$) 51.9; *m/z* (ES+) (Found: $[M+Na]^+$ 199.0858. $C_8H_{17}O_2NaP$ requires $[M+Na]^+$, 199.0864); *m/z* (ES+) 375 ($[2M+Na]^+$, 100%), 199 $[M+Na]^+$ (50). These data correlate well with the experimental data for a similar compound.[120]

Method 2.

Diisopropylamino dimethylphosphine **113** (386 mg, 2.4 mmol, 2.5 equiv) and 1*H*-tetrazole (0.43 M solution in acetonitrile, 5.6 mL, 2.4 mmol, 2.5 equiv) were dissolved in dry dichloromethane (5 mL) under an atmosphere of nitrogen. The mixture was cooled to - 78 °C and dry cyclohexanol (96 mg, 100 μL, 960 μmol, 1.0 equiv) was added. The resulting mixture was allowed to warm to RT and stirred for 1.5 h. ^{31}P NMR analysis indicated the complete conversion to the intermediate phosphinite (δ_P 112.3). The mixture was re-cooled to - 78 °C and 3-chloroperoxybenzoic acid (60% w/w, 414 mg, 2.4 mmol, 2.5 equiv) was added. The resulting mixture was allowed to warm to RT and stirred for 30 min. The 3-chloroperoxybenzoic acid was quenched with a 10% aqueous solution of sodium hydrogen sulfite (5 mL). The layers were separated and the aqueous layer extracted with dichloromethane (3 × 5 mL). The combined organic layers were washed with a saturated solution of sodium hydrogen carbonate (5 mL), brine (5 mL), dried (magnesium sulfate), filtered and concentrated under reduced pressure. Purification by silica gel column chromatography, eluting with methanol/ethyl acetate (10/90) yielded *cyclohexyl dimethylphosphinate* **110** as a deliquescent solid [152 mg yield, 90% (with respect to cyclohexanol)]; R_f 0.50 (methanol/ethyl acetate 30/70).

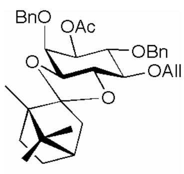
4.1.32. Cyclohexyl dimethylphosphinothioate 111



111

Diisopropylphosphoramidous dichloride **112** (500 mg, 456 μL, 2.5 mmol, 2.0 equiv) was dissolved in dry diethyl ether (20 mL) under an atmosphere of nitrogen. The resulting mixture was cooled to - 78 °C and methyl lithium (1.6 M in hexane, 4.6 mL, 8.4 mmol, 6.8 equiv) was added dropwise over 30 min. The resulting mixture was stirred for 10 min at - 78 °C and then for 30 min at RT when the reaction was adjudged to be incomplete by ^{31}P NMR analysis. The mixture was re-cooled to - 78 °C and methyl lithium (1.6 M in hexane, 0.6 mL, 1.0 mmol, 0.8 equiv) of was added dropwise over 5 min. The mixture was stirred for 10 min at - 78 °C and for 30 min at RT. The reaction was adjudged to be complete by ^{31}P NMR analysis and the reaction mixture was cannulated onto a stirred solution of cyclohexanol (115 mg, 120 mL, 1.0 mmol, 1.0 equiv) and imidazole (157 mg, 2.3 mmol, 2.0 equiv) in dry dichloromethane (15 mL) under an atmosphere of nitrogen at - 78 °C. The resulting mixture was warmed to RT and stirred overnight. The reaction was adjudged to be complete by ^{31}P NMR analysis and sulfur (74 mg, 2.3 mmol, 2.0 equiv) was added. The resulting mixture was stirred for 30 min at RT. The sulfur was quenched with a 10% aqueous solution of sodium hydrogen sulfite (10 mL). The layers were separated and the aqueous layer extracted with dichloromethane (3 × 10 mL). The combined organic layers were washed with a saturated solution of sodium hydrogen carbonate (10 mL), brine (10 mL), dried (magnesium sulfate), filtered and concentrated under reduced pressure. Purification by silica gel column chromatography, eluting with triethylamine/ethyl acetate/petrol ether (1/4/95) gave cyclohexyl dimethylphosphinothioate **111** as a colourless solid [116 mg yield, 53% (with respect to cyclohexanol)]; R_f 0.55 (ethyl acetate/petroleum ether 20/80); mp 59-60 °C (*from ethyl acetate/petroleum ether*, Lit.[121,122] 62 °C); δ_H (300 MHz; $CDCl_3$) 4.50-4.36 (1H, m, OC*H*), 1.88-1.80 (2H, m, cyclohexane ring), 1.76 (6H, d, J_{HP} 13.3, 2 × CH_3), 1.73-1.60 (2H, m, cyclohexane ring), 1.48-1.10 (6H, m, cyclohexane ring); δ_C (125 MHz; C_6D_6) 73.7 [d, J_{CP} 6.2, P(S)O*C*H], 34.0 (d, J_{CP} 4.1, *C*-2 position *C*H_2 and *C*-6 position *C*H_2), 25.2 (*C*-3 position *C*H_2 and *C*-5 position *C*H_2), 24.7 (d, J_{CP} 74.7, 2 × *C*H_3), 23.7 (*C*-4 position *C*H_2); δ_P (121 MHz; $CDCl_3$) 91.0; *m/z* (ES+) [Found: (M) 192.0741. $C_8H_{17}OPS$ requires *M*, 192.0738]; *m/z* (ES+) 111 ($[C_2H_8OPS]^+$, 100%), 54 (10), 67 (20), 77 (15), 92 (35), 95 (20). These data are in good agreement with the literature values.[121,122]

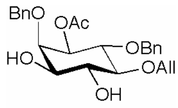
4.1.33. (-)-1D-5-*O*-Allyl-2,6-bis-*O*-benzyl-1-*O*-(acetyl)-3-*O*-*endo*-4-*O*-*exo*-(L-1',7',7'-trimethylbicyclo[2.2.1]hept-2'-ylidene)-*myo*-inositol 79



79

(-)-1D-5-*O*-Allyl-2,6-bis-*O*-benzyl-3-*O*-*endo*-4-*O*-*exo*-(L-1',7',7'-trimethylbicyclo[2.2.1] hept-2'-ylidene)-*myo*-inositol **34** (1.0 g, 1.9 mmol, 1.0 equiv) was dissolved in dry pyridine (10 mL) under an atmosphere of nitrogen and 4-dimethylaminopyridine (71 mg, 580 μmol, 0.3 equiv) was added. The mixture was cooled to 0 °C and acetic anhydride (236 mg, 219 μL, 2.3 mmol, 1.2 equiv) was added dropwise. The mixture was warmed to RT and stirred for 5 h. The acetic anhydride was quenched with water (2 mL) and the solvent removed under reduced pressure. The residue was reconstituted in ethyl acetate (20 mL) and water (20 mL), the layers separated and the aqueous layer extracted with ethyl acetate (3 × 15 mL). The combined organic layers were washed with a saturated aqueous solution of sodium hydrogen carbonate (15 mL), brine (15 mL), dried (magnesium sulfate), filtered and concentrated under reduced pressure. Purification by silica gel column chromatography, eluting with ethyl acetate/petroleum ether (5/95) gave *(-)-1D-5-O-Allyl-2,6-bis-O-benzyl-1-O-(acetyl)-3-O-endo-4-O-exo-(L-1',7',7'-trimethylbicyclo [2.2.1]hept-2'-ylidene)-myo-inositol* **79** (821 mg yield, 74%) as a colourless oil (Found: C, 73.0, H, 7.5; $C_{35}H_{44}O_7$ requires C, 72.9, H, 7.7); R_f 0.4 (ethyl acetate/petroleum ether 20/80); R_f 0.45 (ethyl acetate/hexane 30/70); $[\alpha]_D^{26}$ - 53.0 (*c* 0.72 in $CHCl_3$); ν_{max} (*KBr disc*)/cm^{-1} 3031.8 (w), 2952.0 (s), 2874.7 (s), 1743.9 (s), 1497.6 (w), 1454.2 (m), 1372.1 (m), 1310.2 (m), 1237.0 (s), 1168.9 (m), 1087.9 (s), 1046.7 (s), 925.3 (w), 843.9 (w), 776.6 (w), 736.0 (m), 697.3 (m); δ_H (300 MHz; $CDCl_3$) 7.40-7.25 (10H, m, Ar*H*), 5.96 (1H, ddt *J* 17.2, 10.5, 5.6 C*H*=CH_2), 5.32 (1H, ddt, *J* 17.2, 1.8, 1.5, CH=C*H*H), 5.17 (1H, ddt, *J* 10.5, 1.8, 1.3, CH=CH*H*), 4.90 (1H, d, J_{AB} 11.3, OCH_AH_B), 4.87 (1H, d, $J_{A'B'}$ 12.3, $OCH_{A'}H_{B'}$), 4.85 (1H, dd, *J* 10.0, 3.1, *C*-1 position inositol ring proton), 4.66 (1H, d, J_{AB} 11.3, OCH_AH_B), 4.63 (1H, d, $J_{A'B'}$ 12.3, $OCH_{A'}H_{B'}$), 4.40 (1H, dddd, *J* 12.8, 5.6, 1.8, 1.5, C*H*HCH=CH_2), 4.27, (1H, dd, *J* 3.1, 1.8, inositol ring), 4.20 (1H, dddd, *J* 12.8, 5.6, 1.8, 1.3, CH*H*CH=CH_2), 4.02 (1H, t, *J* 9.7, inositol ring), 3.88 (1H, dd, *J* 10.0, 8.4, inositol ring), 3.57 (1H, dd, *J* 9.7, 8.4, inositol ring), 3.34 (1H, dd, *J* 10.0, 1.8, inositol ring), 2.19-2.13 (1H, m, camphor ring), 1.97 (3H, s, CH_3CO),1.94-1.88 (1H, m, camphor ring), 1.77-1.67 (2H, m, camphor ring), 1.49-1.35 (2H, m, camphor ring), 1.27-1.18 (1H, m, camphor ring), 1.03 (3H, s, CH_3-camphor bridge), 0.87 (3H, s, CH_3-camphor bridge), 0.86 (3H, s, CH_3-camphor bridge); δ_C (75 MHz; $CDCl_3$) 170.6 (*C*=O), 139.2 (Ar*C*), 138.6 (Ar*C*), 135.6 (*C*H=CH_2), 128.7 (Ar*C*), 128.2 (Ar*C*), 128.1 (Ar*C*), 127.9 (Ar*C*), 121.7 (ketyl carbon), 117.0 (CH=CH_2), 81.3 (inositol ring), 81.1 (inositol ring), 77.5 (inositol ring), 76.6 (inositol ring), 76.3 (CH_2), 74.9 (inositol ring), 74.2 (CH_2), 72.3 (inositol ring), 72.1 (CH_2), 53.3 (C_q), 48.7 (C_q), 46.5 (CH_2), 45.3 (*C*H), 29.3 (CH_2), 27.1 (CH_2), 21.3 (CH_3), 20.7 (CH_3), 20.6 (CH_3), 10.1 (CH_3); *m/z* (ES+) [Found: (M+Na)$^+$ 599.2972. $C_{35}H_{44}O_7Na$ requires M^+, 599.2985], *m/z* (ES+) 599 ([M+Na]$^+$, 100%).

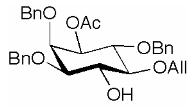
4.1.34. (-)-1D-5-*O*-Allyl-2,6-bis-*O*-benzyl-1-*O*-(acetyl)-*myo*-inositol 80



80

(-)-1D-5-O-Allyl-2,6-bis-O-benzyl-1-O-(acetyl)-3-O-endo-4-O-exo-(L-1',7',7'-trimethyl bicyclo[2.2.1]hept-2'-ylidene)-myo-inositol **79** (740 mg, 1.3 mmol, 1.0 equiv) was dissolved in methanol (20 mL) and dichloromethane (30 mL) under an atmosphere of nitrogen and acetyl chloride (60 mg, 55 µL, 0.8 mmol, 0.6 equiv) was added. The resulting mixture was stirred for 4h, then the generated hydrochloric acid reaction was quenched by the addition of triethylamine (1 mL) and the solvent removed under reduced pressure. Purification by silica gel column chromatography, eluting with ethyl acetate/petroleum ether (30/70, then 50/50) and then ethyl acetate gave *(-)-1D-5-O-allyl-2,6-bis-O-benzyl-1-O-(acetyl)-myo-inositol* **80** (450 mg yield, 79%) as a colourless solid (Found: C, 67.9, H, 6.8; $C_{25}H_{30}O_7$ requires C, 67.9, H, 6.9); R_f 0.1 (ethyl acetate/petroleum ether 50/50); mp 128-129 °C (*from ethyl acetate/petroleum ether*); $[\alpha]_D^{26}$ - 59.3 (*c* 0.53 in $CHCl_3$); ν_{max} (*KBr disc*)/cm^{-1} 3428.1 (s), 3066.5 (w), 2909.7 (m), 1735.4 (s), 1458.1 (w), 1368.9 (m), 1238.1 (s), 1161.8 (m), 1130.0 (w), 1058.3 (s), 926.1 (w), 904.5 (w), 730.2 (m), 696.1 (m), 623.5 (w); δ_H (300 MHz; $CDCl_3$) 7.46-7.29 (10H, m, Ar*H*), 5.96 (1H, ddt *J* 17.2, 10.5, 5.6 C*H*=CH_2), 5.29 (1H, ddt, *J* 17.2, 1.8, 1.5, CH=C*H*H), 5.19 (1H, ddt, *J* 10.5, 1.8, 1.3, CH=CH*H*), 4.88-4.76 (3H, m, 1 × *C*-1 position inositol ring proton, 1 × OCH_AH_B and 1 × OC$H_{A'}H_{B'}$), 4.70 (1H, d, J_{AB} 11.3, OC$H_A$$H_B$), 4.69 (1H, d, $J_{A'B'}$ 11.8, OC$H_{A'}H_{B'}$), 4.40 (1H, m, C*H*HCH=CH_2), 4.28, (1H, m, CH*H*CH=CH_2), 4.05 (1H, t, *J* 2.8, inositol ring), 3.97 (1H, t, *J* 9.5, inositol ring), 3.84 (1H, td, *J* 9.7, 2.3, inositol ring), 3.58-3.52 (1H, m, inositol ring), 3.30 (1H, t, *J* 9.2, inositol ring), 2.56 (1H, br s, O*H*), 2.30 (1H, d, *J* 6.7, O*H*), 1.96, (3H, s, CH_3); δ_C (75 MHz; $CDCl_3$) 170.3 (*C*=O), 138.5 (Ar*C*), 138.3 (Ar*C*), 135.0 (*C*H=CH_2), 128.5 (Ar*C*H), 128.4 (Ar*C*H), 127.82 (Ar*C*H), 127.8 (Ar*C*H), 127.74 (Ar*C*H), 127.7 (Ar*C*H), 117.1 (CH=*C*H_2), 82.6 (inositol ring), 79.5 (inositol ring), 77.8 (inositol ring), 75.4 (*C*H_2), 75.3 (*C*H_2), 74.3 (*C*H_2), 73.9 (inositol ring), 73.5 (inositol ring), 72.1 (inositol ring), 20.9 [C(O)*C*H_3]; *m/z* (ES+) [Found: $(M+Na)^+$ 465.1888. $C_{25}H_{30}O_7Na$ requires M^+, 465.1889], *m/z* (ES+) 465 ($[M+Na]^+$, 100%).

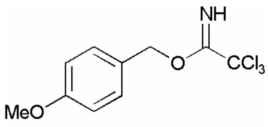
4.1.35. (-)-1D-5-*O*-Allyl-2,3,6-tris-*O*-benzyl-1-*O*-(acetyl)-*myo*-inositol 81



81

(-)-1D-5-*O*-Allyl-2,6-bis-*O*-benzyl-1-*O*-(acetyl)-*myo*-inositol **80** (425 mg, 960 μmol, 1.0 equiv), di-*n*-butyltin oxide (263 mg, 1.1 mmol, 1.1 equiv), tetra-*n*-butylammonium iodide (390 mg, 1.1 mmol, 1.0 equiv) and benzyl bromide (787 mg, 548 μL, 4.6 mmol, 4.8 equiv) were suspended in acetonitrile (50 mL) under an atmosphere of nitrogen. The mixture was heated under reflux for 24 h using soxhlet apparatus filled with 3 Å molecular sieves to remove water generated in the reaction. The reaction mixture was cooled to RT and the solvent was removed under reduced pressure. The residue was suspended in water (20 mL) and extracted with ethyl acetate (3 × 20 mL). The combined organic layers were washed with a saturated aqueous solution of sodium hydrogen carbonate (10 mL) and the formed solid was removed by filtration through Celite®. The filtrate was washed with brine (10 mL), dried (magnesium sulfate), filtered and concentrated under reduced pressure. Purification by silica gel column chromatography, eluting with diethyl ether/petroleum ether (40/60) yielded a mixture of two compounds that was re-columned eluting with diethyl ether/petroleum ether (20/80) to furnish *(-)-1D-5-O-allyl-1-O-(acetyl)-2,3,6-tris-O-benzyl-myo-inositol* **81** as a colourless solid (438 mg yield, 56%) (Found: C, 72.2, H, 6.8; $C_{32}H_{36}O_7$ requires C, 72.2, H, 6.8); R_f 0.6 (diethyl ether/petroleum ether 60/40); mp 56-57 °C (*from diethyl ether/petroleum ether*); $[\alpha]_D^{26}$ - 31.4 (*c* 0.47 in $CHCl_3$); ν_{max} (*KBr disc*)/cm^{-1} 3514.7 (s), 3034.1 (m), 2912.8 (s), 1719.0 (s), 1454.1 (m), 1369.9 (m), 1256.2 (s), 1168.4 (m), 1124.0 (s), 1046.0 (s), 940.1 (m), 917.0 (w), 745.8 (s), 696.0 (s), 624.4 (w), 526.9 (w), 473.7 (w); δ_H (300 MHz; $CDCl_3$) 7.39-7.29 (15H, m, Ar*H*), 6.00 (1H, ddt *J* 17.2, 10.5, 5.6 C*H*=CH_2), 5.32 (1H, ddt, *J* 17.2, 1.8, 1.5, CH=C*H*H), 5.19 (1H, ddt, *J* 10.5, 1.8, 1.3, CH=CH*H*), 4.86 (1H, d, J_{AB} 11.3, OCH_AH$_B$), 4.81 (1H, d, $J_{A'B'}$ 11.8, OC$H_{A'}$H$_{B'}$), 4.76 (1H, dd, *J* 10.2, 12.8, *C*-1 position inositol ring proton), 4.72 (1H, d, *J* 11.3, OCH$_A$$H_B$), 4.70 (1H, d, *J* 11.8, OC$H_{A''}$H$_{B''}$), 4.67 (1H, d, *J* 11.8, OCH$_{A'}$$H_{B'}$), 4.61 (1H, d, *J* 11.8, OCH$_{A''}$$H_{B''}$), 4.28-4.26, (2H, m, inositol ring), 4.07-4.00 (2H, m, CH_2CH=CH_2), 3.92 (1H, t, *J* 9.5, inositol ring), 3.25 (1H, dd, *J* 9.7, 2.3, inositol ring), 3.22 (1H, t, *J* 9.2, inositol ring) 2.49 (1H, d, *J* 2.0, O*H*); δ_C (75 MHz; $CDCl_3$) 170.9 (*C*=O), 139.0 (Ar*C*), 138.8 (Ar*C*), 138.2 (Ar*C*), 135.6 (*C*H=CH_2), 129.0 (Ar*C*H), 128.8 (Ar*C*H), 128.7 (Ar*C*H), 128.4 (Ar*C*H), 128.2 (Ar*C*H), 128.12 (Ar*C*H), 128.1 (Ar*C*H), 117.4 (CH=CH_2), 83.2 (inositol ring), 80.5 (inositol ring), 79.8 (inositol ring), 75.9 (CH_2), 75.0 (CH_2), 74.72 (CH_2), 74.7 (inositol ring), 74.3 (inositol ring), 73.1 (inositol ring), 73.0 (CH_2), 21.4 [C(O)CH_3]; *m/z* (ES+) [Found: $(M+Na)^+$ 555.2349. $C_{32}H_{36}O_7Na$ requires M^+, 555.2359], *m/z* (ES+) 555 ($[M+Na]^+$, 100%), 556 (40).

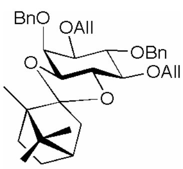
4.1.36. 4-Methoxybenzyl 2,2,2-trichloroacetimidate 84



84

4-Methoxybenzyl alcohol **83** (10.0 g, 72.4 mmol, 10.0 equiv) was dissolved in dichloromethane (80 mL), tetra-*n*-butylammonium hydrogen sulfate (246 mg, 0.7 mmol, 0.01 equiv) and a 50% aqueous solution of potassium hydroxide (80 mL) were added and the resulting mixture cooled at - 10 °C. Trichloroacetonitrile (12.0 g, 8.3 mL, 82.2 mmol, 1.1 equiv) was added dropwise with vigorous stirring over a period of 30 min. The resulting mixture was allowed to warm to RT and stirred for 2 h. The layers were separated and the aqueous layer extracted with diethyl ether (3 × 100 mL). The combined organic layers were dried (magnesium sulfate), filtered and concentrated under reduced pressure. Purification by activated aluminium oxide column chromatography, eluting with ethyl acetate/petroleum ether (5/95) furnished the title compound **84** as a colourless oil (7.4 g yield, 36%); R_f 0.36 (ethyl acetate/petroleum ether 20/80); δ_H (300 MHz; $CDCl_3$) 8.37 (1H, br s, *H*N), 7.39 (2H, d, *J* 8.7, Ar*H*), 6.92, (2H, d, *J* 8.7, Ar*H*), 5.28 (2H, s, OCH_2Ph), 3.83 (3H, s, OCH_3). These data are in good agreement with the literature values.[139]

4.1.37. (-)-1D-1,5-bis-*O*-Allyl-2,6-bis-*O*-benzyl-3-*O*-*endo*-4-*O*-*exo*-(L-1',7',7'-trimethylbicyclo[2.2.1]hept-2'-ylidene)-*myo*-inositol 103

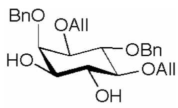


103

(-)-1D-5-*O*-Allyl-2,6-bis-*O*-benzyl-3-*O*-*endo*-4-*O*-*exo*-(L-1’,7’,7’-trimethylbicyclo[2.2.1] hept-2’-ylidene)-*myo*-inositol **34** (2.4 g, 4.5 mmol, 1.0 equiv) was dissolved in dry tetrahydrofuran (20 mL) under an atmosphere of nitrogen, the resulting mixture was cooled to 0 °C and sodium hydride (219 mg, 60% dispersion in mineral oil, 5.4 mmol, 1.2 equiv) was added. The resulting mixture was allowed to warm to RT and stirred for 1 h. The mixture was then re-cooled to 0 °C and imidazole (catalytic amount) and tetra-*n*-butylammonium iodide (catalytic amount) were added, followed by allyl bromide (653 mg, 472 µL, 5.4 mmol, 1.2 equiv) which was added dropwise. The reaction mixture was allowed to warm to RT, then dry *N,N*-dimethyl formamide (30 mL) was added and the resulting mixture stirred overnight. The sodium hydride was quenched with water (2 mL), the solvent removed under reduced pressure and the residue reconstituted in ethyl acetate (15 mL) and water (15 mL). The layers were separated and the aqueous layer extracted with ethyl acetate (3 × 15 mL). The combined organic layers were washed with brine (10 mL), dried (magnesium sulfate), filtered and concentrated under reduced pressure to afford a yellow oil. Purification by silica gel column chromatography, eluting with ethyl acetate/petroleum ether (5/95) yielded *(-)-1D-1,5-bis-O-allyl-2,6-bis-O-benzyl-3-O-endo-4-O-exo-(L-1',7',7'-trimethylbicyclo[2.2.1]hept-2'-ylidene)-myo-inositol* **103** (2.4 g yield, 91%) as a colourless solid. (Found: C, 75.3, H, 8.3; $C_{36}H_{46}O_6$ requires C, 75.2, H, 8.1); R_f 0.45 (ethyl acetate/petroleum ether 20/80); $[\alpha]_D^{26}$ - 23.0 (*c* 0.49 in $CHCl_3$); mp 55-57 °C (*from ethyl acetate/petroleum ether*); ν_{max} (*KBr disc*)/cm^{-1} 3064.4 (w), 3025.2 (w), 2932.8 (s), 2868.3 (s), 1647.6 (w), 1453.9 (m), 1366.6 (m), 1309.5 (m), 1203.4 (m), 1092.7 (s), 1048.9 (s), 921.2 (s), 778.2 (w), 747.9 (s), 697.6 (s), 595.8 (w); δ_H (300 MHz; $CDCl_3$) 7.40-7.25 (10H, m, Ar*H*), 6.06-5.85 (2H, m, $CH_X{=}CH_YH_Z$+ $CH_{X'}{=}CH_{Y'}H_{Z'}$), 5.35 (1H, ddt, *J* 17.1, 1.8, 1.5, $CH_X{=}CH_YH_Z$), 5.30 (1H, ddt, *J* 17.4, 1.8, 1.3, $CH_{X'}{=}CH_{Y'}H_{Z'}$), 5.19 (2H, ddt, *J* 10.2, 1.8, 1.3, $CH_X{=}CH_YH_Z$ + $CH_{X'}{=}CH_{Y'}H_{Z'}$), 4.93 (1H, d, J_{AB} 12.3, OCH_AH_B), 4.88 (1H, d, $J_{A'B'}$ 10.5, $OCH_{A'}H_{B'}$), 4.86 (1H, d, J_{AB} 12.3, OCH_AH_B), 4.84 (1H, d, $J_{A'B'}$ 10.5, $OCH_{A'}H_{B'}$), 4.40 (1H, ddt, *J* 13.1, 5.4, 1.5, $CH_vH_WCH_X{=}CH_YH_Z$), 4.27-4.20, (2H, m, 1 × $CH_VH_WCH_X{=}CH_YH_Z$ + $CH_{V'}H_{W'}CH_{X'}{=}CH_{Y'}H_{Z'}$), 4.09-4.02 (3H, m, 1 × $CH_{V'}H_{W'}CH_{X'}{=}CH_{Y'}H_{Z'}$ + 2 × inositol ring), 3.85 (1H, t, *J* 9.2, inositol ring), 3.51 (1H, dd, *J* 9.5, 8.7, inositol ring), 3.42 (1H, dd, *J* 9.7, 3.0, inositol ring), 3.22 (1H, dd, *J* 9.7, 1.5, inositol ring), 2.16 (1H, dt, *J* 13.6, 3.6, camphor ring), 2.02-1.93 (1H, m, camphor ring), 1.77-1.71 (2H, m, camphor ring), 1.48-1.36 (2H, m, camphor ring), 1.29-1.19 (1H, m, camphor ring), 1.05 (3H, s, CH_3-camphor bridge), 0.91 (3H, s, CH_3-camphor bridge), 0.88 (3H, s, CH_3-camphor bridge); δ_C (75 MHz; $CDCl_3$), 139.1 (Ar*C*), 138.5 (Ar*C*), 135.4 ($CH_X{=}CH_YH_Z$), 134.9 ($CH_{X'}{=}CH_{Y'}H_{Z'}$), 128.3 (Ar*C*H), 128.2 (Ar*C*H), 127.9 (Ar*C*H), 127.55 (Ar*C*H), 127.5 (Ar*C*H), 120.4 (ketyl carbon), 117.1 ($CH_X{=}CH_YH_Z$), 116.4 ($CH_{X'}{=}CH_{Y'}H_{Z'}$), 82.8 (inositol ring), 81.2 (inositol ring), 81.0 (inositol ring), 77.2

(inositol ring), 76.7 (inositol ring), 76.5 (CH_2), 73.3 (CH_2), 71.7 (CH_2), 71.66 (CH_2), 70.8 (inositol ring), 52.9 (C_q), 48.2 (C_q), 46.2 (CH_2), 45.0 (*C*H), 29.0 (CH_2), 26.8 (CH_2), 20.4 (CH_3), 20.2 (CH_3), 9.7 (CH_3); *m/z* (ES+) [Found: $(M+Na)^+$ 597.3171. $C_{36}H_{46}O_6Na$ requires M^+, 597.3192], *m/z* (ES+) 597 ($[M+Na]^+$, 100%).

4.1.38. (-)-1D-1,5-bis-*O*-Allyl-2,6-bis-*O*-benzyl-*myo*-inositol 104

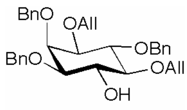


104

(-)-1D-1,5-bis-*O*-Allyl-2,6-bis-*O*-benzyl-3-*O*-endo-4-*O*-exo-(L-1',7',7'-trimethylbicyclo [2.2.1]hept-2'-ylidene)-*myo*-inositol **104** (2.4 g, 4.1 mmol, 1.0 equiv) was dissolved in methanol/dichloromethane 2/3 (50 mL) under an atmosphere of nitrogen. Acetyl chloride (194 mg, 176 µL, 2.5 mmol, 0.6 equiv) was added and the resulting mixture stirred for 4 h. The generated hydrochloric acid was quenched with triethylamine (1 mL), the solvent removed under reduced pressure and the resulting yellow solid adsorbed onto silica and purified by silica gel column chromatography, eluting with ethyl acetate/petroleum ether (30/70), to yield *(-)-1D-1,5-bis-O-allyl-2,6-bis-O-benzyl-myo-inositol* **104** (1.6 g yield, 88%) as a colourless solid. (Found: C, 70.6, H, 7.6; $C_{36}H_{46}O_6$ requires C, 70.9, H, 7.3); R_f 0.55 (ethyl acetate); $[\alpha]_D^{26}$ - 16.8 (*c* 0.64 in $CHCl_3$); mp 119-120 °C (*from ethyl acetate/petroleum ether*); ν_{max} (*KBr disc*)/cm^{-1} 3405.3 (s), 3066.5 (m), 3034.6 (m), 2910.9 (s), 2862.7 (s), 1647.7 (w), 1497.4 (m), 1455.7 (s), 1425.7 (s), 1354.7 (s), 1255.6 (w), 1160.8 (s), 1052.2 (s), 992.8 (s), 928.6 (s), 724.1 (s), 696.4 (s), 576.2 (w), 460.1 (w); δ_H (300 MHz; $CDCl_3$) 7.43-7.28 (10H, m, Ar*H*), 6.05-5.90 (2H, m, $CH_X{=}CH_YH_Z$ + $CH_{X'}{=}CH_{Y'}H_{Z'}$), 5.35 (1H, ddt, *J* 17.1, 1.8, 1.5, $CH_X{=}CH_YH_Z$), 5.30 (1H, ddt, *J* 17.2, 1.8, 1.5, $CH_{X'}{=}CH_{Y'}H_{Z'}$), 5.22 (1H, ddt, *J* 10.5, 1.5, 1.3, $CH_X{=}CH_YH_Z$), 5.14 (1H, ddt, *J* 10.2, 1.8, 1.3, $CH_{X'}{=}CH_{Y'}H_{Z'}$), 5.05 (1H, d, J_{AB} 11.8, OCH_AH_B), 4.90 (1H, d, $J_{A'B'}$ 10.5, $OCH_{A'}H_{B'}$), 4.80 (1H, d, $J_{A'B'}$ 10.5, $OCH_{A'}H_{B'}$), 4.70 (1H, d, J_{AB} 11.8, OCH_AH_B), 4.41 (1H, ddt, *J* 12.3, 5.6, 1.5, $CH_VH_WCH_X{=}CH_YH_Z$), 4.28 (1H, ddt, *J* 12.3, 5.8, 1.3, $CH_VH_WCH_X{=}CH_YH_Z$), 4.19-4.17, (2H, m, $CH_{V'}H_{W'}CH_{X'}{=}CH_{Y'}H_{Z'}$), 4.02 (1H, t, *J* 2.3, inositol ring), 3.92 (1H, t, *J* 9.7, inositol ring), 3.81 (1H, t, *J* 9.5, inositol ring), 3.42-3.34 (2H, m, inositol ring), 3.19 (1H, t, *J* 9.2, inositol ring), 2.69 (2H, br s, 2 × O*H*); δ_C (75 MHz; $CDCl_3$), 138.7 (Ar*C*), 138.67 (Ar*C*), 135.2 ($CH_X{=}CH_YH_Z$), 134.8 ($CH_{X'}{=}CH_{Y'}H_{Z'}$), 128.4 (Ar*C*H), 128.39 (Ar*C*H), 128.2 (Ar*C*H), 127.8 (Ar*C*H), 127.7 (Ar*C*H), 127.68 (Ar*C*H), 116.95 ($CH_X{=}CH_YH_Z$), 116.92 ($CH_{X'}{=}CH_{Y'}H_{Z'}$), 82.6 (inositol ring), 81.4 (inositol ring), 81.0

(inositol ring), 77.2 (inositol ring), 75.8 (CH_2), 74.8 (CH_2), 74.2 (CH_2), 73.8 (inositol ring), 72.1 (inositol ring), 71.9 (CH_2); *m/z* (ES+) [Found: $(M+Na)^+$ 463.2088. $C_{26}H_{32}O_6Na$ requires M^+, 463.2097], *m/z* (ES+) 463 ($[M+Na]^+$, 100%).

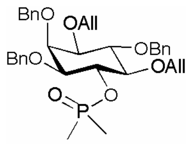
4.1.39. (+)-1D-1,5-bis-*O*-Allyl-2,3,6-tris-*O*-benzyl-*myo*-inositol 100



100

(-)-1D-1,5-bis-*O*-Allyl-2,6-bis-*O*-benzyl-*myo*-inositol **104** (2.0 g, 4.5 mmol, 1.0 equiv), di-*n*-butyltin oxide (1.2 g, 5.0 mmol, 1.1 equiv), tetra-*n*-butylammonium iodide (1.9 g, 4.5 mmol, 1.0 equiv) and benzyl bromide (2.6 mL, 21.8 mmol, 4.8 equiv) were dissolved in acetonitrile (80 mL) under an atmosphere of nitrogen. The mixture was heated under reflux for 24 h, using a soxhlet apparatus filled with 3 Å molecular sieves to remove water generated in the reaction. The reaction mixture was cooled to RT and the solvent was removed under reduced pressure. The residue was reconstituted in ethyl acetate (20 mL) and water (20 mL) the layers separated and the aqueous layer extracted with ethyl acetate (3 × 20 mL). The combined organic layers were washed with a saturated aqueous solution of sodium hydrogen carbonate (20 mL) and the resulting solid was removed by filtration through Celite®. The filtrate was washed with brine (20 mL), dried (magnesium sulfate), filtered and concentrated under reduced pressure to yield a yellow residue. Purification by activated aluminium oxide column chromatography (30 cm path), eluting with ethyl acetate/petroleum ether (50/50) (twice) yielded *(+)-1D-1,5-bis-O-allyl-2,3,6-tris-O-benzyl-myo-inositol* **100** (1.7 g, yield 71%) as a colourless solid. (Found: C, 74.5, H, 7.3; $C_{33}H_{38}O_6$ requires C, 74.7, H, 7.2); R_f 0.23 (ethyl acetate/petroleum ether 30/70); $[\alpha]_D^{26}$ + 2.8 (*c* 0.68 in $CHCl_3$); mp 69-71 °C (*from diethyl ether/petroleum ether*); ν_{max} (*KBr disc*)/cm^{-1} 3530.9 (s), 3258.2 (s), 3064.2 (m), 3030.1 (m), 2893.6 (s), 2862.7 (s), 1648.1 (w), 1497.5 (m), 1454.3 (s), 1350.9 (s), 1210.4 (w), 1128.6 (s), 1069.3 (s), 1027.2 (s), 929.6 (m), 928.6 (s), 755.2 (w), 728.6 (s), 695.4 (s), 565.0 (w); δ_H (300 MHz; $CDCl_3$) 7.44-7.25 (15H, m, Ar*H*), 6.06-5.87 (2H, m, $CH_X=CH_YH_Z$ + $CH_{X'}=CH_{Y'}H_{Z'}$), 5.33 (1H, ddt, *J* 17.2, 1.8, 1.5, $CH_X=CH_YH_Z$), 5.30 (1H, ddt, *J* 17.4, 1.8, 1.5, $CH_{X'}=CH_{Y'}H_{Z'}$), 5.20 (1H, ddt, *J* 10.5, 1.5, 1.3, $CH_X=CH_YH_Z$), 5.18 (1H, ddt, *J* 10.2, 1.8, 1.5, $CH_{X'}=CH_{Y'}H_{Z'}$), 4.90 (1H, d, J_{AB} 12.0, OCH_AH_B), 4.89 (1H, d, $J_{A'B'}$ 10.5, $OCH_{A'}H_{B'}$), 4.82-4.78 (2H, m, OCH_AH_B + $OCH_{A'}H_{B'}$), 4.63 (1H, d, $J_{A''B''}$ 11.8, $OCH_{A''}H_{B''}$), 4.57 (1H, d, $J_{A''B''}$ 11.8, $OCH_{A''}H_{B''}$), 4.43-4.30 (2H, m, C$H_V H_W$CH_X=C$H_Y H_Z$), 4.16-4.09 (3H, m, C$H_{V'} H_{W'}$C$H_{X'}$=C$H_{Y'} H_{Z'}$ + 1 × inositol ring), 4.05 (1H, t, *J* 2.3, inositol ring), 4.00 (1H, t, *J* 9.5, inositol ring), 3.29-3.18 (3H, m, inositol ring), 2.55 (1H, br s, O*H*); δ_C (75 MHz; $CDCl_3$), 139.3 (2 × Ar*C*), 138.4 (Ar*C*), 135.8 (*C*H_X=C$H_Y H_Z$), 135.3 (*C*$H_{X'}$=C$H_{Y'} H_{Z'}$), 128.9 (Ar*C*H), 128.8 (Ar*C*H), 128.6 (Ar*C*H), 128.3 (Ar*C*H), 128.2 (Ar*C*H), 128.16 (Ar*C*H), 128.0 (Ar*C*H), 127.8 (Ar*C*H), 117.2 (CH_X=*C*$H_Y H_Z$ + C$H_{X'}$=*C*$H_{Y'} H_{Z'}$), 83.3 (inositol ring), 81.8 (inositol ring), 81.3 (inositol ring), 80.5 (inositol ring), 76.3 (*C*H_2), 74.6 (*C*H_2), 74.4 (*C*H_2), 74.0 (inositol ring), 73.1 (inositol ring), 72.8 (*C*H_2), 72.2 (*C*H_2); *m/z* (ES+) [Found: $(M+Na)^+$ 553.2563. $C_{33}H_{38}O_6Na$ requires M^+, 553.2566], *m/z* (ES+) 553 ($[M+Na]^+$, 100%).

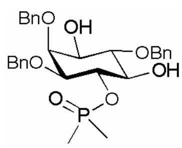
4.1.40. (-)-1D-1,5-bis-*O*-Allyl-2,3,6-tris-*O*-benzyl-4-*O*-dimethylphosphinyl-*myo*-inositol 102



102

Diisopropylamino dimethylphosphine **113** (76 mg, 471 µmol, 2.5 equiv) and 1*H*-tetrazole (0.43 M solution in acetonitrile, 1.1 mL, 471 µmol, 2.5 equiv) were dissolved in dry dichloromethane (3 mL), the resulting mixture was cooled to - 78 °C and (+)-1D-1,5-bis-*O*-allyl-2,3,6-tris-*O*-benzyl-*myo*-inositol **100** (100 mg, 188 µmol, 1.0 equiv) dissolved in dry dichloromethane (2 mL) was added by cannula. The resulting mixture was allowed to warm to RT and stirred overnight. ^{31}P NMR analysis indicated the complete conversion of diisopropylamino dimethylphosphine in the intermediate phosphinite (δ_P 130.0). The mixture was re-cooled to - 78 °C and 3-chloroperoxybenzoic acid (60% w/w, 112 mg, 471 µmol, 2.5 equiv) was added, the resulting mixture warmed to RT and stirred for 30 min. The 3-chloroperoxybenzoic acid was quenched with a 10% aqueous solution of sodium hydrogen sulfite (5 mL), the layers were separated and the aqueous layer was extracted with dichloromethane (3 × 5 mL). The combined organic layers washed with a 10% aqueous solution of sodium hydrogen bicarbonate (5 mL), brine (5 mL), dried (magnesium sulfate), filtered and concentrated under reduced pressure. Purification by silica gel column chromatography, eluting with methanol/ethyl acetate (2/98) yielded *(-)-1D-1,5-bis-O-allyl-2,3,6-tris-O-benzyl-4-O-dimethylphosphinyl-myo-inositol* **102** (107 mg yield, 94%) as a colourless solid; a very pure sample was obtained by crystallisation from diethyl ether/dichloromethane/petroleum ether. (Found: C, 69.3, H, 7.2; $C_{35}H_{43}O_7P$ requires C, 69.3, H, 7.1); R_f 0.38 (ethyl acetate); $[\alpha]_D^{26}$ - 1.9 (*c* 0.27 in $CHCl_3$); mp 122-124 °C (*from diethyl ether/dichloromethane/ petroleum ether*); ν_{max} (*KBr disc*)/cm^{-1} 3064.3 (w), 3031.7 (w), 2823.2 (s), 2851.5 (s), 1454.5 (m), 1302.9 (m), 1216.5 (s), 1130.6 (m), 1096.4 (s), 1050.2 (s), 935.2 (s), 866.9 (m), 736.2 (s), 698.9 (w); δ_H (300 MHz; $CDCl_3$) 7.41-7.28 (15H, m, Ar*H*), 6.04-5.83 (2H, m, CH_X=CH_YH_Z + $CH_{X'}$=$CH_{Y'}H_{Z'}$), 5.33-5.25 (2H, m, CH_X=CH_YH_Z + $CH_{X'}$=$CH_{Y'}H_{Z'}$), 5.18 (1H, ddt, *J* 10.5, 1.8, 1.5, CH_X=CH_YH_Z), 5.15 (1H, ddt, *J* 10.2, 1.5, 1.3, $CH_{X'}$=$CH_{Y'}H_{Z'}$), 4.88-4.74 (4H, m, OCH_AH_B + $OCH_{A'}H_{B'}$), 4.66-4.54 (3H, m, $OCH_{A''}H_{B''}$ and *C*-4 position inositol ring),4.39 (1H, ddt, *J* 12.3, 5.6, 1.5, $CH_VH_WCH_X$=CH_YH_Z), 4.27 (1H, ddt, *J* 12.3, 5.6, 1.5, $CH_VH_WCH_X$=CH_YH_Z), 4.13-4.05 (2H, m, $CH_{V'}H_{W'}CH_{X'}$=$CH_{Y'}H_{Z'}$), 4.00-3.94 (2H, m, inositol ring), 3.33-3.29 (2H, m, inositol ring), 3.24 (1H, dd, *J* 9.8, 2.1, inositol ring), 1.50 [3H, d, J_{HP} 14.1, P(O)C*H*$_3$CH$_3$], 1.49 [3H, d, J_{HP} 14.1, P(O)CH$_3$C*H*$_3$]; δ_C (75 MHz; $CDCl_3$), 139.1 (Ar*C*), 139.0 (Ar*C*), 138.0 (Ar*C*), 135.3 (CH_X=CH_YH_Z), 135.1 ($CH_{X'}$=$CH_{Y'}H_{Z'}$), 128.9 (Ar*C*H), 128.9 (Ar*C*H), 128.8 (Ar*C*H), 128.6 (Ar*C*H), 128.4 (Ar*C*H), 128.3 (Ar*C*H), 128.2 (Ar*C*H), 128.1 (Ar*C*H), 127.9 (Ar*C*H), 117.3 (CH_X=CH_YH_Z), 117.1 ($CH_{X'}$=$CH_{Y'}H_{Z'}$), 81.9 (d, J_{CP} 2.8, inositol ring), 81.7 (inositol ring), 80.7 (inositol ring), 79.4 (d, J_{CP} 2.2, inositol ring), 76.6, (d, J_{CP} 8.3, inositol ring), 76.3 (*C*H$_2$), 74.7 (*C*H$_2$), 74.6 (*C*H$_2$), 73.9 (inositol ring), 73.0 (*C*H$_2$), 72.1 (*C*H$_2$), 17.0 [d, J_{CP} 94.0, P(O)*C*H$_3$CH$_3$], 16.9 [d, J_{CP} 94.0, P(O)CH$_3$*C*H$_3$]; δ_P (121 MHz; $CDCl_3$) 54.8; *m/z* (ES+) [Found: (M+Na)$^+$ 629.2643. $C_{35}H_{43}O_7NaP$ requires *M*$^+$, 629.2644], *m/z* (ES+) 629 ([M+Na]$^+$, 100%).

