Studies of the Synthesis of Cyclodextrins with Novel Stereochemistry

A Thesis Presented for the Degree of
Doctor of Philosophy, Ph. D.

by

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ABSTRACT

Proton-deuterium exchange of the carbinol hydrogens of *gluco*-, *manno*- and *galacto*-methyl-α-D-pyranosides with deuterium oxide catalysed by ultrasonicated Raney nickel had low regioselectivity. 4,6-*O*-Benzylidene-methyl-α-D-glucopyranoside underwent deuteration at C-2 and C-3 and cleavage of the benzylidene group, whereas 4,6-*O*-isopropylidene-methyl-α-D-glucopyranoside was deuterated exclusively at C-2 (^{2}H-NMR), which provides a rapid and economical route to 2-[$^2H_1$]-methyl-α-D-glucopyranoside.

The diol moiety of 4,6-*O*-benzylidene-methyl-α-D-glucopyranoside was cleaved under aqueous (sodium periodate), and non-aqueous (periodic acid) conditions to give a D-erythritol in high yield (80%, 90%). Application of similar conditions to *heptakis*(6-*O*-*tert*-butyldimethylsilyl)-β-cyclodextrin failed to give an isolable product.

The epoxides; methyl 2,3-anhydro-4,6-*O*-benzylidene-methyl-α-D-mannopyranoside and *heptakis*(2,3-anhydro)-β-cyclomannin were prepared *via* the C2-tosylates. Both reacted with 4-*tert*-butylbenzylthiol at the 3-position. The cyclomannin gave an adduct with $^1C_4$-*altro* stereochemistry as determined by NMR and the homogeneity was established by MALDI-MS ($M+K^+$, *m/z* 3109.5, $C_{161}H_{266}O_{28}S_7Si_7K$). The mannopyranoside similarly reacted with L-cysteine to give a $^4C_1$-*altro*-amino acid sugar adduct, but the cyclomannin failed to react.

The benzylidene-, isopropylidene- glucopyranosides and the silylated cyclodextrin mentioned above underwent allylation with allyl bromide, to give 2,3-di-*O*-allyl-glucopyranosides and heptakis(2,3-di-*O*-allyl)-β-CD respectively. Ring closing metathesis (RCM) with Grubb's 2nd generation catalyst of the allylated cyclodextrin yielded a complex mixture, but the allylated glucopyranosides yielded the cyclo-octenes, which were hydrogenated. Unexpectedly, the benzylidene protecting group was partially cleaved during RCM. All NMR spectra were assigned by correlation methods.

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To my Parents, Afshin, Arezoo and Armin

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ABBREVIATIONS

Ac	Acetyl
APCI	Atmospheric pressure chemical ionization
Aq.	Aqueous
Bn	Benzyl
BOC	*tert*-butoxycarbonyl
Bu	Butyl
Bz	Benzoyl
COSY	Correlation spectroscopy
Cys.	Cysteine
Δ	Reflux
$\theta°$	Angle in degrees
DBU	1,8-Diazabicyclo[5.4.0]undec-7-ene
DEPT	Distortionless enhancement of polarisation transfer
DIBAL	Diisobutylaluminum hydride
DMAP	4-Dimethylaminopyridine
DMF	Dimethyl formamide
DMSO	Dimethyl sulfoxide
EI/CI	Electron ionization/ Chemical ionization
ES	Electrospray (ionisation)
Et	Ethyl
Et_2O	Diethyl ether
EtOAc	Ethyl acetate
Equiv. or eq.	Equivalent
FAB-MS	Fast atom bombardment mass spectroscopy
g	gram
HMBC	Heteronuclear multiple-bond correlation
HPLC	High pressure (performance) liquid chromatography
HRMS	High resolution mass spectroscopy
HSQC	Heteronuclear single-quantum correlation
Hz	Hertz
IR	Infrared
J	Coupling constant (in Hz)
MALDI	Matrix-assisted laser desorption ionization
m.p.	Melting point
MS	Mass spectroscopy
Ms	Mesyl
m/z	mass to charge ratio
NBS	*N*-bromosuccinimide
Nu	Nucleophile

OMe	Methoxy
Ph	Phenyl
Ppm	Parts per million
pTsOH	*p*-toluene sulphonic acid
Py	pyridine
R	Specific substituent
r.t.	Room temperature
T	Time
TBDMS	*tert*-butyldimethylsilyl
TBDPS	*tert*-butyldiphenylsilyl
tert	tertiary
TFA	Trifluoroacetic acid
THF	Tetrahydrofuran
TLC	Thin layer chromatography
Ts	Tosyl

PREAMBLE

Compounds are numbered sequentially from the start of the thesis. If an acetylated compound is described, the non-acetylated compound is designated **Xa** and the acetylated compound **Xb**. The meaning of all other Markush formulae is shown in the diagrams.

To avoid numerous very similar formulae and an excessively complicated numbering system all *O*-deuterated alcohols are shown as OH rather than OD, unless the presence of the latter is essential to the argument being made. Otherwise for example, different structures would have to be shown for NMR spectra run in $CDCl_3$ and D_2O.

Compound numbers shown in brackets refer to related structures or to non-explicitly cited structures.

Chapter 1

Carbohydrate deuteration

Introduction

1. Carbohydrates and their importance

1.1. Introduction

Carbohydrates are the most abundant class of organic compounds found in living organisms. They play important structural and functional roles at the surface of biological membranes. They function as receptors for a variety of hormones and play critical roles in cell identification and differentiation. They form the major component of shells of insects, crabs, and lobsters, and are present as parts of basically all cell walls; from microbes to mammals[1].

In the natural world, carbohydrates are created by green plants or micro-organisms by photosynthesis which is a complex process in which sunlight provides the energy to convert CO_2 into glucose. This occurs in organelles called chloroplasts, which is believed were originally free living organisms.

$$6\,CO_2 + 6\,H_2O \xrightarrow{\text{Sunlight}} 6\,O_2 + \underset{\text{Glucose}}{C_6H_{12}O_6}$$

In the nineteenth century, the word *carbohydrate* (literally ''hydrate of carbon'') was applied to soluble sugars that were thought to consist of the carbon and water and hence had the formula $(CH_2O)_x$[2]. Today the general term 'carbohydrate' includes monosaccharides, oligosaccharides and polysaccharides, which contain many functional groups, at least one carbonyl or (hemi)acetal group and several hydroxyl functions per monosaccharide[3].

There are also a number of substances derived from monosaccharides by reduction of the carbonyl group (alditols), by oxidation of one or more terminal groups to form carboxylic acids; e.g. aldonic, uronic and aldaric acids by oxidation at C-1, C-n, and both C-1 and C-n respectively. If sugars have the hydroxyl groups replaced by hydrogen, amine, thiol or other heteroatomic groups they are known as deoxy-sugars[4].

The hexoses, D-glucose, D-mannose and D-galactose and the pentose, ribose are the most biologically important sugars. If n is the number of carbons in an aldose, the number of chiral centres will be at least $n - 2$. All aldoses except glyceraldehyde, exist as internal hemiacetals, consequently the number of chiral centres is $n - 1$. Some alditols (e.g. allitol) contain numerous chiral centres, but nevertheless are achiral because the molecule contains a plane of symmetry.

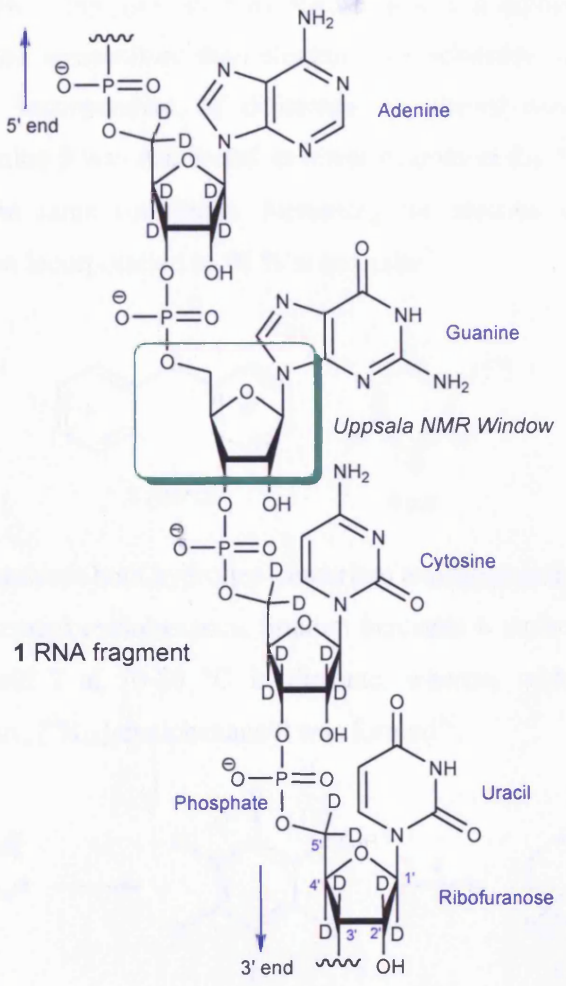
1.2. Deuterium NMR

Substitution by deuterium in specific positions (labeling) is used extensively for the study of chemical and biochemical reaction mechanisms and also to simplify ^{1}H-NMR spectra by removing both overlap of resonances and/or coupling constants.

The detection of deuterium by NMR in natural materials is difficult, because the abundance of deuterium is only 0.015% and the receptivity is low (1.50×10^{-6} relative to ^{1}H = 1 at 99.9845 % abundance). With 100 % enrichment of deuterium the receptivity (9.65×10^{-3}) is comparable to that of ^{13}C (8.78×10^{-3}) at natural abundance (1.1 %). The magnetogyric ratio (γ) of deuterium is 4.1066×10^{7} rad T^{-1} s^{-1} which is about 15 % of that of protons (26.7519×10^{7} rad T^{-1} s^{-1}) and so for example deuterium resonates at 61.4 MHz in a 400 MHz ^{1}H-NMR spectrometer[5]. Deuterium-deuterium coupling constants are very small and about only 2.5 % of proton-proton couplings. However, ^{2}H-^{1}H coupling constants which are about 15 % of the corresponding ^{1}H-^{1}H couplings can be observed in partially deuterated compounds and are typically 1 – 2 Hz. Deuterium is a spin = 1 nucleus (^{1}H spin = ½) hence it usually yields broad signals with line widths between 0.5 - 2 KHz with similar chemical shifts and an identical chemical shift range to those of protons[6,7]. In ^{13}C-NMR spectra, carbons bearing a deuterium appear as a triplet ($^{1}J_{D,C}$ = 20 - 25 Hz) with equal intensities due to coupling to the spin = 1 nucleus. If carbons are present which bear protons and normal proton decoupling is used, the intensity of these carbons will be increased by the nOe effect, which typically results in an increase in signal to noise of about two fold. The carbon bearing deuterium does not benefit from the nOe effect and is split three fold, hence the signal to noise ratio is decreased six fold relative to a similar protonated carbon. In practice, unless the sample is very concentrated the carbon signal "disappears" into the noise.

The NMR spectra of di- and tri- and oligonucleotides which are used in the synthesis of RNA **1** (ribonucleic acid) and DNA (deoxyribonucleic acid), have complex spectra due to overlapping proton resonances particularly from the ribofuranoside moiety. This problem has been overcome by preparing oligonucleotides in which all the ribofuranosides are deuterated except one. By varying the position of the non-deuterated ribofuranoside, a series of ^{1}H-NMR spectra can be acquired, which together show the complete chemical shifts and coupling constants of the whole oligonucleotide. Similarly by preparing an oligonucleotide which bears one non-deuterated ribofuranoside and an adjacent one bearing one or more hydrogens,

intraresidue nOe measurements can be made. This technique has been called the *Uppsala Window*[8,9].