4.1.41. (-)-1D-2,3,6-tris-*O*-Benzyl-4-*O*-dimethylphosphinyl-*myo*-inositol 117

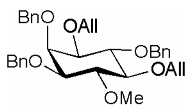


117

(-)-1D-1,5-Bis-*O*-allyl-2,3,6-tris-*O*-benzyl-4-*O*-dimethylphosphinyl-*myo*-inositol **102** (314 mg, 517 µmol, 1.0 equiv), was dissolved methanol/water (4/1, 20 mL) and of 4-toluenesulfonic acid monohydrate (30 mg, 155 µmol, 0.3 equiv) and palladium on activated carbon (loading 10%, 80 mg, 155 µmol, 0.15 equiv) were added. The resulting mixture was heated at 60 °C for 24 h. Analysis by TLC indicated complete consumption of the starting material and the mixture was cooled to RT, the

4-toluenesulfonic acid quenched with triethylamine (1 mL) and the palladium catalyst removed by filtration onto Celite®. The filtrate was concentrated under reduced pressure, the residue reconstituted in water (5 mL) and dichloromethane (5 mL), the layers separated and the aqueous layer extracted with dichloromethane (3 × 5 mL). The combined organic layers were washed with a saturated aqueous solution of sodium hydrogen carbonate (10 mL), brine (10 mL), dried (magnesium sulfate), filtered and concentrated under reduced pressure. The resulting yellow oil was purified three time by silica gel column chromatography, eluting with triethylamine/methanol/dichloromethane (1/2/97) to give *(-)-1D-2,3,6-tris-O-benzyl-4-O-dimethylphosphinyl-myo-inositol* **117** (57 mg yield, 21%) as a colourless gum; R_f 0.56 (methanol/dichloromethane 8/92); ν_{max} (*thin film*)/cm^{-1} 3350.4 (s), 3063.2 (m), 3031.1 (m), 2920.4 (s), 1723.9 (m), 1668.1 (s), 1496.9 (m), 1454.8 (m), 1387.3 (m), 1365.9 (m), 1306.0 (m), 1274.9 (m), 1199.0 (s), 1070.5 (s), 943.8 (s), 876.5 (m), 825.0 (w), 740.5 (s), 700.0 (s), 662.1 (m); δ_H (300 MHz; $CDCl_3$) 7.43-7.28 (15H, m, Ar*H*), 5.11 (1H, d, J_{AB} 11.2, OCH_AH$_B$), 4.87 (1H, d, $J_{A'B'}$ 11.8, OC$H_{A'}$H$_{B'}$), 4.79 (1H, d, $J_{A'B'}$ 11.8, OCH$_{A'}$$H_{B'}$), 4.74 (1H, d, J_{AB} 11.2, OCH$_A$$H_B$), 4.63 (1H, d, $J_{A''B''}$ 11.8, OC$H_{A''}$H$_{B''}$), 4.51 (1H, d, $J_{A''B''}$ 11.8, OCH$_{A''}$$H_{B''}$), 4.09 (1H, t, *J* 2.3, inositol ring), 3.78 (1H, t, *J* 9.2, inositol ring), 3.67 (1H, t, *J* 8.7, inositol ring), 3.49 (1H, dt, *J* 9.5, 2.6, inositol ring), 3.42 (1H, dd, *J* 9.7 2.6, inositol ring), 2.35 (1H, d, *J* 3.8, inositol ring), 1.55 [3H, d, J_{HP} 14.2, P(O)CH_3CH$_3$], 1.53 [3H, d, J_{HP} 14.2, P(O)CH$_3$CH_3]; δ_P (121 MHz; $CDCl_3$) 60.6; *m/z* (ES+) [Found: (M+Na)$^+$ 549.2003. $C_{29}H_{35}O_7NaP$ requires M^+, 540.2018], *m/z* (ES+) 549 ([M+Na]$^+$, 100%).

4.1.42. (-)-1D-1,5-bis-*O*-Allyl-2,3,6-tris-*O*-benzyl-4-*O*-methyl-*myo*-inositol 105

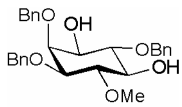


105

(+)-1D-1,5-bis-*O*-Allyl-2,3,6-tris-*O*-benzyl-*myo*-inositol **100** (150 mg, 283 μmol, 1.0 equiv) was dissolved in dry tetrahydrofuran (8 mL) under an atmosphere of nitrogen, the mixture was cooled to 0 °C and sodium hydride (13 mg, 60% dispersion in mineral oil, 311 μmol, 1.1 equiv) was added. The mixture was allowed to warm to RT and stirred for 2 h, then it was re-cooled to 0 °C and methyl iodide (44 mg, 19 μL, 311 μmol, 1.1 equiv) was added. The mixture was warmed to RT and stirred overnight. The sodium hydride was quenched with water (1 mL), the solvent removed under reduced pressure and the residue reconstituted in ethyl acetate

(10 mL) and water (10 mL). The layers were separated and the aqueous layer extracted with ethyl acetate (3 × 10 mL). The combined organic layers were washed with brine (10 mL), dried (magnesium sulfate), filtered and concentrated under reduced pressure. Purification by silica gel column chromatography, eluting with ethyl acetate/petroleum ether (10/90) yielded *(-)-1D-1,5-bis-O-allyl-2,3,6-tris-O-benzyl-O-methyl-myo-inositol* **105** (184 mg yield, 92%) as a colourless waxy solid. (Found: C, 75.2, H, 7.4; $C_{34}H_{40}O_6$ requires C, 75.0, H, 7.4); R_f 0.70 (ethyl acetate/petroleum ether 30/70); mp 35-36 °C (*from ethyl acetate/petroleum ether*); $[\alpha]_D^{26}$ - 4.05 (*c* 0.41 in $CHCl_3$); v_{max} (*KBr disc*)/cm^{-1} 3064.6 (w), 3030.5 (w), 2925.6 (m), 1647.5 (w), 1496.9 (m), 1454.8 (m), 1357.4 (m), 1207.7 (w), 1132.9 (s), 1088.2 (s), 1028.3 (m), 995.6 (w), 924.5 (m), 734.9 (m), 697.2 (m); δ_H (300 MHz; $CDCl_3$) 7.36-7.16 (15H, m, Ar*H*), 5.98-5.76 (2H, m, CH_X=CH_YH_Z + $CH_{X'}$=$CH_{Y'}H_{Z'}$), 5.26-5.18 (2H, m, CH_X=CH_YH_Z + $CH_{X'}$=$CH_{Y'}H_{Z'}$), 5.11-5.06 (2H, m, CH_X=CH_YH_Z + $CH_{X'}$=$CH_{Y'}H_{Z'}$), 4.79 (2H, s, OCH_2Ph), 4.78 (1H, d, $J_{A'B'}$ 10.5, $OCH_{A'}H_{B'}$), 4.70 (1H, d, $J_{A'B'}$ 10.5, $OCH_{A'}H_{B'}$), 4.61 (1H, d, $J_{A''B''}$ 11.8, $OCH_{A''}H_{B''}$) 4.51 (1H, d, $J_{A''B''}$ 11.8, $OCH_{A''}H_{B''}$), 4.25 (2H, dt, *J* 5.6, 1.3, CH_2CH_X=CH_YH_Z), 4.02-3.98 (2H, m, $CH_2CH_{X'}$=$CH_{Y'}H_{Z'}$), 3.90 (1H, t, *J* 2.3, inositol ring), 3.85 (1H, t, *J* 9.4, inositol ring), 3.64 (1H, t, *J* 9.7, inositol ring), 3.58 (2H, s, OCH_3), 3.17-3.09 (3H, m, inositol ring); δ_C (75 MHz; $CDCl_3$), 139.4 (Ar*C*), 139.36 (Ar*C*), 139.1 (Ar*C*), 135.9 (CH_X=CH_YH_Z), 135.4 ($CH_{X'}$=$CH_{Y'}H_{Z'}$), 128.8 (Ar*C*H), 128.76 (Ar*C*H), 128.7 (Ar*C*H), 128.5 (Ar*C*H), 128.2 (Ar*C*H), 128.01 (Ar*C*H), 128.0 (Ar*C*H), 127.8 (Ar*C*H), 127.7 (Ar*C*H), 117.1 (CH_X=CH_YH_Z), 116.9 ($CH_{X'}$=$CH_{Y'}H_{Z'}$), 84.0 (inositol ring), 83.9 (inositol ring), 81.9 (inositol ring), 81.1 (inositol ring), 80.9 (inositol ring), 76.3 (CH_2), 75.0 (CH_2), 74.8 (inositol ring), 74.4 (CH_2), 73.2 (CH_2), 72.8 (CH_2), 61.8 (OCH_3); *m/z* (ES+) [Found: $(M+Na)^+$ 567.2716. $C_{34}H_{40}O_6$Na requires M^+, 567.2723], *m/z* (ES+) 567 ($[M+Na]^+$, 100%).

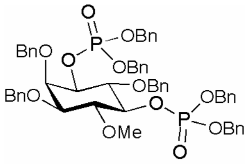
4.1.43. (+)-1D-2,3,6-tris-*O*-Benzyl-4-*O*-methyl-*myo*-inositol 106



106

(-)-1D-1,5-bis-*O*-Allyl-2,3,6-tris-*O*-benzyl-*O*-methyl-*myo*-inositol **105** (80 mg, 147 µmol, 1.0 equiv), Wilkinson's catalyst (41 mg, 44 µmol, 0.3 equiv) and Hunig's base (38 mg, 51 µL, 294 µmol, 2.0 equiv) were suspended in ethanol (8 mL) and the resulting mixture heated under reflux for 3 h. The mixture was then cooled to 0 °C and filtered through Celite® and the filtrate concentrated under reduced pressure. The resulting red residue was dissolved in methanol/dichloromethane (2/3, 8 mL) and acetyl chloride (7 mg, 6 µL, 88 µmol, 0.6 equiv) was added and the mixture stirred for 2 h. The generated hydrochloric acid was quenched with triethylamine (1 mL), the solvent removed under reduced pressure, the residue reconstituted in ethyl acetate (5 mL) and water (5 mL)the layers separated and the aqueous layer extracted with ethyl acetate (3 × 5 mL). The combined organic layers were washed with a saturated aqueous solution of sodium hydrogen carbonate (5 mL), brine (5 mL), dried (magnesium sulfate), filtered and concentrated under reduced pressure. Purification by silica gel column chromatography (twice), eluting with ethyl acetate/petroleum ether (30/70), yielded *(+)-1D-2,3,6-tris-O-benzyl-O-methyl-myo-inositol* **106** (54 mg yield, 79%) as a colourless solid. (Found: C, 72.5, H, 6.9; $C_{28}H_{32}O_6$ requires C, 72.4, H, 6.9); R_f 0.5 (ethyl acetate/petroleum ether 50/50); mp 80-81 °C (*from ethyl acetate/petroleum ether*); $[\alpha]_D^{25}$ + 2.1 (*c* 0.45 in $CHCl_3$); ν_{max} (*KBr disc*)/cm^{-1} 3474.9 (s), 3032.1 (w), 2914.8 (m), 1719.3 (w), 1605.0 (w), 1496.9 (m), 1454.8 (m), 1357.7 (m), 1206.2 (w), 1119.6 (s), 1070.8 (s), 1027.4 (s), 934.3 (w), 869.9 (w), 727.0 (s), 696.2 (s), 572.0 (w), 518.1 (w); δ_H (300 MHz; $CDCl_3$) 7.41-7.29 (15H, m, Ar*H*), 4.99 (1H, d, *J* 11.5, OCH_AH_B), 4.90 (1H, d, *J* 11.5, $OCH_{A'}H_{B'}$), 4.82 (1H, d, *J* 11.5, $OCH_{A'}H_{B'}$), 4.71 (1H, d, *J* 11.5, OCH_AH_B), 4.67 (1H, d, *J* 11.5, OCH_2Ph), 4.03 (1H, t, *J* 2.3, inositol ring), 3.71-3.59 (5H, m, 2 × inositol ring + 3 × OCH_3), 3.47 (1H, dd, *J* 9.5, 2.8, inositol ring), 3.44 (1H, t, *J* 9.0, inositol ring), 3.36 (1H, dd, *J* 9.7, 2.3, inositol ring), 2.30 (2H, br s, O*H*); δ_C (75 MHz; $CDCl_3$), 138.7 (Ar*C*), 138.6 (Ar*C*), 138.2 (Ar*C*), 128.6 (Ar*C*H), 128.5 (Ar*C*H), 128.4 (Ar*C*H), 128.1 (Ar*C*H), 127.8 (Ar*C*H), 127.79 (Ar*C*H), 127.74 (Ar*C*H), 127.7 (Ar*C*H), 127.5 (Ar*C*H), 82.9 (inositol ring), 81.7 (inositol ring), 80.8 (inositol ring), 77.2 (inositol ring), 75.0 (CH_2), 74.9 (inositol ring), 74.7 (CH_2), 72.6 (CH_2), 72.2 (inositol ring), 61.4 (OCH_3); *m/z* (ES+) [Found: $(M+Na)^+$ 487.2088. $C_{28}H_{32}O_6Na$ requires M^+, 487.2097], *m/z* (ES+) 487 ($[M+Na]^+$, 100%).

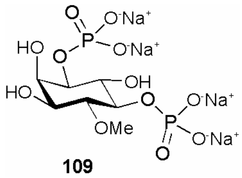
4.1.44. (+)-1D-2,3,6-tris-*O*-Benzyl-4-*O*-methyl-*myo*-inositol 1,5-bis(dibenzylphosphate) 107



107

Bis(benzyloxy)-*N,N*-diisopropylamino phosphine **92** (353 mg, 1.0 mmol, 5.0 equiv) was stirred with 1*H*-tetrazole (0.43 M solution in acetonitrile, 2.4 mL, 1.0 mmol, 5.0 equiv) for 30 min under an atmosphere of nitrogen. (+)-1D-2,3,6-tris-*O*-benzyl-4-*O*-methyl-*myo*-inositol **106** (95 mg, 204 μmol, 1.0 equiv) dissolved in dry dichloromethane (8 mL) was added by cannula and the resulting mixture stirred overnight. The mixture was cooled to - 78 °C and 3-chloroperoxybenzoic acid (176 mg, 1.0 mmol, 5.0 equiv) was added. The resulting mixture was allowed to warm to RT and stirred for 30 min. The 3-chloroperoxybenzoic acid was quenched with a 10% aqueous solution of sodium hydrogen sulfite (5 mL). The layers were separated and the aqueous layer was extracted with dichloromethane (3 × 10 mL). The combined organic layers were washed with a saturated aqueous solution of sodium hydrogen carbonate (5 mL), brine (5 mL), dried (magnesium sulfate), filtered and concentrated under reduced pressure. Purification by silica gel column chromatography, eluting with ethyl acetate/petroleum ether (30/70, then 40/60, then 50/50), yielded *(+)-1D-2,3,6-tris-O-benzyl-4-O-methyl-myo-inositol 1,5-bis(dibenzylphosphate)* **107** (132 mg yield, 66%) as a colourless gum. (Found: C, 68.25, H, 5.8; $C_{56}H_{58}O_{12}P_2$ requires C, 68.3, H, 5.9); R_f 0.37 (ethyl acetate/petroleum ether 50/50); $[\alpha]_D^{25}$ + 7.6 (*c* 0.2 in $CHCl_3$); ν_{max} (*thin film*)/cm^{-1} 3064.4 (w), 3033.3 (w), 2933.0 (m), 1497.5 (m), 1455.5 (s), 1379.8 (m), 1269.5 (s), 1214.3 (m), 1124.9 (m), 1091.8 (s), 1013.9 (s), 881.1 (m), 800.0 (w), 736.5 (s), 696.6 (s); δ_H (300 MHz; $CDCl_3$) 7.31-7.00 (35H, m, Ar*H*), 4.98-4.50 (14H, s, 7 × OCH_2Ph), 4.35-4.24 (2H, m, 2 × inositol ring), 4.18-4.12 (1H, m, inositol ring), 4.00 (1H, t, *J* 9.4, inositol ring), 3.70 (1H, t, *J* 9.4, inositol ring), 3.47 (3H, s, OCH_3), 3.28 (1H, d, *J* 9.7, 2.3, inositol ring); δ_C (75 MHz; $CDCl_3$), 139.0 (Ar*C*), 138.6 (Ar*C*), 138.3 (Ar*C*), 136.6 [d, J_{CP} 7.8, P(O)($OCH_2C_AC_5H_5$], 136.4 [d, J_{CP} 7.8, P(O)($OCH_2C_BC_5H_5$)], 136.1 [d, J_{CP} 1.7, P(O)($OCH_2C_CC_5H_5$)], 136.0 [d, J_{CP} 1.7, P(O)($OCH_2C_DC_5H_5$)], 129.0 (Ar*C*H), 128.95 (Ar*C*H), 128.9 (Ar*C*H), 128.86 (Ar*C*H), 128.8 (Ar*C*H), 128.7 (Ar*C*H), 128.6 (Ar*C*H), 128.3 (Ar*C*H), 128.2 (Ar*C*H), 128.1 (Ar*C*H), 128.02 (Ar*C*H), 128.0 (Ar*C*H), 127.93 (Ar*C*H), 127.9 (Ar*C*H), 127.8 (Ar*C*H), 127.7 (Ar*C*H), 81.4 (d, J_{CP} 1.7, inositol ring), 80.6 (dd, J_{CP} 6.9, 1.6, inositol ring), 80.3 (inositol ring), 78.7 (dd, J_{CP} 7.7, 4.5, inositol ring), 78.4 (d, J_{CP} 5.5, inositol ring), 76.4 (inositol ring), 75.5 (CH_2), 75.1 (CH_2), 73.3 (CH_2), 69.9 [d, J_{CP} 5.6, P(O)OC_AH_2Ph], 69.7 [d, J_{CP} 5.4, P(O)OC_BH_2Ph], 69.5 [d, J_{CP} 5.3, P(O)OC_CH_2Ph], 69.4 [d, J_{CP} 5.2, P(O)OC_DH_2Ph], 61.5 (OCH_3); δ_P (121 MHz; $CDCl_3$) 0.15, - 0.56; *m/z* (ES+) [Found: $(M+Na)^+$ 1007.3288. $C_{56}H_{58}O_{12}NaP_2$ requires M^+, 1007.3301]; *m/z* (ES+) 1007 ($[M+Na]^+$, 100%).

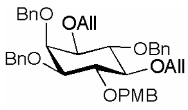
4.1.45. (-)-1D-4-*O*-Methyl-*myo*-inositol 1,5-bisphosphate (sodium salt) 109



109

(+)-1D-2,3,6-tris-*O*-Benzyl-4-*O*-methyl-*myo*-inositol 1,5-bis(dibenzylphosphate) **107** (11 mg, 11 µmol, 1.0 equiv) was dissolved in *tert*-butanol/water (6/1, 3.5 mL), sodium hydrogen carbonate (4 mg, 43 µmol, 4.0 equiv) and palladium black (23 mg, 213 µmol, 20.0 equiv) were added and the flask flushed three times with hydrogen, then stirred for 4h at RT under an atmosphere of hydrogen. The organic layer was removed by filtration, the dark residue washed with water (3 mL) and the collected aqueous layer lyophilized to yield *(-)-1D-4-O-methyl-myo-inositol 1,5-bisphosphate (sodium salt)* **109** as a colourless solid (4 mg yield, 82%). $[\alpha]_D^{22}$ - 4.6 (*c* 0.2 in H_2O); ν_{max} (*KBr disc*)/cm^{-1} 3423.1 (s), 1686.1 (s), 1650.3 (w), 1384.5 (s), 1205.6 (w), 1133.8 (s), 1085.3 (s), 1029.4 (s), 973.2 (s), 917.5 (w), 804.9 (m), 724.8 (m), 595.8 (w), 551.6 (m); δ_H (300 MHz; D_2O) 4.10 (1H, br s, inositol ring), 3.74-3.65 (3H, m, inositol ring), 3.45-3.39 (4H, m, 1 × inositol ring and CH_3), 3.23 (1H, dd, *J* 10.2, 8.5, inositol ring); δ_C (75 MHz; D_2O), 81.3 (d, J_{CP} 5.5, inositol ring), 78.0 (dd, J_{CP} 6.1, 1.1, inositol ring), 74.6 (d, J_{CP} 5.5, inositol ring), 72.2 (d, J_{CP} 6.1, inositol ring), 70.9 (d, J_{CP} 1.1, inositol ring), 69.6 (d, J_{CP} 1.7, inositol ring), 60.2 (OCH_3); δ_P (121 MHz; D_2O) 3.56, 2.99; *m/z* (ES-) [Found: $(M)^-$ 374.9855. $C_7H_{14}O_{12}NaP_2$ requires M^-, 374.9858]; *m/z* 352 ($[C_7H_{15}O_{12}P_2]^-$ 100%), 375 [$C_7H_{14}NaO_{12}P_2$] (70), 273 [$C_7H_{14}O_9P$] (10).

4.1.46. (+)-1D-1,5-bis-*O*-Allyl-2,3,6-tris-*O*-benzyl-4-*O*-(4-methoxybenzyl)-*myo*-inositol 101

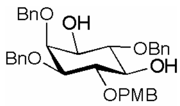


101

(+)-1D-1,5-bis-*O*-Allyl-2,3,6-tris-*O*-benzyl-*myo*-inositol **100** (2.3 g, 4.3 mmol, 1.0 equiv) was dissolved in dry *N,N*-dimethyl formamide (80 mL) under an atmosphere of nitrogen, the mixture was cooled to 0 °C and sodium hydride (191 mg, 60% dispersion in mineral oil, 4.8 mmol, 1.1 equiv) was added. The resulting mixture was allowed to warm to RT and stirred for 1h, then re-cooled to 0 °C and tetra-*n*-butylammonium iodide (80 mg, 216 μmol, 0.05 equiv) and 4-methoxybenzyl chloride (747 mg, 647 μL, 4.8 mmol, 1.1 equiv) were added. The mixture was allowed to warm to RT and stirred overnight. The sodium hydride was quenched with water (3 mL), the solvent removed under reduced pressure and the residue reconstituted in ethyl acetate (20 mL) and water (20 mL). The layers were separated and the aqueous layer extracted with ethyl acetate (3 × 10 mL). The combined organic layers were washed with brine (20 mL), dried (magnesium sulfate), filtered and concentrated under reduced pressure to give a pale yellow oil. Purification by silica gel column chromatography, eluting with ethyl acetate/petroleum ether (10/90) yielded *(+)-1D-1,5-bis-O-allyl-2,3,6-tris-O-benzyl-4-O-(4-methoxybenzyl)-myo-inositol* **101** (2.7 g yield, 95%) as a colourless solid (Found: C, 75.7, H, 7.4; $C_{41}H_{46}O_7$ requires C, 75.7, H, 7.1); R_f 0.6 (ethyl acetate/petroleum ether 30/70); $[\alpha]_D^{25}$ + 6.4 (*c* 0.6 in $CHCl_3$); mp 58-59 °C (*from ethyl acetate/petroleum ether*); ν_{max} (*KBr disc*)/cm^{-1} 3058.8 (w), 3031.8 (w), 2921.3 (m), 1725.6 (w), 1613.9 (m), 1514.1 (s), 1454.3 (m), 1359.1 (m), 1302.1 (w), 1250.3 (s), 1172.6 (w), 1074.4 (s), 1035.6 (s), 917.3 (m), 821.8 (m), 744.7 (s), 697.4 (s), 605.7 (w); δ_H (300 MHz; $CDCl_3$) 7.37-7.16 (17H, m, 15 × Ar*H* and 2 × $OCH_2C_6H_4OCH_3$), 6.76 (2H, d, *J* 8.7, $OCH_2C_6H_4OCH_3$), 5.99-5.77 (2H, m, CH_X=CH_YH_Z + $CH_{X'}$=$CH_{Y'}H_{Z'}$), 5.26-5.19 (2H, m, 1 × CH_X=CH_YH_Z and 1 × $CH_{X'}$=$CH_{Y'}H_{Z'}$), 5.12-5.07 (2H, m, 1 × CH_X=CH_YH_Z and 1 × $CH_{X'}$=$CH_{Y'}H_{Z'}$), 4.81-4-77 (3H, m, 2 × OCH_AH_B and 1 × $OCH_{A'}H_{B'}$), 4.73 (1H, d, $J_{A''B''}$ 10.2, $OCH_{A''}H_{B''}$), 4.71 (1H, d, $J_{A'B'}$ 10.5, $OCH_{A'}H_{B'}$), 4.67 (1H, d, $J_{A''B''}$ 10.2, $OCH_{A''}H_{B''}$), 4.61 (1H, d, $J_{A'''B'''}$ 11.8, $OCH_{A'''}H_{B'''}$), 4.53 (1H, d, $J_{A'''B'''}$ 11.8, $OCH_{A'''}H_{B'''}$), 4.28 (2H, dt, *J* 5.6, 1.5, 2 × $CH_VH_WCH_X$=CH_YH_Z), 4.03-3.99 (2H, m, $CH_{V'}H_{W'}CH_{X'}$=$CH_{Y'}H_{Z'}$) 3.96-3.86 (3H, m, inositol ring), 3.72 (3H, s, OCH_3), 3.24-3.18 (2H, m, 2 × inositol ring), 3.12 (1H, dd, *J* 10.0, 2.3, inositol ring); δ_C (75

MHz; $CDCl_3$) 159.6 (Ar*C*OCH_3), 139.5 (Ar*C*), 139.4 (Ar*C*), 139.0 (Ar*C*), 135.9 (CH_X=CH_YH_Z), 135.4 ($CH_{X'}$=$CH_{Y'}H_{Z'}$), 131.5 (Ar*C*H), 130.3 (Ar*C*H), 128.83 (Ar*C*H), 128.8 (Ar*C*H), 128.7 (Ar*C*H), 128.6 (Ar*C*H), 128.3 (Ar*C*H), 128.03 (Ar*C*H), 128.0 (Ar*C*H), 127.8 (Ar*C*H), 117.1 (CH_X=CH_YH_Z), 117.0 ($CH_{X'}$=$CH_{Y'}H_{Z'}$), 114.2 (Ar*C*H), 83.8 (inositol ring), 82.1 (inositol ring), 81.8 (inositol ring), 81.3 (inositol ring), 81.0 (inositol ring), 76.4 (CH_2), 76.1 (CH_2), 75.1 (CH_2), 74.8 (inositol ring), 74.5 (CH_2), 73.3 (CH_2), 72.1 (CH_2), 55.7 (OCH_3); *m/z* (ES+) [Found: $(M+Na)^+$ 673.3165. $C_{41}H_{46}O_7$Na requires M^+, 673.3141], *m/z* (ES+) 673 ($[M+Na]^+$, 100%).

4.1.47. (-)-1D-2,3,6-tris-*O*-Benzyl-4-*O*-(4-methoxybenzyl)-*myo*-inositol 120

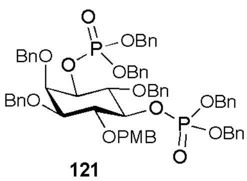


120

Wilkinson's catalyst (22 mg, 24 μmol, 0.4 equiv) was dissolved in dry tetrahydrofuran (0.5 mL) under an atmosphere of nitrogen, *n*-butyl lithium (1.6 M solution in hexanes, 23 μL, 36 μmol, 1.7 equiv) was added and the resulting mixture stirred for 10 min at RT. The mixture was then cannulated onto a solution of (+)-1D-1,5-bis-*O*-allyl-2,3,6-tris-*O*-benzyl-4-*O*-(4-methoxybenzyl)-*myo*-inositol **101** (40 mg, 61 μmol, 1.0 equiv) in dry tetrahydrofuran (0.5 mL) under an atmosphere of nitrogen, and the resulting mixture heated under reflux for 6 h. The mixture was cooled to RT, and the solvent removed under reduced pressure to give a dark red residue. ^{1}H NMR analysis indicated that the allyl groups had completely isomerised. The residue was suspended in ethanol and the resulting mixture filtered through Celite® (to remove most of the Wilkinson's catalyst) and the solvent removed under reduced pressure. The resulting residue was dissolved in a mixture methanol/dichloromethane (2/3, 1 mL) under an atmosphere of nitrogen, acetyl chloride (3 mg, 3 μL, 37 μmol, 0.6 equiv) was added and the resulting mixture stirred for 3 h. The generated hydrochloric acid was quenched with triethylamine (50 μL), the solvent removed under reduced pressure, the residue adsorbed onto silica gel and purified by column chromatography, eluting with ethyl acetate/petroleum ether (20/80) to yield *(-)-1D-2,3,6-tris-O-benzyl-4-O-(4-methoxybenzyl)-myo-inositol* **120** (31 mg yield, 89%) as a colourless gum. (Found: C, 73.25, H, 6.75; $C_{35}H_{38}O_7$ requires C, 73.66, H, 6.7); R_f 0.54 (ethyl acetate/petroleum ether 50/50); $[\alpha]_D^{25}$ - 6.7 (*c* 0.56 in $CHCl_3$); ν_{max} (*thin film*)/cm^{-1} 3555.0 (m), 3449.2 (m), 3055.3 (m), 2924.8 (s), 1612.8 (m), 1586.1 (w), 1514.1 (s), 1455.0 (s), 1364.3 (m), 1265.7 (s), 1250.2

(m), 1113.2 (m), 1069.2 (s), 1028.0 (m), 933.9 (w), 822.7 (w), 737.3 (s), 701.9 (s); δ_H (300 MHz; $CDCl_3$) 7.29-7.18 (17H, m, 15 × Ar*H* and 2 × $OCH_2C_6H_4OCH_3$), 6.79 (2H, d, *J* 8.7, $OCH_2C_6H_4OCH_3$), 4.92 (1H, d, J_{AB} 11.5, OCH_AH_B), 4.84 (1H, d, $J_{A'B'}$ 11.0, $OCH_{A'}H_{B'}$), 4.83 (1H, d, $J_{A''B''}$ 11.4, $OCH_{A''}H_{B''}$), 4.73-4.65 (2H, m, 1 × OCH_AH_B and 1 × $OCH_{A''}H_{B''}$), 4.62 (2H, s, $OCH_{A'''}H_{B'''}$), 4.61 (1H, d, $J_{A'B'}$ 11.0, $OCH_{A'}H_{B'}$), 3.98 (1H, t, *J* 2.6, inositol ring), 3.81 (1H, *J* 9.2, inositol ring), 3.73 (3H, s, OCH_3), 3.61 (1H, d, *J* 9.5, inositol ring), 3.46-3-34 (3H, m, 3 × inositol ring), 2.39 (1H, d, *J* 2.0, OH_X), 2.22 (1H, d, *J* 6.4, OH_Y); δ_C (75 MHz; $CDCl_3$) 159.7 (Ar*C*OCH_3), 139.2 (Ar*C*), 139.1 (Ar*C*), 138.6 (Ar*C*), 131.2 (Ar*C*), 130.2 (Ar*C*H), 128.95 (Ar*C*H), 128.9 (Ar*C*H), 128.8 (Ar*C*H), 128.5 (Ar*C*H), 128.22 (Ar*C*H), 128.2 (Ar*C*H), 128.1 (Ar*C*H), 128.0 (Ar*C*H), 114.4 (Ar*C*H), 82.1 (inositol ring), 81.4 (inositol ring), 81.3 (inositol ring), 77.9 (CH_2), 77.5 (inositol ring), 77.1 (CH_2), 75.4 (inositol ring), 73.1 (CH_2), 72.6 (inositol ring), 55.7 (OCH_3); *m/z* (ES+) [Found: $(M+Na)^+$ 593.2504. $C_{35}H_{38}O_7Na$ requires M^+, 593.2515], *m/z* (ES+) 593 ($[M+Na]^+$, 100%).

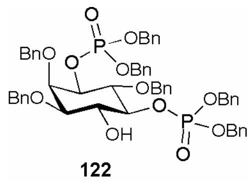
4.1.48. (+)-1D-2,3,6-tris-*O*-Benzyl-4-*O*-(4-methoxybenzyl)-*myo*-inositol 1,5-bis(dibenzylphosphate) 121



121

Bis(benzyloxy)-*N,N*-diisopropylamino phosphine **92** (3.0 g, 8.8 mmol, 5.0 equiv) was stirred with 1*H*-tetrazole (613 mg, 8.8 mmol, 5.0 equiv) for 10 min under an atmosphere of nitrogen at RT. (-)-1D-2,3,6-tris-*O*-Benzyl-4-*O*-(4-methoxybenzyl)-*myo*-inositol **120** (1.0 g, 1.8 mmol, 1.0 equiv) dissolved in dry dichloromethane (20 mL) was added by cannula and the resulting mixture stirred overnight. The mixture was cooled to - 78 °C and 3-chloroperoxybenzoic acid (1.5 g, 8.8 mmol, 5.0 equiv) was added. The resulting mixture was allowed to warm to RT and stirred for 30 min. The 3-chloroperoxybenzoic acid was quenched with a 10% aqueous solution of sodium hydrogen sulfite (20 mL). The layers were separated and the aqueous layer was extracted with dichloromethane (3 × 10 mL). The combined organic layers were washed with a saturated aqueous solution of sodium hydrogen carbonate (10 mL), brine (10 mL), dried (magnesium sulfate), filtered and concentrated under reduced pressure. Purification by silica gel column chromatography, eluting with ethyl acetate/petroleum ether (30/70, then 50/50), yielded *(+)-1D-2,3,6-tris-O-benzyl-4-O-(4-methoxybenzyl)-myo-inositol 1,5-bis(dibenzylphosphate)* **121** (1.4 g yield, 75%) as a colourless gum. (Found: C, 69.7, H, 5.8; $C_{63}H_{64}O_{13}P_2$ requires C, 69.35, H, 5.9); R_f 0.39 (ethyl acetate/petroleum 50/50), $[\alpha]_D^{25}$ + 7.5 (*c* 0.3 in $CHCl_3$); ν_{max} (*thin film*)/cm^{-1} 3064.2 (m), 3033.0 (m), 2934.8 (m), 1612.8 (m), 1586.4 (w), 1514.3 (s), 1497.6 (m), 1455.6 (s), 1364.6 (m), 1250.1 (s), 1214.6 (m), 1073.8 (w), 1012.2 (s), 880.7 (m), 823.0 (w), 737.3 (s), 696.6 (s); δ_H (300 MHz; $CDCl_3$) 7.30-6.93 (37H, m, 35 × Ar*H* and 2 × $OCH_2C_6H_4OCH_3$), 6.67 (2H, d, *J* 8.7, $OCH_2C_6H_4OCH_3$), 4.87-4.62 (14H, m, 7 × CH_2), 4.48 (1H, d, J_{AB} 11.5, OCH_AH_B), 4.43 (1H, d, J_{AB} 11.5, OCH_AH_B), 4.34 (1H, dd, J_{HP} 18.2, *J* 9.0, inositol ring), 4.26 (1H, t, *J* 2.3, inositol ring), 4.18-4.12 (1H, m, inositol ring), 4.03-3.93 (2H, m, 2 × inositol ring), 3.67 (3H, s, OCH_3), 3.31 (1H, dd, *J* 9.7, 2.3, inositol ring); δ_C (75 MHz; $CDCl_3$) 159.3 ($ArCOCH_3$), 139.0 (Ar*C*), 138.6 (Ar*C*), 138.2 (Ar*C*), 136.5 [d, J_{CP} 4.8, $P(O)(OCH_2C_AC_5H_5)$], 136.4 [d, J_{CP} 4.8, $P(O)(OCH_2C_BC_5H_5$], 136.1 [d, J_{CP} 2.3, $P(O)(OCH_2C_CC_5H_5$], 136.0 [d, J_{CP} 1.8, $P(O)(OCH_2C_DC_5H_5$], 131.0 (Ar*C*), 129.8 (Ar*C*H), 129.0 (Ar*C*H), 128.96 (Ar*C*H), 128.8 (Ar*C*H), 128.75 (Ar*C*H), 128.7 (Ar*C*H), 128.5 (Ar*C*H), 128.2 (Ar*C*H), 128.12 (Ar*C*H), 128.1 (Ar*C*H), 128.0 (Ar*C*H), 127.9 (Ar*C*H), 127.7 (Ar*C*H), 113.9 (Ar*C*H), 80.9 (dd, J_{CP} 7.0, 1.5, inositol ring), 80.4 (inositol ring), 79.1 (d, J_{CP} 2.8, inositol ring), 78.8 (dd, J_{CP} 7.5, 3.3, inositol ring), 78.5 (d, J_{CP} 5.9, inositol ring), 76.3 (inositol ring), 75.5 (CH_2), 75.1 (CH_2), 75.0 (CH_2), 73.1 (CH_2), 69.9 (d, J_{CP} 5.7, $P(O)OC_AH_2Ph$), 69.7 (d, J_{CP} 5.5, $P(O)OC_BH_2Ph$), 69.6 (d, J_{CP} 4.9, 2 × $P(O)OCH_2Ph$), 55.6 (OCH_3); δ_P (121 MHz; $CDCl_3$) - 0.22, - 0.61; *m/z* (ES+) [Found: $(M+Na)^+$ 1113.3711. $C_{63}H_{64}O_{13}NaP_2$ requires M^+, 1113.3720], *m/z* (ES+) 1113 ($[M+Na]^+$, 100%).

4.1.49. (+)-1D-2,3,6-tris-*O*-Benzyl-*myo*-inositol 1,5-bis(dibenzylphosphate) 122

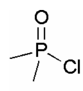


122

(+)-1D-2,3,6-tris-*O*-benzyl-4-*O*-(4-methoxybenzyl)-*myo*-inositol 1,5 bis(dibenzyl phosphate) **121** (327 mg, 0.3 mmol, 1.0 equiv) was dissolved in acetonitrile/water (4/1, 5 mL) and ceric ammonium nitrate (987 mg, 1.8 mmol, 6.0 equiv) was added at RT. The resulting orange solution was stirred for 2h. The solvent was removed under reduced pressure, the residue reconstituted in ethyl acetate (5 mL) and water (5 mL), the layers separated and the aqueous layer extracted with ethyl acetate

(3 × 5 mL). The combined organic layers were washed with a saturated aqueous solution of sodium hydrogen carbonate (10 mL), dried (magnesium sulfate), filtered and concentrated under reduced pressure to give an orange residue. Silica gel column chromatography eluting with ethyl acetate/petroleum ether (50/50), followed by crystallisation from diethyl ether/dichloromethane/petroleum ether, yielded *(+)-1D-2,3,6-tris-O-benzyl-myo-inositol 1,5-bis(dibenzylphosphate)* **122** (232 mg yield, 73%) as a colourless solid (Found: C, 68.1, H, 5.65; $C_{55}H_{56}O_{12}P_2$ requires C, 68.0, H, 5.8); R_f 0.24 (ethyl acetate/petroleum 50/50), $[\alpha]_D^{25}$ + 1.6 (*c* 0.6 in $CHCl_3$); mp 125-126 °C; ν_{max} (*thin film*)/cm^{-1} 3397.3 (s), 3064.6 (m), 3030.5 (m), 2938.6 (m), 2890.7 (m), 1497.5 (m), 1455.5 (s), 1367.4 (m), 1269.4 (s), 1240.1 (s), 1216.2 (m), 1162.8 (m), 1129.1 (m), 1068.7 (s), 1013.4 (s), 888.8 (m), 737.0 (s), 695.3 (s), 589.3 (w), 554.7 (w), 502.2 (m); δ_H (300 MHz; $CDCl_3$) 7.37-7.12 (35H, m, Ar*H*), 5.05-4.70 (12H, m, 6 × CH_2), 4.62 (1H, d, J_{AB} 11.8, OC$H_A$$H_B$), 4.57 (1H, d, J_{AB} 11.8, OC$H_A$$H_B$), 4.30 (1H, t, *J* 2.3, inositol ring), 4.25-4.17 (3H, m, inositol ring), 4.08-3.98 (1H, m, inositol ring), 3.87 (1H, br s, O*H*), 3.25 (1H, dd, *J* 9.2, 2.0, inositol ring); δ_C (75 MHz; $CDCl_3$) 138.6 (Ar*C*), 138.0 (Ar*C*), 137.8 (Ar*C*), 135.8-135.6 [m, 4 × P(O)($OCH_2CC_5H_5$)] 128.6 (Ar*C*H), 128.52 (Ar*C*H), 128.5 (Ar*C*H), 128.46 (Ar*C*H), 128.4 (Ar*C*H), 128.25 (Ar*C*H), 128.2 (Ar*C*H), 127.8 (Ar*C*H), 127.7 (Ar*C*H), 127.6 (Ar*C*H), 127.4 (Ar*C*H), 82.4 (dd, J_{CP} 6.1, 1.9, inositol ring), 79.1 (inositol ring), 78.3-78.0 (m, 2 × inositol ring), 76.0, (inositol ring), 75.2 (CH_2), 75.1 (CH_2), 72.9 (CH_2), 72.0 (inositol ring), 69.6 [d, J_{CP} 5.2, 2 × P(O)OC_AH_2Ph)], 69.5 [d, J_{CP} 5.8, P(O)OC_AH_2Ph], 69.3 [d, J_{CP} 5.5, P(O)OC_BH_2Ph]; δ_P (121 MHz; $CDCl_3$) 1.34, - 0.49; *m/z* (ES+) [Found: $(M+Na)^+$ 993.3147. $C_{55}H_{56}O_{12}NaP_2$ requires M^+, 993.3145], *m/z* (ES+) 993 ($[M+Na]^+$, 100%).