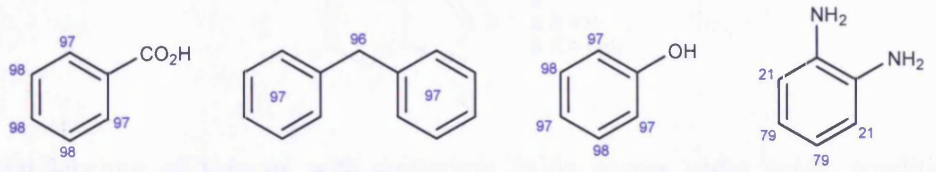
Scheme: The presence of a single non-deuterated nucleotide in deuterated RNA oligomers enables the assignment of chemical shifts and coupling constants[8].

1.2.1. Proton-deuterium exchange reactions of non-carbohydrates

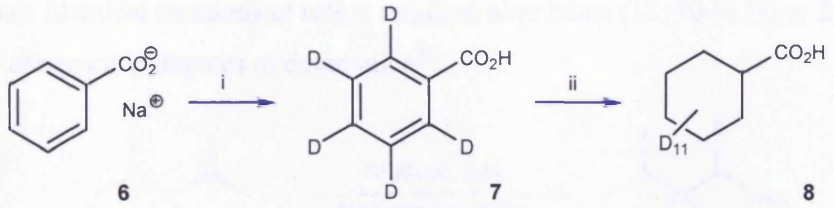
Deuteration can be achieved by chemical reaction (e.g. reduction of ketones to alcohols) or by exchange reactions. Deuterated reagents (e.g. $NaBD_4$) are expensive; consequently reagents which undergo exchange with a cheap source of deuterium (usually D_2O) are particularly useful. For example aromatic ketones are selectively reduced to deuterium labeled alcohols by a titanocene complex in D_2O[10].

Hydrogen on the surface of platinum on charcoal (Pt/C) undergoes rapid exchange with D_2O, hence it is possible to deuterate substrates using this catalyst without using deuterium gas. For

example, hydrogen-deuterium exchange of aromatic and alkyl side protons with D_2O is catalysed by 5 % Pt/C under hydrogen in a sealed tube. Deuteration of electron deficient/neutral aromatic compounds such as benzoic acid **2** or diphenylmethane **3** needs to be run in higher reaction temperature than electron rich substrates such as phenol **4** or *o*-phenylene diamine **5**. Incorporation of deuterium in phenol was high even at room temperature, while diamine **5** was deuterated in lower degrees at the 3 and 6 positions of the aromatic ring under the same conditions. Increasing the reaction temperature to 80 °C, increased the deuteration incorporation to 98 % at each site[11].



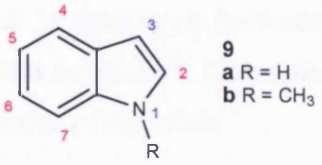
Raney nickel in D_2O catalyses both hydrogen-deuterium exchange and deuterium reduction of aromatics to fully deuterated cyclohexanes. Sodium benzoate **6** underwent exchange to give pentadeuterobenzoic acid **7** at 70-80 °C in dioxane, whereas with the same catalyst in deuterium oxide at reflux, [$^2H_{11}$]-cyclohexane **8** was formed[12].



Scheme: Reaction condition, i) deuterated Raney nickel, dioxane, 70 - 80 °C, 70% ii) deuterated Raney nickel, D_2O, reflux, 70 %[12]

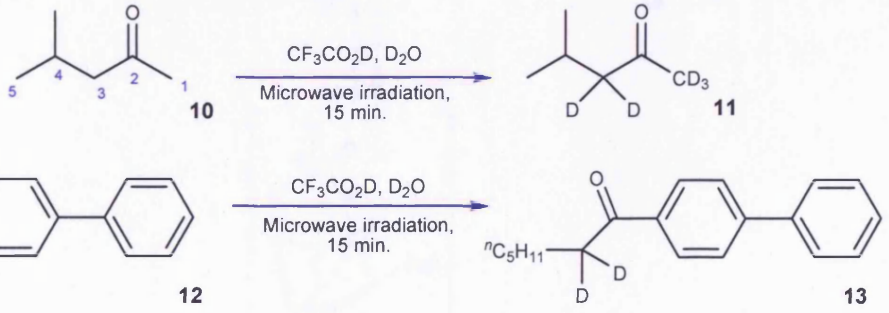
In recent years, the use of microwave irradiation has increased as an efficient source of energy in synthetic chemistry. Domestic and industrial microwaves have frequencies of 2.45 GHz with wavelengths of around 12.2 cm. In microwave heating, the reactants and solvents directly heats without heating the reaction vessel. The energy from the microwave irradiation is absorbed by molecules of solvents and solutes in the reaction mixture, by dipole rotation or ionic conduction which results in an increase in temperature[13]. Hydrogen-deuterium exchange of indole **9a** and *N*-methylindole **9b** catalysed by deuterated Raney nickel is promoted by microwave irradiation. D_2O, CD_3OD, acetone-d_6 and chloroform-*d* were tested as solvents/deuterium donors. More polar solvents, which have stronger interactions with microwaves such as deuterium oxide and methanol-d_4, gave the highest degree of deuterium

incorporation, but without regioselectivity. Whereas in $CDCl_3$, indole **9a** was deuterated regioselectively at C-3 (26 %) and *N*-methylindole **9b** was deuterated at both C-3 (89 %) and the *N*-methyl group (65 %) after 10 and 6 minutes microwave irradiation respectively. Higher deuterium incorporation was observed at both sites (*N*-methyl group, 86 %, C-3, 80 %) when *N*-methylindole **9b** was deuterated in D_2O and catalysed by Raney nickel at 40°C, but this required a reaction time of one week. In the case of indole **9a** a lower degree of deuterium incorporation was obtained at C-3 (75 % D)[14].



9
a R = H
b R = CH_3

Deuterium labeling of ketones with deuterium oxide occurs under acidic conditions *via* enolisation and can be enhanced by microwave irradiation. 4-Methyl-2-pentanone **10** and 4-*n*-heptanoylbiphenyl **12** were dissolved in trifluoroacetic acid-d_1 and a large excess of D_2O and irradiated for 15 minutes. For 4-methyl-2-pentanone **10** both the α-keto-methyl and -methylene groups were deuterated (85 % incorporation), whereas with 4-*n*-heptanoyl-biphenyl **12** incorporation occurred exclusively at the α-keto-methylene group (75 % incorporation). Identical reactions at reflux required nine hours (**11**, 90 % D) or 24 hours (**13**, 68 % D) for comparable degrees of deuteration[15].

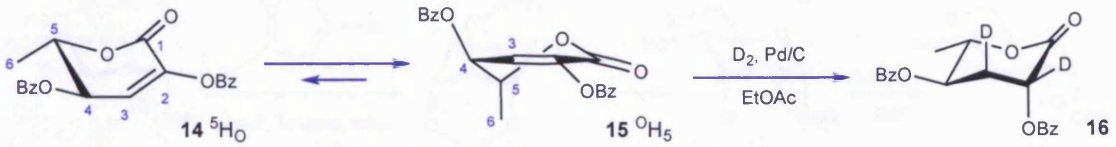


1.2.2. The synthesis of deuterated carbohydrates

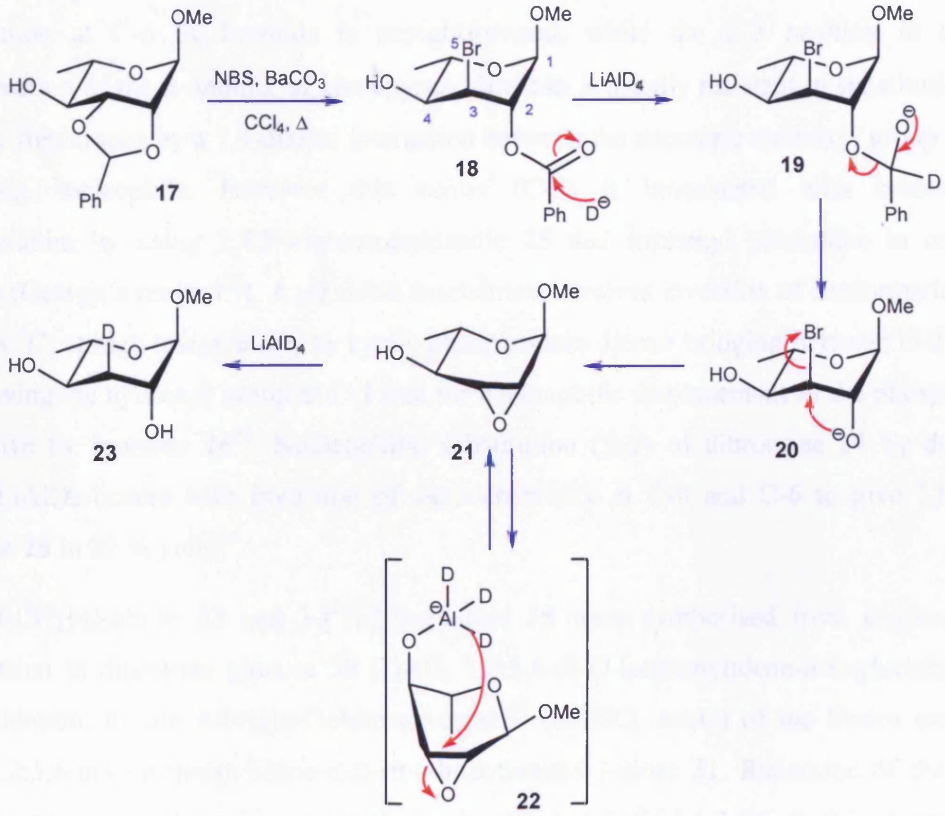
1.2.2.1. Synthesis of deuterated carbohydrates, by reduction

Catalytic deuteration of unsaturated sugars is a convenient method to deuterate alkenes and is most valuable if addition is selective for one face of the alkene group. For instance reduction of the 2,3-unsaturated lactone **14, 15** using deuterium gas catalysed by Pd/C results in stereospecific 2*R*,3*S*-deuterium labeling and formation of 2,3-[$^2H_2$]-2,4-di-*O*-benzoyl-3,6-

dideoxy-L-*arabino*-hexono-1,5-lactone **16** (80% yield)[16]. This is remarkable because it occurs *syn* to the allylic substituent. However the ^{1}H-NMR coupling constants $^3J_{3,4}$ and $^3J_{4,5}$ are both 5 Hz, which is consistant with a $^OH_5$ conformation **15** in which the 4-*O*-benzoate is pseudo-axial and the 6-methyl group is axial which disfavours approach to the catalytic surface[17].

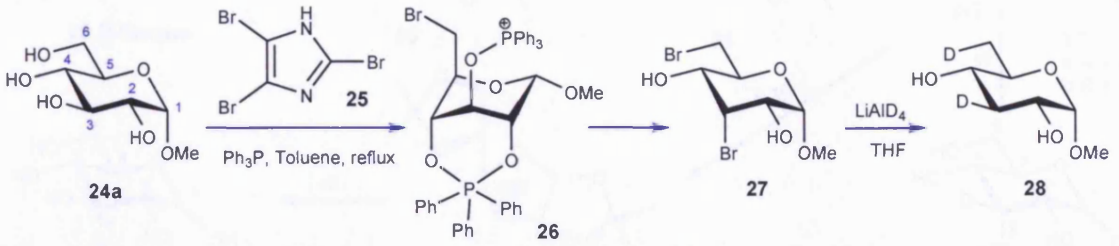


Organohalides are easily reduced to deuterated hydrocarbons by reduction with lithium aluminium deuteride or tri-*n*-butyltin deuteride[18]. Consequently to apply this methodology to sugars, the major issue is incorporation of the halide[19].