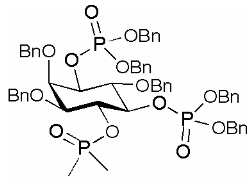
4.1.50. Dimethylphosphinic chloride 134



134

Tetramethyl diphosphine disulfide **133** (400 mg, 2.3 mmol, 1.0 equiv) was suspended in dry toluene (3 mL), under an atmosphere of nitrogen. The mixture was cooled to 0 °C and thionyl chloride (1.2 g, 0.8 mL, 10.3 mmol, 4.8 equiv) was added dropwise. The resulting mixture was allowed to warm to RT and stirred for 30 min, then heated under reflux for 1 h. ^{31}P NMR analysis indicated the complete consumption of the starting material, when the reaction mixture was cooled to RT and the solvent removed under reduced pressure, keeping the product under an atmosphere of nitrogen. The resulting yellow residue was purified using Kugelrohr distillation. The desired product **134** distilled at 140-150 °C (18 mbar) and was trapped by keeping the receiving flask at - 78 °C. The title compound, obtained as a slightly yellow deliquescent solid, was stored in the freezer under an atmosphere of nitrogen (143 mg yield, 59%); δ_H (300 MHz; $CDCl_3$) 1.97 (6H, d, J_{HP} 13.7); δ_P (121 MHz; $CDCl_3$) 61.4. These data are in good agreement with the literature values.[140]

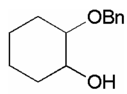
4.1.51. (+)-1D-2,3,6-tris-*O*-Benzyl-4-*O*-dimethylphosphinyl-*myo*-inositol 1,5-bis(dibenzylphosphate) 119



119

(+)-1D-2,3,6-tris-*O*-Benzyl-*myo*-inositol 1,5-bis(dibenzylphosphate) **122** (40 mg, 41 μmol, 1.0 equiv) was dissolved in dry *N,N*-dimethyl formamide (1 mL) under an atmosphere of nitrogen. 2,6-Lutidine (22 mg, 24 μL, 206 μmol, 5.0 equiv) was added and the resulting mixture was cooled to - 42 °C. Dimethylphosphinic chloride (19 mg, 165 μmol, 4.0 equiv) dissolved in dry *N,N*-dimethyl formamide (0.5 mL) was added by cannula. The resulting mixture was allowed to warm to RT and stirred for 22 h. The solvent was removed under reduced pressure, the residue adsorbed onto silica gel and purified by silica gel column chromatography, eluting with methanol/ethyl acetate (1/99) (three times) to give *(+)-1D-2,3,6-tris-O-benzyl-4-O-dimethylphosphinyl-myo-inositol 1,5-bis(dibenzylphosphate)* **199** (33 mg yield, 76%) as a colourless solid. A very pure sample was obtained by crystallisation from ethyl acetate/petroleum ether (Found: C, 65.05, H, 5.6; $C_{57}H_{61}O_{13}P_3$ requires C, 65.4, H, 5.9); R_f 0.52 (methanol/ethyl acetate 5/95)**;** $[\alpha]_D^{22}$ + 6.2 (*c* 0.85 in $CHCl_3$); mp 105-106 °C (*from ethyl acetate/petroleum ether*); ν_{max} (*KBr disc*)/cm^{-1} 3058.8 (m), 3033.4 (m), 2924.4 (m), 2879.5 (m), 1498.2 (m), 1455.4 (m), 1381.0 (w), 1262.8 (s), 1215.8 (s), 1124.5 (w), 1017.2 (s), 939.9 (w), 872.2 (m), 736.4 (s), 695.9 (s), 594.5 (w), 507.5 (w); δ_H (300 MHz; $CDCl_3$) 7.32-6.95 (35H, m, Ar*H*), 5.05 (1H, dd, J_{AB} 11.8, J_{HP} 6.1, OCH_AH_B), 4.92-4.59 (12H, m, 11 × OCH_2 and 1 × inositol ring), 4.44 (1H, d, $J_{A'B'}$ 11.3, $OCH_{A'}H_{B'}$), 4.38-4.32 (2H, m, 1 × $OCH_{A'}H_{B'}$ and 1 × inositol ring), 4.28 (1H, t, *J* 2.6, inositol ring), 4.21-4.15 (1H, m, inositol ring), 4.01 (1H, t, *J* 9.5, inositol ring), 3.29 (1H, dd, *J* 10.0, 2.0, inositol ring), 1.40 [3H, d, J_{HP} 14.0, $P(O)CH_3CH_3$], 1.26 [3H, d, J_{HP} 14.0, $P(O)CH_3CH_3$]; δ_C (75 MHz; $CDCl_3$) 138.24 (Ar*C*), 138.2 (Ar*C*), 136.9 (ArC), 136.0 [d, J_{CP} 7.1, P(O)(OCH$_2C_A$C$_5$H$_5$)],135.9 [d, J_{CP} 5.8, P(O)(OCH$_2C_B$C$_5$H$_5$)] 135.6-135.5 [m, 2 × P(O)(OCH$_2C$C$_5$H$_5$)], 128.6 (ArCH), 128.52 (ArCH), 128.5 (ArCH), 128.4 (ArCH), 128.3 (ArCH), 128.2 (ArCH), 128.1 (ArCH), 128.06 (ArCH), 127.9 (ArCH), 127.8 (ArCH), 127.7 (ArCH), 127.6 (ArCH), 127.4 (ArCH), 127.2 (ArCH), 79.4-79.3 (m, inositol ring), 78.1-77.9 (m, 3 × inositol ring), 75.3 (CH$_2$), 74.9 (CH$_2$), 74.8 (inositol ring), 73.3-73.2 (m, inositol ring), 72.2 (CH$_2$), 69.6 [d, J_{CP} 6.2, P(O)OC_AH$_2$Ph)], 69.5 [d, J_{CP} 5.5, P(O)OC_BH$_2$Ph)], 69.3 [d, J_{CP} 4.9, 2 × P(O)OCH$_2$Ph)], 17.6 (d, J_{CP} 69.7, P(O)CH$_3$CH$_3$], 16.3 (d, J_{CP} 73.5, P(O)CH$_3C$H$_3$]; δ_P (121 MHz; CDCl$_3$) 57.3, - 0.17, - 0.54; *m/z* (ES+) [Found: (M+Na)$^+$ 1069.3218. $C_{57}H_{61}O_{13}NaP_3$ requires M^+, 1069.3223]; *m/z* (ES+) 1069 ([M+Na]$^+$, 100%).

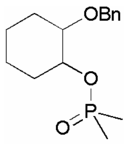
4.1.52. (±)-1-*O*-Benzyl-1,2-*trans*-dihydroxycyclohexane 130



130

(±)-1,2-*trans*-Dihydroxycyclohexane **55** (5.0 g, 43.0 mmol, 1.0 equiv) was dissolved in dry tetrahydrofuran (300 mL) under an atmosphere of nitrogen. The mixture was cooled to 0 ºC and sodium hydride (60% w/w, 1.9 g, 47.3 mmol, 1.1 equiv) was added portionwise over 10 min. The resulting mixture was allowed to warm to RT and stirred for 1.5 h. The mixture was re-cooled to 0 ºC and benzyl bromide (8.1 g, 5.6 mL, 47.3 mmol, 1.1 equiv) was added dropwise. The mixture was warmed to RT and stirred for 1 h. Dry *N,N*-dimethyl formamide (53 mL) was added and the mixture stirred overnight. The sodium hydride was quenched with water (20 mL), the solvent removed under reduced pressure and the residue reconstituted in ethyl acetate (50 mL) and water (50 mL). The layers were separated and the aqueous layer extracted with ethyl acetate (3 × 50 mL). The combined organic layers were washed with brine (20 mL), dried (magnesium sulfate), filtered and concentrated under reduced pressure. Purification by silica gel column chromatography, eluting with ethyl acetate/petroleum ether (30/70) yielded the (±)-1-*O*-benzyl-1,2-*trans*-dihydroxycyclohexane **130** as colourless oil (3.5 g yield, 40%); R_f 0.55 (ethyl acetate/petroleum ether 50/50); δ_H (300 MHz; CDCl$_3$) 7.38-7.28 (5H, m, ArH), 5.07 (1H, d, J 11.5, OCH_AH$_B$Ph), 4.48 (1H, d, J 11.5, OCH$_A H_B$Ph), 3.53-3.45 (1H, m, CHOBn), 3.23-3.15 (1H, m, CHOH), 2.18-1.98 (2H, m, CH_2CHOBn), 1.78-1.68 (2H, m, CH_2CHOH), 1.32-1.18 (4H, m, CH_2CH_2). These data are in good agreement with the literature values.[141]

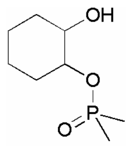
4.1.53. (±)-1-*O*-Benzyl-2-*O*-dimethylphosphinyl-1,2-*trans*-dihydroxycyclohexane 131



131

Diisopropylamino dimethylphosphine **113** (1.2 g, 7.3 mmol, 2.5 equiv) and 1*H*-tetrazole (0.43 M solution in acetonitrile, 16.9 mL, 7.3 mmol, 2.5 equiv) were dissolved in dry dichloromethane (10 mL), the resulting mixture was cooled to - 78 °C and (±)-1-*O*-benzyl-1,2-*trans*-dihydroxycyclohexane (600 mg, 2.9 mmol, 1.0 equiv) dissolved in dry dichloromethane (5 mL) was added by cannula. The resulting mixture was allowed to warm to RT and stirred overnight. The mixture was re-cooled to - 78 °C and 3-chloroperoxybenzoic acid (1.3 g, 7.3 mmol, 2.5 equiv) was added, the resulting mixture warmed to RT and stirred for 30 min. The 3-chloroperoxybenzoic acid was quenched with a 10% aqueous solution of sodium hydrogen sulfite (10 mL), the layers separated and the aqueous layer extracted with dichloromethane (3 × 10 mL). The combined organic layers washed with a saturated aqueous solution of sodium hydrogen carbonate (10 mL), brine (10 mL), dried (magnesium sulfate), filtered and concentrated under reduced pressure. Purification by silica gel column chromatography, eluting with methanol/ethyl acetate (2/98) furnished *(±)-1-O-benzyl-2-O-dimethylphosphinyl-1,2-trans-dihydroxycyclohexane* **131** (771 mg yield, 89%) as a colourless oil; R_f 0.3 (methanol/ethyl acetate 5/95); δ_H (300 MHz; $CDCl_3$) 7.35-7.28 (5H, m, Ar*H*), 4.65 (1H, d, *J* 11.8, OC$H_A$$H_B$Ph), 4.53 (1H, d, *J* 11.8, OC$H_A$$H_B$Ph), 4.28-4.17 (1H, m, C*H*OBn), 3.38-3.30 (1H, m, C*H*OH), 2.20-2.04 (2H, m, CH_2CHOBn), 1.73-1.64 (2H, m, CH_2CHOH), 1.55-1.20 [10H, m, 6 × P(O)CH_3CH_3 and 4 × CH_2CH_2]; δ_P (121 MHz; $CDCl_3$) 54.4; *m/z* (ES+) 305 ([M+Na]$^+$, 100%).

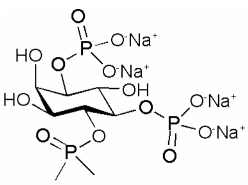
4.1.54. (±)-1-*O*-Dimethylphosphinyl-1,2-*trans*-dihydroxycyclohexane 70



70

(±)-1-*O*-Benzyl-2-*O*-dimethylphosphinyl-1,2-*trans*-dihydroxycyclohexane **131** (50 mg, 177 μmol, 1.0 equiv) was dissolved was dissolved in *tert*-butanol/water (6/1, 2 mL), sodium hydrogen carbonate (60 mg, 708 μmol, 4.0 equiv) and palladium black (377 mg, 3.5 mmol, 20.0 equiv) were added and the flask flushed three times with hydrogen, then stirred for 2 h at RT under an atmosphere of hydrogen. The catalyst was removed by filtration and the collected organic layer concentrated under reduced pressure to furnish *(±)-1-O-dimethylphosphinyl-1,2-trans-dihydroxycyclohexane* **70** (31 mg yield, 92%) as a colourless oil; R_f 0.12 (methanol/ethyl acetate 5/95); δ_H (300 MHz; $CDCl_3$) 3.95-3.84 (1H, m, C*H*OBn), 3.46-3.38 (1H, m, C*H*OH), 2.85-2.05 (2H, m, CH_2CHOBn), 1.68-1.62 (2H, m, CH_2CHOH), 1.49 [6H, d, J_{HP} 13.6, P(O)CH_3CH_3], 1.42-1.12 (4H, m, CH_2CH_2); δ_P (121 MHz; $CDCl_3$) 56.1; *m/z* (ES+) 215 ($[M+Na]^+$, 100%).

4.1.55. (+)-1D-4-*O*-Dimethylphosphinyl-*myo*-inositol 1,5-bisphosphate (sodium salt) 32

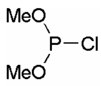


32

(+)-1D-2,3,6-tris-*O*-Benzyl-4-*O*-dimethylphosphinyl-*myo*-inositol 1,5-bis(dibenzyl phosphate) **119** (71 mg, 68 μmol, 1.0 equiv) was dissolved in *tert*-butanol/water (6/1, 12 mL), sodium hydrogen carbonate (23 mg, 271 μmol, 4.0 equiv) and palladium black (145 mg, 1.4 mmol, 20.0 equiv) were added and the flask flushed three times with hydrogen, then stirred for 7 h at RT under an atmosphere of hydrogen. The organic layer was removed by filtration, the dark residue washed with water (4 × 3 mL) and the collected aqueous layer lyophilized to yield *(+)-1D-4-O-dimethylphosphinyl-myo-inositol 1,5-bisphosphate (sodium salt)* **32** as a colourless solid (32 mg yield, 93%); $[\alpha]_D^{22}$ + 0.81 (*c* 0.6 in H_2O); ν_{max} (*KBr disc*)/cm^{-1} 3423.3 (s), 2198.8 (m), 1655.3 (w), 1309.1 (w), 1188.8 (s), 1116.1 (s), 1053.1 (s), 950.1 (s), 920.3 (m), 883.9 (m), 811.2 (w), 721.7 (w), 513.8 (m); δ_H (300 MHz; D_2O) 4.29-4.15

(2H, m, inositol ring), 3.97-3.77 (3H, m, inositol ring), 3.63 (1H, dd, *J* 9.7, 2.8, inositol ring), 1.58 [3H, d, J_{HP} 11.0, P(O)C*H*$_3$CH$_3$], 1.53 [3H, d, J_{HP} 11.0, P(O)CH$_3$C*H*$_3$]; δ_C (75 MHz; D_2O) 76.8 (dd, J_{CP} 7.7, 6.3, inositol ring), 76.1-75.9 (m, inositol ring), 74.4 (d, J_{CP} 5.5, inositol ring), 72.2 (d, J_{CP} 7.2, inositol ring), 70.8 (inositol ring), 69.2 (inositol ring), 15.3 (d, J_{CP} 95.0, P(O)*C*H$_3$CH$_3$), 15.1 (d, J_{CP} 95.0, P(O)CH$_3$*C*H$_3$); δ_P (121 MHz; D_2O) 67.1, 1.63, 1.41; *m/z* (MALDI - matrix 3AQ, internal calculation on glucose sulfate and ATP) [Found: $(C_8H_{18}O_{13}P_3)^-$ 414.9939. $C_8H_{18}O_{13}P_3$ requires M^-, 414.9960]; *m/z* (MALDI - matrix 3AQ, external calculation on glucose sulfate and ATP) 415 [$(C_8H_{18}O_{13}P_3)^-$].

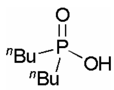
4.1.56. Dimethyl chlorophosphite 144



144

Trimethylphosphite (27.1 mL, 28.4 g, 229.2 mmol, 2.0 equiv) was placed in a flask under an atmosphere of nitrogen and warmed to 60 °C. Phosphorus trichloride (10.0 mL, 15.7 g, 114.6 mmol, 1.0 equiv) was added dropwise over a period of 30 min with stirring. The resulting mixture was stirred for a further 30 min at 60 °C, then cooled to RT. ^{31}P NMR analysis confirmed the presence of the desired compound in the mixture. Purification by distillation under reduced pressure afforded dimethyl chlorophosphite **144** (8.2 g yield, 28%) as a colourless oil; bp 40 °C (101-107 mbar) (Lit.[142] 30 °C, 46.7 mbar); δ_P (121 MHz; d$_6$-acetone) 170.2. These data are in good agreement with the literature values.[142]

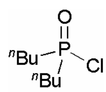
4.1.57. Di-*n*-butylphosphinic acid 136



136

Magnesium turnings (5.3 g, 218.9 mmol, 4.0 equiv) were placed in a three-necked flask under an atmosphere of nitrogen. Iodine (3 pellets) was added and the magnesium turnings shaken for 20 min at RT. Dry diethyl ether (100 mL) was added to the flask and *n*-butyl bromide (23.5 mL, 30.0 g, 218.0 mmol, 4.0 equiv) dissolved in dry diethyl ether (80 mL) was slowly added, cooling down the reaction mixture with an ice-bath when the reaction was too vigorous. The resulting mixture was then heated under reflux for 30 min to complete the formation of the Grignard reagent, then cooled to 0 °C. Thiophosphoryl chloride (5.5 mL, 9.3 g, 54.7 mmol, 1.0 equiv) dissolved in dry diethyl ether (10 mL) was carefully added dropwise with stirring, as the reaction was very vigorous. The resulting mixture was then heated under reflux for 1 h, cooled to 0 °C and poured onto water ice. The aqueous layer was acidified to pH 2 using concentrated hydrochloric acid, the layers were separated and the aqueous layer extracted with diethyl ether (3 × 50 mL). The combined organic layers were dried (magnesium sulfate), filtered and concentrated under reduced pressure to furnish a crude oil (12.0 g). This material was placed in a flask fitted with a condenser, then cooled to 0 °C and nitric acid (30% aqueous solution, 60 mL) slowly added with stirring. The resulting mixture was heated to 70 °C for 1 h, then cooled to RT and diethyl ether (50 mL) added. The layers were separated and the organic layer washed with water (3 × 50 mL), then extracted with a 10% solution of sodium hydroxide (2 × 50 mL). The combined aqueous layers were acidified to pH 2 by careful addition of concentrated sulfuric acid, then extracted with diethyl ether (3 × 50 mL). The combined organic layers were washed with water (3 × 50 mL), dried (magnesium sulfate), filtered and concentrated to furnish a slurry which was kept under reduced pressure at 80 °C for 3 h to give a yellow solid. Crystallisation from warm petroleum ether gave di-*n*-butylphosphinic acid **136** (3.1 g yield, 31% with respect to thiophosphoryl chloride) as a colourless solid; mp 69-70 °C (*from petroleum ether*) [Lit.[143] 70.5-71 °C (*from hexane*)]; δ_H (300 MHz; $CDCl_3$) 11.73 (1H, br s, O*H*), 1.75-1.52 [8H, m, $P(O)(CH_2C\mathit{H}_2C\mathit{H}_2CH_3)_2$], 1.47-1.36 [4H, m, $P(O)(C\mathit{H}_2CH_2CH_2CH_3)_2$], 0.98 [6H, t, *J* 7.3, $P(O)(CH_2CH_2CH_2C\mathit{H}_3)_2$]; δ_P (121 MHz; $CDCl_3$) 62.2. These data are in good agreement with the literature values.[129,143]

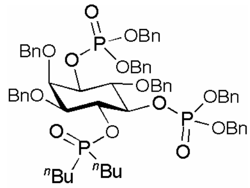
4.1.58. Di-*n*-butylphosphinyl chloride 137



137

Di-*n*-butylphosphinic acid **136** (500 mg, 2.8 mmol, 1.0 equiv) was dissolved in dry toluene (4 mL) under and atmosphere of nitrogen. The resulting mixture was cooled to 0 °C and thionyl chloride (225 µL, 366 mg, 3.1 mmol, 1.1 equiv) was added and the mixture heated under reflux for 30 min. The solvent was removed under reduced pressure and the residue purified by distillation under reduced pressure to give di-*n*-butylphosphinic chloride **137** (461 mg yield, 84%) as a colourless oil; bp 100-105 °C (5 mbar) (Lit.[128] 103-105 °C, 0.7 mbar); δ_P (121 MHz; d_8-toluene) 69.3. These data are in good agreement with the literature values.[128]

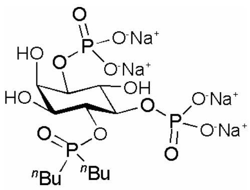
4.1.59. (+)-1D-2,3,6-tris-*O*-Benzyl-4-*O*-di-*n*-butylphosphinyl-*myo*-inositol 1,5-bis(dibenzylphosphate) 138



138

(+)-1D-2,3,6-tris-*O*-Benzyl-*myo*-inositol 1,5-bis(dibenzylphosphate) **122** (100 mg, 103 μmol, 1.0 equiv) was dissolved in dry *N,N*-dimethyl formamide (4 mL) under an atmosphere of nitrogen and the mixture cooled to - 42 °C. 4-Dimethylaminopyridine (catalytic amount) was added, followed by dry triethylamine (72 μL, 52 mg, 515 μmol, 5.0 equiv) and di-*n*-butylphosphinyl chloride (79 μL, 82 mg, 416 μmol, 4.0 equiv). The mixture was allowed to warm to RT and stirred overnight. The di-*n*-butylphosphinic chloride was quenched with water (0.5 mL), the solvent was removed under reduced pressure and the residue reconstituted in ethyl acetate (2 mL) and water (2 mL). The layers were separated and the aqueous layer extracted with ethyl acetate (3 × 5 mL). The combined organic layers were dried (magnesium sulfate), filtered and concentrated under reduced pressure. Purification by silica gel column chromatography, eluting with ethyl acetate/petroleum ether (60/40) gave *(+)-1D-2,3,6-tris-O-benzyl-4-O-di-n-butylphosphinyl-myo-inositol 1,5-bis(dibenzylphosphate)* **138** as a colourless solid which was recrystallised from ethyl acetate and petroleum ether (76 mg yield, 65%); R_f 0.25 (ethyl acetate/petroleum ether 60/40)**;** $[\alpha]_D^{25}$ + 0.5 (*c* 0.27 in $CHCl_3$); mp 104-105 °C (*from ethyl acetate/petroleum ether*); ν_{max} (*KBr disc*)/cm^{-1} 3064.4 (m), 3030.8 (m), 2930.0 (m), 1457.3 (w), 1381.8 (w), 1261.3 (m), 1211.2 (w), 1160.8 (w), 1127.3 (w), 1037.8 (s), 1015.5 (s), 881.1 (w), 867.1 (w), 135.7 (m), 695.9 (s), 593.0 (w); δ_H (300 MHz; $CDCl_3$) 7.28-6.91 (35H, m, Ar*H*), 5.11 (1H, dd, J_{AB} 11.8, J_{HP} 6.1, OCH_AH_B), 4.92-4.57 (12H, m, 11 × OCH_2 and 1 × inositol ring, 4.45 (1H, d, $J_{A'B'}$ 11.5, $OCH_{A'}H_{B'}$), 4.35-4.30 (3H, m, 1 × , $OCH_{A'}H_{B'}$ and 2 × inositol ring), 4.23-4.16 (1H, m, inositol ring), 4.02 (1H, t, *J* 9.5, inositol ring), 3.30 (1H, dd, *J* 9.7, 1.8, inositol ring), 1.73-0.91 [12H, m, $P(O)(C_3H_6CH_3)_A(C_3H_6CH_3)_B$], 0.71 [3H, t, *J* 7.2 $P(O)(C_3H_6CH_3)_A(C_3H_6CH_3)_B$], 0.65 [3H, t, *J* 7.2 $P(O)(C_3H_6CH_3)_A(C_3H_6CH_3)_B$]; δ_C (75 MHz; $CDCl_3$) 138.3 (Ar*C*), 138.2 (Ar*C*), 137.8 (Ar*C*), 136.2 [d, J_{CP} 8.1, $P(O)(OCH_2C_AC_5H_5)$],135.9 [d, J_{CP} 6.6, $P(O)(OCH_2C_BC_5H_5)$] 135.6 [d, J_{CP} 7.5, $P(O)(OCH_2C_CC_5H_5)$], 135.5 [d, J_{CP} 6.9, $P(O)(OCH_2C_DC_5H_5)$], 128.6 (Ar*C*H), 128.5, 128.4 (Ar*C*H), 128.31 (Ar*C*H), 128.3 (Ar*C*H), 128.13 (Ar*C*H), 128.1 (Ar*C*H), 127.84 (Ar*C*H), 127.8 (Ar*C*H), 127.7 (Ar*C*H), 127.64 (Ar*C*H), 127.6 (Ar*C*H), 127.3 (Ar*C*H), 127.2 (Ar*C*H), 79.6 (d, J_{CP} 6.1, inositol ring), 78.2-77.9 (m, 3 × inositol ring), 75.3 (CH_2), 74.72 (inositol ring), 74.7 (CH_2), 73.2 (inositol ring), 72.0 (CH_2), 69.5 [d, J_{CP} 4.2, 2 × P(O)OCH_2Ph)], 69.3 [d, J_{CP} 6.7, P(O)OC_AH_2Ph)], 69.2 [d, J_{CP} 5.4, P(O)OC_BH_2Ph)], 28.7 [d, J_{CP} 30.9, P(O)($CH_2C_3H_7$)$_A$($CH_2C_3H_7$)$_B$], 27.5 [d, J_{CP} 33.3, P(O)($CH_2C_3H_7$)$_A$($CH_2C_3H_7$)$_B$], 24.5 [d, J_{CP} 2.5, P(O)($CH_2CH_2C_2H_5$)$_A$($CH_2CH_2C_2H_5$)$_B$], 24.4 [d, J_{CP} 3.6, P(O)($CH_2CH_2C_2H_5$)$_A$($CH_2CH_2C_2H_5$)$_B$], 24.1-23.8 [m, P(O)($CH_2C_2H_4CH_3$)$_A$($CH_2C_2H_4CH_3$)$_B$], 13.7 [P(O)($C_3H_6CH_3$)$_A$($C_3H_6CH_3$)$_B$], 13.6 [P(O)($C_3H_6CH_3$)$_A$($C_3H_6CH_3$)$_B$]; δ_P (121 MHz; $CDCl_3$) 62.1, - 0.37, - 0.60; *m/z* (ES+) 1153 ($[M+Na]^+$, 100%).

4.1.60. (-)-1D-4-*O*-Di-*n*-butylphosphinyl-*myo*-inositol 1,5-bisphosphate (sodium salt) 123

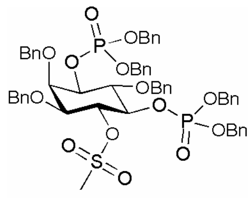


123

(+)-1D-2,3,6-tris-*O*-Benzyl-4-*O*-di-*n*-butylphosphinyl-*myo*-inositol 1,5-bis(dibenzyl phosphate) **138** (79 mg, 70 μmol, 1.0 equiv) was dissolved in *tert*-butanol/water (5/1, 12 mL), sodium hydrogen carbonate (24 mg, 280 μmol, 4.0 equiv) and palladium black (149 mg, 1.4 mmol, 20.0 equiv) were added and the flask flushed three times with hydrogen, then stirred for 8 h at RT under an atmosphere of hydrogen. The organic layer was removed by filtration, the dark residue washed with water (3 × 5 mL) and the collected aqueous layer lyophilized to yield *(-)-1D-4-O-di-n-butylphosphinyl-myo-inositol 1,5-bisphosphate (sodium salt)* **123** as a colourless solid (39 mg yield, 95%); $[\alpha]_D^{25}$ - 1.43 (*c* 0.5 in H_2O); ν_{max} (*KBr disc*)/cm^{-1} 3428.6 (s), 2959.6 (s), 2930.0 (s), 2868.3 (s), 1650.3 (m), 1457.3, (w), 1376.2 (w), 1236.4 (w), 1114.6 (s), 972.2 (s), 900.7 (w), 800.0 (w), 724.5 (w), 576.2 (w), 537.1 (w); δ_H (300 MHz; D_2O) 4.28-4.25 (1H, m, inositol ring), 4.20 (1H, t, *J* 9.2, inositol ring), 3.86-3.72 (3H, m, inositol ring), 3.63 (1H, dd, *J* 9.7, 2.8, inositol ring),1.97-1.77 [4C, m, P(O)C$H_2C_2H_4CH_3$)$_2$], 1.51-1.22 [8H, m, P(O)$CH_2C_2H_4CH_3$)$_2$], 0.82-0.76 (6H, m, P(O)$CH_2C_2H_4CH_3$)$_2$], δ_C (75 MHz; D_2O) 76.4 (dd, J_{CP} 8.3, 6.6, inositol ring), 76.2-76.1 (m, inositol ring), 74.3 (d, J_{CP} 5.5, inositol ring), 72.2 (d, J_{CP} 7.7, inositol ring), 70.9 (inositol ring), 69.4 (inositol ring), 23.6-23-1 [m, P(O)$C_3H_6CH_3$)$_2$], 13.0 [d,

J_{CP} 1.1, P(O)(C$_3$H$_6$CH$_3$)$_A$(C$_3$H$_6$CH$_3$)$_B$], 12.8 [d, J_{CP} 1.1, P(O)(C$_3$H$_6$CH$_3$)$_A$(C$_3$H$_6$*C*H$_3$)$_B$]; δ_P (121 MHz; D$_2$O) 70.3, 3.9, 3.0; *m/z* (ES+) [Found: $[C_{14}H_{29}O_{13}Na_3P_3]^+$ 567.0522. $C_{14}H_{29}O_{13}Na_3P_3$ requires M^+, 567.0514]; *m/z* (ES+) 567 $[C_{14}H_{29}O_{13}Na_3P_3]^+$, 100%).

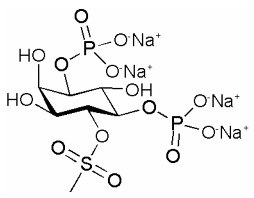
4.1.61. (+)-1D-2,3,6-tris-*O*-Benzyl-4-*O*-methylsulfonyl-*myo*-inositol 1,5-bis(dibenzylphosphate) 139



139

(+)-1D-2,3,6-tris-*O*-Benzyl-*myo*-inositol 1,5-bis(dibenzylphosphate) **122** (70 mg, 72 µmol, 1.0 equiv) was dissolved in dry dichloromethane (4 mL) under an atmosphere of nitrogen and the mixture cooled to - 78 °C. 4-Dimethylaminopyridine (catalytic amount) was added, followed by dry triethylamine (50 µL, 36 mg, 360 µmol, 5.0 equiv) and methanesulfonyl chloride (22 µL, 33 mg, 288 µmol, 4.0 equiv). The mixture was allowed to warm to RT and stirred for 2 days. The methanesulfonyl chloride was quenched with a saturated aqueous solution of sodium hydrogen carbonate (2 mL). The layers were separated and the aqueous layer extracted with dichloromethane (3 × 5 mL). The combined organic layers were dried (magnesium sulfate), filtered and concentrated under reduced pressure. Purification by silica gel column chromatography, eluting with ethyl acetate/petroleum ether (50/50, then 70/30) gave *(+)-1D-2,3,6-tris-O-benzyl-4-O-methylsulfonyl-myo-inositol 1,5-bis(dibenzylphosphate)* **139** (42 mg yield, 56%) as a colourless gum (Found: C, 64.0; H, 5.3. $C_{56}H_{58}O_{14}P_2S$ requires C, 64.1; H, 5.6); R_f 0.25 (ethyl acetate/petroleum ether 50/50); $[\alpha]_D^{25}$ + 1.5 (*c* 0.94 in CHCl$_3$); ν_{max} (*thin film*)/cm^{-1} 3064.4 (s), 3033.2 (s), 2931.9 (s), 1956.3 (w), 1884.7 (w), 1813.1 (w), 1726.8 (m), 1606.2 (w), 1497.6 (s), 1455.6 (s), 1355.1 (s), 1272.3 (s), 1214.7 (m), 1176.6 (m), 1124.5 (w), 1099.3 (w), 1014.2 (m), 880.9 (m), 847.7 (w), 736.2 (s), 696.5 (s), 599.6 (m); δ_H (300 MHz; CDCl$_3$) 7.39-7.00 (35H, m, Ar*H*), 5.14-4.41 (16H, m, 14 × OC*H*$_2$ and 2 × inositol ring, 4.37 (1H, 7, *J* 2.3, inositol ring), 4.26-4.17 (1H, m, inositol ring), 4.09 (1H, t, *J* 9.5, inositol ring), 3.40 (1H, dd, *J* 10.2, 2.0, inositol ring), 2.91 [3H, 2, S(O)$_2$C*H*$_3$]; δ_C (75 MHz; CDCl$_3$) 138.1 (Ar*C*), 138.0 (Ar*C*), 136.6 (Ar*C*), 135.9 [d, J_{CP} 7.7, P(O)(OCH$_2$*C*$_A$C$_5$H$_5$)], 135.7 [d, J_{CP} 6.8, P(O)(OCH$_2$*C*$_B$C$_5$H$_5$)] 135.5 [d, J_{CP} 7.0, P(O)(OCH$_2$*C*$_C$C$_5$H$_5$)], 128.6 (Ar*C*H), 128.4 (Ar*C*H), 128.34 (Ar*C*H), 128.3 (Ar*C*H), 128.2 (Ar*C*H), 128.1 (Ar*C*H), 128.01 (Ar*C*H), 128.0 (Ar*C*H), 127.9 (Ar*C*H), 127.85 (Ar*C*H), 127.8 (Ar*C*H), 127.3 (Ar*C*H), 79.9 (d, J_{CP} 4.5, inositol ring), 78.1 (dd, J_{CP} 8.1, 1.5, inositol ring), 77.7-77.6 (m, inositol ring), 77.1 (inositol ring), 75.3 (CH_2), 74.9 (CH_2), 74.8 (inositol ring), 72.6 (CH_2), 69.9 [d, J_{CP} 5.5, P(O)OC_AH_2Ph)], 69.6 [d, J_{CP} 5.6, P(O)OC_BH_2Ph)], 69.5-69.4 [d, J_{CP} 5.4, 2 × P(O)OCH_2Ph)], 39.3 [$S(O)_2CH_3$]; δ_P (121 MHz; $CDCl_3$) - 0.22, - 0.53; *m/z* (ES+) 1071 ($[M+Na]^+$, 100%).

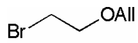
4.1.62. (+)-1D-4-*O*-Methylsulfonyl-*myo*-inositol 1,5-bisphosphate (sodium salt) 124



124

(+)-1D-2,3,6-tris-*O*-benzyl-4-*O*-methylsulfonyl-*myo*-inositol1,5-bis(dibenzyl phosphate) **139** (73 mg, 70 µmol, 1.0 equiv) was dissolved in *tert*-butanol/water (10/1, 11 mL), sodium hydrogen carbonate (24 mg, 280 µmol, 4.0 equiv) and palladium black (149 mg, 1.4 mmol, 20.0 equiv) were added and the flask flushed three times with hydrogen, then stirred for 8 h at RT under an atmosphere of hydrogen. The organic layer was removed by filtration, the dark residue washed with water (3 × 5 mL) and the collected aqueous layer lyophilized to yield *(+)-1D-4-O-methylsulfonyl-myo-inositol 1,5-bisphosphate (sodium salt)* **124** as a colourless solid (32 mg yield, 91%); $[\alpha]_D^{22}$ + 1.64 (*c* 0.3 in H_2O); v_{max} (*KBr disc*)/cm^{-1} 3448.9 (s), 2969.2 (s), 2924.4 (s), 1655.1 (m), 1340.9 (m), 1158.0 (s), 1107.8 (s), 973.7 (s), 942.6 (w), 869.9 (w), 802.8 (w), 724.5 (w), 598.6 (w), 539.9 (w); δ_H (300 MHz; D_2O) 4.60 (1H, t, *J* 9.5, inositol ring), 4.33 (1H, m, inositol ring), 4.00-3.91 (1H, m, inositol ring), 3.82-2.80 (2H, m, inositol ring), 3.75 (1H, dd, *J* 10.2, 3.0, inositol ring), 3.25 [3C, s, S(O)CH_3]; δ_C (75 MHz; D_2O) 83.8 (d, J_{CP}, 6.6, inositol ring), 75.0 (d, J_{CP} 5.5, inositol ring), 74.2 (d, J_{CP} 5.5, inositol ring), 72.3 (d, J_{CP} 7.7, inositol ring), 70.8 (inositol ring), 68.3 (inositol ring), 38.9 [S(O)CH_3]; δ_P (121 MHz; D_2O) 4.7, 4.1; *m/z* (ES-); 343, (100%), 439 $[C_7H_{14}NaO_{14}P_2S]^-$ (5), 417 $[C_7H_{15}O_{14}P_2S]^-$, (15),365 (20), 321 (40), 303 (15), 208 (15).

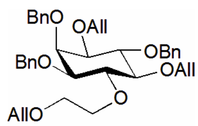
4.1.63. 2-Allyloxyethyl bromide 143



143

Phosphorus tribromide (5.6 mL, 16.7 g, 60.0 mmol, 0.35 equiv) was placed in a flask under an atmosphere of nitrogen and cooled to 0 °C. A mixture of 2-allyloxyethanol (18.2 mL, 17.4 g, 17.0 mmol, 1.0 equiv) and dry pyridine (4.8 mL, 4.7 g, 60.0 mmol, 0.35 equiv) was added dropwise with stirring over a period of 1.5 h. The resulting mixture was stirred for 30 min at 0 °C and for 2 h at RT. Distillation under reduced pressure furnished 2-allyloxyethyl bromide **143** (9.8 g yield, 35%) as a colourless oil; bp 30-35 °C (6-8 mbar); δ_H (300 MHz; $CDCl_3$) 5.92 (1H, ddt *J* 17.4, 10.5, 5.6 C*H*=CH_2), 5.30 (1H, ddt, *J* 17.4, 1.7, 1.5, CH=C*H*H), 5.22 (1H, ddt, *J* 10.5, 1.7, 1.2, CH=CH*H*), 4.34-4.21 (2H, ddd, *J* 5.6, 1.5, 1.2 C*H*HCH=CH_2), 3.77 (2H, t, *J* 6.1, O$CH_2$$CH_2$Br), 3.45 (2H, t, *J* 6.1, O$CH_2$$CH_2$Br). These data are in good agreement with the literature values.[144]

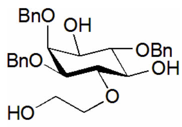
4.1.64. (-)-1D-1,5-Bis-*O*-allyl-4-*O*-(2-allyloxy)ethyl-2,3,6-tris-*O*-benzyl-*myo*-inositol 140



140

(+)-1D-1,5-Bis-*O*-allyl-2,3,6-tris-*O*-benzyl-*myo*-inositol **100** (170 mg, 320 μmol, 1.0 equiv) was dissolved in dry *N,N*-dimethyl formamide (5 mL) under an atmosphere of nitrogen. The resulting mixture was cooled to 0 °C and sodium hydride (60% w/w, 15 mg, 384 μmol, 1.2 equiv) added with stirring. The mixture was allowed to warm to RT and stirred for 2 h, then re-cooled to 0 °C and tetra-*n*-butylammonium iodide (catalytic amount) and 2-allyloxyethyl bromide (48 μL, 63 mg, 384 μmol, 1.2 equiv) were added. The resulting mixture was allowed to warm to RT and stirred overnight. The sodium hydride was quenched with water (0.5 mL), the solvent removed under reduced pressure and the residue reconstituted with ethyl acetate (10 mL) and water (10 mL). The layers were separated and the aqueous layer extracted with ethyl acetate (3 × 5 mL). The combined organic layers were washed with brine (10 mL), dried (magnesium sulfate), filtered and concentrated under reduced pressure. Purification by silica gel column chromatography, eluting with ethyl acetate/petroleum ether (10/90) gave *(-)-1D-1,5-bis-O-allyl-4-O-(2-allyloxy)ethyl-2,3,6-tris-O-benzyl-myo-inositol* **140** (158 mg yield, 80%) as a colourless oil; R_f 0.6 (ethyl acetate/petroleum ether 30/70); $[\alpha]_D^{25}$ - 7.8 (*c* 0.51 in $CHCl_3$); ν_{max} (*thin film*)/cm^{-1} 3064.0 (m), 3031.6 (m), 2984.1 (s), 2869.3 (s), 1647.2 (w), 1496.9 (w), 1454.9 (m), 1421.2 (w), 1266.0 (s), 1208.6 (w), 1131.9 (m), 1086.6 (s), 1028.1 (m), 996.0 (w), 926.4 (m), 737.3 (s), 699.3 (m); δ_H (300 MHz; $CDCl_3$) 7.43-7.29 (15H, m, Ar*H*), 6.06-5.83 (3H, m, $CH_X{=}CH_YH_Z$ + $CH_{X'}{=}CH_{Y'}H_{Z'}$ + $CH_{X''}{=}CH_{Y''}H_{Z''}$), 5.32-5.12 (6H, m, $CH_X{=}CH_YH_Z$ + $CH_{X'}{=}CH_{Y'}H_{Z'}$ + $CH_{X''}{=}CH_{Y''}H_{Z''}$), 4.86 (2H, s, CH_AH_B), 4.85 (1H, d, $J_{A'B'}$ 10.2, $OCH_{A'}H_{B'}$), 4.78 (1H, d, $J_{A'B'}$ 10.2, $OCH_{A'}H_{B'}$), 4.72 (1H, d, $J_{A''B''}$ 11.8, $OCH_{A''}H_{B''}$), 4.59 (1H, d, $J_{A''B''}$ 11.8, $OCH_{A''}H_{B''}$), 4.43-4.28 (2H, m, $CH_VH_WCH_X{=}CH_YH_Z$), 4.10-4.06 (2H, m, $CH_{V'}H_{W'}CH_{X'}{=}CH_{Y'}H_{Z'}$), 4.02-3.89 (6H, m, 2 × $CH_{V''}H_{W''}CH_{X''}{=}CH_{Y''}H_{Z''}$ + 2 × OCH_2CH_2OAll + 2 × inositol ring), 3.82 (1H, t, *J* 9.5, inositol ring), 3.61 (2H, t, *J* 4.9, OCH_2CH_2OAll), 3.29 (1H, d, *J* 9.2, inositol ring), 3.27 (1H, dd, *J* 9.7, 5.9, inositol ring), 3.18 (1H, dd, *J* 9.7, 2.3, inositol ring); δ_C (75 MHz; $CDCl_3$) 139.05 (Ar*C*), 139.0 (Ar*C*), 138.7 (Ar*C*), 135.6 ($CH_X{=}CH_YH_Z$), 135.0 ($CH_{X'}{=}CH_{Y'}H_{Z'}$), 134.9 ($CH_{X''}{=}CH_{Y''}H_{Z''}$), 128.33 (Ar*C*H), 128.3 (Ar*C*H), 128.1 (Ar*C*H), 127.8 (Ar*C*H), 127.6 (Ar*C*H), 127.5 (Ar*C*H), 127.3 (Ar*C*H), 116.8 ($CH_X{=}CH_YH_Z$), 116.6 ($CH_{X'}{=}CH_{Y'}H_{Z'}$), 116.4 ($CH_{X''}{=}CH_{Y''}H_{Z''}$), 83.1 (inositol ring), 82.5 (inositol ring), 81.6 (inositol ring), 80.53 (inositol ring), 80.5 (inositol ring), 75.9 (CH_2), 74.6 (CH_2), 74.5 (inositol ring), 74.0 (CH_2), 72.9 (CH_2), 72.7 (CH_2), 72.0 (CH_2), 71.7 (CH_2), 69.9 (CH_2); *m/z* (ES+) [Found: $(M+Na)^+$ 637.3140. $C_{38}H_{46}O_7Na$ requires M^+, 637.3141], *m/z* (ES+) 637 ($[M+Na]^+$, 100%).