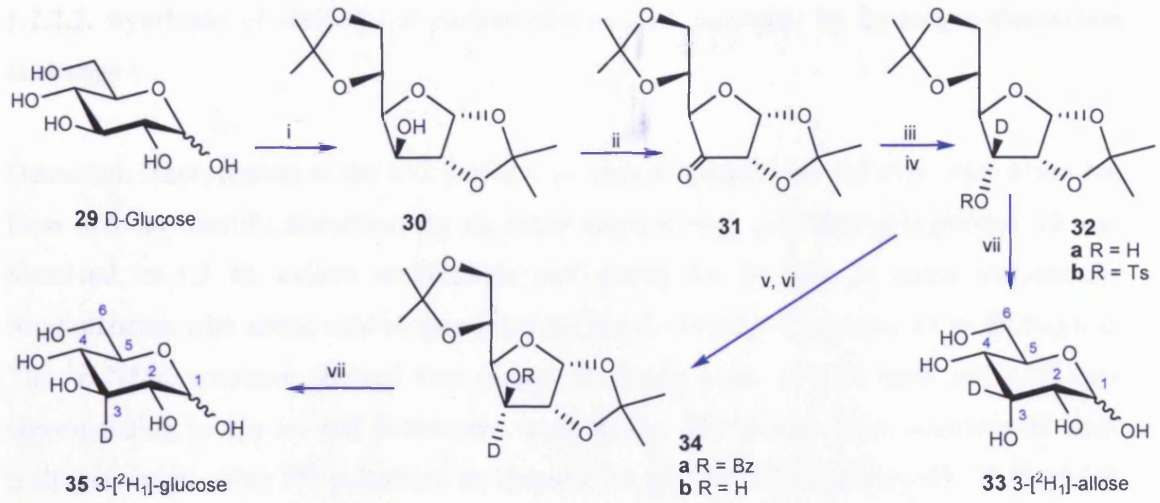
Treatment of the benzylidene sugar **17** with NBS (Hannessian-Hullar reaction[20,21]) gave the bromobenzoate-sugar **18** which was reduced by lithium aluminium deuteride with retention of configuration. The benzoyl group is reduced by $LiAlD_4$ to the alkoxide **20**, which undergoes intramolecular nucleophilic displacement of the bromo-substituent to give the epoxide **21**, facilitated by the *trans*-relationship between the bromo- and alkoxide-substituents. Attack of deuterium occurs axially from the unhindered top face of the ring to C-3, to give 3-[$^2H_1$]-3,6-

dideoxy-α-L-*arabino*-hexopyranoside **23**. This can be rationalized as *trans*-diaxial ring opening or intramolecular donation of hydride from a 4-*O*-aluminum alkoxide **22**, albeit this requires a ring flip which places the substituents at C-4 and C-5 in axial positions.



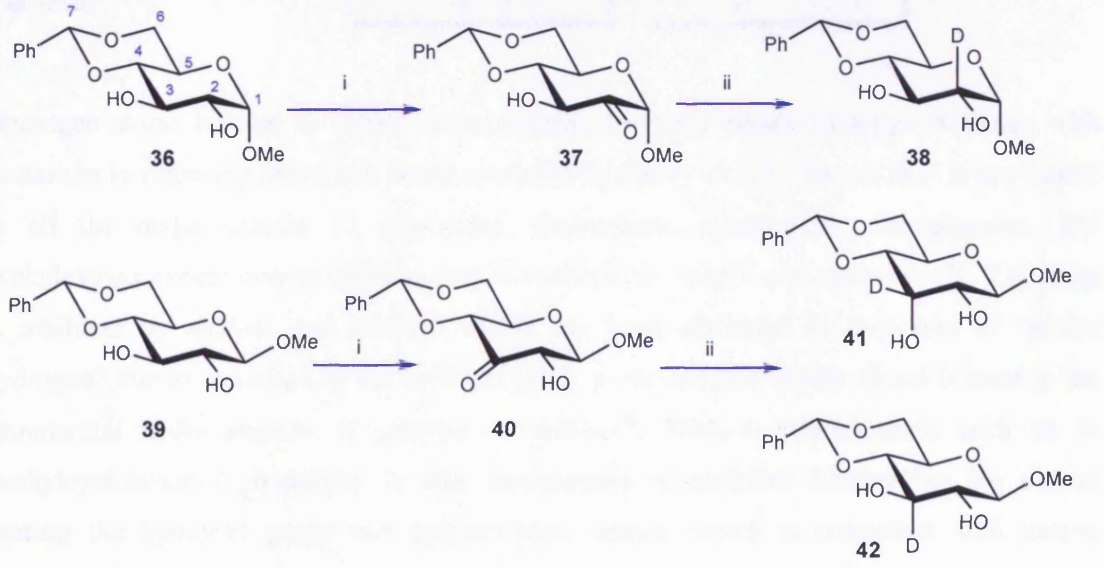
Deuterium was incorporated at both C-3 and C-6 positions of 3,6-[$^2H_2$]-paratose **28** by reduction of 3,6-dibromo-allopyranoside **27** with lithium aluminium deuteride. Nucleophilic substitution at C-6 by bromide is straightforward, while the C-3 position in the $^4C_1$ conformation of the α-anomer of glucopyranoside **24a** is usually resistant to substitution, due to steric hinderance by a 1,3-diaxial interaction between the anomeric methoxyl group and the incoming nucleophile. However this center (C-3) is brominated with inversion of configuration by using 2,4,5-tribromoimidazole **25** and triphenyl phosphine in refluxing toluene (Garegg's reagent[22]). A plausible mechanism involves inversion of conformation from $^4C_1$ to a $^1C_4$ at high temperature, by cyclic phosphonium diester bridging between *O*-2 and *O*-4[23], leaving the hydroxyl group at C-3 free for nucleophilic displacement as the phosphonium derivative by bromide **26**[24]. Nucleophilic substitution (S_N2) of dibromide **27** by deuteride using $LiAlD_4$ occurs with inversion of stereochemistry at C-3 and C-6 to give 3,6-[$^2H_2$]-paratose **28** in 22 % yield[16].