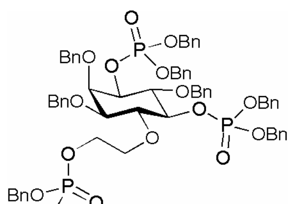
4.1.65. (-)-1D-4-*O*-(2-Hydroxy)ethyl-2,3,6-tris-*O*-benzyl-*myo*-inositol 141



141

Wilkinson's catalyst (53 mg, 49 µmol, 0.1 equiv) was dissolved in dry tetrahydrofuran (1.0 mL) under an atmosphere of nitrogen, *n*-butyl lithium (1.6 M solution in hexanes, 308 µL, 207 µmol, 0.4 equiv) was added and the resulting mixture stirred for 10 min at RT. The mixture was then cannulated onto a solution of (-)-1D-1,5-bis-*O*-allyl-4-*O*-(2-allyloxy)ethyl-2,3,6-tris-*O*-benzyl-*myo*-inositol **140** (303 mg, 493 µmol, 1.0 equiv) in dry tetrahydrofuran (0.5 mL) under an atmosphere of nitrogen and the resulting mixture heated under reflux for 6 h. The mixture was cooled to RT and the solvent removed under reduced pressure to give a dark red residue. ^{1}H NMR analysis indicated that the allyl groups had completely isomerised. The residue was dissolved in a mixture methanol/dichloromethane (2/3, 5 mL) under an atmosphere of nitrogen, acetyl chloride (21 µL, 23 mg, 296 µmol, 0.6 equiv) was added and the resulting mixture stirred for 2 h. The generated hydrochloric acid was quenched with triethylamine (0.2 mL), the solvent removed under reduced pressure, the residue adsorbed onto silica gel and purified using silica gel column chromatography, eluting with ethyl acetate/petroleum ether (60/40), *to give (-)-1D-4-O-(2-hydroxy)ethyl-2,3,6-tris-O-benzyl-myo-inositol* **141** (195 mg yield, 80%) as a colourless solid. A very pure sample was obtained by crystallisation from ethyl acetate and petroleum ether (Found: C, 70.4, H, 6.8; $C_{29}H_{34}O_7$ requires C, 70.4, H, 6.9); R_f 0.46 (ethyl acetate/petroleum ether 60/40)**;** $[\alpha]_D^{25}$ - 0.45 (*c* 1.1 in $CHCl_3$); mp 92-93 °C (*from ethyl acetate/petroleum ether*); ν_{max} (*KBr disc)*/cm^{-1} 3398.6 (s), 3064.4 (w), 3025.2 (w), 2911.9 (m), 2873.9 (m), 1496.9 (w), 1454.7 (m), 1364.5 (m), 1249.1 (w), 1208.9 (w), 1131.7 (s), 1085.6 (s), 2068.5 (s), 1023.2 (s), 928.7 (w), 723.3 (s), 969.8 (s), 607.0 (w), 539.9 (w); δ_H (300 MHz; $CDCl_3$) 7.37-7.29 (15H, m, Ar*H*), 5.00 (1H, d, J_{AB} 11.8, OCH_AH_B), 4.89 (1H, d, $J_{A'B'}$ 11.3, $OCH_{A'}H_{B'}$), 4.80 (1H, d, $J_{A'B'}$ 11.3, $OCH_{A'}H_{B'}$), 4.70 (1H, d, $J_{A'B'}$ 11.8, OCH_AH_B), 4.69 (2H, s, $OCH_{A''}H_{B''}$), 4.05-3.46 (9H, m, 4 × OCH_2H_2OH and 5 × inositol ring), 3.37 (1H, dd, *J* 9.7, 2.3, inositol ring), 3.30 (1H, br s, O*H*), 3.06 (1H, br s, O*H*), 2.26 (1H, d, *J* 7,4, O*H*); δ_C (75 MHz; $CDCl_3$) 138.5 (Ar*C*), 138.49 (Ar*C*), 137.8 (Ar*C*), 128.6 (Ar*C*H), 128.55 (Ar*C*H), 128.5 (Ar*C*H), 128.1 (Ar*C*H), 127.95 (Ar*C*H), 127.93 (Ar*C*H), 127.9 (Ar*C*H), 127.8 (Ar*C*H), 127.7 (Ar*C*H), 82.1 (inositol ring), 81.5 (inositol ring), 80.6 (inositol ring), 77.2 (inositol ring), 75.0 (CH_2), 74.91 (CH_2), 74.9 (inositol ring), 74.7 (CH_2), 72.8 (CH_2), 72.3 (inositol ring), 62.2 (CH_2OH); *m/z* (ES+) [Found: $(M+Na)^+$ 517.2192. $C_{29}H_{34}O_7Na$ requires M^+, 517.2202], *m/z* (ES+) 517 ($[M+Na]^+$, 100%).

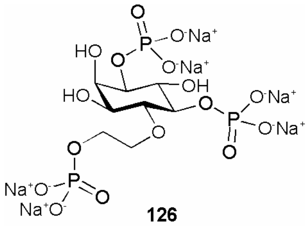
4.1.66. (+)-1D-4-*O*-(2-Dibenzylphosphoryloxy)ethyl-2,3,6-tris-*O*-benzyl-*myo*-inositol 1,5-bis(dibenzylphosphate) 142



142

Bis(benzyloxy)-*N,N*-diisopropylamino phosphine **92** (1 mg, 2.9 mmol, 7.5 equiv) was stirred with 1*H*-tetrazole (0.43 M solution in acetonitrile, 6.9 mL, 2.9 mmol, 7.5 equiv) for 30 min under an atmosphere of nitrogen. (-)-1D-4-*O*-(2-Hydroxy)ethyl-2,3,6-tris-*O*-benzyl-*myo*-inositol **141** (195 mg, 394 µmol, 1.0 equiv) dissolved in dry dichloromethane (8 mL) was added by cannula and the resulting mixture stirred overnight. The mixture was cooled to - 78 °C and 3-chloroperoxybenzoic acid (510 mg, 2.9 mmol, 7.5 equiv) was added. The resulting mixture was allowed to warm to RT and stirred for 30 min. The 3-chloroperoxybenzoic acid was quenched with a 10% aqueous solution of sodium hydrogen sulfite (10 mL). The layers were separated and the aqueous layer was extracted with dichloromethane (3 × 10 mL). The combined organic layers were washed with a saturated aqueous solution of sodium hydrogen carbonate (10 mL), dried (magnesium sulfate), filtered and concentrated under reduced pressure. Purification by silica gel column chromatography, eluting with ethyl acetate/petroleum ether (40/60, then 60/40), gave *(+)-1D-4-O-(2-dibenzylphosphoryloxy)ethyl-2,3,6-tris-O-benzyl-myo-inositol 1,5-bis(dibenzylphosphate)* **142** (232 mg yield, 46%) as a colourless oil (Found: C, 66.7, H, 5.8; $C_{71}H_{73}O_{16}P_3$ requires C, 66.9, H, 5.8); R_f 0.54 (ethyl acetate/petroleum ether 80/20); $[\alpha]_D^{25}$ + 9.7 (*c* 0.88 in $CHCl_3$); ν_{max} (*thin film*)/cm^{-1} 3064.3 (w), 3033.2 (w), 2948.6 (m), 2885.2 (m), 1497.5 (m), 1455.6 (s), 1273.7 (s), 1214.7 (m), 1012.1 (s), 920.3 (w), 881.7 (m), 736.5 (s), 696.4 (s); δ_H (300 MHz; $CDCl_3$) 7.28-6.97 (45H, m, Ar*H*), 4.98-4.50 (14H, s, 7 × OCH_2Ph), 4.89-4.59 (19H, m, 18 × OCH_2Ph and 1 × inositol ring), 4.46 (1H, d, J_{AB} 11.8, OCH_AH_B), 4.37 (1H, d, J_{AB} 11.8, OCH_AH_B), 4.15 (1H, t, *J* 2.0, inositol ring), 4.09-3.84 (5H, m, 2 × OCH_2CH_2O and 3 × inositol ring), 3.74 (2H, t, *J* 9.5, OCH_2CH_2O), 3.09 (1H, dd, *J* 9.7, 2.0, inositol ring); δ_C (75 MHz; $CDCl_3$), 138.5 (Ar*C*), 138.1 (Ar*C*), 137.7 (Ar*C*), 136-135.5 [m, 6 × P(O)(OC$H_2CC_5H_5$], 128.52 (Ar*C*H), 128.5 (Ar*C*H), 128.4 (Ar*C*H), 128.3 (Ar*C*H), 128.24 (Ar*C*H), 128.2 (Ar*C*H), 128.1 (Ar*C*H), 127.84 (Ar*C*H), 127.8 (Ar*C*H), 127.7 (Ar*C*H), 127.6 (Ar*C*H), 127.5 (Ar*C*H), 127.4 (Ar*C*H), 127.3 (Ar*C*H), 80.4 (d, J_{CP} 6.6, inositol ring), 79.9 (d, J_{CP} 1.7, inositol ring), 79.3 (inositol ring), 78.1 (dd, J_{CP} 11.4, 4.0, inositol ring), 77.9 (d, J_{CP} 5.9, inositol ring), 76.0 (inositol ring), 75.1 (CH_2), 74.6 (CH_2), 72.7 (CH_2), 71.6 (d, J_{CP} 7.8, OCH_2CH_2O), 69.4 [d, J_{CP} 5.9, P(O)OCH_2Ph], 69.3-69.2 [m, 3 × P(O)OCH_2Ph], 69.0 [d, J_{CP} 5.7, 2 × P(O)OCH_2Ph], 66.9 (d, J_{CP} 6.1, OCH_2CH_2O); δ_P (121 MHz; $CDCl_3$) 0.38, - 0.35, - 0.61; *m/z* (ES+) 1297 ($[M+Na]^+$, 100%).

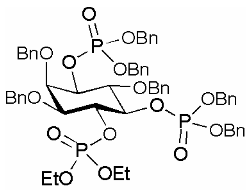
4.1.67. (-)-1D-4-*O*-(2-Phosphoryloxy)ethyl-*myo*-inositol 1,5-bisphosphate (sodium salt) 126



126

(+)-1D-4-*O*-(2-Dibenzylphosphoryloxy)ethyl-2,3,6-tris-*O*-benzyl-*myo*-inositol **142** 1,5-bis (dibenzylphosphate) (97 mg, 76 µmol, 1.0 equiv) was dissolved in *tert*-butanol/water (5/1, 12 mL), sodium hydrogen carbonate (38 mg, 455 µmol, 6.0 equiv) and palladium black (162 mg, 1.5 mmol, 20.0 equiv) were added and the flask flushed three times with hydrogen, then stirred for 8 h at RT under an atmosphere of hydrogen. The organic layer was removed by filtration, the dark residue washed with water (3 × 5 mL) and the collected aqueous layer lyophilized to yield *(-)-1D-4-O-(2-phosphoryloxy)ethyl-myo-inositol 1,5-bisphosphate (sodium salt)* **126** as a colourless solid (40 mg yield, 89%); $[\alpha]_D^{25}$ - 2.95 (*c* 0.44 in H_2O); v_{max} (*KBr disc*)/cm^{-1} 3290.0 (s), 2963.6 (m), 2930.0 (m), 1655.1 (s), 1639.2 (s), 1093.2 (s), 978.3 (s), 802.8 (w), 721.7 (w), 550.5 (m); δ_H (300 MHz; D_2O) 4.30 (1H, br s, inositol ring), 4.11-4.06 (1H, m, inositol ring), 3.81-3.69 (6H, m, 4 × OCH_2CH_2O and 2 × inositol ring), 3.58-3.45 (2H, m, inositol ring); δ_C (75 MHz; D_2O) 81.3 (d, J_{CP}, 6.0, inositol ring), 78.4 (d, J_{CP} 5.4, inositol ring), 74.8 (d, J_{CP} 5.7, inositol ring), 73.2 (d, J_{CP} 7.3, OCH$_2$CH$_2$O), 72.8 (d, J_{CP} 7.0, inositol ring), 70.9 (inositol ring), 69.9 (inositol ring), 64.2 [d, J_{CP} 4.5, OCH$_2$$CH_2$O]; δ_P (121 MHz; D_2O) 5.2, 4.9, 4.1; *m/z* (ES+); 289 (100%), 597 $[M+H]^+$ (50).

4.1.68. (-)-1D-2,3,6-Tris-*O*-benzyl-4-*O*-diethylphosphoryl-*myo*-inositol 1,5-bis(dibenzylphosphate) 146

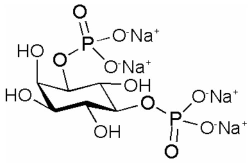


146

(+)-1D-2,3,6-tris-*O*-Benzyl-*myo*-inositol 1,5-bis(dibenzylphosphate) **122** (100 mg, 103 µmol, 1.0 equiv) was dissolved in dry dichloromethane (2 mL) under an atmosphere of nitrogen and the mixture cooled to - 78 °C. Dry triethylamine (57 µL, 42 mg, 412 µmol, 4.0 equiv) was added, followed by diethylchlorophosphite (45 µL, 48 mg, 309 μmol, 3.0 equiv). The mixture was allowed to warm to RT and stirred for 4 h. TLC anaysis indicated complete consumption of the starting material and the presence of a less polar spot. The mixture was re-cooled to - 78 °C and 3-chloroperoxybenzoic acid (53 mg, 309 μmol, 3.0 equiv) added. The resulting mixture was allowed to wam to RT and stirred for 30 min. The 3-chloroperoxybenzoic acid was quenched with a 10% aqueous solution of sodium hydrogen sulfite (2 mL) and the mixture stirred for 30 min, then the layers were separated and the aqueous layer extracted with dichloromethane (3 × 5 mL). The combined organic layers were washed with asaturated aqueous soluton of sodium hydrogen carbonate (5 mL), dried (magnesium sulfate), filtered and concentrated under reduced pressure. Purification by silica gel column chromatography, eluting with ethyl acetate/petroleum ether (60/40) gave *(-)-1D-2,3,6-tris-O-benzyl-4-O-diethylphosphoryl-myo-inositol 1,5-bis(dibenzylphosphate)* **146** (59 mg yield, 52%) as a colourless solid (Found: C, 63.8; H, 5.9. $C_{59}H_{65}O_{15}P_3$ requires C, 64.0; H, 5.9). A very pure sample was obtained by crystallisation from diethyl ether, ethyl acetate and petroleum ether; R_f 0.48 (ethyl acetate/petroleum ether 80/20); $[\alpha]_D^{25}$ - 1.3 (*c* 1.1 in $CHCl_3$); mp 94-95 °C (*from diethyl ether/ethyl acetate/petroleum ether*); ν_{max} (*thin film*)/cm^{-1} 3064.4 (w), 3036.4 (w), 2937.3 (m), 1498.1 (m), 1455.5 (m), 1382.4 (w), 1261.5 (s), 1216.1 (w), 1160.7 (m), 1104.9 (m), 1037.8 (s), 10238. (s), 877.1 (m), 730.6 (m), 695.9 (m), 497.3 (w); δ_H (300 MHz; $CDCl_3$) 7.33-6.89 (35H, m, Ar*H*), 5.05 (1H, dd, J_{AB} 11.8, J_{HP} 6.6, OCH_AH_B), 4.90 (1H, dd, J_{AB} 11.8, J_{HP} 6.6, OCH_AH_B), 4.84-4.37 (14H, m, 12 × OCH_2 and 2 × inositol ring), 4.26 (1H, t, *J* 2.0, inositol ring), 4.21-4.15 (1H, m, inositol ring), 4.06-3.82 [5H, m, 4 × $P(O)(CH_2CH_3)_2$ and 1 × inositol ring], 3.35 (1H, dd, *J* 10.0, 2.3, inositol ring), 1.06 [3H, td, *J* 7.2, J_{HP} 1.3, $P(O)(CH_2CH_3)_A(CH_2CH_3)_B$], 1.00 [3H, td, *J* 7.2, J_{HP} 1.3, $P(O)(CH_2CH_3)_A(CH_2CH_3)_B$]; δ_C (75 MHz; $CDCl_3$) 138.3 (Ar*C*), 138.29 (Ar*C*) 137.5 (Ar*C*), 136.2 [d, J_{CP} 8.1, $P(O)(OCH_2C_AC_5H_5)$],135.9 [d, J_{CP} 7.5, $P(O)(OCH_2C_BC_5H_5)$] 135.6 [d, J_{CP} 2.7, $P(O)(OCH_2C_AC_5H_5)$], 135.5 [d, J_{CP} 2.4, $P(O)(OCH_2C_AC_5H_5)$], 128.6 (Ar*C*H), 128.5 (Ar*C*H), 128.32 (Ar*C*H), 128.3 (Ar*C*H), 128.2 (Ar*C*H), 128.1 (Ar*C*H), 127.9 (Ar*C*H), 127.8 (Ar*C*H), 127.7 (Ar*C*H), 127.6 (Ar*C*H), 127.1 (Ar*C*H), 79.2-79.0 (m, inositol ring), 78.0-77.9 (m, 2 × inositol ring), 77.6 (inositol ring), 77.4 (inositol ring), 75.3 (CH_2), 75.2 (inositol ring), 74.5 (CH_2), 72.3 (CH_2), 69.5-69.1 [m, 4 × $P(O)OCH_2Ph$)], 64.1 [d, J_{CP} 6.4, $P(O)(CH_2CH_3)_A(CH_2CH_3)_B$], 63.8 [d, J_{CP} 5.9, $P(O)(CH_2CH_3)_A(CH_2CH_3)_B$], 16.0 [$P(O)(CH_2CH_3)_A(CH_2CH_3)_B$], 15.9

$[P(O)(CH_2CH_3)_A(CH_2CH_3)_B]$; δ_P (121 MHz; $CDCl_3$) - 1.67, - 1.70, - 1.88; *m/z* (ES+) 1129 ($[M+Na]^+$, 100%).

4.1.69. (+)-1D-*myo*-Inositol 1,5-bisphosphate (sodium salt) 125



125

(+)-1D-2,3,6-tris-*O*-Benzyl-*myo*-inositol 1,5-bis(dibenzylphosphate) **122** (100 mg, 103 μmol, 1.0 equiv) (92 mg, 94 μmol, 1.0 equiv) was dissolved in *tert*-butanol/water (5/1, 10 mL), sodium hydrogen carbonate (32 mg, 377 μmol, 4.0 equiv) and palladium black (201 mg, 1.9 mmol, 20.0 equiv) were added and the flask flushed three times with hydrogen, then stirred for 8 h at RT under an atmosphere of hydrogen. The organic layer was removed by filtration, the dark residue washed with water (3 × 5 mL) and the collected aqueous layer lyophilized to yield (+)-1D-*myo*-inositol 1,5-bisphosphate (sodium salt) **125** as a colourless solid (37 mg yield, 92%); $[\alpha]_D^{25}$ + 5.7 (*c* 0.53 in H_2O) [Lit.[145] + 6.0 (*c* 0.5 in H_2O)]; ν_{max} (*KBr disc*)/cm^{-1} 3423.5 (s), 2930.0 (m), 1655.1 (m), 1560.7 (w), 1376.3 (w), 1094.1 (s), 968.2 (s), 897.9 (w), 808.6 (m), 718.9 (m), 568.0 (m); δ_H (300 MHz; D_2O) 4.18 (1H, t, *J* 2.8, inositol ring), 3.87-3.81 (1H, m, inositol ring), 3.75-3.64 (3H, m, inositol ring), 3.53 (1H, dd, *J* 9.5, 2.8, inositol ring); δ_C (75 MHz; D_2O) 78.3 (d, J_{CP} 5.6, inositol ring), 74.6 (d, J_{CP} 5.1, inositol ring), 72.6 (d, J_{CP} 1.5, inositol ring), 71.9 (t, J_{CP} 5.0, inositol ring), 71.6 (inositol ring), 71.0 (inositol ring); δ_P (121 MHz; D_2O) 5.4, 4.5; *m/z* (ES-) 259 ($[C_6H_{12}O_9P]^-$ 100%), 405 $[C_6H_{10}Na_3O_{12}P_2]^-$ (15), 383 $[C_6H_{11}Na_2O_{12}P_2]^-$ (20), 361 $[C_6H_{12}NaO_{12}P_2]^-$ (50), 339 $[C_6H_{13}O_{12}P_2]^-$ (45), 281 $[C_6H_{11}NaO_9P]^-$ (70). These data are in good agreement with the literature values.[145]

References

5 References

(1) J. Scherer, *Justus Liebigs Ann. Chem.*, 1850, **73**, 322-328.
(2) J. Folch, *J. Biol. Chem.*, 1949, **177**, 505-519.
(3) M. R. Hokin and L. E. Hokin, *J. Biol. Chem.*, 1953, **203**, 967-977.
(4) R. C. M. Dawson, *Biochim. Biophys. Acta*, 1954, **14**, 374-375.
(5) R. H. Michell, *Phosphoinositides and Receptors Mechanism*, Ed. J. W. J. Putney, Alan R. Liss, New York, 1986.
(6) M. R. Hokin and L. E. Hokin, *Metabolism and Physiological Significance of Lipids*, Ed. D. N. Rhodes, John Wiley, London, 1964.
(7) J. Durell and J. T. Garland, *Ann. NY Acad. Sci.*, 1969, **165**, 743-754.
(8) A. A. Abdel-Latif, R. A. Akhtar, and J. N. Hawthorne, *Biochem. J.*, 1977, **162**, 61-63.
(9) R. A. Akhtar and A. A. Abdel-Latif, *Biochem. J.*, 1980, **192**, 783-791.
(10) R. H. Michell, *Biochim. Biophys. Acta*, 1975, **415**, 81-147.
(11) M. J. Berridge and J. N. Fain, *Biochem. J.*, 1979, **178**, 59-69.
(12) Y. Takai, A. Kishimoto, U. Kikkawa, T. Mori, and Y. Nishizuka, *Biochem. Biophys. Res. Commun.*, 1979, **91**, 1218-1224.
(13) R. H. Michell, C. J. Kirk, L. M. Jones, C. P. Downes, and J. A. Creba, *Philos. Trans. Roy. Soc. Lond. B*, 1981, **296**, 123-138.
(14) H. Streb, R. F. Irvine, M. J. Berridge, and I. Schulz, *Nature*, 1983, **306**, 67-69.
(15) R. V. Tomlinson and C. E. Ballou, *J. Biol. Chem.*, 1961, **236**, 1902-1906.
(16) M. J. Berridge and R. F. Irvine, *Nature*, 1989, **341**, 197-205.
(17) D. J. Cosgrove, *Inositol Phosphates, Their Chemistry, Biochemistry and Physiology*, Elsevier, Oxford, 1980.
(18) S. K. Fisher, J. E. Novak, and B. W. Agranoff, *J. Neurochem.*, 2002, **82**, 736.
(19) B. W. Agranoff, *Trends Biochem. Sci.*, 1978, **3**, N283-N285.
(20) R. F. Irvine and M. J. Schell, *Nat. Rev. Mol. Cell Biol.*, 2001, **2**, 327-338.
(21) IUB Nomenclature Committee, *Biochem. J.*, 1989, **258**, 1-2.
(22) S. Ringer, *J. Physiol.*, 1883, **4**, 29-43.
(23) A. Weber, *J. Biol. Chem.*, 1959, **234**, 2764-2769.
(24) S. Ebashi, *J. Biochem.*, 1961, **50**, 236-244.
(25) S. Ebashi and F. Lipmann, *J. Cell Biol.*, 1962, **14**, 389-400.
(26) W. Hasselbach and M. Makinose, *Biochem. Z.*, 1961, **333**, 518-528.
(27) M. J. Berridge, P. Lipp, and M. D. Bootman, *Nat. Rev. Mol. Cell Biol.*, 2000, **1**, 11-21.
(28) M. J. Berridge, M. D. Bootman, and H. L. Roderick, *Nat. Rev. Mol. Cell Biol.*, 2003, **4**, 517-529.
(29) T. A. White, M. S. Kannan, and T. F. Walseth, *FASEB J.*, 2003, **17**, 482-484.
(30) I. Bosanac, J.-R. Alattia, T. K. Mal, J. Chan, S. Talarico, F. K. Tong, K. I. Tong, F. Yoshikawa, T. Furuichi, M. Iwai, T. Michikawa, K. Mikoshiba, and M. Ikura, *Nature*, 2002, **420**, 696-700.
(31) E. C. Thrower, R. E. Hagar, and B. E. Ehrlich, *Trends Pharmacol. Sci.*, 2001, **22**, 580-586.
(32) I. Bosanac, H. Yamazaki, T. Matsu-ura, T. Michikawa, K. Mikoshiba, and M. Ikura, *Mol. Cell.*, 2005, **17**, 193-203.
(33) C. W. Taylor, P. C. A. da Fonseca, and E. P. Morris, *Trends Biochem. Sci.*, 2004, **29**, 210-219.
(34) G. A. Mignery and T. C. Südhof, *EMBO J.*, 1990, **9**, 3893-3898.
(35) S. Patel, S. K. Joseph, and A. P. Thomas, *Cell Calcium*, 1999, **25**, 247-264.
(36) T. Meyer, D. Holowka, and L. Stryer, *Science*, 1988, **240**, 653-656.
(37) C. W. Taylor and A. J. Laude, *Cell Calcium*, 2002, **32**, 321-334.
(38) E. A. Finch, T. J. Turner, and S. M. Goldin, *Science*, 1991, **252**, 443-446.
(39) K. Hamada, A. Terauchi, and K. Mikoshiba, *J. Biol. Chem.*, 2003, **278**, 52881-52889.
(40) Serysheva, II, D. J. Bare, S. J. Ludtke, C. S. Kettlun, W. Chiu, and G. A. Mignery, *J. Biol. Chem.*, 2003, **278**, 21319-21322.
(41) P. C. A. da Fonseca, S. A. Morris, E. P. Nerou, C. W. Taylor, and E. P. Morris, *Proc. Nat. Acad. Sci. (USA)*, 2003, **100**, 3936-3941.
(42) V. G. Veresov and S. V. Konev, *Biochem. Biophys. Res. Comm.*, 2006, **341**, 1277-1285.
(43) I. Sienaert, N. Nadif Kasri, S. Vanlingen, J. B. Parys, G. Callewaert, L. Missiaen, and H. De Smedt, *Biochem. J.*, 2002, **365**, 269-277.
(44) V. I. Shvets, *Russ. Chem Rev. (Engl. Trans.)*, 1974, **43**, 488-493.
(45) R. Gigg, *Chem. Phys. Lip.*, 1980, **26**, 287-404.
(46) T. Posternak, *The Cyclitols*, Holden-Day Inc., San Francisco, 1965.

(47) S. Ozaki, Y. Watanabe, T. Ogasawara, Y. Kondo, N. Shiotani, H. Nishii, and T. Matsuki, *Tetrahedron Lett.*, 1986, **27**, 3157-3160.
(48) B. V. L. Potter and D. Lampe, *Angew. Chem. Int. Ed. Engl.*, 1995, **34**, 1933-1972.
(49) M. Takahashi, T. Kagasaki, T. Hosoya, and S. Takahashi, *J. Antibiot.*, 1993, **46**, 1643-1647.
(50) W. Li, C. Schultz, J. Llopis, and R. Y. Tsien, *Tetrahedron*, 1997, **53**, 12017-12040.
(51) A. M. Cooke, R. Gigg, and B. V. L. Potter, *J. Chem. Soc. Chem. Comm.*, 1987, 1525-1526.
(52) C. W. Taylor, M. J. Berridge, K. D. Brown, A. M. Cooke, and B. V. L. Potter, *Biochem. Biophys. Res. Comm.*, 1988, **150**, 626.632.
(53) C. W. Taylor, M. J. Berridge, A. M. Cooke, and B. V. L. Potter, *Biochem. J.*, 1989, **259**, 645-650.
(54) D. Lampe, C. S. Liu, and B. V. L. Potter, *J. Med. Chem.*, 1994, **37**, 907-912.
(55) N. J. Noble, A. M. Cooke, and B. V. L. Potter, *Carbohydr. Res.*, 1992, **234**, 177-187.
(56) A. M. Cooke, N. J. Noble, S. Payne, R. Gigg, and B. V. L. Potter, *J. Chem. Soc. Chem. Commun.*, 1989, 269-271.
(57) S. J. Mills, A. M. Riley, C. T. Murphy, A. J. Bullock, J. Westwick, and B. V. L. Potter, *Bioorg. Med. Chem. Lett.*, 1995, **5**, 203-208.
(58) D. Lampe, S. J. Mills, and B. V. L. Potter, *J. Chem. Soc. Perkin Trans. 1*, 1992, 2899-2906.
(59) R. J. Mourey, V. A. Estevez, J. F. Marecek, R. K. Barrow, G. D. Prestwich, and S. H. Snyder, *Biochemistry*, 1993, **32**, 1719-1726.
(60) Y. Watanabe, M. Hirata, T. Ogasawara, T. Koga, and S. Ozaki, *Bioorg. Med. Chem. Lett.*, 1991, **1**, 399-402.
(61) D. L. Kalinosky, S. B. Aldinger, A. G. Boyle, T. Huque, J. F. Marecek, G. D. Prestwich, and D. Restrepo, *Biochem. J.*, 1992, **281**, 449-456.
(62) M. Takahashi, T. Kagasaki, and S. Takahashi, *J. Biol. Chem.*, 1994, **269**, 369-372.
(63) J. S. Marchant, M. D. Beecroft, A. M. Riley, D. J. Jenkins, R. D. Marwood, C. W. Taylor, and B. V. L. Potter, *Biochemistry*, 1997, **36**, 12780-12790.
(64) C. T. Murphy, A. M. Riley, C. J. Lindley, D. J. Jenkins, J. Westwick, and B. V. L. Potter, *Mol. Pharmacol.*, 1997, **52**, 741-748.
(65) S. Shuto, K. Tatani, Y. Ueno, and A. Matsuda, *J. Org. Chem.*, 1998, **63**, 8815-8824.
(66) M. Kashiwayanagi, K. Tatani, S. Shuto, and A. Matsuda, *Eur. J. Neurosci.*, 2000, **12**, 606-612.
(67) N. C. R. van Straten, G. A. van der Marel, and J. H. van Boom, *Tetrahedron*, 1997, **53**, 6523-6538.
(68) M. D. Beecroft, J. S. Marchant, A. M. Riley, N. C. R. van Straten, A. G. van Marel, J. H. van Boom, B. V. L. Potter, and C. W. Taylor, *Mol. Pharmacol.*, 1999, **55**, 109.
(69) R. D. Marwood, S. Shuto, D. J. Jenkins, and B. V. L. Potter, *Chem. Commun.*, 2000, **3**, 219-220.
(70) A. B. Holmes *et al, unpublished data*.
(71) T. K. Ghosh, P. S. Eis, J. M. Mullaney, C. L. Ebert, and D. L. Gill, *J. Biol. Chem.*, 1988, **263**, 11075-11079.
(72) E. P. Nerou, A. M. Riley, B. V. L. Potter, and C. W. Taylor, *Biochem. J.*, 2001, **355**, 59-69.
(73) M. A. Tones, M. D. Bootman, B. F. Higgins, D. A. Lane, G. F. Pay, and U. Lindahl, *FEBS Letter*, 1989, **252**, 105-108.
(74) L. L. Dasso and C. W. Taylor, *Biochem. J.*, 1991, **280**, 791-795.
(75) K. Takazawa and C. Erneux, *Biochem. J.*, 1989, **261**, 1059-1060.
(76) I. Bezprozvanny, K. Ondrias, E. Kaftan, D. A. Stoyanovsky, and B. E. Ehrlich, *Mol. Biol. Cell*, 1993, **4**, 347-352.
(77) J. Gafni, J. A. Munsch, T. H. Lam, M. C. Catlin, L. G. Costa, T. F. Molinski, and I. N. Pessah, *Neuron*, 1997, **19**, 723-733.
(78) J. Venkateswara Rao, D. Desaiah, P. J. Vig, and Y. Venkateswarlu, *Toxicology*, 1998, **129**, 103-112.
(79) L. M. Broad, T. R. Cannon, and C. W. Taylor, *J. Physiol.*, 1999, **517**, 121-134.
(80) R. Schaloske, C. Schlatterer, and D. Malchow, *J. Biol. Chem.*, 2000, **275**, 8404-8408.
(81) U. K. Misra, G. Gawdi, and S. V. Pizzo, *J. Cell. Biochem.*, 1997, **64**, 225-232.
(82) A. P. Passos and C. R. Garcia, *Biochem. Biophys. Res. Comm.*, 1998, **245**, 155-160.
(83) J. P. Sieb, M. Milone, and A. G. Engel, *Brain Res.*, 1996, **712**, 179-189.
(84) J. A. Ballestero, P. V. Plazas, S. Kracun, M. E. Gomez-Casati, J. Taranda, C. V. Rothlin, E. Katz, N. S. Millar, and A. B. Elgoyhen, *Mol. Pharm.*, 2005, **68**, 822-829.
(85) R. E. Phillips, S. Looareesuwan, N. J. White, P. Chanthavanich, J. Karbwang, W. Supanaranond, R. C. Turner, and D. A. Warrell, *Brit. Med. J.*, 1986, **292**, 1319-1321.
(86) K. Bokvist, P. Rorsman, and P. A. Smith, *J. Physiol.*, 1990, **423**, 327-342.
(87) B. Baune, V. Furlan, A. M. Taburet, and R. Farinotti, *Drug Met. Disp.*, 1999, **27**, 565-568.

(88) K. J. Föhr, Y. Wahl, R. Engling, T. P. Kemmer, and M. Gratzl, *Cell Calcium*, 1991, **12**, 735-742.

(89) K. J. Föhr, J. Scott, M. Ahnert-Hilger, and M. Gratzl, *Biochem. J.*, 1989, **262**, 83-89.

(90) J. Strupish, R. J. H. Wojcikiewicz, R. A. J. Chaliss, S. T. Safrany, A. L. Willcocks, B. V. L. Potter, and S. R. Nahorski, *Biochem. J.*, 1991, **277**, 294.

(91) T. Maruyama, T. Kanaji, S. Nakade, T. Kanno, and K. Mikoshiba, *Jpn. J. Biochem.*, 1997, **122**, 498-505.

(92) C. M. Peppiatt, T. J. Collins, L. Mackenzie, S. J. Conway, A. B. Holmes, M. D. Bootman, M. J. Berridge, S. J. T., and H. L. Roderick, *Cell Calcium*, 2003, **34**, 97-108.

(93) L. Missiaen, G. Callewaert, H. De Smedt, and J. B. Parys, *Cell Calcium*, 2001, **29**, 111-116.

(94) M. D. Bootman, T. J. Collins, L. Mackenzie, H. L. Roderick, M. J. Berridge, and C. M. Peppiatt, *FASEB J.*, 2002, **16**, 1145-1150.

(95) H. Iwaski, Y. Mori, Y. Hara, K. Uchida, H. Zhou, and K. Mikoshiba, *Recept. Channels*, 2001, 429-439.

(96) M. Prakriya and R. S. Lewis, *J. Physiol.*, 2001, **536**, 3-19.

(97) H. T. Ma, K. Venkatachalam, J. B. Parys, and D. L. Gill, *J. Biol. Chem.*, 2002, **277**, 6915-6922.

(98) F. J. Braun, L. M. Broad, D. L. Armstrong, and J. W. Putney, *J. Biol. Chem.*, 2001, **276**, 1063-1070.

(99) J. G. Bilmen, L. L. Wooton, R. E. Godfrey, O. S. Smart, and F. Michelangeli, *Eur. J. Biochem.*, 2002, **269**, 3678-3687.

(100) H. Z. Hu, Q. H. Gu, C. B. Wang, C. K. Colton, J. S. Tang, M. Kinoshita-Kawada, L. Y. Lee, J. D. Wood, and M. X. Zhu, *J. Biol. Chem.*, 2004, **279**, 35741-35748.

(101) C. E. Dreef, W. Schieblerb, G. A. van der Marel, and J. H. van Boom, *Tetrahedron Lett.*, 1991, **32**, 6021-6024.

(102) C. E. Dreef, J. P. Jansze, C. J. J. Elie, G. A. van der Marel, and J. H. van Boom, *Carbohydr. Res.*, 1992, **234**, 37-50.

(103) J. Desai, J. Gigg, R. Gigg, and S. Payne, *Carbohydr. Res.*, 1992, **225**, 209-228.

(104) G. F. Painter, S. J. A. Grove, I. H. Gilbert, A. B. Holmes, P. R. Raithby, M. L. Hill, P. T. Hawkins, and L. R. Stephens, *J. Chem. Soc. Perkin Trans. 1*, 1999, 923-935.

(105) S. J. A. Grove, I. H. Gilbert, A. B. Holmes, G. F. Painter, and M. L. Hill, *Chem. Comm.*, 1997, 1633-1634.

(106) D. C. Billington, R. Baker, J. J. Kulagowski, I. M. Mawer, J. P. Vacca, S. J. DeSolms, and J. R. Huff, *J. Chem. Soc., Perkin Trans. 1*, 1989, 1423-1429.

(107) I. H. Gilbert, A. B. Holmes, and R. C. Young, *Tetrahedron Lett.*, 1990, **31**, 2633-2634.

(108) I. H. Gilbert, A. B. Holmes, M. J. Pestchanker, and R. C. Young, *Carbohydr. Res.*, 1992, **234**, 117-130.

(109) Z. Y. Lim, T. J. W., A. B. Holmes, M. Manifava, and N. T. Ktistakis, *J. Chem. Soc. Perkin Trans. 1*, 2002, 1067-1075.

(110) Z. Y. Lim, PhD Thesis, University of Cambridge, 2001.

(111) M. Naruto, K. Ohno, N. Naruse, and H. Takeuchi, *Tetrahedron Lett.*, 1979, 251-254.

(112) C. A. A. van Boeckel and T. Beetz, *Tetrahedron Lett.*, 1983, **24**, 3775-3778.

(113) A. S. Campbell and B. Fraser-Reid, *J. Am. Chem. Soc.*, 1995, **117**, 10387-10388.

(114) Y. Watanabe, M. Tomioka, and S. Ozaki, *Tetrahedron*, 1995, **51**, 8969-8976.

(115) A. Chaudhary, G. Dormán, and G. D. Prestwich, *Tetrahedron Lett.*, 1994, **35**, 7521-7524.

(116) T. W. Greene and P. G. M. Wuts, *Substituted benzyl ethers*, in *Protective groups in organic synthesis (third edition)*, Ed. P. G. M. Wuts, 1999, John Wiley & Sons, Inc., New York, p. 86-87.

(117) E. J. Nurminen, J. K. Mattinen, and H. Lönnberg, *J. Chem. Soc., Perkin Trans. 2*, 1998, 1621-1628.

(118) E. J. Nurminen, J. K. Mattinen, and H. Lönnberg, *J. Chem. Soc., Perkin Trans. 2*, 2001, 2159-2165.

(119) S. David, *Selective O-substitution and oxidation using stannylene acetals and stannyl ethers*, in *Preparative carbohydrate chemistry*, Ed. S. Hanessian, 1997, M. Dekker Inc., New York, p. 69-83.

(120) R. M. Moriarty, C. Condeiu, A. Tao, and O. Prakash, *Tetrahedron Lett.*, 1997, **38**, 2401-2404.

(121) G. Schrader, W. Lorentz, R. Coelln, and H. Schloer, *Chemical Abstracts*, 1966, **64**,15923b.

(122) G. Schrader, W. Lorentz, R. Coelln, and H. Schloer, *Farbenfabriken Bayer Aktiengesellschaft*, 1966, US 3232830, 21.

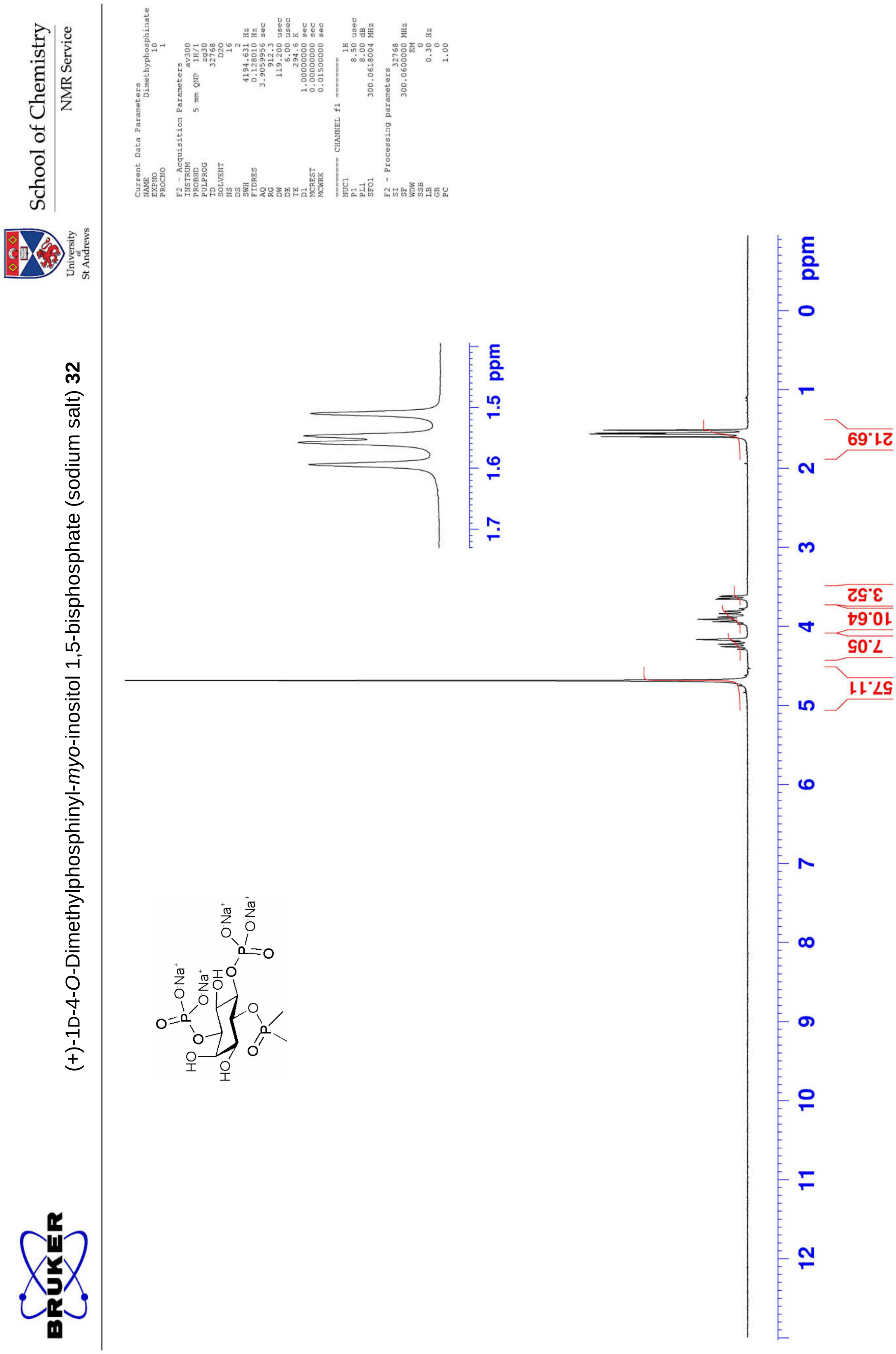
(123) R. K. Kanjolia, D. K. Srivastava, C. L. Watkins, and L. K. Krannick, *Inorg. Chem.*, 1989, **28**, 3341-3345.

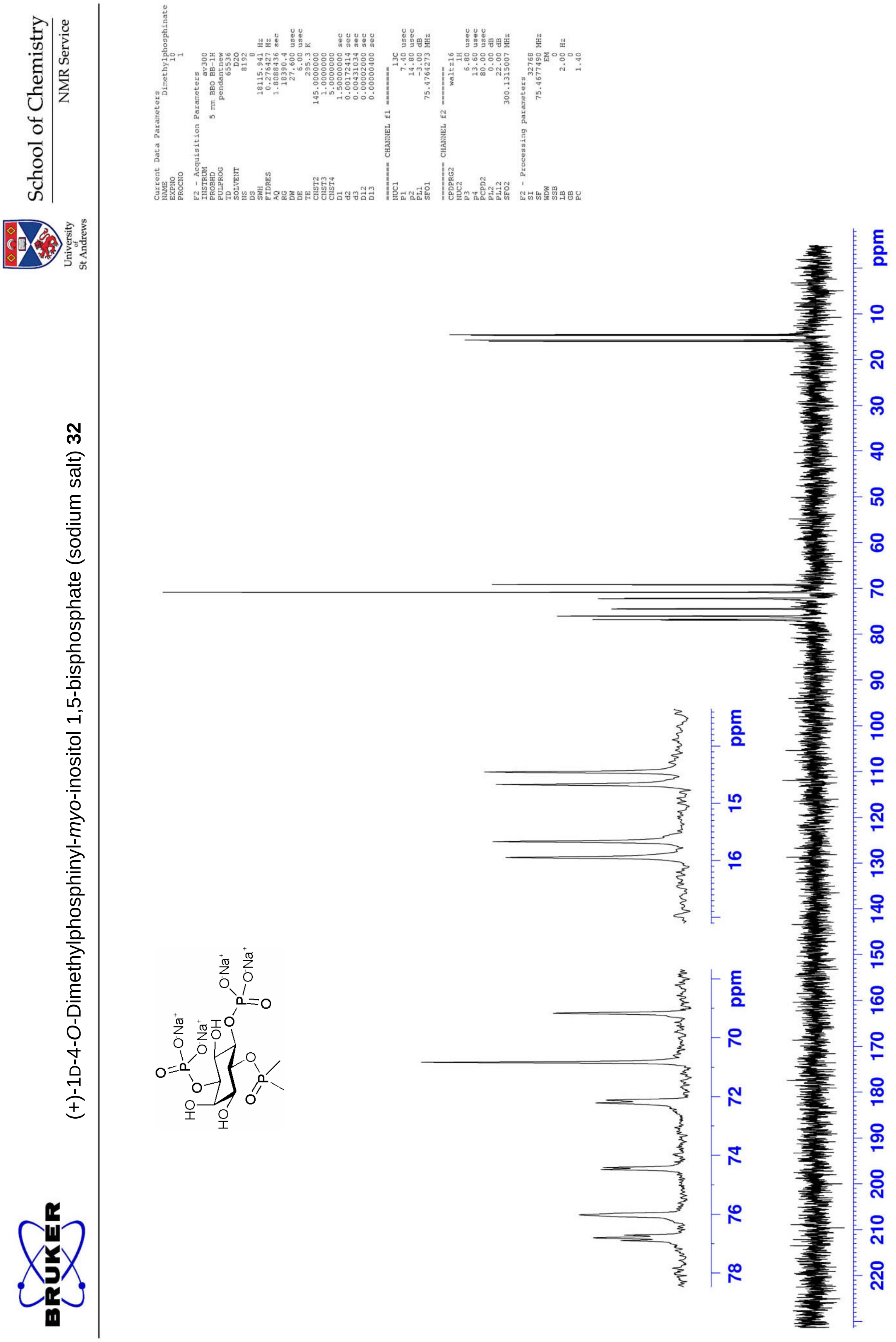
(124) D.-S. Wang and C.-S. Chen, *J. Org. Chem.*, 1996, **61**, 5905-5910.

(125) A. Dahlén, A. Sundgren, M. Lahmann, S. Oscarson, and O. Hilmersson, *Org. Lett.*, 2003, **5**, 4085-4088.
(126) J. Gigg and R. Gigg, *J. Chem. Soc. C.*, 1966, 82-86.
(127) G. J. Boons, A. Burton, and S. Isles, *Chem. Comm.*, 1996, 141-142.
(128) R. Ramage, B. Atrash, D. Hopton, and M. J. Parrott, *J. Chem. Soc. Perkin Trans. 1*, 1985, 1217-1226.
(129) P. J. Christen and L. M. van der Linde, *Rec. Trav. Chim. Pays-Bas.*, 1959, **78**, 543-550.
(130) T. Wada, R. Kato, and T. Hata, *J. Org. Chem.*, 1991, **56**, 1243-1250.
(131) I. F. Pickersgill, P. G. Devitt, and T. P. Kee, *Synth. Comm.*, 1993, **23**, 1643-1650.
(132) C. J. Cobley, K. Gardner, J. Klosin, C. Praquin, C. Hill, G. T. Whiteker, A. Zanotti-Gerosa, J. L. Petersen, and K. A. Abboud, *J. Org. Chem.*, 2004, **69**, 4031-4040.
(133) W. L. F. Armarego and C. L. L. Chai, *Purification of laboratory chemicals*, Butterworth Heinemann, 2003.
(134) H. W. Lee and Y. Kishi, *J. Org. Chem.*, 1985, **50**, 4402-4404.
(135) G. F. Painter, S. J. A. Grove, I. H. Gilbert, A. B. Holmes, P. R. Raithby, M. L. Hill, P. T. Hawkins, and L. R. Stephens, *J. Chem. Soc., Perkin Trans. 1*, 1999, **8**, 923-935.
(136) K. B. Wiberg and W. C. Cunningham, *J. Org. Chem.*, 1990, **55(2)**, 679-684.
(137) J. T. Gupton, S. J. Duranceau, J. F. Miller, and M. L. Kosiba, *Synth. Comm.*, 1988, **18**, 937-947.
(138) T. Tanaka, S. Tamatsukuri, and M. Ikehara, *Tetrahedron Lett.*, 1986, 199-202.
(139) N. Nakajima, K. Horita, R. Abe, and O. Yonemitsu, *Tetrahedron Lett.*, 1988, **29**, 4139-4142.
(140) K. A. Pollart and H. J. Hardwood, *J. Org. Chem.*, 1962, **27**, 4444-4447.
(141) F. Calvani, P. Crotti, C. Gardelli, and M. Pineschi, *Tetrahedron*, 1994, **50**, 12999-13022.
(142) A. E. Lippman, *J. Org. Chem.*, 1965, **30**, 3217-3218.
(143) G. M. Kosolapoff, *J. Am. Chem. Soc.*, 1949, **71**, 369-370.
(144) J. A. Rilatt and W. Kitching, *Organometallics*, 1982, **1**, 1089-1093.
(145) P. Westerduin, H. A. M. Willems, and C. A. A. van Boeckel, *Tetrahedron Lett.*, 1990, **31**, 6915-6918.

Appendix 1

Appendix 1 - Selected NMR Spectra



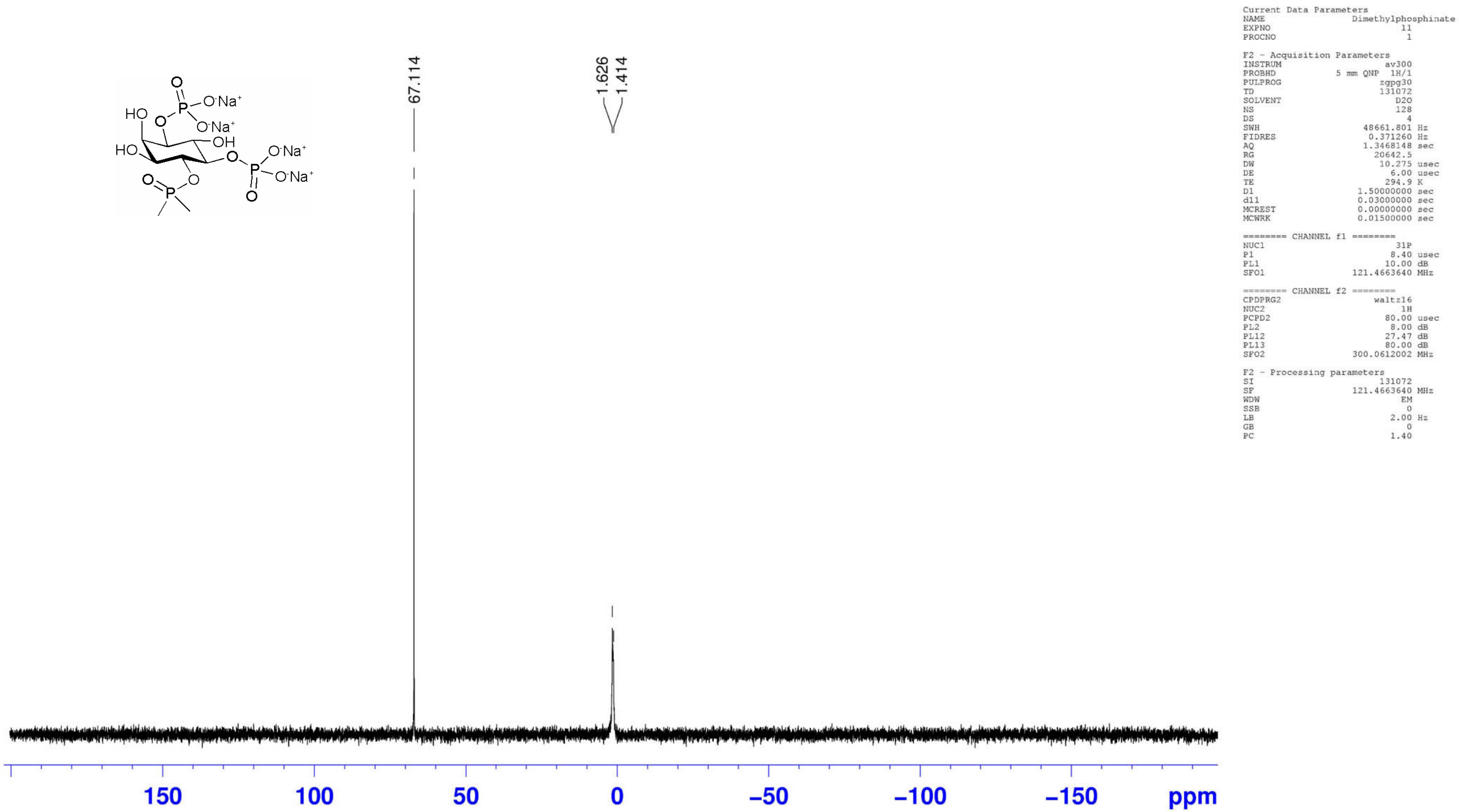


(+)-1D-4-*O*-Dimethylphosphinyl-*myo*-inositol 1,5-bisphosphate (sodium salt) **32**

BRUKER

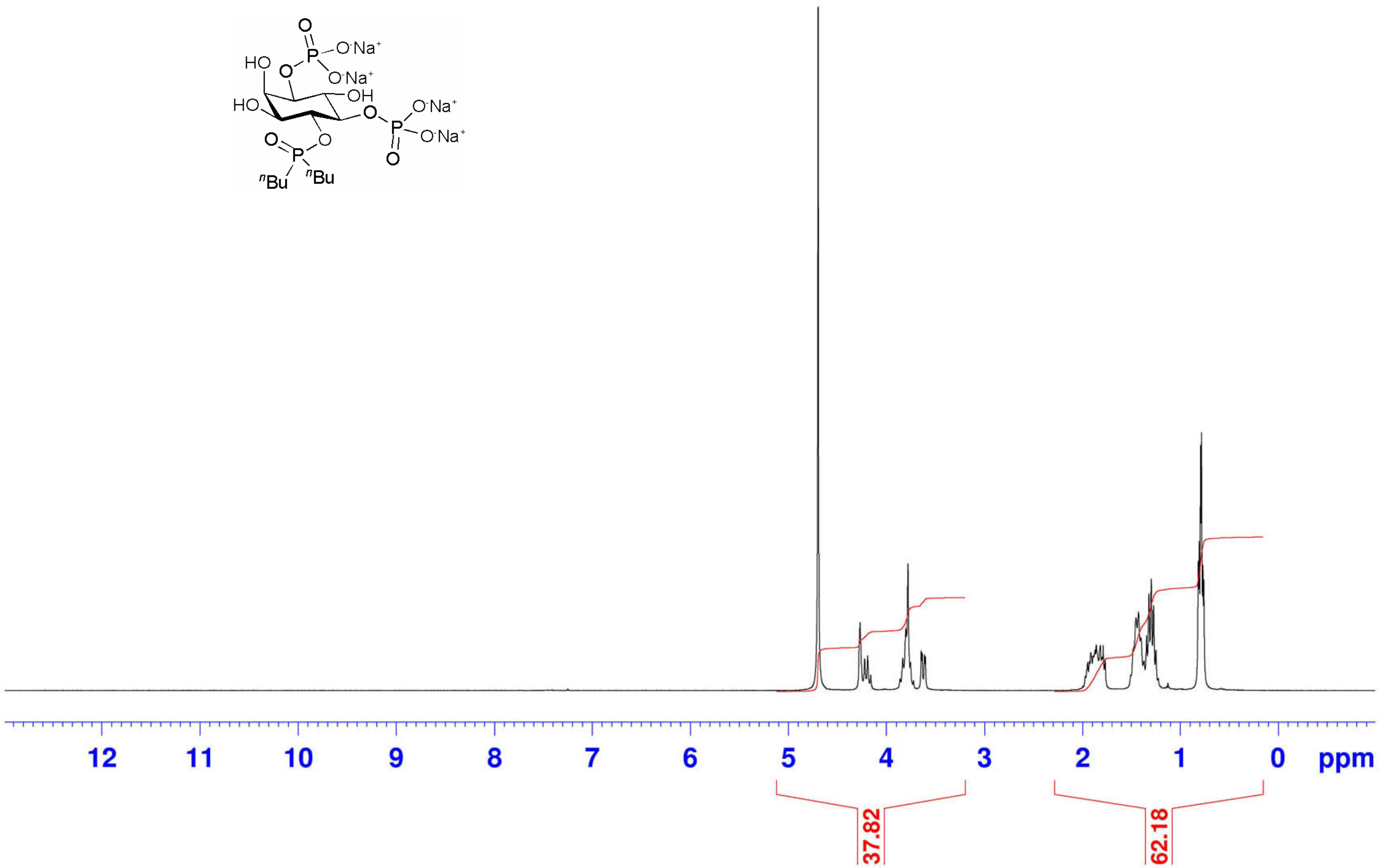
School of Chemistry
NMR Service
University of St Andrews

(+)-1D-4-*O*-Dimethylphosphinyl-*myo*-inositol 1,5-bisphosphate (sodium salt) **32**



(-)-1D-4-*O*-Di-*n*-butylphosphinyl-*myo*-inositol 1,5-bisphosphate (sodium salt) **123**

School of Chemistry
NMR Service



Current Data Parameters
NAME Dibutyl
EXPNO 10
PROCNO 1

F2 - Acquisition Parameters
INSTRUM av300
PROBHD 5 mm BBO BB-1H
PULPROG zg30
TD 32768
SOLVENT D2O
NS 16
DS 2
SWH 4194.631 Hz
FIDRES 0.128010 Hz
AQ 3.9059956 sec
RG 362
DW 119.200 usec
DE 6.00 usec
TE 295.5 K
D1 1.00000000 sec
MCREST 0.00000000 sec
MCWRK 0.01500000 sec

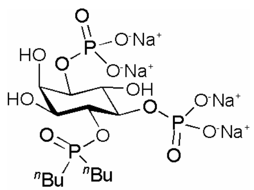
======== CHANNEL f1 ========
NUC1 1H
P1 7.10 usec
PL1 0.00 dB
SFO1 300.1318008 MHz

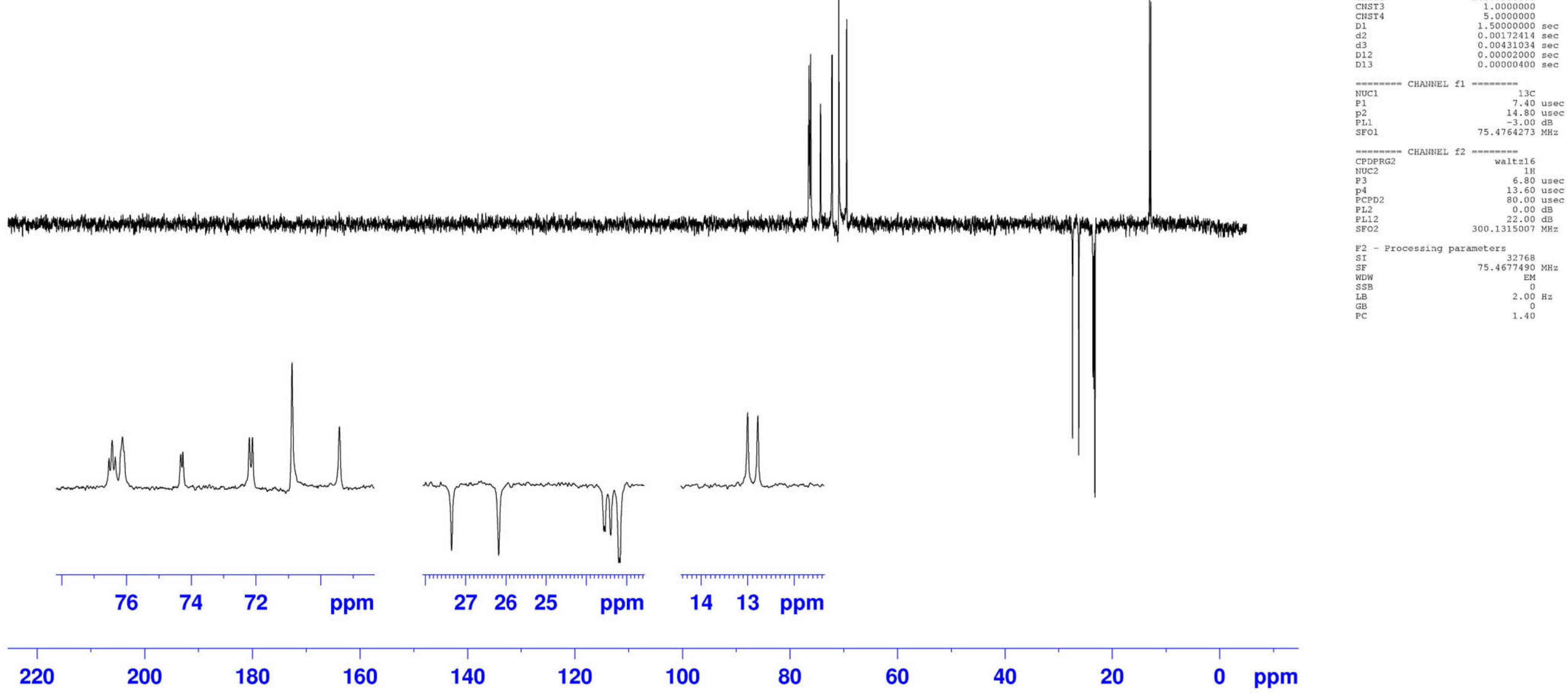
F2 - Processing parameters
SI 32768
SF 300.1300000 MHz
WDW EM
SSB 0
LB 0.30 Hz
GB 0
PC 1.00

BRUKER

School of Chemistry
NMR Service
University of St Andrews

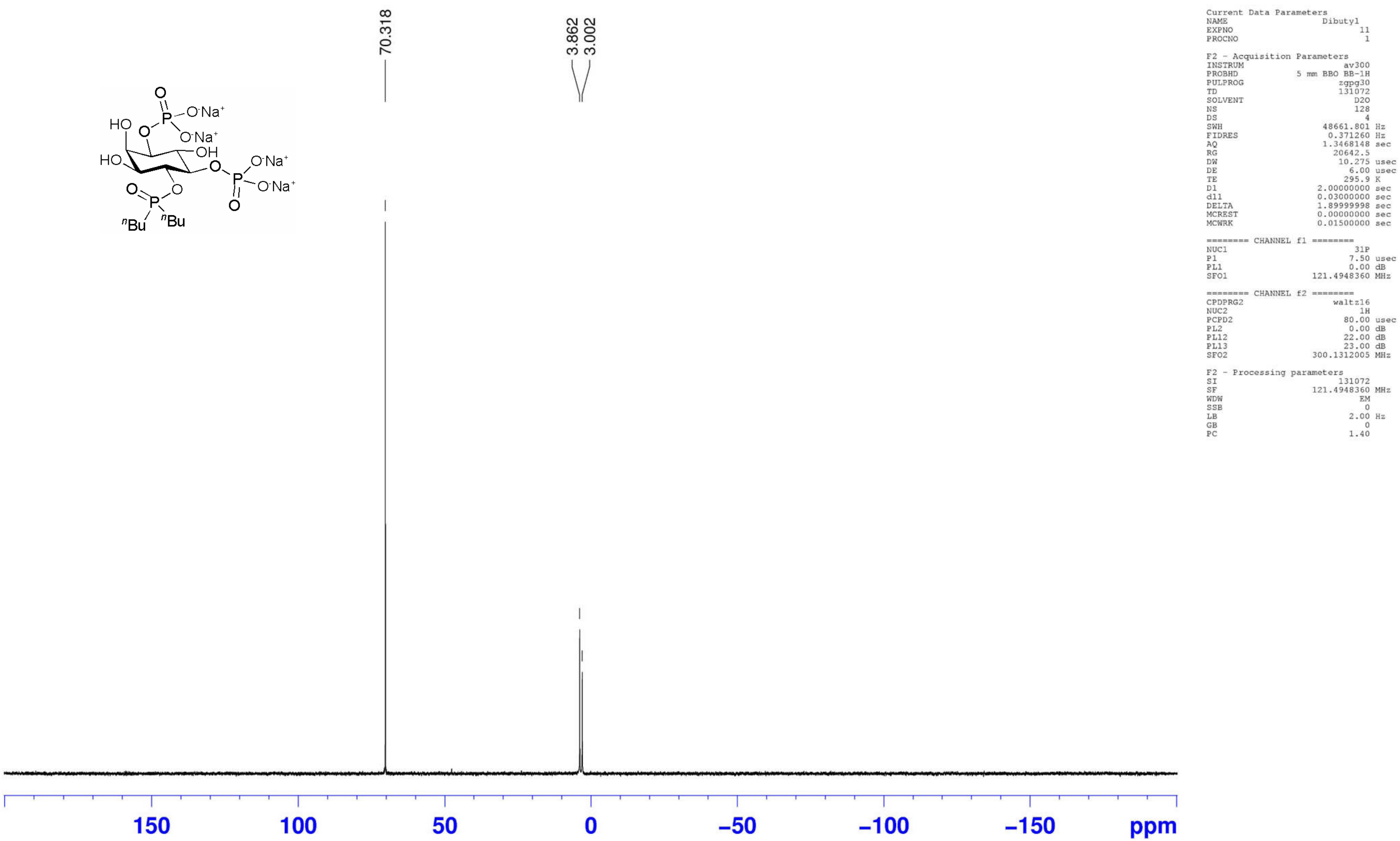
(-)-1D-4-*O*-Di-*n*-butylphosphinyl-*myo*-inositol 1,5-bisphosphate (sodium salt) **123**





(-)-1D-4-*O*-Di-*n*-butylphosphinyl-*myo*-inositol 1,5-bisphosphate (sodium salt) **123**

School of Chemistry
NMR Service

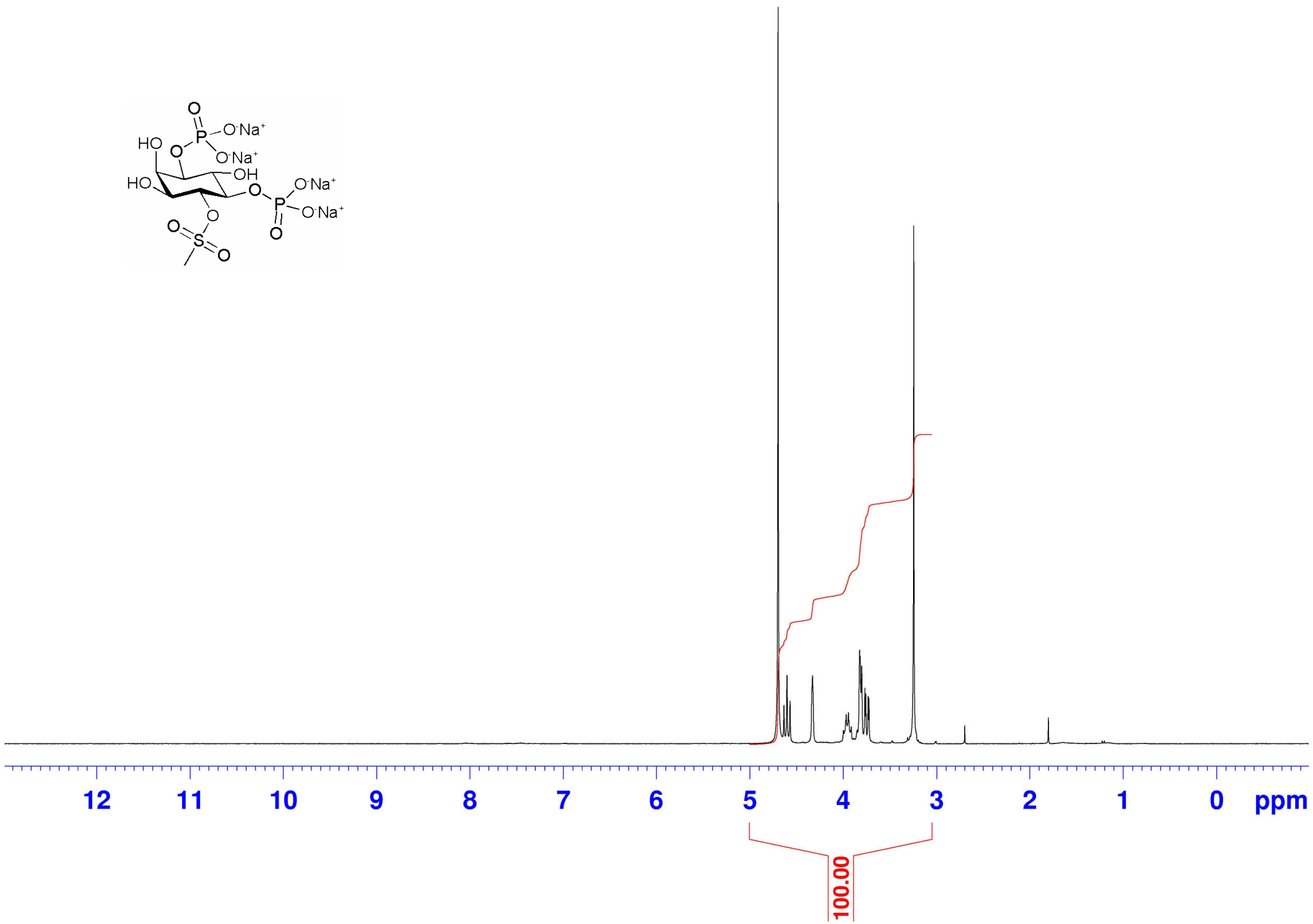


School of Chemistry
NMR Service

University of St Andrews

BRUKER

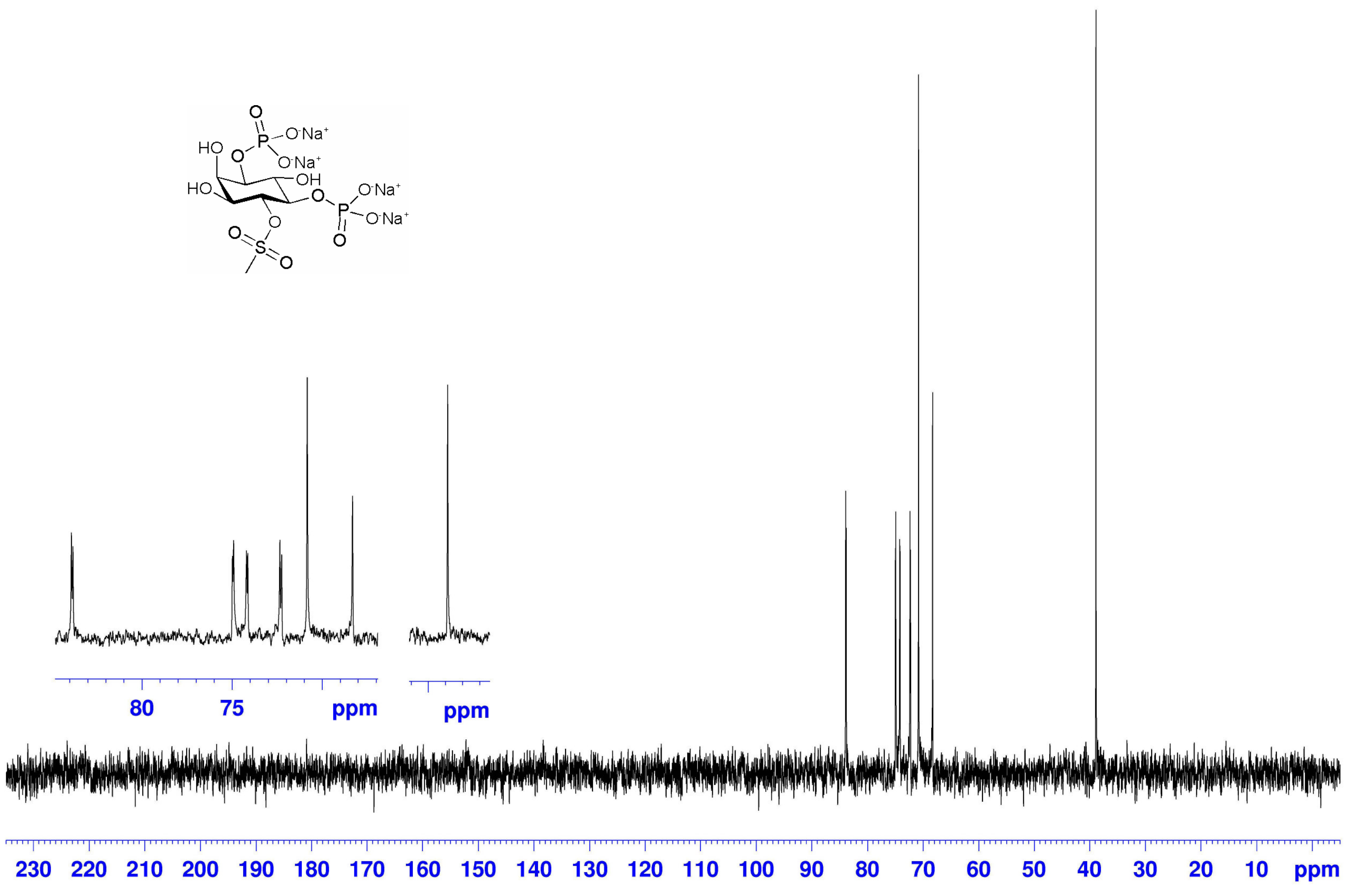
(+)-1D-4-*O*-Methylsulfonyl-*myo*-inositol 1,5-bisphosphate (sodium salt) **124**




```
Current Data Parameters
NAME                 Mesyl
EXPNO                   10
PROCNO                   1

F2 - Acquisition Parameters
INSTRUM              av300
PROBHD     5 mm BBO BB-1H
PULPROG               zg30
TD                   32768
SOLVENT                D2O
NS                      16
DS                       2
SWH               4194.631 Hz
FIDRES            0.128010 Hz
AQ               3.9059956 sec
RG                   724.1
DW                 119.200 usec
DE                    6.00 usec
TE                   295.5 K
D1             1.00000000 sec
MCREST         0.00000000 sec
MCWRK          0.01500000 sec

======== CHANNEL f1 ========
NUC1                    1H
P1                    7.10 usec
PL1                   0.00 dB
SFO1           300.1318008 MHz

F2 - Processing parameters
SI                   32768
SF             300.1300000 MHz
WDW                     EM
SSB                      0
LB                    0.30 Hz
GB                       0
PC                    1.00
```

(+)-1D-4-*O*-Methylsulfonyl-*myo*-inositol 1,5-bisphosphate (sodium salt) **124**

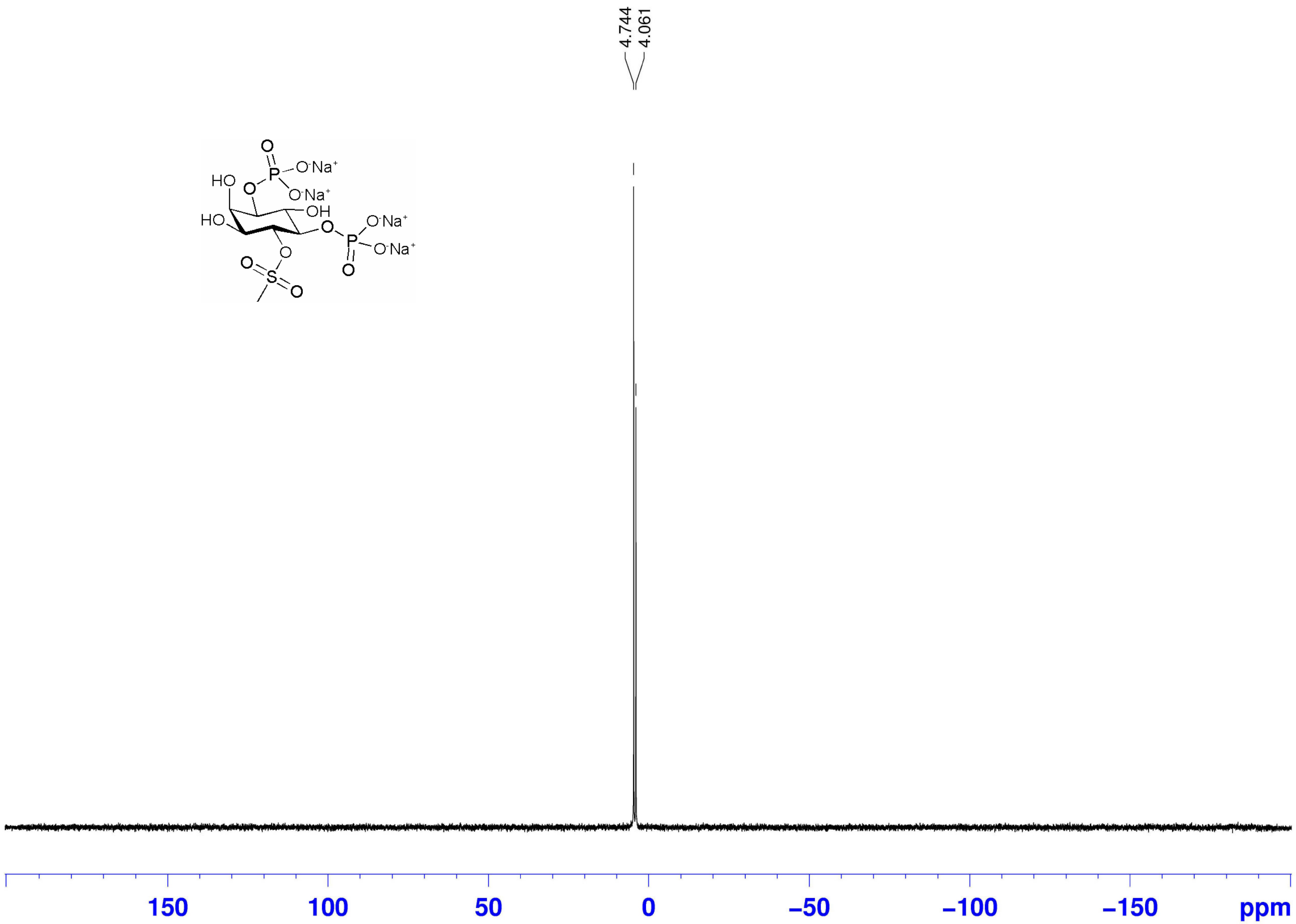
School of Chemistry
NMR Service
University of St Andrews




```
Current Data Parameters
NAME                Mesyl
EXPNO                  11
PROCNO                  1

F2 - Acquisition Parameters
INSTRUM             av300
PROBHD      5 mm BBO BB-1H
PULPROG        pendantnew
TD                  65536
SOLVENT               D2O
NS                   3072
DS                      8
SWH             18115.941 Hz
FIDRES           0.276427 Hz
AQ              1.8088436 sec
RG                18390.4
DW                 27.600 usec
DE                   6.00 usec
TE                  295.9 K
CNST2         145.0000000
CNST3           1.0000000
CNST4           5.0000000
D1             1.50000000 sec
d2             0.00172414 sec
d3             0.00431034 sec
D12            0.00002000 sec
D13            0.00000400 sec

======== CHANNEL f1 ========
NUC1                  13C
P1                   7.40 usec
p2                  14.80 usec
PL1                 -3.00 dB
SFO1           75.4764273 MHz

======== CHANNEL f2 ========
CPDPRG2           waltz16
NUC2                   1H
P3                   6.80 usec
p4                  13.60 usec
PCPD2               80.00 usec
PL2                  0.00 dB
PL12                22.00 dB
SFO2          300.1315007 MHz

F2 - Processing parameters
SI                  32768
SF             75.4677490 MHz
WDW                    EM
SSB                     0
LB                   2.00 Hz
GB                      0
PC                   1.40
```

School of Chemistry
NMR Service

(+)-1D-4-*O*-Methylsulfonyl-*myo*-inositol 1,5-bisphosphate (sodium salt) **124**