Both 3-[$^2H_1$]-D-allose **33** and 3-[$^2H_1$]-D-glucose **35** were synthesised from D-glucose **29**. Conversion to diacetone glucose **30** (DAG, 1,2:5,6-di-*O*-isopropylidene-α-D-glucofuranose) and oxidation, by the Albright-Goldman variant[25] (DMSO, Ac_2O) of the Swern oxidation, gave 1,2:5,6-di-*O*-isopropylidene-α-D-ribo-hexofuranos-3-ulose **31**. Reduction of the ketose **31** with sodium borodeuteride stereoselectively afforded 3-[$^2H_1$]-1,2:5,6-di-*O*-isopropylidene-α-D-allofuranose **32a** by delivery of deuteride from the less hindered "top" face and acidic hydrolysis yielded 3-[$^2H_1$]-D-allose **33**. Alternatively the stereochemistry at C-3 was inverted, by conversion to the toluene-*p*-sulfonate **32b** and treatment with sodium benzoate in hot DMF. This resulted in S_N2 inversion at C-3 to give the D-*gluco*-derivative **34b**. Alkaline hydrolysis of the benzoate **34b** and acidic hydrolysis of the isopropylidene groups gave 3-[$^2H_1$]-D-glucose **35**[26].



Scheme: Reaction conditions: i) Acetone, HCl[27], ii) dimethyl sulfoxide, acetic anhydride, iii) $NaBD_4$, MeOH, 69 %, iv) *p*-TsCl, pyridine, quantitative yield, v) NaOBz, DMF, reflux, 24 hr., vi) NaOMe, MeOH, 2 hr., 85 %, vii) Amberlyst resin, water, heating, 5 hr, (**31**, quantitative yield; **29**, 83 %)[26].

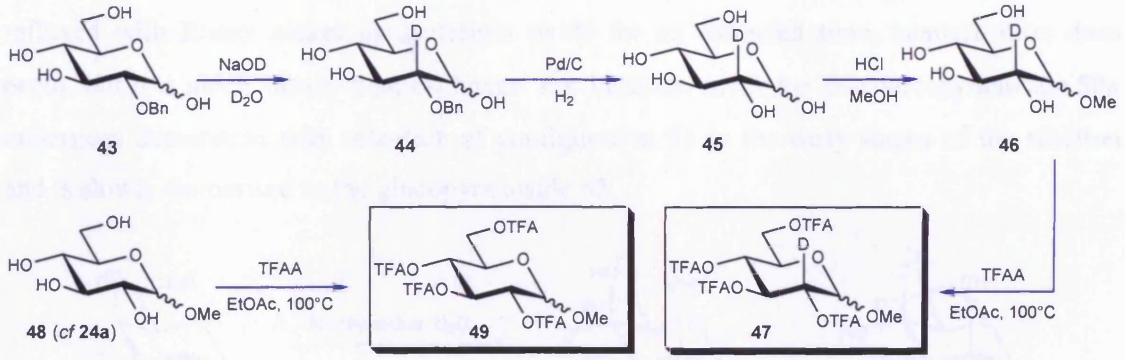
Hydrogen-deuterium exchange of the α- and β- anomers of methyl 4,6-*O*-benzylidene-D-glucopyranoside **36**, **39**, in a redox procedure using *bis*-tributyltin oxide-bromine and sodium borodeuteride, results in deuteration at C-2 and C-3 respectively. The anomers **36**, **39** were regioselectively oxidized to the ketones **37**, **40** with an excess of $(Bu_3Sn)_2O$ in refluxing chloroform followed by brominolysis of the cooled mixture (93%, 63% yield respectively). Reduction of the α-anomer **37** by $NaBD_4$ gave 2-[$^2H_1$]-4,6-*O*-benzylidene-α-D-glucopyranoside **38** as the only product in quantitative yield, whereas the β-anomer **40** was reduced to a mixture of 3-[$^2H_1$]-β-D-alloside **41** and 3-[$^2H_1$]-β-D-glucoside **42** in a 2:1 ratio[28].



Scheme: Reaction conditions: i), $(Bu_3Sn)_2O$, $CHCl_3$, reflux, Br_2, 0° C, (**37**, 93 %; **40**, 63 %), ii) $NaBD_4$[28].

1.2.2.2. Synthesis of deuterated carbohydrates and inositols, by hydrogen-deuterium exchange

Deuterium incorporation at the C-2 position of methyl pyranosides and their derivatives has been used to identify stereoisomers by mass spectrometry. 2-*O*-Benzyl-D-glucose **43** was dissolved in 1.7 M sodium deuteroxide and stirred for 10 days at room temperature. Neutralization with acetic acid-d_1 generated 2-[$^2H_1$]-2-*O*-benzyl-D-glucose **44** in 89 % yield. The ^{1}H-NMR spectrum showed two singlet resonance peaks at 5.26 ppm and 4.70 ppm corresponding to the α- and β-anomers respectively. The benzyl ether anomers **44** were hydrogenolysed using 5% palladium on charcoal to give 2-[$^2H_1$]-D-glucose **45** (75 % yield), which in turn was reacted with acidic methanol to obtain a mixture of α- and β-methyl glucopyranosides **46** after purification. Treatment of the anomers **46** with trifluoroacetic anhydride in ethyl acetate at 100°C gave the deuterated trifluoroacetates **47** which were characterized by mass spectra and compared with the non-deuterated trifluoroacetates of α- and methyl-β-D-glucopyranoside **49**[29,30].