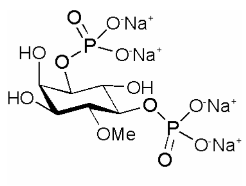


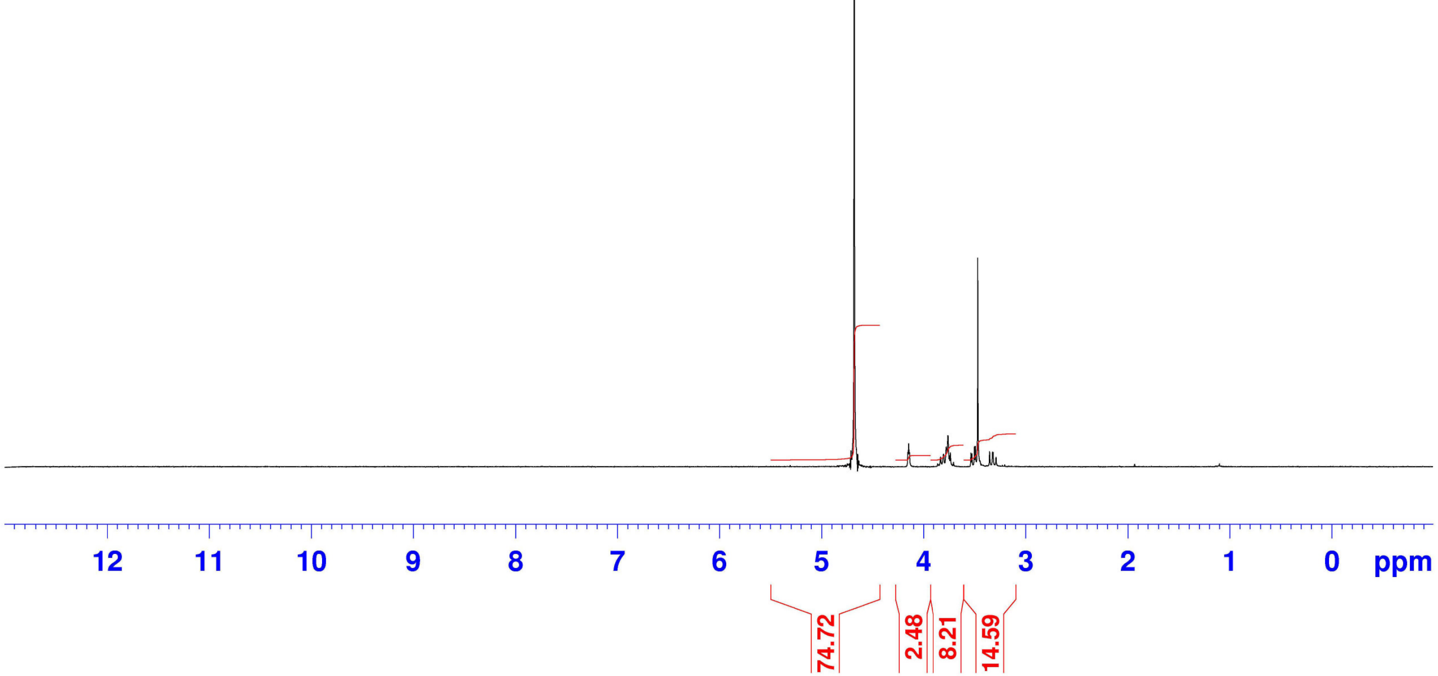

```
Current Data Parameters
NAME              Mesyl
EXPNO                11
PROCNO                1

F2 - Acquisition Parameters
INSTRUM           av300
PROBHD     5 mm QNP  1H/1
PULPROG         zgpg30
TD               131072
SOLVENT             D2O
NS                  128
DS                    4
SWH           48661.801 Hz
FIDRES         0.371260 Hz
AQ            1.3468148 sec
RG              20642.5
DW               10.275 usec
DE                 6.00 usec
TE                295.2 K
D1           1.50000000 sec
d11          0.03000000 sec
MCREST       0.00000000 sec
MCWRK        0.01500000 sec

======== CHANNEL f1 ========
NUC1                31P
P1                 8.40 usec
PL1               10.00 dB
SFO1        121.4663640 MHz

======== CHANNEL f2 ========
CPDPRG2         waltz16
NUC2                 1H
PCPD2             80.00 usec
PL2                8.00 dB
PL12              27.47 dB
PL13              80.00 dB
SFO2        300.0612002 MHz

F2 - Processing parameters
SI               131072
SF          121.4663640 MHz
WDW                  EM
SSB                   0
LB                 2.00 Hz
GB                    0
PC                 1.40
```

BRUKER

School of Chemistry
NMR Service
University of St Andrews

(-)-1D-4-*O*-Methyl-*myo*-inositol 1,5-bisphosphate (sodium salt) **109**

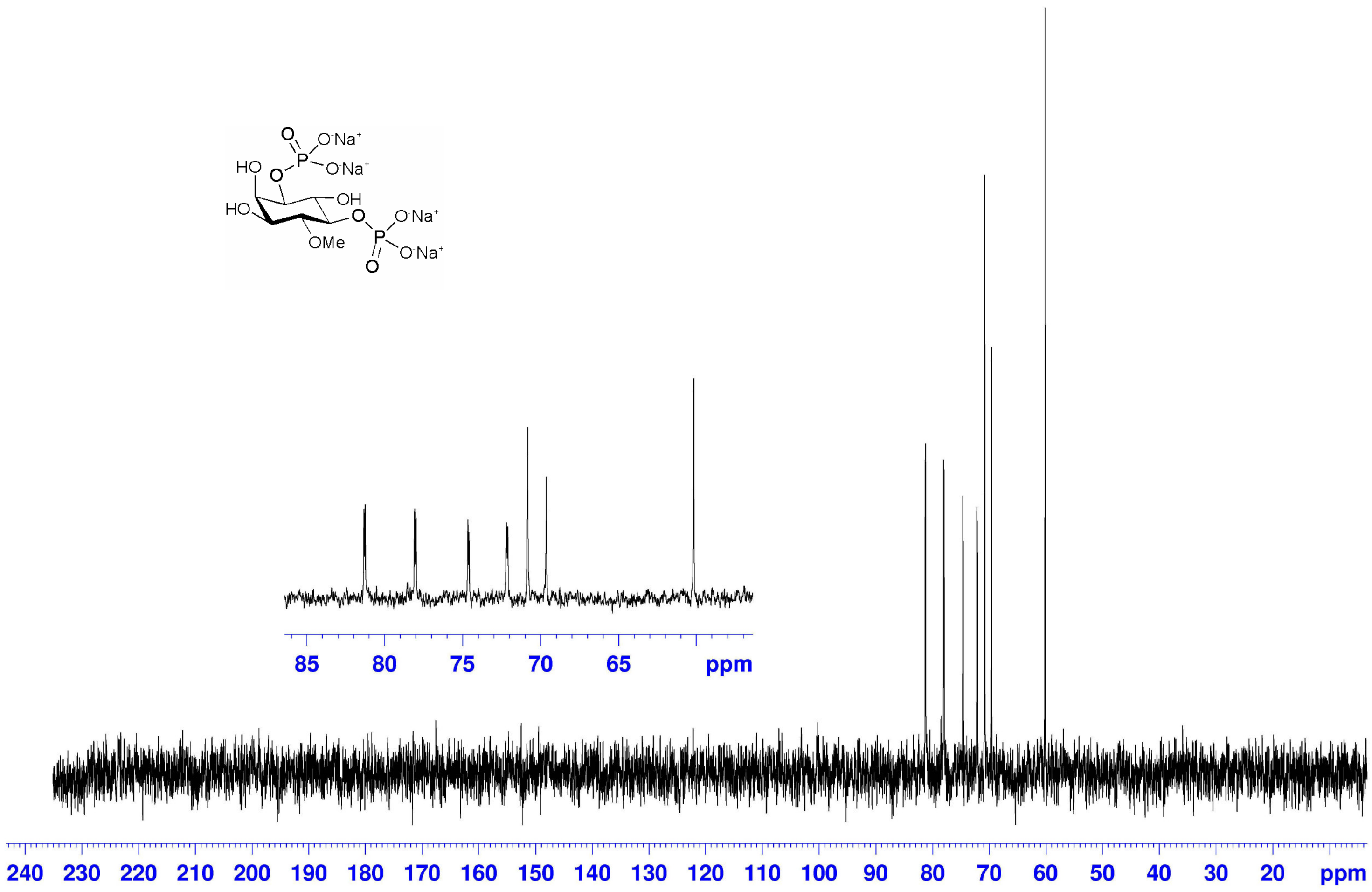





```
Current Data Parameters
NAME                 Methyl
EXPNO                    10
PROCNO                    1

F2 - Acquisition Parameters
INSTRUM               av300
PROBHD      5 mm QNP  1H/1
PULPROG                zg30
TD                    32768
SOLVENT                 D2O
NS                       16
DS                        2
SWH                4194.631 Hz
FIDRES             0.128010 Hz
AQ                3.9059956 sec
RG                    912.3
DW                  119.200 usec
DE                     6.00 usec
TE                    294.4 K
D1               1.00000000 sec
MCREST           0.00000000 sec
MCWRK            0.01500000 sec

======== CHANNEL f1 ========
NUC1                     1H
P1                     8.50 usec
PL1                    8.00 dB
SFO1           300.0618004 MHz

F2 - Processing parameters
SI                    32768
SF             300.0600000 MHz
WDW                      EM
SSB                       0
LB                     0.30 Hz
GB                        0
PC                     1.00
```

School of Chemistry
NMR Service

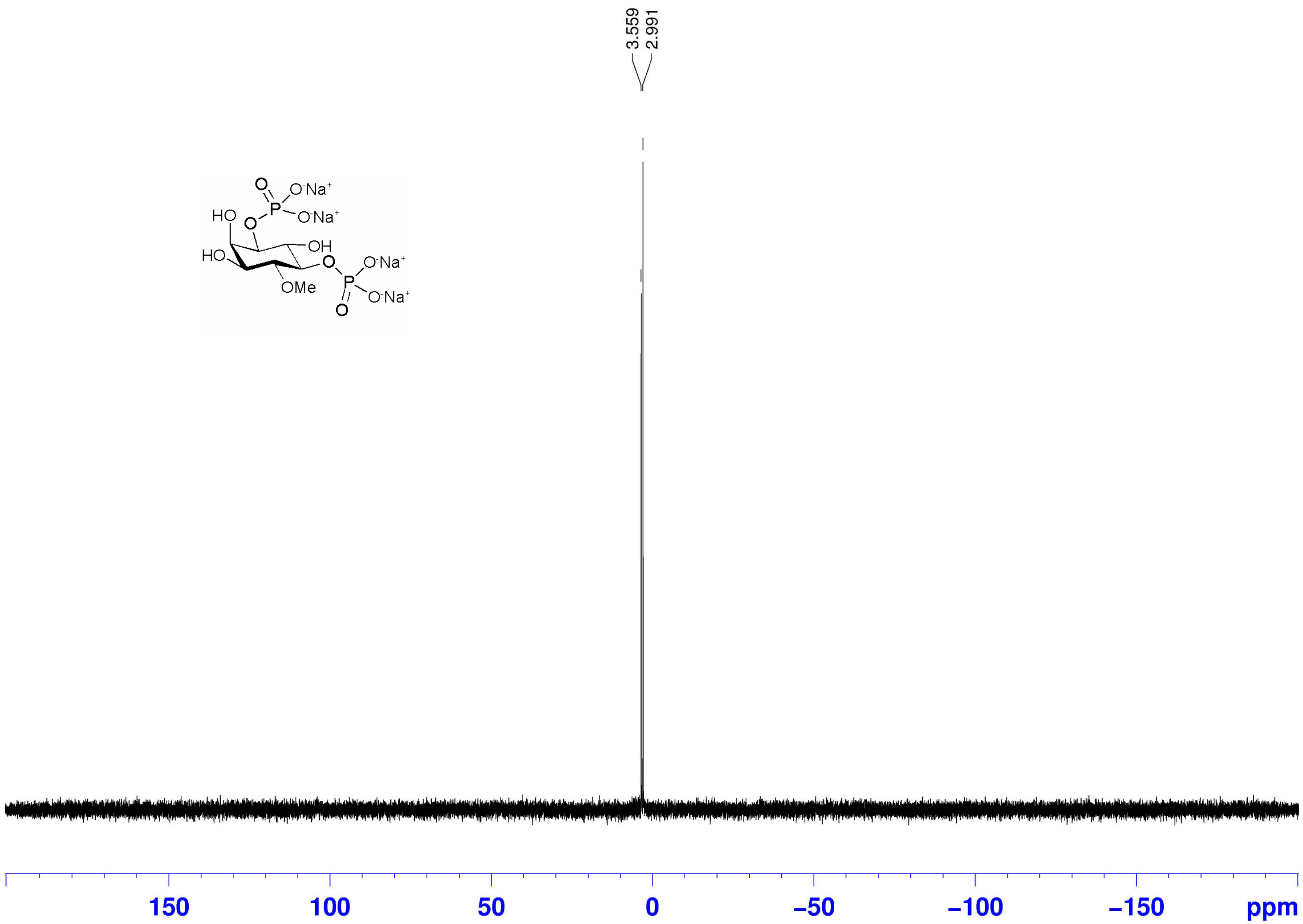
(-)-1D-4-*O*-Methyl-*myo*-inositol 1,5-bisphosphate (sodium salt) **109**




```
Current Data Parameters
NAME                 Methyl
EXPNO                    10
PROCNO                    1

F2 - Acquisition Parameters
INSTRUM               av300
PROBHD         5 mm BBO BB-1H
PULPROG          pendantnew
TD                    65536
SOLVENT                 D2O
NS                     6000
DS                        8
SWH              18115.941 Hz
FIDRES            0.276427 Hz
AQ               1.8088436 sec
RG                  18390.4
DW                   27.600 usec
DE                     6.00 usec
TE                    295.5 K
CNST2          145.0000000
CNST3            1.0000000
CNST4            5.0000000
D1              1.50000000 sec
d2              0.00172414 sec
d3              0.00431034 sec
D12             0.00002000 sec
D13             0.00000400 sec

======== CHANNEL f1 ========
NUC1                    13C
P1                     7.40 usec
p2                    14.80 usec
PL1                   -3.00 dB
SFO1             75.4764273 MHz

======== CHANNEL f2 ========
CPDPRG2             waltz16
NUC2                     1H
P3                     6.80 usec
p4                    13.60 usec
PCPD2                 80.00 usec
PL2                    0.00 dB
PL12                  22.00 dB
SFO2            300.1315007 MHz

F2 - Processing parameters
SI                    32768
SF               75.4677490 MHz
WDW                      EM
SSB                       0
LB                     2.00 Hz
GB                        0
PC                     1.40
```

School of Chemistry
NMR Service
University of St Andrews

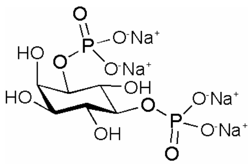
(-)-1D-4-*O*-Methyl-*myo*-inositol 1,5-bisphosphate (sodium salt) **109**

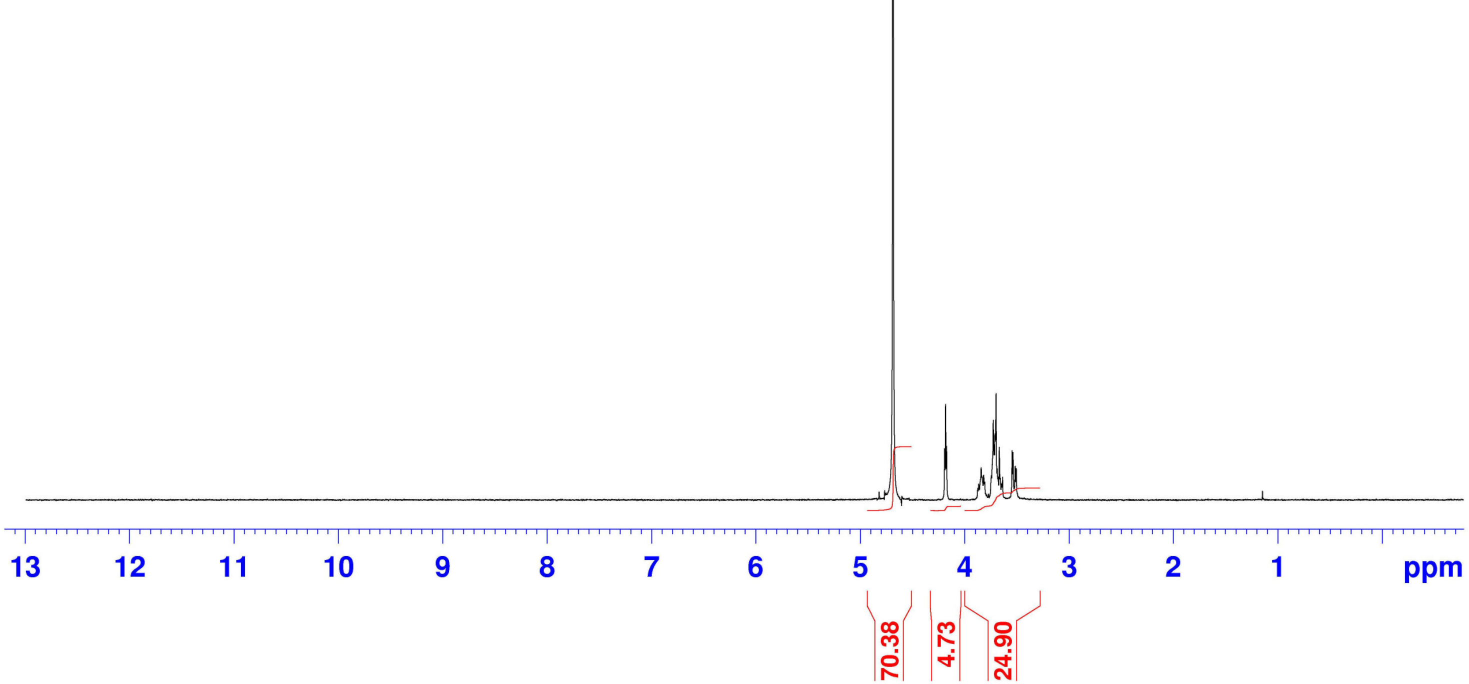



```
Current Data Parameters
NAME                  Methyl
EXPNO                     11
PROCNO                     1

F2 - Acquisition Parameters
INSTRUM                av300
PROBHD        5 mm QNP  1H/1
PULPROG               zgpg30
TD                    131072
SOLVENT                  D2O
NS                       128
DS                         4
SWH               48661.801 Hz
FIDRES             0.371260 Hz
AQ                1.3468148 sec
RG                   20642.5
DW                    10.275 usec
DE                      6.00 usec
TE                     294.6 K
D1                2.00000000 sec
d11               0.03000000 sec
MCREST            0.00000000 sec
MCWRK             0.01500000 sec

======== CHANNEL f1 ========
NUC1                     31P
P1                      8.40 usec
PL1                    10.00 dB
SFO1             121.4663640 MHz

======== CHANNEL f2 ========
CPDPRG2              waltz16
NUC2                      1H
PCPD2                  80.00 usec
PL2                     8.00 dB
PL12                   27.47 dB
PL13                   80.00 dB
SFO2             300.0612002 MHz

F2 - Processing parameters
SI                    131072
SF               121.4663640 MHz
WDW                       EM
SSB                        0
LB                      1.00 Hz
GB                         0
PC                      1.40
```

School of Chemistry
NMR Service

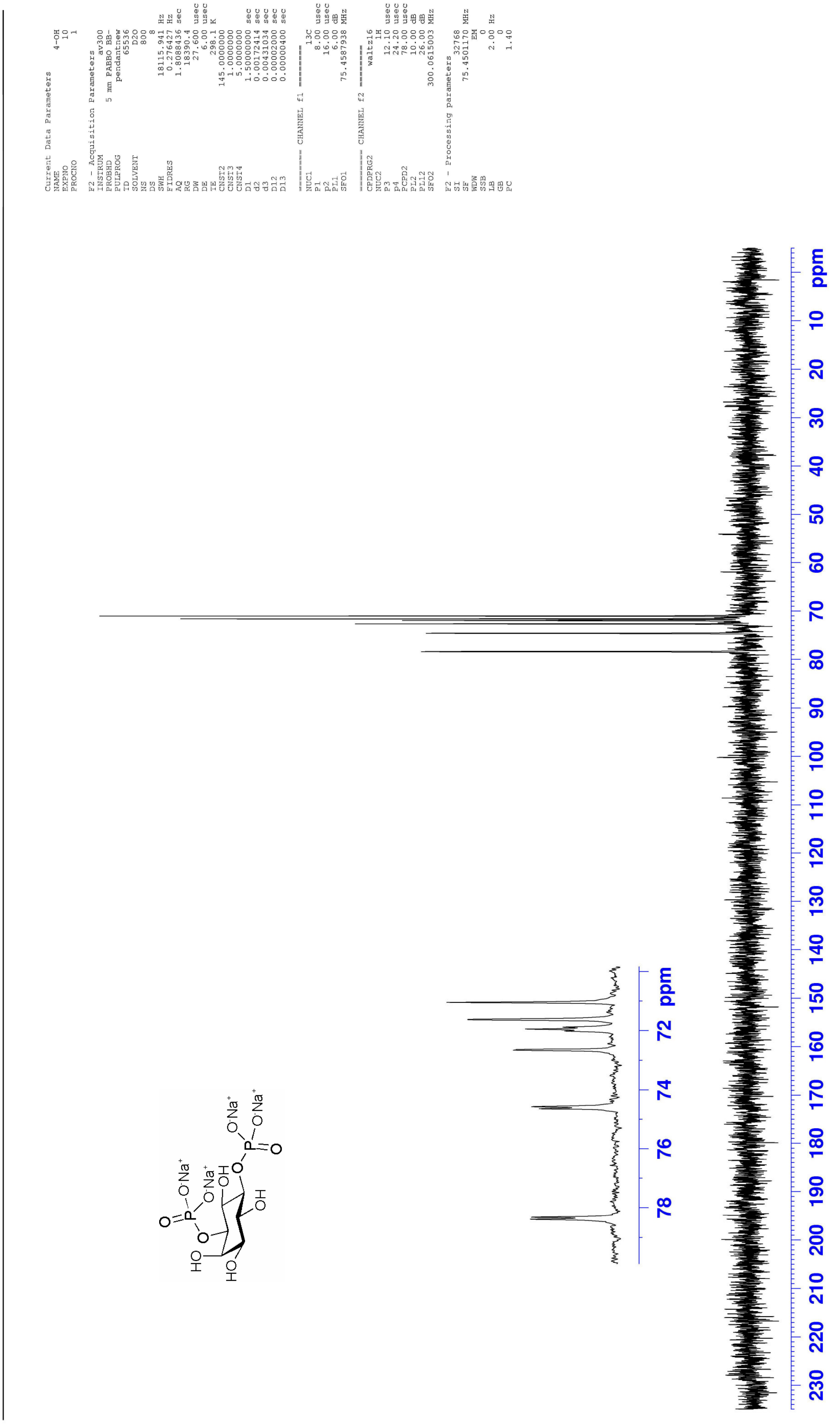
(+)-1D-*myo*-inositol 1,5-bisphosphate (sodium salt) **125**





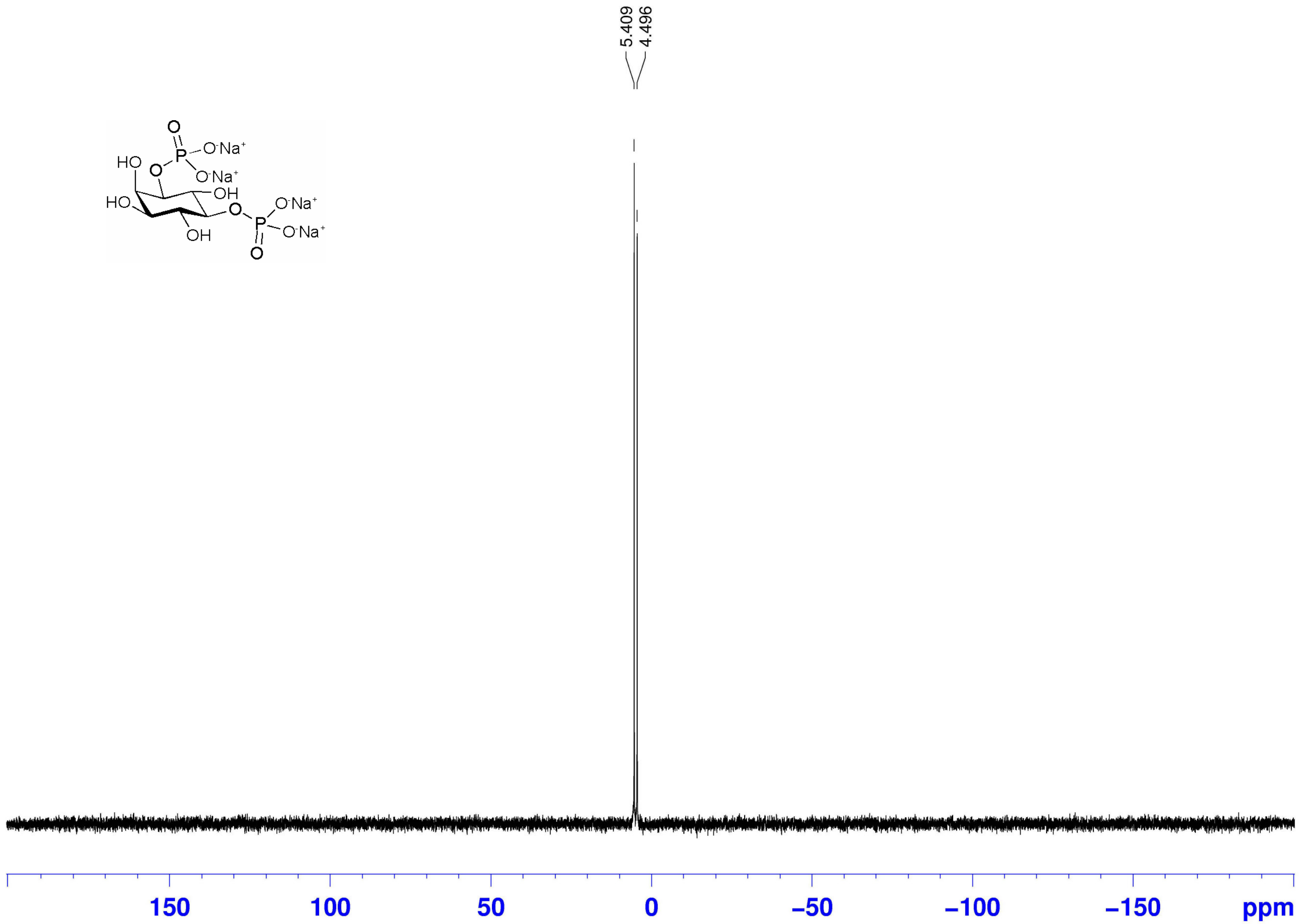

```
Current Data Parameters
NAME                4-OH
EXPNO                 10
PROCNO                 1

F2 - Acquisition Parameters
INSTRUM            av300
PROBHD    5 mm PABBO BB-
PULPROG             zg30
TD                 32768
SOLVENT              D2O
NS                    16
DS                     2
SWH             4194.631 Hz
FIDRES          0.128010 Hz
AQ             3.9059956 sec
RG                   512
DW               119.200 usec
DE                  6.00 usec
TE                 298.2 K
D1            1.00000000 sec
TD0                    1

======== CHANNEL f1 ========
NUC1                  1H
P1                 12.30 usec
PL1                10.00 dB
SFO1         300.0618004 MHz

F2 - Processing parameters
SI                 32768
SF           300.0600000 MHz
WDW                   EM
SSB                    0
LB                  0.30 Hz
GB                     0
PC                  1.00
```

(+)-1D-*myo*-inositol 1,5-bisphosphate (sodium salt) **125**



(+)-1D-*myo*-inositol 1,5-bisphosphate (sodium salt) **125**

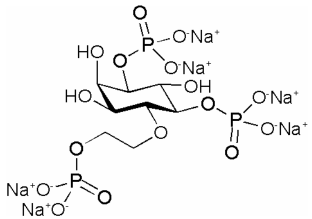
School of Chemistry
NMR Service
University of St Andrews

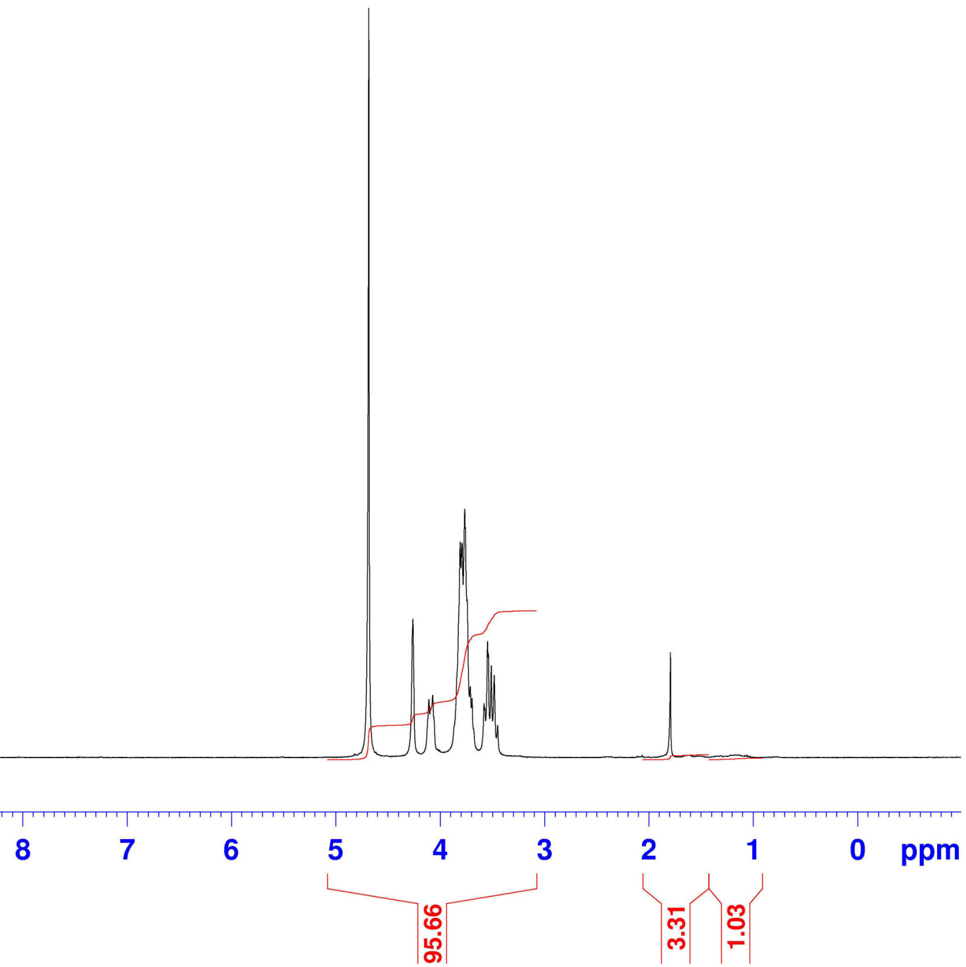



```
Current Data Parameters
NAME                  4-OH
EXPNO                   11
PROCNO                   1

F2 - Acquisition Parameters
INSTRUM              av300
PROBHD        5 mm PABBO BB-
PULPROG             zgpg30
TD                  131072
SOLVENT                D2O
NS                     128
DS                       4
SWH              48661.801 Hz
FIDRES            0.371260 Hz
AQ               1.3468148 sec
RG                 20642.5
DW                  10.275 usec
DE                    6.00 usec
TE                   298.1 K
D1              1.50000000 sec
d11             0.03000000 sec
DELTA           1.39999998 sec
TD0                      1

======== CHANNEL f1 ========
NUC1                   31P
P1                    9.70 usec
PL1                  15.00 dB
SFO1           121.4663640 MHz

======== CHANNEL f2 ========
CPDPRG2            waltz16
NUC2                    1H
PCPD2                78.00 usec
PL2                  10.00 dB
PL12                 26.00 dB
PL13                 27.00 dB
SFO2           300.0612002 MHz

F2 - Processing parameters
SI                  131072
SF             121.4663640 MHz
WDW                     EM
SSB                      0
LB                    2.00 Hz
GB                       0
PC                    1.40
```

School of Chemistry
NMR Service

(-)-1D-4-*O*-(2-Phosphoryloxy)ethyl-*myo*-inositol 1,5-bisphosphate (sodium salt) **126**





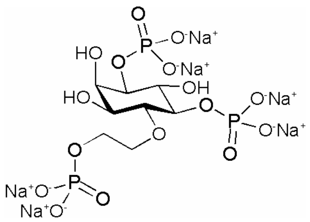

```
Current Data Parameters
NAME                    Ext
EXPNO                    10
PROCNO                    1

F2 - Acquisition Parameters
INSTRUM               av300
PROBHD       5 mm PABBO BB-
PULPROG                zg30
TD                    32768
SOLVENT                 D2O
NS                       16
DS                        2
SWH              4194.631 Hz
FIDRES           0.128010 Hz
AQ              3.9059956 sec
RG                      362
DW                119.200 usec
DE                  6.00 usec
TE                 298.2 K
D1            1.00000000 sec
TD0                       1

======== CHANNEL f1 ========
NUC1                     1H
P1                  12.30 usec
PL1                 10.00 dB
SFO1          300.0618004 MHz

F2 - Processing parameters
SI                    32768
SF           300.0600000 MHz
WDW                      EM
SSB                       0
LB                   0.30 Hz
GB                        0
PC                     1.00
```

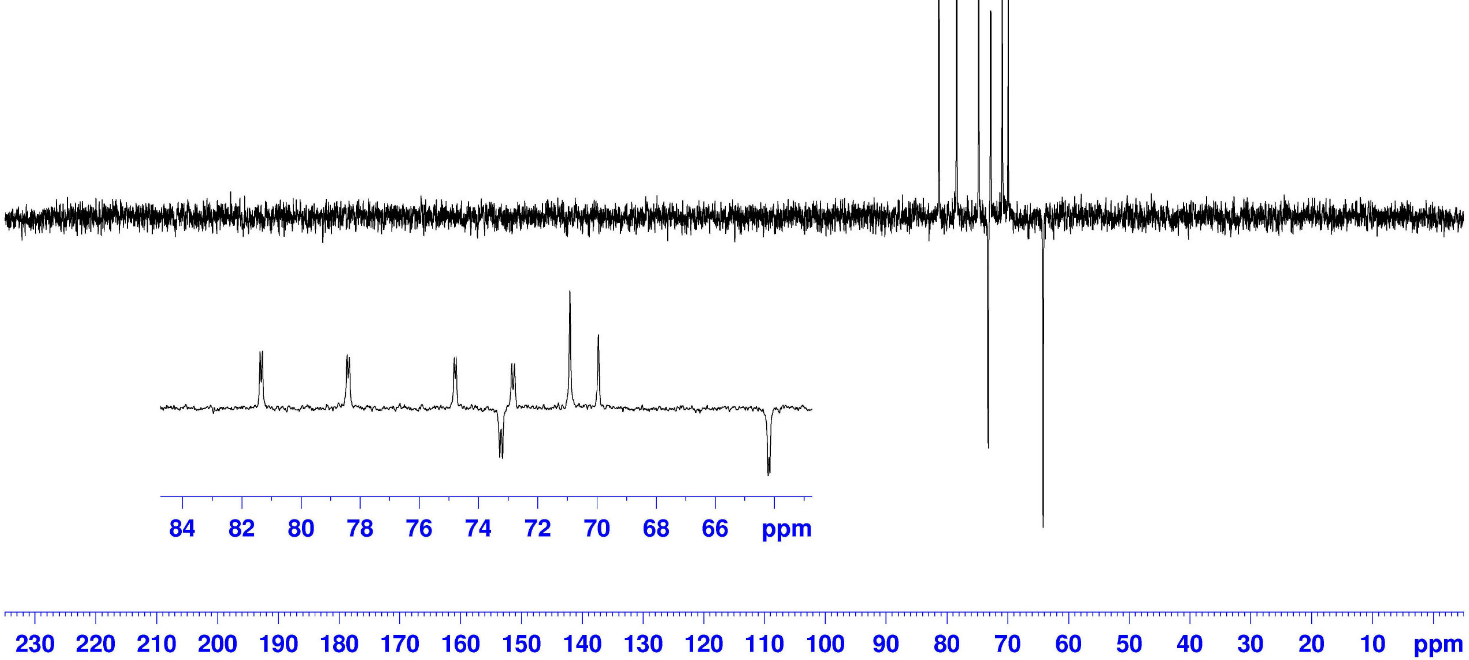
BRUKER

(-)-1D-4-*O*-(2-Phosphoryloxy)ethyl-*myo*-inositol 1,5-bisphosphate (sodium salt) **126**

University of St Andrews

School of Chemistry
NMR Service



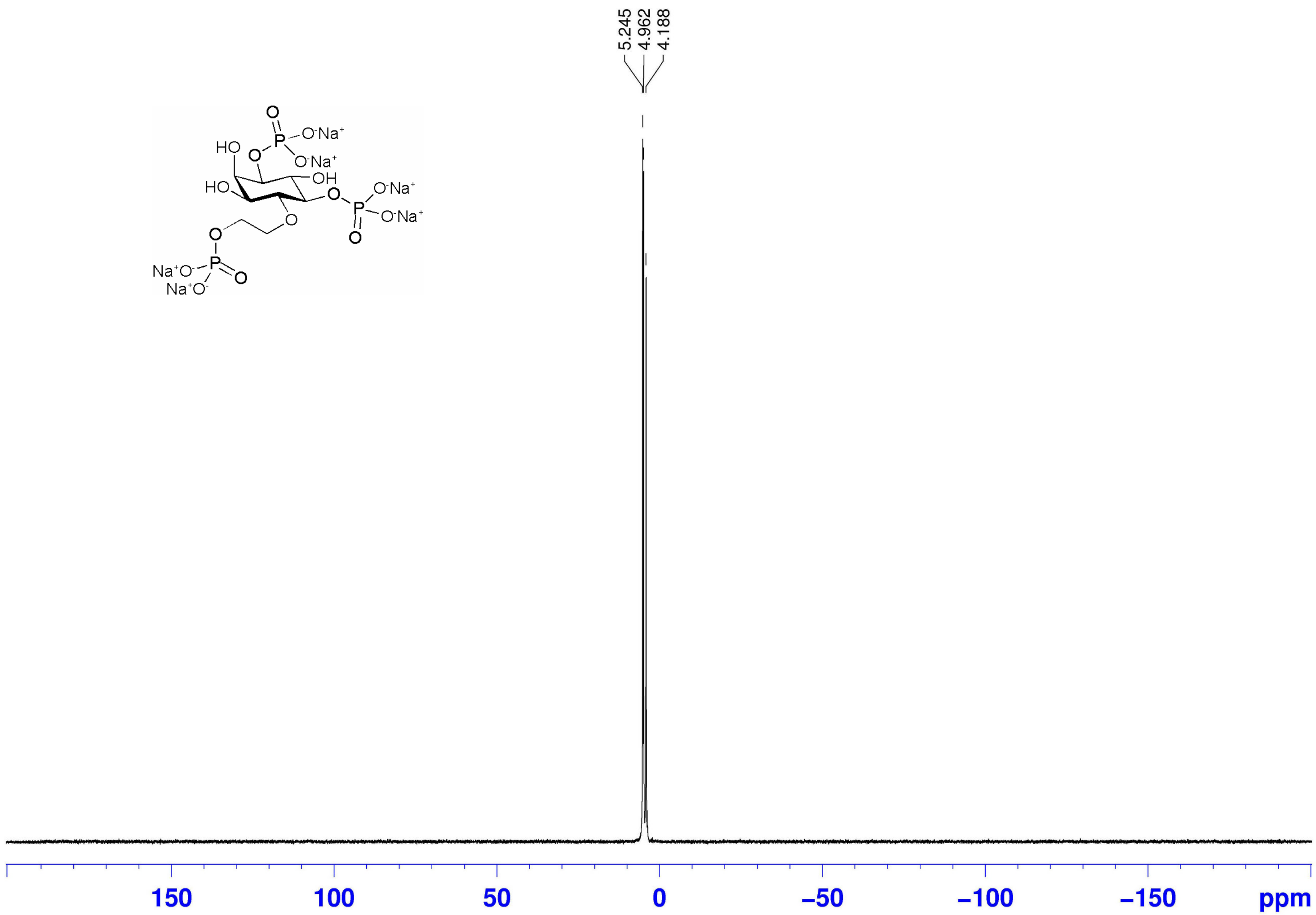


Current Data Parameters		
NAME	Ext	
EXPNO	10	
PROCNO	1	
F2 - Acquisition Parameters		
INSTRUM	av300	
PROBHD	5 mm PABBO BB-	
PULPROG	pendantnew	
TD	65536	
SOLVENT	D2O	
NS	800	
DS	8	
SWH	18115.941	Hz
FIDRES	0.276427	Hz
AQ	1.8088436	sec
RG	18390.4	
DW	27.600	usec
DE	6.00	usec
TE	298.2	K
CNST2	145.0000000	
CNST3	1.0000000	
CNST4	5.0000000	
D1	1.50000000	sec
d2	0.00172414	sec
d3	0.00431034	sec
D12	0.00002000	sec
D13	0.00000400	sec
======== CHANNEL f1 ========		
NUC1	13C	
P1	8.00	usec
p2	16.00	usec
PL1	6.00	dB
SFO1	75.4587938	MHz
======== CHANNEL f2 ========		
CPDPRG2	waltz16	
NUC2	1H	
P3	12.10	usec
p4	24.20	usec
PCPD2	78.00	usec
PL2	10.00	dB
PL12	26.00	dB
SFO2	300.0615003	MHz
F2 - Processing parameters		
SI	32768	
SF	75.4501170	MHz
WDW	EM	
SSB	0	
LB	2.00	Hz
GB	0	
PC	1.40	

School of Chemistry
NMR Service

University of St Andrews

(-)-1D-4-*O*-(2-Phosphoryloxy)ethyl-*myo*-inositol 1,5-bisphosphate (sodium salt) **126**

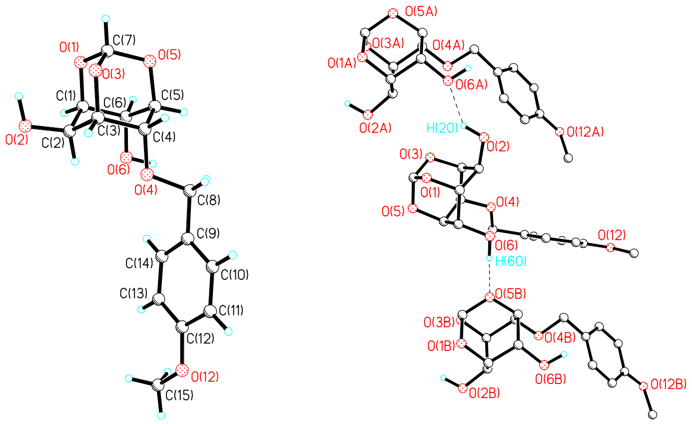



```
Current Data Parameters
NAME                    Ext
EXPNO                    11
PROCNO                    1

F2 - Acquisition Parameters
INSTRUM               av300
PROBHD        5 mm PABBO BB-
PULPROG             zgpg30
TD                  131072
SOLVENT                D2O
NS                     128
DS                       4
SWH              48661.801 Hz
FIDRES            0.371260 Hz
AQ               1.3468148 sec
RG                    8192
DW                  10.275 usec
DE                    6.00 usec
TE                   298.2 K
D1              1.50000000 sec
d11             0.03000000 sec
DELTA           1.39999998 sec
TD0                      1

======== CHANNEL f1 ========
NUC1                   31P
P1                    9.70 usec
PL1                  15.00 dB
SFO1           121.4663640 MHz

======== CHANNEL f2 ========
CPDPRG2            waltz16
NUC2                    1H
PCPD2                78.00 usec
PL2                  10.00 dB
PL12                 26.00 dB
PL13                 27.00 dB
SFO2           300.0612002 MHz

F2 - Processing parameters
SI                  131072
SF             121.4663640 MHz
WDW                     EM
SSB                      0
LB                    2.00 Hz
GB                       0
PC                    1.40
```

Appendix 2

Appendix 2 - Crystallographic Data

6-[(4'-Methoxy)benzyloxy]-2,4,10-trioxatricyclo[3.3.1.1^{3,7}]decane-8,9-diol 38



Crystal structure of compound **38.**

Crystal data and structure refinement for 38			
Empirical formula	$C_{15} H_{18} O_7$	Index ranges	-21<=h<=20, -9<=k<=9, -11<=l<=11
Formula weight	310.29	Reflections collected	8472
Temperature	125(2) K	Independent reflections	2487 [R(int) = 0.0478]
Wavelength	0.71073 Å	Completeness to theta = 25.38°	97.6 %
Crystal system	Monoclinic	Absorption correction	MULTISCAN
Space group	P2(1)/c	Max. and min. transmission	1.00000 and 0.889515
Unit cell dimensions	a = 17.782(5) Å α= 90° b = 8.040(2) Å β=93.521(5)° c = 9.693(3) Å γ = 90°	Refinement method	Full-matrix least-squares on F^2
Volume	1383.1(6) $Å^3$	Data / restraints / parameters	2487 / 2 / 209
Z	4	Goodness-of-fit on F^2	0.937
Density (calculated)	1.490 Mg/m^3	Final R indices [I>2sigma(I)]	R1 = 0.0389, wR2 = 0.0789
Absorption coefficient	0.119 mm^{-1}	R indices (all data)	R1 = 0.0777, wR2 = 0.0909
F(000)	656	Extinction coefficient	0.017(2)
Crystal size	.1 × .1 × .02 mm^3	Largest diff. peak and hole	0.207 and -0.179 e.$Å^{-3}$
Theta range for data collection	2.78 to 25.38°.		

Atomic coordinates ($\times 10^4$) and equivalent isotropic displacement parameters ($Å^2 \times 10^3$) for 38. U(eq) is defined as one third of the trace of the orthogonalized U^{ij} tensor

	x	y	z	U(eq)
O(1)	4568(1)	4234(2)	1390(1)	21(1)
C(1)	4337(1)	3951(2)	2784(2)	18(1)
C(2)	3625(1)	2907(2)	2709(2)	17(1)
O(2)	3765(1)	1248(2)	2273(1)	20(1)
C(3)	3038(1)	3790(2)	1765(2)	19(1)
O(3)	3340(1)	4100(2)	436(1)	21(1)
C(4)	2835(1)	5482(2)	2363(2)	20(1)
O(4)	2471(1)	5154(2)	3593(1)	23(1)
C(5)	3567(1)	6491(2)	2542(2)	20(1)
O(5)	3842(1)	6638(2)	1158(1)	21(1)
C(6)	4199(1)	5646(2)	3430(2)	19(1)
O(6)	4023(1)	5364(2)	4818(1)	23(1)
C(7)	3999(1)	5050(2)	585(2)	21(1)
C(8)	2032(1)	6519(2)	4038(2)	26(1)
C(9)	1655(1)	5993(2)	5314(2)	22(1)
C(10)	897(1)	6324(3)	5483(2)	27(1)
C(11)	560(1)	5834(3)	6658(2)	30(1)
C(12)	971(1)	4965(2)	7692(2)	24(1)
C(13)	1725(1)	4629(2)	7552(2)	23(1)
C(14)	2060(1)	5159(2)	6364(2)	22(1)
O(12)	582(1)	4528(2)	8812(1)	31(1)
C(15)	999(1)	3757(3)	9950(2)	34(1)

Bond lengths [Å] and angles [°] for 38

O(1)-C(7)	1.402(3)	C(6)-H(6A)	1.0000	O(1)-C(1)-C(2)	108.96(15)
O(1)-C(1)	1.453(2)	O(6)-H(6O)	0.9799(11)	O(1)-C(1)-C(6)	107.65(14)
C(1)-C(2)	1.517(3)	C(7)-H(7A)	1.0000	C(2)-C(1)-C(6)	111.03(15)
C(1)-C(6)	1.526(2)	C(8)-C(9)	1.502(3)	O(1)-C(1)-H(1A)	109.7
C(1)-H(1A)	1.0000	C(8)-H(8A)	0.9900	C(2)-C(1)-H(1A)	109.7
C(2)-O(2)	1.426(2)	C(8)-H(8B)	0.9900	C(6)-C(1)-H(1A)	109.7
C(2)-C(3)	1.521(3)	C(9)-C(14)	1.383(3)	O(2)-C(2)-C(1)	111.80(15)
C(2)-H(2A)	1.0000	C(9)-C(10)	1.393(3)	O(2)-C(2)-C(3)	112.60(16)
O(2)-H(2O)	0.9798(11)	C(10)-C(11)	1.377(3)	C(1)-C(2)-C(3)	108.08(15)
C(3)-O(3)	1.447(2)	C(10)-H(10A)	0.9500	O(2)-C(2)-H(2A)	108.1
C(3)-C(4)	1.531(3)	C(11)-C(12)	1.392(3)	C(1)-C(2)-H(2A)	108.1
C(3)-H(3A)	1.0000	C(11)-H(11A)	0.9500	C(3)-C(2)-H(2A)	108.1
O(3)-C(7)	1.400(2)	C(12)-O(12)	1.369(2)	C(2)-O(2)-H(2O)	110.3(15)
C(4)-O(4)	1.415(2)	C(12)-C(13)	1.382(3)	O(3)-C(3)-C(2)	109.74(15)
C(4)-C(5)	1.535(3)	C(13)-C(14)	1.396(3)	O(3)-C(3)-C(4)	107.06(14)
C(4)-H(4A)	1.0000	C(13)-H(13A)	0.9500	C(2)-C(3)-C(4)	110.87(16)
O(4)-C(8)	1.429(2)	C(14)-H(14A)	0.9500	O(3)-C(3)-H(3A)	109.7
C(5)-O(5)	1.461(2)	O(12)-C(15)	1.432(3)	C(2)-C(3)-H(3A)	109.7
C(5)-C(6)	1.532(3)	C(15)-H(15A)	0.9800	C(4)-C(3)-H(3A)	109.7
C(5)-H(5A)	1.0000	C(15)-H(15B)	0.9800	C(7)-O(3)-C(3)	110.84(14)
O(5)-C(7)	1.426(2)	C(15)-H(15C)	0.9800	O(4)-C(4)-C(3)	106.51(14)
C(6)-O(6)	1.418(2)	C(7)-O(1)-C(1)	110.84(14)	O(4)-C(4)-C(5)	115.56(16)
C(3)-C(4)-C(5)	107.07(16)	C(11)-C(10)-C(9)	121.1(2)	O(12)-C(15)-H(15A)	109.5
O(4)-C(4)-H(4A)	109.2	C(11)-C(10)-H(10A)	119.5	O(12)-C(15)-H(15B)	109.5
C(3)-C(4)-H(4A)	109.2	C(9)-C(10)-H(10A)	119.5	H(15A)-C(15)-H(15B)	109.5
C(5)-C(4)-H(4A)	109.2	C(10)-C(11)-C(12)	120.2(2)	O(12)-C(15)-H(15C)	109.5
C(4)-O(4)-C(8)	113.47(14)	C(10)-C(11)-H(11A)	119.9	H(15A)-C(15)-H(15C)	109.5
O(5)-C(5)-C(6)	106.06(15)	C(12)-C(11)-H(11A)	119.9	H(15B)-C(15)-H(15C)	109.5
O(5)-C(5)-C(4)	105.60(15)	O(12)-C(12)-C(13)	124.66(19)	H(8A)-C(8)-H(8B)	108.4
C(6)-C(5)-C(4)	114.67(16)	O(12)-C(12)-C(11)	115.52(19)	C(14)-C(9)-C(10)	118.01(18)
O(5)-C(5)-H(5A)	110.1	C(13)-C(12)-C(11)	119.81(18)	C(14)-C(9)-C(8)	120.26(19)

C(6)-C(5)-H(5A)	110.1	C(12)-C(13)-C(14)	119.2(2)	C(10)-C(9)-C(8)	121.73(19)
C(4)-C(5)-H(5A)	110.1	C(12)-C(13)-H(13A)	120.4	C(1)-C(6)-H(6A)	109.4
C(7)-O(5)-C(5)	111.69(14)	C(14)-C(13)-H(13A)	120.4	C(5)-C(6)-H(6A)	109.4
O(6)-C(6)-C(1)	107.31(14)	C(9)-C(14)-C(13)	121.7(2)	C(6)-O(6)-H(6O)	108.7(14)
O(6)-C(6)-C(5)	113.98(15)	C(9)-C(14)-H(14A)	119.1	C(13)-C(14)-H(14A)	119.1
C(1)-C(6)-C(5)	107.21(16)	O(6)-C(6)-H(6A)	109.4	C(12)-O(12)-C(15)	117.28(17)

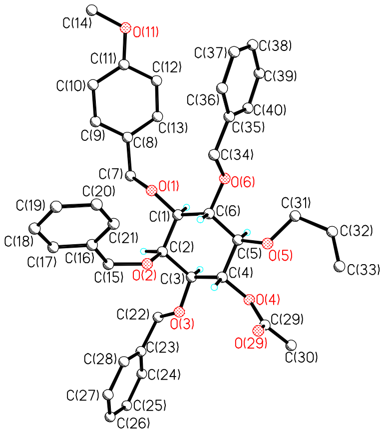
Anisotropic displacement parameters ($Å^2 \times 10^3$) for 38. The anisotropic displacement factor exponent takes the form: $-2\pi^2[h^2 a^{*2} U^{11} + ... + 2 h k a^* b^* U^{12}]$						
	U^{11}	U^{22}	U^{33}	U^{23}	U^{13}	U^{12}
O(1)	26(1)	21(1)	16(1)	1(1)	5(1)	0(1)
C(1)	24(1)	18(1)	13(1)	-1(1)	4(1)	2(1)
C(2)	27(1)	11(1)	14(1)	-2(1)	5(1)	1(1)
O(2)	31(1)	11(1)	19(1)	-2(1)	2(1)	2(1)
C(3)	23(1)	20(1)	14(1)	0(1)	4(1)	-2(1)
O(3)	29(1)	20(1)	15(1)	0(1)	1(1)	-1(1)
C(4)	24(1)	19(1)	17(1)	3(1)	4(1)	2(1)
O(4)	28(1)	18(1)	22(1)	2(1)	11(1)	6(1)
C(5)	35(1)	12(1)	14(1)	0(1)	7(1)	1(1)
O(5)	36(1)	14(1)	13(1)	1(1)	7(1)	-1(1)
C(6)	27(1)	17(1)	12(1)	-2(1)	3(1)	-4(1)
O(6)	40(1)	16(1)	12(1)	0(1)	3(1)	0(1)
C(7)	31(1)	17(1)	16(1)	0(1)	5(1)	0(1)
C(8)	30(1)	20(1)	27(1)	2(1)	7(1)	8(1)
C(9)	27(1)	17(1)	23(1)	-3(1)	3(1)	1(1)
C(10)	27(1)	31(1)	24(1)	2(1)	1(1)	5(1)
C(11)	19(1)	42(1)	30(1)	2(1)	4(1)	6(1)
C(12)	28(1)	22(1)	22(1)	-5(1)	7(1)	-3(1)
C(13)	28(1)	19(1)	24(1)	-2(1)	2(1)	2(1)
C(14)	21(1)	20(1)	26(1)	-4(1)	5(1)	3(1)
O(12)	29(1)	39(1)	25(1)	6(1)	6(1)	0(1)
C(15)	36(2)	41(1)	25(1)	7(1)	2(1)	0(1)

Hydrogen coordinates ($\times 10^4$) and isotropic displacement parameters ($Å^2 \times 10^3$) for 38				
	x	y	z	U(eq)
H(1A)	4746	3354	3338	22
H(2A)	3433	2857	3656	20
H(2O)	3907(14)	1240(30)	1313(8)	57(8)
H(3A)	2575	3086	1640	23
H(4A)	2475	6068	1693	24
H(5A)	3462	7619	2920	24
H(6A)	4668	6331	3418	22
H(6O)	3952(14)	6441(14)	5270(20)	60(8)
H(7A)	4186	5232	-356	26
H(8A)	1647	6831	3301	31
H(8B)	2359	7494	4244	31
H(10A)	608	6897	4776	33
H(11A)	45	6090	6763	36
H(13A)	2011	4044	8255	28
H(14A)	2580	4941	6273	27
H(15A)	1210	2703	9646	51
H(15B)	663	3545	10695	51
H(15C)	1408	4496	10287	51

Torsion angles [°] for 38			
C(7)-O(1)-C(1)-C(2)	59.05(18)	C(2)-C(1)-C(6)-C(5)	-58.27(19)
C(7)-O(1)-C(1)-C(6)	-61.45(19)	O(5)-C(5)-C(6)-O(6)	-178.66(14)
O(1)-C(1)-C(2)-O(2)	69.08(19)	C(4)-C(5)-C(6)-O(6)	-62.5(2)
C(6)-C(1)-C(2)-O(2)	-172.52(15)	O(5)-C(5)-C(6)-C(1)	-60.06(18)
O(1)-C(1)-C(2)-C(3)	-55.40(18)	C(4)-C(5)-C(6)-C(1)	56.05(19)
C(6)-C(1)-C(2)-C(3)	63.00(19)	C(3)-O(3)-C(7)-O(1)	61.39(18)
O(2)-C(2)-C(3)-O(3)	-68.85(19)	C(3)-O(3)-C(7)-O(5)	-62.29(19)
C(1)-C(2)-C(3)-O(3)	55.14(19)	C(1)-O(1)-C(7)-O(3)	-62.16(19)
O(2)-C(2)-C(3)-C(4)	173.09(14)	C(1)-O(1)-C(7)-O(5)	61.27(19)
C(1)-C(2)-C(3)-C(4)	-62.92(18)	C(5)-O(5)-C(7)-O(3)	62.5(2)
C(2)-C(3)-O(3)-C(7)	-57.90(19)	C(5)-O(5)-C(7)-O(1)	-61.9(2)
C(4)-C(3)-O(3)-C(7)	62.50(19)	C(4)-O(4)-C(8)-C(9)	177.93(17)
O(3)-C(3)-C(4)-O(4)	174.23(15)	O(4)-C(8)-C(9)-C(14)	45.3(3)
C(2)-C(3)-C(4)-O(4)	-66.1(2)	O(4)-C(8)-C(9)-C(10)	-134.8(2)
O(3)-C(3)-C(4)-C(5)	-61.59(19)	C(14)-C(9)-C(10)-C(11)	-0.1(3)
C(2)-C(3)-C(4)-C(5)	58.08(19)	C(8)-C(9)-C(10)-C(11)	180.0(2)
C(3)-C(4)-O(4)-C(8)	-160.61(17)	C(9)-C(10)-C(11)-C(12)	-1.3(3)
C(5)-C(4)-O(4)-C(8)	80.6(2)	C(10)-C(11)-C(12)-O(12)	-179.59(19)
O(4)-C(4)-C(5)-O(5)	178.82(14)	C(10)-C(11)-C(12)-C(13)	1.5(3)
C(3)-C(4)-C(5)-O(5)	60.37(18)	O(12)-C(12)-C(13)-C(14)	-179.23(18)
O(4)-C(4)-C(5)-C(6)	62.4(2)	C(11)-C(12)-C(13)-C(14)	-0.5(3)
C(3)-C(4)-C(5)-C(6)	-56.0(2)	C(10)-C(9)-C(14)-C(13)	1.2(3)
C(6)-C(5)-O(5)-C(7)	60.90(19)	C(8)-C(9)-C(14)-C(13)	-178.90(18)
C(4)-C(5)-O(5)-C(7)	-61.20(19)	C(12)-C(13)-C(14)-C(9)	-0.9(3)
O(1)-C(1)-C(6)-O(6)	-176.25(15)	C(13)-C(12)-O(12)-C(15)	3.9(3)
C(2)-C(1)-C(6)-O(6)	64.6(2)	C(11)-C(12)-O(12)-C(15)	-174.87(19)
O(1)-C(1)-C(6)-C(5)	60.92(19)		

Hydrogen bonds for 38 [Å and °]				
D-H...A	d(D-H)	d(H...A)	d(D...A)	<(DHA)
O(2)-H(2O)...O(6)#1	0.9798(11)	1.960(17)	2.7728(18)	139(2)
O(6)-H(6O)...O(5)#2	0.9799(11)	1.787(2)	2.7660(18)	177(2)
Symmetry transformations used to generate equivalent atoms: #1 x,-y+1/2,z-1/2 #2 x,-y+3/2,z+1/2				

(+)-1D-4-*O*-Acetyl-5-*O*-allyl-1-*O*-4-methoxybenzyl-2,3,6-tris-*O*-benzyl-*myo*-inositol 51



Crystal structure of compound **51.**

Crystal data and structure refinement for 51			
Empirical formula	$C_{40}H_{44}O_8$	Index ranges	-10<=h<=16, -6<=k<=8, -21<=l<=21
Formula weight	652.75	Reflections collected	14320
Temperature	93(2) K	Independent reflections	5785 [R(int) = 0.0331]
Wavelength	0.71073 Å	Completeness to theta = 25.35°	98.9 %
Crystal system	Monoclinic	Absorption correction	MULTISCAN
Space group	P2(1)	Max. and min. transmission	1.0000 and 0.9144
Unit cell dimensions	a = 13.384(3) Å α= 90°. b = 7.3188(15) Å β= 96.621(4)°. c = 17.493(4) Å γ = 90°.	Refinement method	Full-matrix least-squares on F^2
Volume	1702.1(6) $Å^3$	Data / restraints / parameters	5785 / 1 / 437
Z	2	Goodness-of-fit on F^2	1.124
Density (calculated)	1.274 Mg/m^3	Final R indices [I>2sigma(I)]	R1 = 0.0458, wR2 = 0.0901
Absorption coefficient	0.088 mm^{-1}	R indices (all data)	R1 = 0.0548, wR2 = 0.0966
F(000)	696	Absolute structure parameter	0.6(9)
Crystal size	0.2000 × 0.0300 × 0.0300 mm^3	Extinction coefficient	0.0165(18)
Theta range for data collection	2.03 to 25.35°.	Largest diff. peak and hole	0.215 and -0.212 e.$Å^{-3}$

Atomic coordinates (× 10^4) and equivalent isotropic displacement parameters (Å^2 × 10^3) for 51. U(eq) is defined as one third of the trace of the orthogonalized U^{ij} tensor.

	x	y	z	U(eq)
C(1)	9229(2)	804(4)	2319(1)	21(1)
O(1)	8513(1)	51(2)	2768(1)	23(1)
C(2)	10248(2)	1031(4)	2799(1)	20(1)
O(2)	10141(1)	2161(2)	3455(1)	21(1)
C(3)	10987(2)	1948(4)	2314(1)	19(1)
O(3)	11929(1)	2321(2)	2746(1)	21(1)
C(4)	10573(2)	3765(4)	2007(1)	18(1)
O(4)	11273(1)	4492(2)	1509(1)	20(1)
C(5)	9550(2)	3549(4)	1531(1)	19(1)
O(5)	9249(1)	5346(2)	1295(1)	22(1)
C(6)	8806(2)	2629(3)	2005(1)	18(1)
O(6)	7893(1)	2324(3)	1511(1)	22(1)
C(7)	8501(2)	-1914(4)	2813(2)	26(1)
C(8)	7416(2)	-2476(4)	2710(1)	21(1)
C(9)	6957(2)	-3198(4)	3307(1)	23(1)
C(10)	5924(2)	-3609(4)	3222(2)	23(1)
C(11)	5358(2)	-3233(4)	2530(2)	21(1)
O(11)	4337(1)	-3511(3)	2382(1)	26(1)
C(12)	5811(2)	-2504(4)	1916(1)	23(1)
C(13)	6832(2)	-2146(4)	2015(2)	23(1)
C(14)	3852(2)	-4203(4)	3008(2)	30(1)
C(15)	10198(2)	1229(4)	4173(1)	26(1)
C(16)	9330(2)	1679(4)	4618(1)	19(1)
C(17)	9355(2)	1048(4)	5370(1)	23(1)
C(18)	8570(2)	1405(4)	5804(2)	29(1)
C(19)	7750(2)	2410(4)	5487(1)	26(1)
C(20)	7718(2)	3052(4)	4742(2)	26(1)
C(21)	8503(2)	2668(4)	4307(1)	22(1)
C(22)	12548(2)	731(4)	2904(1)	24(1)
C(23)	13395(2)	1232(3)	3503(1)	20(1)
C(24)	14384(2)	806(4)	3411(2)	25(1)
C(25)	15165(2)	1320(4)	3960(2)	30(1)
C(26)	14961(2)	2280(4)	4603(2)	31(1)
C(27)	13976(2)	2701(4)	4703(2)	29(1)
C(28)	13198(2)	2187(4)	4156(1)	25(1)
C(29)	11567(2)	6238(4)	1611(1)	21(1)
O(29)	11276(1)	7254(3)	2083(1)	29(1)
C(30)	12299(2)	6736(4)	1060(2)	30(1)
C(31)	8525(2)	5481(4)	626(1)	28(1)
C(32)	8693(2)	7192(4)	203(1)	28(1)
C(33)	9416(2)	8381(4)	381(2)	29(1)
C(34)	7002(2)	2679(4)	1870(1)	22(1)
C(35)	6077(2)	2161(4)	1340(1)	22(1)
C(36)	5141(2)	2451(4)	1593(1)	25(1)
C(37)	4268(2)	1936(4)	1147(2)	30(1)
C(38)	4318(2)	1093(4)	440(2)	32(1)
C(39)	5248(2)	811(4)	180(2)	29(1)
C(40)	6120(2)	1343(4)	628(1)	23(1)

Bond lengths [Å] and angles [°] for 51					
C(1)-O(1)	1.418(3)	C(12)-H(12A)	0.9500	C(30)-H(30C)	0.9800
C(1)-C(2)	1.527(3)	C(13)-H(13A)	0.9500	C(31)-C(32)	1.485(4)
C(1)-C(6)	1.528(4)	C(14)-H(14A)	0.9800	C(31)-H(31A)	0.9900
C(1)-H(1A)	1.0000	C(14)-H(14B)	0.9800	C(31)-H(31B)	0.9900
O(1)-C(7)	1.441(3)	C(14)-H(14C)	0.9800	C(32)-C(33)	1.312(4)
C(2)-O(2)	1.434(3)	C(15)-C(16)	1.507(3)	C(32)-H(32A)	0.9500
C(2)-C(3)	1.531(3)	C(15)-H(15A)	0.9900	C(33)-H(33A)	0.9500
C(2)-H(2A)	1.0000	C(15)-H(15B)	0.9900	C(33)-H(33B)	0.9500
O(2)-C(15)	1.425(3)	C(16)-C(21)	1.380(3)	C(34)-C(35)	1.507(3)
C(3)-O(3)	1.419(3)	C(16)-C(17)	1.390(3)	C(34)-H(34A)	0.9900
C(3)-C(4)	1.516(3)	C(17)-C(18)	1.390(3)	C(34)-H(34B)	0.9900
C(3)-H(3A)	1.0000	C(17)-H(17A)	0.9500	C(35)-C(40)	1.389(3)
O(3)-C(22)	1.436(3)	C(18)-C(19)	1.383(4)	C(35)-C(36)	1.392(3)
C(4)-O(4)	1.453(3)	C(18)-H(18A)	0.9500	C(36)-C(37)	1.382(3)
C(4)-C(5)	1.526(3)	C(19)-C(20)	1.381(3)	C(36)-H(36A)	0.9500
C(4)-H(4A)	1.0000	C(19)-H(19A)	0.9500	C(37)-C(38)	1.389(4)
O(4)-C(29)	1.343(3)	C(20)-C(21)	1.395(3)	C(37)-H(37A)	0.9500
C(5)-O(5)	1.423(3)	C(20)-H(20A)	0.9500	C(38)-C(39)	1.390(4)
C(5)-C(6)	1.525(3)	C(21)-H(21A)	0.9500	C(38)-H(38A)	0.9500
C(5)-H(5A)	1.0000	C(22)-C(23)	1.498(3)	C(39)-C(40)	1.385(3)
O(5)-C(31)	1.433(3)	C(22)-H(22A)	0.9900	C(39)-H(39A)	0.9500
C(6)-O(6)	1.431(3)	C(22)-H(22B)	0.9900	C(40)-H(40A)	0.9500
C(6)-H(6A)	1.0000	C(23)-C(24)	1.388(3)	O(1)-C(1)-C(2)	110.80(19)
O(6)-C(34)	1.435(3)	C(23)-C(28)	1.391(4)	O(1)-C(1)-C(6)	107.05(19)
C(7)-C(8)	1.501(3)	C(24)-C(25)	1.386(4)	C(2)-C(1)-C(6)	111.9(2)
C(7)-H(7A)	0.9900	C(24)-H(24A)	0.9500	O(1)-C(1)-H(1A)	109.0
C(7)-H(7B)	0.9900	C(25)-C(26)	1.380(4)	C(2)-C(1)-H(1A)	109.0
C(8)-C(9)	1.377(3)	C(25)-H(25A)	0.9500	C(6)-C(1)-H(1A)	109.0
C(8)-C(13)	1.388(3)	C(26)-C(27)	1.385(4)	C(1)-O(1)-C(7)	115.54(19)
C(9)-C(10)	1.407(3)	C(26)-H(26A)	0.9500	O(2)-C(2)-C(1)	109.66(19)
C(9)-H(9A)	0.9500	C(27)-C(28)	1.383(3)	O(2)-C(2)-C(3)	108.8(2)
C(10)-C(11)	1.379(3)	C(27)-H(27A)	0.9500	C(1)-C(2)-C(3)	109.66(19)
C(10)-H(10A)	0.9500	C(28)-H(28A)	0.9500	O(2)-C(2)-H(2A)	109.6
C(11)-O(11)	1.376(3)	C(29)-O(29)	1.209(3)	C(1)-C(2)-H(2A)	109.6
C(11)-C(12)	1.399(3)	C(29)-C(30)	1.497(3)	C(3)-C(2)-H(2A)	109.6
O(11)-C(14)	1.429(3)	C(30)-H(30A)	0.9800	C(15)-O(2)-C(2)	115.4(2)
C(12)-C(13)	1.382(3)	C(30)-H(30B)	0.9800	O(3)-C(3)-C(4)	106.63(19)
O(3)-C(3)-C(2)	112.33(18)	C(3)-C(4)-H(4A)	109.9	O(6)-C(6)-C(5)	107.81(18)
C(4)-C(3)-C(2)	110.32(19)	C(5)-C(4)-H(4A)	109.9	O(6)-C(6)-C(1)	109.60(19)
O(3)-C(3)-H(3A)	109.2	C(29)-O(4)-C(4)	117.86(19)	C(5)-C(6)-C(1)	110.08(19)
C(4)-C(3)-H(3A)	109.2	O(5)-C(5)-C(6)	112.6(2)	O(6)-C(6)-H(6A)	109.8
C(2)-C(3)-H(3A)	109.2	O(5)-C(5)-C(4)	105.63(19)	C(5)-C(6)-H(6A)	109.8
C(3)-O(3)-C(22)	113.79(18)	C(6)-C(5)-C(4)	110.44(18)	C(1)-C(6)-H(6A)	109.8

O(4)-C(4)-C(3)	107.36(18)	O(5)-C(5)-H(5A)	109.4	C(6)-O(6)-C(34)	113.66(17)
O(4)-C(4)-C(5)	108.05(18)	C(6)-C(5)-H(5A)	109.4	O(1)-C(7)-C(8)	106.5(2)
C(3)-C(4)-C(5)	111.8(2)	C(4)-C(5)-H(5A)	109.4	O(1)-C(7)-H(7A)	110.4
O(4)-C(4)-H(4A)	109.9	C(5)-O(5)-C(31)	116.28(19)	C(8)-C(7)-H(7A)	110.4
C(8)-C(9)-H(9A)	119.3	H(14A)-C(14)-H(14C)	109.5	O(1)-C(7)-H(7B)	110.4
C(10)-C(9)-H(9A)	119.3	H(14B)-C(14)-H(14C)	109.5	C(8)-C(7)-H(7B)	110.4
C(11)-C(10)-C(9)	118.9(2)	O(2)-C(15)-C(16)	112.6(2)	H(7A)-C(7)-H(7B)	108.6
C(11)-C(10)-H(10A)	120.5	O(2)-C(15)-H(15A)	109.1	C(9)-C(8)-C(13)	118.4(2)
C(9)-C(10)-H(10A)	120.5	C(16)-C(15)-H(15A)	109.1	C(9)-C(8)-C(7)	121.6(2)
O(11)-C(11)-C(10)	124.6(2)	O(2)-C(15)-H(15B)1	109.1	C(13)-C(8)-C(7)	119.9(2)
O(11)-C(11)-C(12)	114.9(2)	C(16)-C(15)-H(15B)	109.1	C(8)-C(9)-C(10)	121.4(2)
C(10)-C(11)-C(12)	120.5(2)	H(15A)-C(15)-H(15B)	107.8	C(18)-C(19)-H(19A)	120.1
C(11)-O(11)-C(14)	115.98(19)	C(21)-C(16)-C(17)	118.5(2)	C(19)-C(20)-C(21)	120.2(2)
C(13)-C(12)-C(11)	119.1(2)	C(21)-C(16)-C(15)	122.8(2)	C(19)-C(20)-H(20A)	119.9
C(13)-C(12)-H(12A)	120.5	C(17)-C(16)-C(15)	118.7(2)	C(21)-C(20)-H(20A)	119.9
C(11)-C(12)-H(12A)	120.5	C(18)-C(17)-C(16)	121.1(2)	C(16)-C(21)-C(20)	120.7(2)
C(12)-C(13)-C(8)	121.7(2)	C(18)-C(17)-H(17A)	119.4	C(16)-C(21)-H(21A)	119.6
C(12)-C(13)-H(13A)	119.2	C(16)-C(17)-H(17A)	119.4	C(20)-C(21)-H(21A)	119.6
C(8)-C(13)-H(13A)	119.2	C(19)-C(18)-C(17)	119.7(2)	O(3)-C(22)-C(23)	108.0(2)
O(11)-C(14)-H(14A)	109.5	C(19)-C(18)-H(18A)	120.1	O(3)-C(22)-H(22A)	110.1
O(11)-C(14)-H(14B)	109.5	C(17)-C(18)-H(18A)	120.1	C(23)-C(22)-H(22A)	110.1
H(14A)-C(14)-H(14B)	109.5	C(20)-C(19)-C(18)	119.7(2)	O(3)-C(22)-H(22B)	110.1
O(11)-C(14)-H(14C)	109.5	C(20)-C(19)-H(19A)	120.1	C(23)-C(22)-H(22B)	110.1
H(22A)-C(22)-H(22B)	108.4	C(26)-C(27)-H(27A)	119.9	C(32)-C(31)-H(31A)	109.7
C(24)-C(23)-C(28)	118.8(2)	C(27)-C(28)-C(23)	120.4(2)	O(5)-C(31)-H(31B)	109.7
C(24)-C(23)-C(22)	121.4(2)	C(27)-C(28)-H(28A)	119.8	C(32)-C(31)-H(31B)	109.7
C(28)-C(23)-C(22)	119.8(2)	C(23)-C(28)-H(28A)	119.8	H(31A)-C(31)-H(31B)	108.2
C(25)-C(24)-C(23)	120.9(3)	O(29)-C(29)-O(4)	124.3(2)	C(33)-C(32)-C(31)	126.2(2)
C(25)-C(24)-H(24A)	119.6	O(29)-C(29)-C(30)	125.2(2)	C(33)-C(32)-H(32A)	116.9
C(23)-C(24)-H(24A)	119.6	O(4)-C(29)-C(30)	110.4(2)	C(31)-C(32)-H(32A)	116.9
C(26)-C(25)-C(24)	119.9(2)	C(29)-C(30)-H(30A)	109.5	C(32)-C(33)-H(33A)	120.0
C(26)-C(25)-H(25A)	120.1	C(29)-C(30)-H(30B)	109.5	C(32)-C(33)-H(33B)	120.0
C(24)-C(25)-H(25A)	120.1	H(30A)-C(30)-H(30B)	109.5	H(33A)-C(33)-H(33B)	120.0
C(25)-C(26)-C(27)	119.8(2)	C(29)-C(30)-H(30C)	109.5	O(6)-C(34)-C(35)	110.46(19)
C(25)-C(26)-H(26A)	120.1	H(30A)-C(30)-H(30C)	109.5	O(6)-C(34)-H(34A)	109.6
C(27)-C(26)-H(26A)	120.1	H(30B)-C(30)-H(30C)	109.5	C(35)-C(34)-H(34A)	109.6
C(28)-C(27)-C(26)	120.2(3)	O(5)-C(31)-C(32)	109.8(2)	O(6)-C(34)-H(34B)	109.6
C(28)-C(27)-H(27A)	119.9	O(5)-C(31)-H(31A)	109.7	C(35)-C(34)-H(34B)	109.6
H(34A)-C(34)-H(34B)	108.1	C(35)-C(36)-H(36A)	119.6	C(39)-C(38)-H(38A)	120.2
C(40)-C(35)-C(36)	118.9(2)	C(36)-C(37)-C(38)	120.0(2)	C(40)-C(39)-C(38)	120.1(3)
C(40)-C(35)-C(34)	123.0(2)	C(36)-C(37)-H(37A)	120.0	C(40)-C(39)-H(39A)	120.0
C(36)-C(35)-C(34)	118.1(2)	C(38)-C(37)-H(37A)	120.0	C(38)-C(39)-H(39A)	120.0
C(37)-C(36)-C(35)	120.8(2)	C(37)-C(38)-C(39)	119.6(2)	C(39)-C(40)-C(35)	120.6(2)
C(37)-C(36)-H(36A)	119.6	C(37)-C(38)-H(38A)	120.2	C(39)-C(40)-H(40A)	119.7

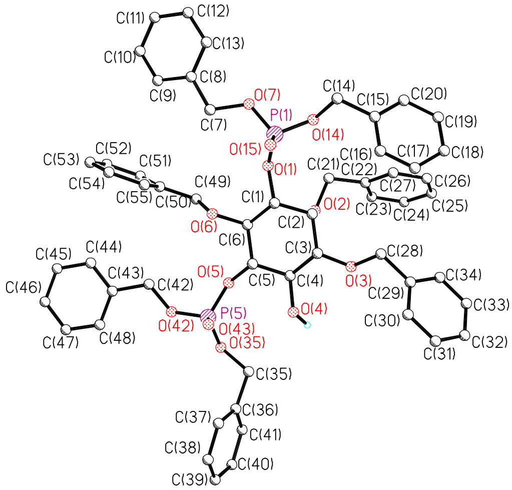
Anisotropic displacement parameters ($Å^2 \times 10^3$) for 51. The anisotropic displacement factor exponent takes the form: $-2\pi^2[h^2 a^{*2}U^{11} + ... + 2 h k a^* b^* U^{12}]$						
	U^{11}	U^{22}	U^{33}	U^{23}	U^{13}	U^{12}
C(1)	21(1)	18(2)	23(1)	-3(1)	5(1)	-6(1)
O(1)	21(1)	21(1)	28(1)	-1(1)	9(1)	-4(1)
C(2)	21(1)	20(2)	19(1)	-1(1)	2(1)	0(1)
O(2)	26(1)	23(1)	14(1)	-1(1)	4(1)	0(1)
C(3)	14(1)	23(2)	19(1)	-1(1)	1(1)	-2(1)
O(3)	17(1)	19(1)	24(1)	-2(1)	-4(1)	-2(1)
C(4)	17(1)	22(2)	14(1)	-1(1)	4(1)	-4(1)
O(4)	20(1)	23(1)	18(1)	0(1)	4(1)	-7(1)
C(5)	22(1)	18(2)	17(1)	1(1)	1(1)	-2(1)
O(5)	23(1)	22(1)	20(1)	3(1)	-4(1)	0(1)
C(6)	14(1)	20(2)	18(1)	-5(1)	0(1)	-3(1)
O(6)	14(1)	31(1)	20(1)	-3(1)	1(1)	0(1)
C(7)	19(1)	23(2)	36(2)	6(1)	4(1)	1(1)
C(8)	20(1)	14(2)	29(1)	3(1)	2(1)	-2(1)
C(9)	25(1)	20(2)	25(1)	3(1)	1(1)	-3(1)
C(10)	22(1)	22(2)	26(1)	1(1)	6(1)	-2(1)
C(11)	18(1)	15(1)	29(1)	-4(1)	5(1)	-3(1)
O(11)	19(1)	29(1)	29(1)	5(1)	3(1)	-3(1)
C(12)	24(1)	23(2)	21(1)	1(1)	1(1)	-3(1)
C(13)	24(1)	20(2)	28(1)	2(1)	9(1)	0(1)
C(14)	23(1)	31(2)	39(2)	6(1)	8(1)	-5(1)
C(15)	24(1)	34(2)	20(1)	6(1)	2(1)	5(1)
C(16)	20(1)	19(2)	20(1)	-2(1)	2(1)	-3(1)
C(17)	21(1)	25(2)	24(1)	2(1)	3(1)	4(1)
C(18)	32(1)	35(2)	21(1)	7(1)	7(1)	2(1)
C(19)	22(1)	32(2)	26(1)	1(1)	7(1)	-1(1)
C(20)	18(1)	29(2)	28(1)	2(1)	0(1)	2(1)
C(21)	23(1)	26(2)	17(1)	3(1)	3(1)	1(1)
C(22)	19(1)	25(2)	26(1)	-4(1)	-2(1)	1(1)
C(23)	19(1)	15(1)	24(1)	5(1)	-2(1)	-1(1)
C(24)	25(1)	19(2)	32(1)	6(1)	4(1)	2(1)
C(25)	21(1)	32(2)	36(2)	5(1)	-4(1)	3(1)
C(26)	22(1)	36(2)	33(2)	10(1)	-8(1)	-5(1)
C(27)	31(1)	34(2)	21(1)	2(1)	0(1)	-7(1)
C(28)	20(1)	27(2)	28(1)	4(1)	3(1)	-6(1)
C(29)	20(1)	22(2)	19(1)	4(1)	-3(1)	-5(1)
O(29)	33(1)	27(1)	28(1)	-5(1)	6(1)	-7(1)
C(30)	30(1)	34(2)	27(1)	6(1)	6(1)	-11(1)
C(31)	24(1)	34(2)	23(1)	4(1)	-6(1)	-2(1)
C(32)	24(1)	37(2)	21(1)	7(1)	-2(1)	7(1)
C(33)	32(2)	29(2)	26(1)	5(1)	3(1)	5(1)
C(34)	16(1)	28(2)	24(1)	-2(1)	3(1)	-1(1)
C(35)	22(1)	21(2)	22(1)	2(1)	1(1)	-1(1)
C(36)	22(1)	26(2)	26(1)	-1(1)	4(1)	0(1)
C(37)	20(1)	33(2)	39(2)	4(1)	4(1)	0(1)
C(38)	22(1)	33(2)	39(2)	-1(1)	-8(1)	-3(1)
C(39)	32(2)	27(2)	25(1)	-3(1)	-4(1)	-2(1)
C(40)	19(1)	26(2)	23(1)	-2(1)	0(1)	1(1)