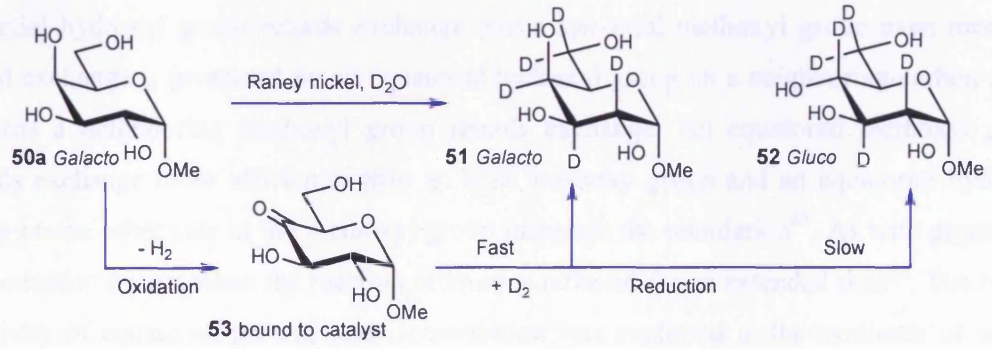
Hydrogen atoms bonded to carbon atoms bearing hydroxyl groups undergo exchange with deuterium in refluxing deuterium oxide catalysed by Raney nickel. The reaction is applicable to all the major classes of glycosides: furanosides, pyranosides, disaccharides, and cyclodextrins except polysaccharides and is economical, simple and convenient[31]. Exchange is inhibited by aldoses and ketoses, which has been attributed to depletion of "active hydrogen" due to reduction of the carbonyl group to the alcohol. Raney nickel is used in the commercial hydrogenation of glucose to sorbitol[32]. With simple alcohols such as 2-methylcyclohexanol, deuterium is also incorporated at positions adjacent to the carbon bearing the hydroxyl group and epimerization occurs, which is consistant with ketone formation and enolisation.

Raney nickel was invented by Murray Raney in 1926[33] for "*the hydrogenation of oils, fats, waxes and the like*"[34]. Raney nickel catalyst is manufactured by treating an alloy of consisting of equal weights of aluminum and nickel[35] with sodium hydroxide[36,37]. The aluminum dissolves to give sodium aluminate, hydrogen and the catalyst which is formed has a highly porous structure with a high surface area (about 100 $m^2\ g^{-1}$)[38,39], which typically contains 85 % nickel by weight and a 2:1 molar ratio of nickel:aluminium.

$$2Al + 2NaOH + 6H_2O \rightarrow 2Na[Al(OH)_4] + 3H_2$$

During the course of activation hydrogen is formed on the surface of the catalyst. Hydrogen seems to be bound in two forms; one of which is tightly bound atomic hydrogen bonded to three nickel atoms, and the other is more weakly bound as chemisorbed molecular hydrogen[40,41].

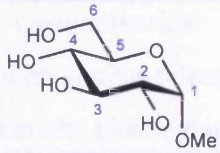
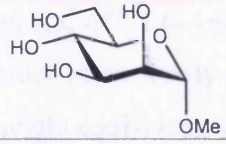
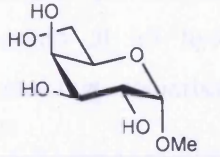
Different hydrogen atoms in the same molecule are exchanged at different rates and replacement occurs with retention of configuration. However, when the reaction mixture is refluxed with Raney nickel in deuterium oxide for an extended time, isomerization does occur, but it is much slower than exchange. For instance, methyl-α-D-galactopyranoside **50a** undergoes deuteration with retention of configuration **51** in the early stages of the reaction and is slowly isomerised to the glucopyranoside **52**.



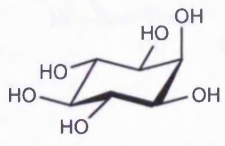
The exact mechanism of the Raney nickel exchange reactions are still in doubt, though they may be explained by a mechanism in which the hydroxyl groups are oxidised to the corresponding ketone **53** and then reduced with incorporation of deuterium[42]. However, in the early stages of the reaction, exchange occurs with retention of configuration and this cannot be explained by the intermediacy of a free ketone. It may be that in the early stages of the reaction, a catalyst bound ketone undergoes exchange with retention of configuration and that isomerisation results from reduction of ketone with incorporation of deuterium, which has

become unbound from the surface and then rebound (dehydrogenation-redeuteration process). Hence under these circumstances galactopyranosides (and mannopyranosides) give the thermodynamically more stable glucopyranosides[43]. The deuteration of several methyl pyranosides and furanosides over Raney nickel in different periods of time has been investigated. Some selectivity was achieved, but the exchange method was not highly regioselective and it was not possible to isolate pure products (Table 1[31,44])

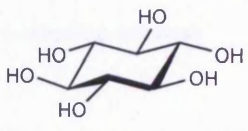
Table 1. The rate of H-D exchange of methyl hexopyranosides **24a**, **54a**, **50a** over Raney nickel in D_2O.

Hexopyranoside	Structure	Rate of exchange	Explanation
Methyl-α-D-glucopyranoside **24a**		H-2, 4 > 3 > 6*R* > 6*S*	O-Me is *trans* to H-2, H-4 is unhindered
Methyl-α-D-mannopyranoside **54a**		H-4 > 2 > 3, 6*R* > 6*S*	O-Me is *cis* to H-2, H-4 is *syn*-axial with OH-2
Methyl-α-D-galactopyranoside **50a**		H-4 > 2 > 3 > 6	H-4 is equatorial, O-Me is *trans* to H-2

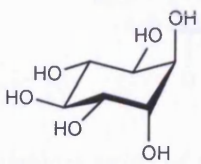
The factors affecting the rate of exchange in inositols **55** - **58** with Raney nickel catalyst can be summarized as follows. Equatorial protons exchange more rapidly than axial protons and a *syn*-axial hydroxyl group retards exchange and a *syn*-axial methoxyl group even more so. Rapid exchange is promoted by an equatorial hydroxyl group on a neighboring carbon atom, whereas a neighboring methoxyl group retards exchange. An equatorial methoxyl group retards exchange more efficiently than an axial methoxy group and an equatorial hydroxyl group on the other side to the methoxyl group increases the retardation[45]. As with glycosides isomerisation occurs when the reaction mixture is refluxed for an extended time[46]. The higher reactivity of equatorial protons and isomerisation was exploited in the synthesis of *scyllo*-inositol **56** from *myo*-inositol **55**. When *myo*-inositol **55** was refluxed with Raney nickel in water for 24 hours, 20 – 30 % was converted to *scyllo*-inositol **56**, which was isolated in 20 % yield[47].