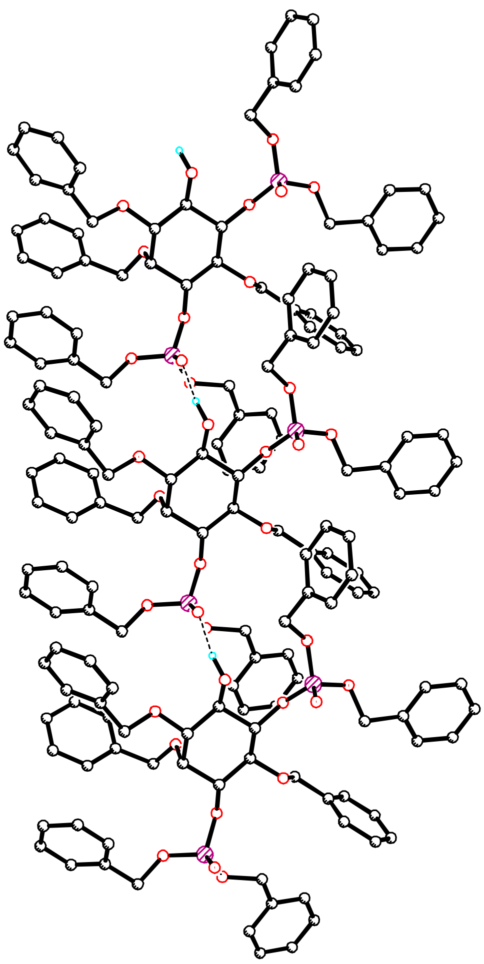
Hydrogen coordinates (× 10^4) and isotropic displacement parameters ($Å^2$ × 10^3) for 51				
	x	y	z	U(eq)
H(1A)	9306	-33	1878	25
H(2A)	10513	-194	2976	24
H(3A)	11095	1134	1871	22
H(4A)	10515	4626	2444	21
H(5A)	9627	2789	1066	23
H(6A)	8672	3443	2441	21
H(7A)	8831	-2329	3319	31
H(7B)	8862	-2454	2404	31
H(9A)	7347	-3423	3787	28
H(10A)	5620	-4137	3634	28
H(12A)	5423	-2258	1438	27
H(13A)	7141	-1661	1597	28
H(14A)	4150	-5381	3174	46
H(14B)	3133	-4367	2841	46
H(14C)	3939	-3337	3438	46
H(15A)	10835	1561	4487	31
H(15B)	10208	-105	4080	31
H(17A)	9918	363	5591	28
H(18A)	8596	959	6316	35
H(19A)	7211	2658	5780	32
H(20A)	7160	3757	4525	31
H(21A)	8469	3092	3791	26
H(22A)	12816	316	2429	29
H(22B)	12147	-274	3094	29
H(24A)	14529	153	2968	30
H(25A)	15839	1013	3893	36
H(26A)	15495	2652	4976	37
H(27A)	13834	3345	5150	35
H(28A)	12524	2489	4227	30
H(30A)	12386	8065	1056	45
H(30B)	12044	6316	542	45
H(30C)	12948	6152	1222	45
H(31A)	7837	5479	782	33
H(31B)	8590	4415	287	33
H(32A)	8233	7450	-239	33
H(33A)	9894	8183	817	35
H(33B)	9460	9440	73	35
H(34A)	6972	3993	2001	27
H(34B)	7024	1969	2354	27
H(36A)	5102	3010	2079	30
H(37A)	3633	2159	1322	36
H(38A)	3719	711	137	39
H(39A)	5286	254	-306	34
H(40A)	6754	1146	447	27

Torsion angles [°] for 51			
C(2)-C(1)-O(1)-C(7)	-85.9(3)	C(4)-C(5)-C(6)-O(6)	-175.12(19)
C(6)-C(1)-O(1)-C(7)	151.8(2)	O(5)-C(5)-C(6)-C(1)	-173.42(18)
O(1)-C(1)-C(2)-O(2)	-57.4(3)	C(4)-C(5)-C(6)-C(1)	-55.6(3)
C(6)-C(1)-C(2)-O(2)	62.0(2)	O(1)-C(1)-C(6)-O(6)	-63.1(2)
O(1)-C(1)-C(2)-C(3)	-176.7(2)	C(2)-C(1)-C(6)-O(6)	175.36(18)
C(6)-C(1)-C(2)-C(3)	-57.3(3)	O(1)-C(1)-C(6)-C(5)	178.52(19)
C(1)-C(2)-O(2)-C(15)	106.6(2)	C(2)-C(1)-C(6)-C(5)	56.9(2)
C(3)-C(2)-O(2)-C(15)	-133.5(2)	C(5)-C(6)-O(6)-C(34)	-139.6(2)
O(2)-C(2)-C(3)-O(3)	55.7(3)	C(1)-C(6)-O(6)-C(34)	100.6(2)

C(1)-C(2)-C(3)-O(3)	175.6(2)	C(1)-O(1)-C(7)-C(8)	-136.1(2)
O(2)-C(2)-C(3)-C(4)	-63.1(2)	O(1)-C(7)-C(8)-C(9)	-110.4(3)
C(1)-C(2)-C(3)-C(4)	56.8(3)	O(1)-C(7)-C(8)-C(13)	64.8(3)
C(4)-C(3)-O(3)-C(22)	-164.11(18)	C(13)-C(8)-C(9)-C(10)	0.4(4)
C(2)-C(3)-O(3)-C(22)	74.9(2)	C(7)-C(8)-C(9)-C(10)	175.7(2)
O(3)-C(3)-C(4)-O(4)	61.9(2)	C(8)-C(9)-C(10)-C(11)	-1.7(4)
C(2)-C(3)-C(4)-O(4)	-175.86(18)	C(9)-C(10)-C(11)-O(11)	-177.7(2)
O(3)-C(3)-C(4)-C(5)	-179.77(18)	C(9)-C(10)-C(11)-C(12)	1.8(4)
C(2)-C(3)-C(4)-C(5)	-57.5(2)	C(10)-C(11)-O(11)-C(14)	1.4(4)
C(3)-C(4)-O(4)-C(29)	-131.5(2)	C(12)-C(11)-O(11)-C(14)	-178.2(2)
C(5)-C(4)-O(4)-C(29)	107.8(2)	O(11)-C(11)-C(12)-C(13)	178.8(2)
O(4)-C(4)-C(5)-O(5)	-63.1(2)	C(10)-C(11)-C(12)-C(13)	-0.7(4)
C(3)-C(4)-C(5)-O(5)	179.00(18)	C(11)-C(12)-C(13)-C(8)	-0.6(4)
O(4)-C(4)-C(5)-C(6)	174.9(2)	C(9)-C(8)-C(13)-C(12)	0.7(4)
C(3)-C(4)-C(5)-C(6)	57.0(3)	C(7)-C(8)-C(13)-C(12)	-174.6(2)
C(6)-C(5)-O(5)-C(31)	-81.8(2)	C(2)-O(2)-C(15)-C(16)	-129.3(2)
C(4)-C(5)-O(5)-C(31)	157.55(19)	O(2)-C(15)-C(16)-C(21)	9.0(4)
O(5)-C(5)-C(6)-O(6)	67.1(2)	O(2)-C(15)-C(16)-C(17)	-172.0(2)
C(21)-C(16)-C(17)-C(18)	-0.2(4)	C(24)-C(23)-C(28)-C(27)	0.0(4)
C(15)-C(16)-C(17)-C(18)	-179.3(2)	C(22)-C(23)-C(28)-C(27)	-178.3(2)
C(16)-C(17)-C(18)-C(19)	-0.3(4)	C(4)-O(4)-C(29)-O(29)	-1.1(3)
C(17)-C(18)-C(19)-C(20)	0.0(4)	C(4)-O(4)-C(29)-C(30)	178.87(19)
C(18)-C(19)-C(20)-C(21)	0.8(4)	C(5)-O(5)-C(31)-C(32)	-148.6(2)
C(17)-C(16)-C(21)-C(20)	1.0(4)	O(5)-C(31)-C(32)-C(33)	3.0(4)
C(15)-C(16)-C(21)-C(20)	-179.9(2)	C(6)-O(6)-C(34)-C(35)	-174.7(2)
C(19)-C(20)-C(21)-C(16)	-1.4(4)	O(6)-C(34)-C(35)-C(40)	3.9(4)
C(3)-O(3)-C(22)-C(23)	-167.30(19)	O(6)-C(34)-C(35)-C(36)	-178.8(2)
O(3)-C(22)-C(23)-C(24)	-131.3(2)	C(40)-C(35)-C(36)-C(37)	0.0(4)
O(3)-C(22)-C(23)-C(28)	47.0(3)	C(34)-C(35)-C(36)-C(37)	-177.4(3)
C(28)-C(23)-C(24)-C(25)	0.0(4)	C(35)-C(36)-C(37)-C(38)	1.0(4)
C(22)-C(23)-C(24)-C(25)	178.2(2)	C(36)-C(37)-C(38)-C(39)	-1.5(4)
C(23)-C(24)-C(25)-C(26)	-0.5(4)	C(37)-C(38)-C(39)-C(40)	1.0(4)
C(24)-C(25)-C(26)-C(27)	1.0(4)	C(38)-C(39)-C(40)-C(35)	0.0(4)
C(25)-C(26)-C(27)-C(28)	-1.0(4)	C(36)-C(35)-C(40)-C(39)	-0.5(4)
C(26)-C(27)-C(28)-C(23)	0.5(4)	C(34)-C(35)-C(40)-C(39)	176.8(3)

(+)-1D-2,3,6-tris-*O*-Benzyl-*myo*-inositol 1,5-bis(dibenzylphosphate) 122





Crystal structure of compound **122.**

Crystal data and structure refinement for 122			
Empirical formula	$C_{55}H_{56}O_{12}P_2$	Theta range for data collection	2.24 to 25.35°
Formula weight	970.94	Index ranges	-10<=h<=10, -18<=k<=18, -19<=l<=20
Temperature	93(2) K	Reflections collected	22443
Wavelength	0.71073 Å	Independent reflections	8543 [R(int) = 0.0440]
Crystal system	Monoclinic	Completeness to theta = 25.00°	97.1 %
Space group	P2(1)	Absorption correction	Multiscan
Unit cell dimensions	a = 9.0930(13) Å α = 90°. b = 15.335(2) Å β = 90.224(2)°. c = 17.377(3) Å γ = 90°.	Max. and min. transmission	1.0000 and 0.8454
Volume	2423.1(6) $Å^3$	Refinement method	Full-matrix least-squares on F^2
		Data / restraints / parameters	8543 / 2 / 627
Z	2	Goodness-of-fit on F^2	0.987
Density (calculated)	1.331 Mg/m^3	Final R indices [I>2sigma(I)]	R1 = 0.0300, wR2 = 0.0766
Absorption coefficient	0.155 mm^{-1}	R indices (all data)	R1 = 0.0305, wR2 = 0.0774
F(000)	1024	Absolute structure parameter	0.03(4)
Crystal size	0.1500 x 0.1500 x 0.1500 mm^3	Largest diff. peak and hole	0.175 and -0.218 $e.Å^{-3}$

Atomic coordinates ($\times 10^4$) and equivalent isotropic displacement parameters ($Å^2 \times 10^3$) for 122.
U(eq) is defined as one third of the trace of the orthogonalized U^{ij} tensor.

	x	y	z	U(eq)
P(1)	-8428(1)	-5979(1)	-8383(1)	15(1)
O(1)	-6857(1)	-6104(1)	-8014(1)	17(1)
C(1)	-5693(2)	-5471(1)	-8139(1)	15(1)
O(2)	-4022(1)	-6536(1)	-8684(1)	20(1)
C(2)	-4749(2)	-5731(1)	-8830(1)	18(1)
O(3)	-2635(1)	-5174(1)	-9561(1)	21(1)
C(3)	-3585(2)	-5021(1)	-8934(1)	17(1)
O(4)	-1584(1)	-4264(1)	-8296(1)	19(1)
C(4)	-2607(2)	-4951(1)	-8218(1)	15(1)
P(5)	-1968(1)	-3921(1)	-6457(1)	15(1)
O(5)	-2620(1)	-4782(1)	-6834(1)	16(1)
C(5)	-3550(2)	-4763(1)	-7512(1)	16(1)
O(6)	-5622(1)	-5257(1)	-6754(1)	17(1)
C(6)	-4743(2)	-5452(1)	-7407(1)	15(1)
O(7)	-9435(1)	-6512(1)	-7825(1)	18(1)
C(7)	-9643(2)	-6181(1)	-7046(1)	20(1)
C(8)	-10731(2)	-6751(1)	-6637(1)	19(1)
C(9)	-10665(2)	-6814(1)	-5840(1)	27(1)
C(10)	-11676(2)	-7317(1)	-5445(1)	34(1)
C(11)	-12766(2)	-7757(1)	-5843(1)	31(1)
C(12)	-12840(2)	-7694(1)	-6638(1)	29(1)

C(13)	-11830(2)	-7191(1)	-7037(1)	23(1)
O(14)	-8393(1)	-6557(1)	-9122(1)	20(1)
C(14)	-9392(2)	-6398(1)	-9778(1)	25(1)
O(15)	-8857(1)	-5067(1)	-8510(1)	20(1)
C(15)	-8478(2)	-6331(1)	-10496(1)	21(1)
C(16)	-7632(2)	-5591(1)	-10626(1)	25(1)
C(17)	-6795(2)	-5521(1)	-11280(1)	30(1)
C(18)	-6790(2)	-6194(1)	-11821(1)	31(1)
C(19)	-7620(2)	-6931(1)	-11695(1)	31(1)
C(20)	-8467(2)	-7003(1)	-11033(1)	26(1)
C(21)	-4761(2)	-7312(1)	-8955(1)	22(1)
C(22)	-4050(2)	-7641(1)	-9682(1)	20(1)
C(23)	-2772(2)	-8126(1)	-9643(1)	23(1)
C(24)	-2073(2)	-8400(1)	-10309(1)	28(1)
C(25)	-2663(2)	-8187(1)	-11021(1)	31(1)
C(26)	-3945(2)	-7711(1)	-11067(1)	34(1)
C(27)	-4643(2)	-7440(1)	-10400(1)	27(1)
C(28)	-3326(2)	-5048(1)	-10301(1)	27(1)
C(29)	-2162(2)	-4782(1)	-10862(1)	21(1)
C(30)	-1634(2)	-3926(1)	-10861(1)	26(1)
C(31)	-567(2)	-3669(1)	-11376(1)	28(1)
C(32)	12(2)	-4266(1)	-11893(1)	28(1)
C(33)	-506(2)	-5113(1)	-11902(1)	28(1)
C(34)	-1587(2)	-5370(1)	-11389(1)	25(1)
O(35)	-255(1)	-3966(1)	-6513(1)	20(1)
C(35)	524(2)	-3542(1)	-7150(1)	22(1)
C(36)	1565(2)	-2857(1)	-6860(1)	18(1)
C(37)	1061(2)	-2016(1)	-6712(1)	26(1)
C(38)	2043(2)	-1371(1)	-6484(1)	33(1)
C(39)	3513(2)	-1563(1)	-6403(1)	31(1)
C(40)	4034(2)	-2392(1)	-6544(1)	26(1)
C(41)	3049(2)	-3042(1)	-6777(1)	21(1)
O(42)	-2107(1)	-4106(1)	-5571(1)	21(1)
C(42)	-3559(2)	-4146(2)	-5245(1)	33(1)
O(43)	-2656(1)	-3119(1)	-6743(1)	21(1)
C(43)	-3464(2)	-4020(1)	-4391(1)	21(1)
C(44)	-4515(2)	-4409(1)	-3928(1)	20(1)
C(45)	-4465(2)	-4293(1)	-3134(1)	24(1)
C(46)	-3377(2)	-3788(1)	-2802(1)	26(1)
C(47)	-2336(2)	-3391(1)	-3262(1)	27(1)
C(48)	-2373(2)	-3504(1)	-4056(1)	26(1)
C(49)	-5649(2)	-5964(1)	-6219(1)	18(1)
C(50)	-6622(2)	-5759(1)	-5544(1)	17(1)
C(51)	-6627(2)	-6343(1)	-4925(1)	21(1)
C(52)	-7501(2)	-6191(1)	-4290(1)	25(1)
C(53)	-8390(2)	-5452(1)	-4258(1)	25(1)
C(54)	-8411(2)	-4880(1)	-4875(1)	23(1)
C(55)	-7525(2)	-5029(1)	-5516(1)	19(1)

Bond lengths [Å] and angles [°] for 122					
P(1)-O(15)	1.4689(12)	C(22)-C(23)	1.381(2)	O(15)-P(1)-O(1)	114.67(6)
P(1)-O(14)	1.5611(11)	C(22)-C(27)	1.392(2)	O(14)-P(1)-O(1)	104.14(6)
P(1)-O(7)	1.5668(11)	C(23)-C(24)	1.389(2)	O(7)-P(1)-O(1)	102.48(6)
P(1)-O(1)	1.5750(11)	C(24)-C(25)	1.385(3)	C(1)-O(1)-P(1)	121.18(9)
O(1)-C(1)	1.4531(18)	C(25)-C(26)	1.378(3)	O(1)-C(1)-C(2)	110.70(12)
C(1)-C(2)	1.532(2)	C(26)-C(27)	1.387(3)	O(1)-C(1)-C(6)	107.29(11)
C(1)-C(6)	1.534(2)	C(28)-C(29)	1.499(2)	C(2)-C(1)-C(6)	109.82(12)
O(2)-C(2)	1.4236(19)	C(29)-C(34)	1.389(2)	C(2)-O(2)-C(21)	116.21(11)
O(2)-C(21)	1.4449(19)	C(29)-C(30)	1.398(3)	O(2)-C(2)-C(1)	110.32(12)
C(2)-C(3)	1.530(2)	C(30)-C(31)	1.380(3)	O(2)-C(2)-C(3)	108.55(12)
O(3)-C(3)	1.4112(18)	C(31)-C(32)	1.387(3)	C(1)-C(2)-C(3)	107.34(12)
O(3)-C(28)	1.4418(18)	C(32)-C(33)	1.382(3)	C(3)-O(3)-C(28)	113.59(12)
C(3)-C(4)	1.531(2)	C(33)-C(34)	1.386(3)	O(3)-C(3)-C(4)	106.45(12)
O(4)-C(4)	1.4121(18)	O(35)-C(35)	1.4691(19)	O(3)-C(3)-C(2)	113.51(12)
C(4)-C(5)	1.528(2)	C(35)-C(36)	1.500(2)	C(4)-C(3)-C(2)	110.65(12)
P(5)-O(43)	1.4657(11)	C(36)-C(41)	1.386(2)	O(4)-C(4)-C(5)	107.92(12)
P(5)-O(35)	1.5625(12)	C(36)-C(37)	1.393(2)	O(4)-C(4)-C(3)	110.79(12)
P(5)-O(42)	1.5707(11)	C(37)-C(38)	1.389(3)	C(5)-C(4)-C(3)	109.90(12)
P(5)-O(5)	1.5879(11)	C(38)-C(39)	1.375(3)	O(43)-P(5)-O(35)	116.19(7)
O(5)-C(5)	1.4470(17)	C(39)-C(40)	1.378(3)	O(43)-P(5)-O(42)	116.64(6)
C(5)-C(6)	1.526(2)	C(40)-C(41)	1.400(2)	O(35)-P(5)-O(42)	97.85(6)
O(6)-C(6)	1.4218(18)	O(42)-C(42)	1.441(2)	O(43)-P(5)-O(5)	113.53(6)
O(6)-C(49)	1.4295(18)	C(42)-C(43)	1.499(2)	O(35)-P(5)-O(5)	107.94(6)
O(7)-C(7)	1.4589(18)	C(43)-C(44)	1.386(2)	O(42)-P(5)-O(5)	102.83(6)
C(7)-C(8)	1.501(2)	C(43)-C(48)	1.394(2)	C(5)-O(5)-P(5)	122.37(9)
C(8)-C(13)	1.389(2)	C(44)-C(45)	1.392(2)	O(5)-C(5)-C(6)	107.63(12)
C(8)-C(9)	1.390(2)	C(45)-C(46)	1.381(3)	O(5)-C(5)-C(4)	108.82(12)
C(9)-C(10)	1.384(3)	C(46)-C(47)	1.383(3)	C(6)-C(5)-C(4)	111.52(12)
C(10)-C(11)	1.382(3)	C(47)-C(48)	1.390(2)	C(6)-O(6)-C(49)	111.76(11)
C(11)-C(12)	1.387(3)	C(49)-C(50)	1.505(2)	O(6)-C(6)-C(5)	110.60(11)
C(12)-C(13)	1.387(2)	C(50)-C(55)	1.390(2)	O(6)-C(6)-C(1)	110.39(12)
O(14)-C(14)	1.4763(19)	C(50)-C(51)	1.400(2)	C(5)-C(6)-C(1)	108.24(12)
C(14)-C(15)	1.505(2)	C(51)-C(52)	1.383(2)	C(7)-O(7)-P(1)	118.05(9)
C(15)-C(16)	1.390(2)	C(52)-C(53)	1.393(3)	O(7)-C(7)-C(8)	108.88(12)
C(15)-C(20)	1.390(2)	C(53)-C(54)	1.386(2)	C(13)-C(8)-C(9)	119.52(15)
C(16)-C(17)	1.375(3)	C(54)-C(55)	1.396(2)	C(13)-C(8)-C(7)	121.38(14)
C(17)-C(18)	1.395(3)	O(15)-P(1)-O(14)	115.07(6)	C(9)-C(8)-C(7)	119.05(14)
C(18)-C(19)	1.378(3)	O(15)-P(1)-O(7)	115.75(6)	C(10)-C(9)-C(8)	120.42(16)
C(19)-C(20)	1.392(3)	O(14)-P(1)-O(7)	103.07(6)	C(11)-C(10)-C(9)	120.08(18)
C(21)-C(22)	1.507(2)	C(38)-C(39)-C(40)	120.89(16)	C(26)-C(27)-C(22)	120.43(17)
C(10)-C(11)-C(12)	119.75(16)	C(39)-C(40)-C(41)	119.22(16)	O(3)-C(28)-C(29)	108.13(13)
C(13)-C(12)-C(11)	120.39(17)	C(36)-C(41)-C(40)	120.42(15)	C(34)-C(29)-C(30)	118.71(16)
C(12)-C(13)-C(8)	119.84(16)	C(42)-O(42)-P(5)	118.07(10)	C(34)-C(29)-C(28)	121.35(16)

C(14)-O(14)-P(1)	121.86(10)	O(42)-C(42)-C(43)	109.53(13)	C(30)-C(29)-C(28)	119.94(16)
O(14)-C(14)-C(15)	108.12(13)	C(44)-C(43)-C(48)	119.56(14)	C(31)-C(30)-C(29)	120.63(17)
C(16)-C(15)-C(20)	119.38(16)	C(44)-C(43)-C(42)	118.76(14)	C(30)-C(31)-C(32)	120.06(17)
C(16)-C(15)-C(14)	119.93(15)	C(48)-C(43)-C(42)	121.65(15)	C(33)-C(32)-C(31)	119.82(16)
C(20)-C(15)-C(14)	120.68(15)	C(43)-C(44)-C(45)	120.00(15)	C(32)-C(33)-C(34)	120.14(16)
C(17)-C(16)-C(15)	120.48(16)	C(46)-C(45)-C(44)	120.42(16)	C(33)-C(34)-C(29)	120.62(16)
C(16)-C(17)-C(18)	120.13(18)	C(47)-C(46)-C(45)	119.73(15)	C(35)-O(35)-P(5)	120.68(10)
C(19)-C(18)-C(17)	119.81(17)	C(46)-C(47)-C(48)	120.34(16)	O(35)-C(35)-C(36)	111.28(12)
C(18)-C(19)-C(20)	120.09(16)	C(47)-C(48)-C(43)	119.93(16)	C(41)-C(36)-C(37)	119.43(15)
C(15)-C(20)-C(19)	120.12(17)	O(6)-C(49)-C(50)	111.11(12)	C(41)-C(36)-C(35)	120.31(14)
O(2)-C(21)-C(22)	110.45(13)	C(55)-C(50)-C(51)	119.00(15)	C(37)-C(36)-C(35)	120.17(15)
C(23)-C(22)-C(27)	119.08(15)	C(55)-C(50)-C(49)	123.04(14)	C(38)-C(37)-C(36)	119.97(16)
C(23)-C(22)-C(21)	120.12(15)	C(51)-C(50)-C(49)	117.95(14)	C(39)-C(38)-C(37)	120.07(17)
C(27)-C(22)-C(21)	120.76(15)	C(52)-C(51)-C(50)	120.63(15)	C(50)-C(55)-C(54)	120.19(15)
C(22)-C(23)-C(24)	120.63(16)	C(51)-C(52)-C(53)	120.24(15)	C(26)-C(25)-C(24)	120.14(17)
C(25)-C(24)-C(23)	119.76(16)	C(54)-C(53)-C(52)	119.44(15)	C(25)-C(26)-C(27)	119.95(17)
C(53)-C(54)-C(55)	120.47(16)				

Anisotropic displacement parameters ($Å^2 \times 10^3$) for 122. The anisotropic displacement factor exponent takes the form: $-2\pi^2[h^2 a^{*2} U^{11} + ... + 2 h k a^* b^* U^{12}]$

	U^{11}	U^{22}	U^{33}	U^{23}	U^{13}	U^{12}
P(1)	13(1)	17(1)	15(1)	-2(1)	0(1)	-1(1)
O(1)	14(1)	18(1)	18(1)	0(1)	0(1)	-2(1)
C(1)	14(1)	16(1)	16(1)	0(1)	1(1)	-2(1)
O(2)	19(1)	20(1)	22(1)	-6(1)	-2(1)	2(1)
C(2)	17(1)	22(1)	15(1)	-3(1)	-1(1)	2(1)
O(3)	16(1)	36(1)	11(1)	0(1)	0(1)	1(1)
C(3)	15(1)	23(1)	14(1)	-1(1)	0(1)	1(1)
O(4)	15(1)	21(1)	21(1)	-1(1)	1(1)	-3(1)
C(4)	14(1)	18(1)	14(1)	-2(1)	-1(1)	-2(1)
P(5)	15(1)	16(1)	13(1)	-1(1)	0(1)	-1(1)
O(5)	18(1)	17(1)	14(1)	1(1)	-3(1)	-2(1)
C(5)	16(1)	19(1)	13(1)	0(1)	-3(1)	4(1)
O(6)	20(1)	17(1)	14(1)	1(1)	3(1)	1(1)
C(6)	16(1)	17(1)	13(1)	-1(1)	0(1)	1(1)
O(7)	18(1)	20(1)	17(1)	-3(1)	2(1)	-4(1)
C(7)	22(1)	22(1)	17(1)	-6(1)	2(1)	-4(1)
C(8)	19(1)	17(1)	21(1)	0(1)	3(1)	1(1)
C(9)	28(1)	31(1)	21(1)	0(1)	1(1)	-5(1)
C(10)	39(1)	37(1)	26(1)	6(1)	8(1)	-4(1)
C(11)	30(1)	24(1)	38(1)	8(1)	13(1)	-3(1)
C(12)	23(1)	24(1)	40(1)	3(1)	2(1)	-7(1)
C(13)	20(1)	22(1)	26(1)	0(1)	-1(1)	-2(1)
O(14)	20(1)	25(1)	16(1)	-3(1)	-2(1)	1(1)
C(14)	19(1)	37(1)	18(1)	-6(1)	-3(1)	-1(1)
O(15)	15(1)	20(1)	24(1)	0(1)	1(1)	-1(1)
C(15)	19(1)	27(1)	17(1)	-1(1)	-5(1)	3(1)
C(16)	27(1)	23(1)	25(1)	-3(1)	-7(1)	2(1)
C(17)	30(1)	32(1)	28(1)	8(1)	-7(1)	-3(1)
C(18)	28(1)	46(1)	20(1)	3(1)	1(1)	3(1)
C(19)	32(1)	38(1)	21(1)	-9(1)	-6(1)	5(1)

C(20)	27(1)	27(1)	23(1)	-4(1)	-6(1)	-2(1)
C(21)	22(1)	21(1)	24(1)	-6(1)	3(1)	-3(1)
C(22)	22(1)	16(1)	23(1)	-4(1)	0(1)	-2(1)
C(23)	28(1)	21(1)	22(1)	-1(1)	-1(1)	1(1)
C(24)	26(1)	21(1)	36(1)	-5(1)	7(1)	4(1)
C(25)	42(1)	24(1)	27(1)	-7(1)	11(1)	0(1)
C(26)	47(1)	32(1)	22(1)	-4(1)	-5(1)	7(1)
C(27)	28(1)	26(1)	26(1)	-6(1)	-4(1)	8(1)
C(28)	21(1)	48(1)	13(1)	2(1)	-5(1)	-5(1)
C(29)	19(1)	32(1)	13(1)	3(1)	-5(1)	-2(1)
C(30)	26(1)	31(1)	21(1)	-4(1)	-5(1)	2(1)
C(31)	27(1)	28(1)	30(1)	6(1)	-6(1)	-4(1)
C(32)	20(1)	40(1)	23(1)	11(1)	1(1)	0(1)
C(33)	26(1)	37(1)	21(1)	-1(1)	0(1)	7(1)
C(34)	30(1)	24(1)	21(1)	1(1)	-2(1)	-3(1)
O(35)	18(1)	22(1)	21(1)	2(1)	0(1)	-3(1)
C(35)	21(1)	25(1)	19(1)	-3(1)	4(1)	-8(1)
C(36)	19(1)	19(1)	15(1)	-1(1)	1(1)	-2(1)
C(37)	18(1)	25(1)	35(1)	-3(1)	-4(1)	3(1)
C(38)	32(1)	17(1)	50(1)	-5(1)	-10(1)	3(1)
C(39)	27(1)	22(1)	44(1)	1(1)	-8(1)	-8(1)
C(40)	17(1)	26(1)	34(1)	4(1)	-3(1)	-3(1)
C(41)	21(1)	18(1)	23(1)	1(1)	1(1)	1(1)
O(42)	19(1)	29(1)	14(1)	-1(1)	-2(1)	-3(1)
C(42)	23(1)	60(1)	16(1)	-2(1)	3(1)	-14(1)
O(43)	24(1)	19(1)	21(1)	-1(1)	-1(1)	2(1)
C(43)	24(1)	26(1)	15(1)	0(1)	-2(1)	-2(1)
C(44)	23(1)	19(1)	20(1)	1(1)	0(1)	2(1)
C(45)	28(1)	26(1)	19(1)	7(1)	5(1)	5(1)
C(46)	32(1)	34(1)	14(1)	-1(1)	0(1)	7(1)
C(47)	34(1)	27(1)	21(1)	-4(1)	-8(1)	-2(1)
C(48)	28(1)	31(1)	19(1)	2(1)	-1(1)	-8(1)
C(49)	21(1)	17(1)	17(1)	1(1)	0(1)	1(1)
C(50)	16(1)	20(1)	14(1)	-1(1)	-2(1)	-3(1)
C(51)	21(1)	20(1)	23(1)	4(1)	-2(1)	-1(1)
C(52)	28(1)	28(1)	19(1)	7(1)	1(1)	-5(1)
C(53)	25(1)	32(1)	18(1)	-2(1)	5(1)	-6(1)
C(54)	21(1)	25(1)	24(1)	-2(1)	2(1)	-1(1)
C(55)	21(1)	19(1)	18(1)	1(1)	-1(1)	0(1)

Hydrogen coordinates (× 10^4) and isotropic displacement parameters (Å^2 × 10^3) for 122

	x	y	z	U(eq)
H(1A)	-6132	-4881	-8229	18
H(2A)	-5373	-5778	-9303	21
H(3A)	-4093	-4450	-9016	21
H(4O)	-617(11)	-4456(15)	-8472(12)	38(6)
H(4A)	-2067	-5513	-8141	18
H(5A)	-4013	-4175	-7564	19
H(6A)	-4271	-6035	-7334	18
H(7A)	-10012	-5574	-7066	24
H(7B)	-8693	-6183	-6765	24
H(9A)	-9921	-6509	-5564	32
H(10A)	-11620	-7360	-4901	41
H(11A)	-13462	-8101	-5572	37
H(12A)	-13588	-7998	-6912	35
H(13A)	-11889	-7147	-7581	27
H(14A)	-9948	-5851	-9698	29
H(14B)	-10104	-6884	-9828	29

H(16A)	-7631	-5131	-10260	30
H(17A)	-6220	-5012	-11365	36
H(18A)	-6215	-6143	-12274	38
H(19A)	-7616	-7391	-12061	37
H(20A)	-9038	-7512	-10947	31
H(21A)	-5809	-7179	-9057	27
H(21B)	-4713	-7770	-8554	27
H(23A)	-2367	-8272	-9155	28
H(24A)	-1193	-8732	-10277	33
H(25A)	-2183	-8370	-11478	37
H(26A)	-4352	-7569	-11556	41
H(27A)	-5531	-7116	-10434	32
H(28A)	-4090	-4591	-10266	33
H(28B)	-3799	-5597	-10472	33
H(30A)	-2013	-3517	-10502	31
H(31A)	-229	-3083	-11377	34
H(32A)	763	-4093	-12239	33
H(33A)	-121	-5520	-12260	34
H(34A)	-1937	-5953	-11397	30
H(35A)	-200	-3271	-7503	26
H(35B)	1080	-3985	-7443	26
H(37A)	45	-1883	-6766	31
H(38A)	1700	-797	-6385	40
H(39A)	4178	-1118	-6248	37
H(40A)	5050	-2520	-6483	31
H(41A)	3399	-3613	-6880	25
H(42A)	-4012	-4718	-5361	40
H(42B)	-4185	-3685	-5475	40
H(44A)	-5269	-4755	-4153	24
H(45A)	-5183	-4564	-2818	29
H(46A)	-3344	-3714	-2259	32
H(47A)	-1593	-3039	-3035	33
H(48A)	-1656	-3229	-4370	31
H(49A)	-4637	-6081	-6033	22
H(49B)	-6014	-6495	-6480	22
H(51A)	-6024	-6848	-4942	26
H(52A)	-7496	-6592	-3873	30
H(53A)	-8976	-5342	-3818	30
H(54A)	-9033	-4382	-4862	28
H(55A)	-7540	-4630	-5934	23

Torsion angles [°] for 122			
O(15)-P(1)-O(1)-C(1)	-28.76(12)	C(18)-C(19)-C(20)-C(15)	0.1(3)
O(14)-P(1)-O(1)-C(1)	97.86(11)	C(2)-O(2)-C(21)-C(22)	102.83(15)
O(7)-P(1)-O(1)-C(1)	-155.01(10)	O(2)-C(21)-C(22)-C(23)	80.15(18)
P(1)-O(1)-C(1)-C(2)	-92.86(13)	O(2)-C(21)-C(22)-C(27)	-97.85(18)
P(1)-O(1)-C(1)-C(6)	147.33(10)	C(27)-C(22)-C(23)-C(24)	0.9(3)
C(21)-O(2)-C(2)-C(1)	93.83(14)	C(21)-C(22)-C(23)-C(24)	-177.19(15)
C(21)-O(2)-C(2)-C(3)	-148.81(12)	C(22)-C(23)-C(24)-C(25)	-0.1(3)
O(1)-C(1)-C(2)-O(2)	-63.48(15)	C(23)-C(24)-C(25)-C(26)	-0.5(3)
C(6)-C(1)-C(2)-O(2)	54.80(15)	C(24)-C(25)-C(26)-C(27)	0.3(3)
O(1)-C(1)-C(2)-C(3)	178.42(11)	C(25)-C(26)-C(27)-C(22)	0.4(3)
C(6)-C(1)-C(2)-C(3)	-63.30(15)	C(23)-C(22)-C(27)-C(26)	-1.0(3)
C(28)-O(3)-C(3)-C(4)	-165.88(13)	C(21)-C(22)-C(27)-C(26)	177.00(16)
C(28)-O(3)-C(3)-C(2)	72.15(17)	C(3)-O(3)-C(28)-C(29)	151.76(14)
O(2)-C(2)-C(3)-O(3)	61.21(15)	O(3)-C(28)-C(29)-C(34)	102.50(18)
C(1)-C(2)-C(3)-O(3)	-179.55(11)	O(3)-C(28)-C(29)-C(30)	-77.39(19)
O(2)-C(2)-C(3)-C(4)	-58.39(15)	C(34)-C(29)-C(30)-C(31)	0.4(2)

C(1)-C(2)-C(3)-C(4)	60.86(15)	C(28)-C(29)-C(30)-C(31)	-179.75(14)
O(3)-C(3)-C(4)-O(4)	59.49(15)	C(29)-C(30)-C(31)-C(32)	-1.2(2)
C(2)-C(3)-C(4)-O(4)	-176.75(12)	C(30)-C(31)-C(32)-C(33)	1.5(3)
O(3)-C(3)-C(4)-C(5)	178.67(12)	C(31)-C(32)-C(33)-C(34)	-1.0(3)
C(2)-C(3)-C(4)-C(5)	-57.57(16)	C(32)-C(33)-C(34)-C(29)	0.1(3)
O(43)-P(5)-O(5)-C(5)	13.32(13)	C(30)-C(29)-C(34)-C(33)	0.2(2)
O(35)-P(5)-O(5)-C(5)	-116.99(11)	C(28)-C(29)-C(34)-C(33)	-179.70(15)
O(42)-P(5)-O(5)-C(5)	140.25(11)	O(43)-P(5)-O(35)-C(35)	-33.11(13)
P(5)-O(5)-C(5)-C(6)	-141.40(10)	O(42)-P(5)-O(35)-C(35)	-158.02(11)
P(5)-O(5)-C(5)-C(4)	97.60(13)	O(5)-P(5)-O(35)-C(35)	95.70(11)
O(4)-C(4)-C(5)-O(5)	-64.37(15)	P(5)-O(35)-C(35)-C(36)	119.15(13)
C(3)-C(4)-C(5)-O(5)	174.71(12)	O(35)-C(35)-C(36)-C(41)	98.57(17)
O(4)-C(4)-C(5)-C(6)	177.05(11)	O(35)-C(35)-C(36)-C(37)	-84.93(19)
C(3)-C(4)-C(5)-C(6)	56.13(16)	C(41)-C(36)-C(37)-C(38)	0.1(3)
C(49)-O(6)-C(6)-C(5)	-124.00(13)	C(35)-C(36)-C(37)-C(38)	-176.44(17)
C(49)-O(6)-C(6)-C(1)	116.24(13)	C(36)-C(37)-C(38)-C(39)	-0.2(3)
O(5)-C(5)-C(6)-O(6)	61.66(15)	C(37)-C(38)-C(39)-C(40)	-0.1(3)
C(4)-C(5)-C(6)-O(6)	-179.05(11)	C(38)-C(39)-C(40)-C(41)	0.5(3)
O(5)-C(5)-C(6)-C(1)	-177.28(11)	C(37)-C(36)-C(41)-C(40)	0.3(2)
C(4)-C(5)-C(6)-C(1)	-58.00(15)	C(35)-C(36)-C(41)-C(40)	176.79(15)
O(1)-C(1)-C(6)-O(6)	-56.52(15)	C(39)-C(40)-C(41)-C(36)	-0.6(3)
C(2)-C(1)-C(6)-O(6)	-176.90(11)	O(43)-P(5)-O(42)-C(42)	56.67(14)
O(1)-C(1)-C(6)-C(5)	-177.70(11)	O(35)-P(5)-O(42)-C(42)	-178.75(13)
C(2)-C(1)-C(6)-C(5)	61.92(15)	O(5)-P(5)-O(42)-C(42)	-68.24(13)
O(15)-P(1)-O(7)-C(7)	-57.06(12)	P(5)-O(42)-C(42)-C(43)	-160.74(12)
O(14)-P(1)-O(7)-C(7)	176.43(10)	O(42)-C(42)-C(43)-C(44)	-150.70(15)
O(1)-P(1)-O(7)-C(7)	68.48(11)	O(42)-C(42)-C(43)-C(48)	31.1(2)
P(1)-O(7)-C(7)-C(8)	175.62(10)	C(48)-C(43)-C(44)-C(45)	-0.9(2)
O(7)-C(7)-C(8)-C(13)	-29.1(2)	C(42)-C(43)-C(44)-C(45)	-179.10(16)
O(7)-C(7)-C(8)-C(9)	153.24(14)	C(43)-C(44)-C(45)-C(46)	0.3(2)
C(13)-C(8)-C(9)-C(10)	0.7(3)	C(44)-C(45)-C(46)-C(47)	0.4(3)
C(7)-C(8)-C(9)-C(10)	178.36(16)	C(45)-C(46)-C(47)-C(48)	-0.6(3)
C(8)-C(9)-C(10)-C(11)	-0.4(3)	C(46)-C(47)-C(48)-C(43)	0.0(3)
C(9)-C(10)-C(11)-C(12)	0.2(3)	C(44)-C(43)-C(48)-C(47)	0.7(3)
C(10)-C(11)-C(12)-C(13)	-0.2(3)	C(42)-C(43)-C(48)-C(47)	178.87(17)
C(11)-C(12)-C(13)-C(8)	0.4(3)	C(6)-O(6)-C(49)-C(50)	-178.16(11)
C(9)-C(8)-C(13)-C(12)	-0.7(3)	O(6)-C(49)-C(50)-C(55)	8.5(2)
C(7)-C(8)-C(13)-C(12)	-178.29(15)	O(6)-C(49)-C(50)-C(51)	-172.57(13)
O(15)-P(1)-O(14)-C(14)	-31.02(14)	C(55)-C(50)-C(51)-C(52)	-0.9(2)
O(7)-P(1)-O(14)-C(14)	95.92(12)	C(49)-C(50)-C(51)-C(52)	-179.83(15)
O(1)-P(1)-O(14)-C(14)	-157.38(12)	C(50)-C(51)-C(52)-C(53)	0.0(2)
P(1)-O(14)-C(14)-C(15)	127.49(12)	C(51)-C(52)-C(53)-C(54)	1.2(3)
O(14)-C(14)-C(15)-C(16)	-73.47(19)	C(52)-C(53)-C(54)-C(55)	-1.5(2)
O(14)-C(14)-C(15)-C(20)	106.48(17)	C(51)-C(50)-C(55)-C(54)	0.6(2)
C(20)-C(15)-C(16)-C(17)	0.3(2)	C(49)-C(50)-C(55)-C(54)	179.48(15)
C(14)-C(15)-C(16)-C(17)	-179.75(15)	C(53)-C(54)-C(55)-C(50)	0.6(2)
C(15)-C(16)-C(17)-C(18)	0.0(3)	C(16)-C(15)-C(20)-C(19)	-0.3(2)
C(16)-C(17)-C(18)-C(19)	-0.2(3)	C(14)-C(15)-C(20)-C(19)	179.73(15)
C(17)-C(18)-C(19)-C(20)	0.2(3)		

Hydrogen bonds for 122 [Å and °]				
D-H...A	d(D-H)	d(H...A)	d(D...A)	<(DHA)
O(4)-H(4O)...O(15)#1	0.977(3)	1.855(8)	2.7939(16)	160(2)
Symmetry transformations used to generate equivalent atoms: #1 x+1,y,z				