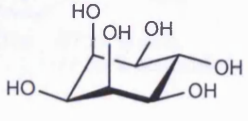
55 *myo*-inositol



56 *scyllo*-inositol

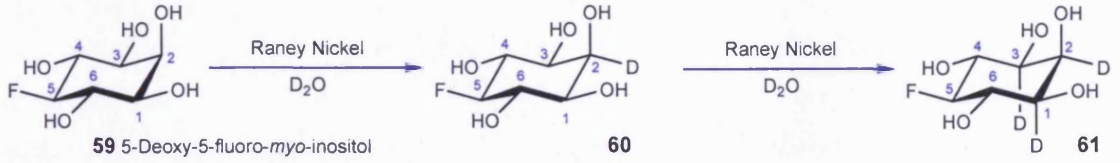


57 *chiro*-inositol



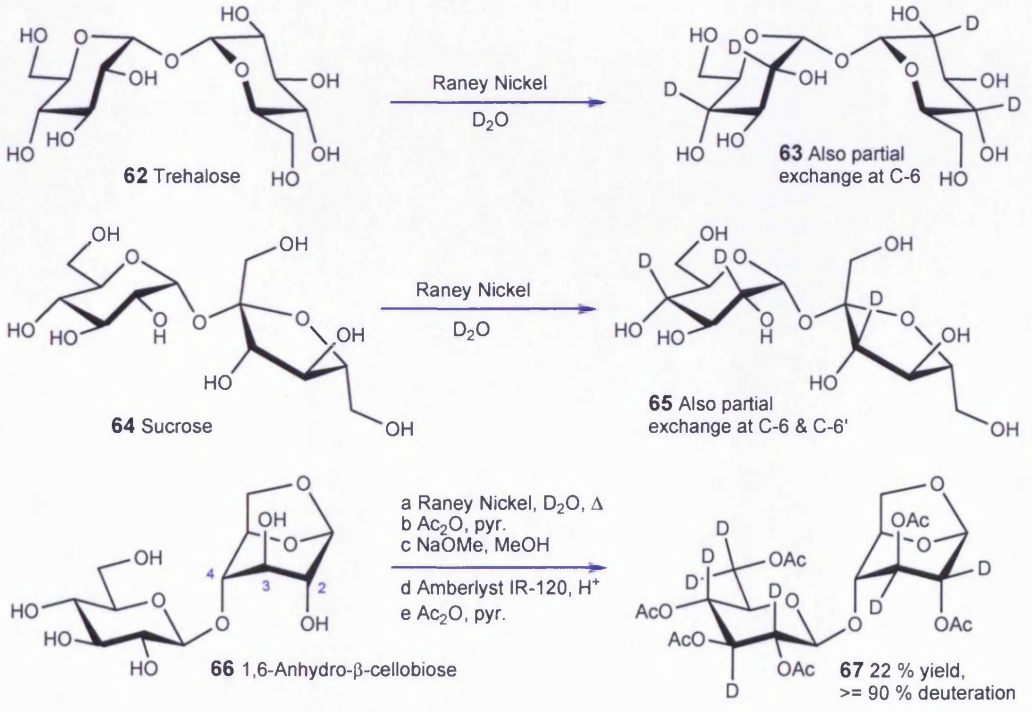
58 *epi*-inositol

Similarly the fluorinated *myo*-inositol **59**, underwent preferential exchange of the sole equatorial proton at C-2 (**60**) followed by slow exchange at the two flanking positions, but no exchange occurred at C-4, C-5 or C-6 (**61**)[48].



59 5-Deoxy-5-fluoro-*myo*-inositol **60** **61**

Raney nickel catalysed hydrogen-deuterium exchange has also been applied to dissacharides and higher sugars. The C_2-symmetrical di-α-glucopyranoside trehalose is selectively deuterated at C-2 and C-4 with similar selectivity to methyl-α-D-glucopyranoside **24a**[49]. The same selectivity is shown with the glucopyranoside moiety of sucrose, whereas the furanoside ring is selectively deuterated at C-3[50]. 1,6-Anhydroglucose (cf. **66**) is deuterated preferentially at C-3, then C-4 and then much more slowly at C-2[51]. Exhaustive deuteration of 1,6-anhydro-β-cellobiose **66** with five weight equivalents of Raney at reflux for 24 hours resulted in >= 90 % incorporation of deuterium at all hydroxylated sites. The acetylation-deacetylation purification sequence indicates that isomerisation to other stereoisomers was likely a problem and the yield was very low[52].



Scheme: Raney nickel catalysed deuteration with deuterium oxide of the disaccharides: trehalose[49], sucrose[50] and 1,6-anhydro-β-cellobiose[52].

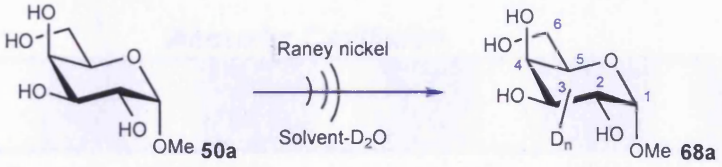
Chapter 2

Results and Discussion

2. Raney nickel catalysed deuteration of sugars using ultasound

2.1. Introduction

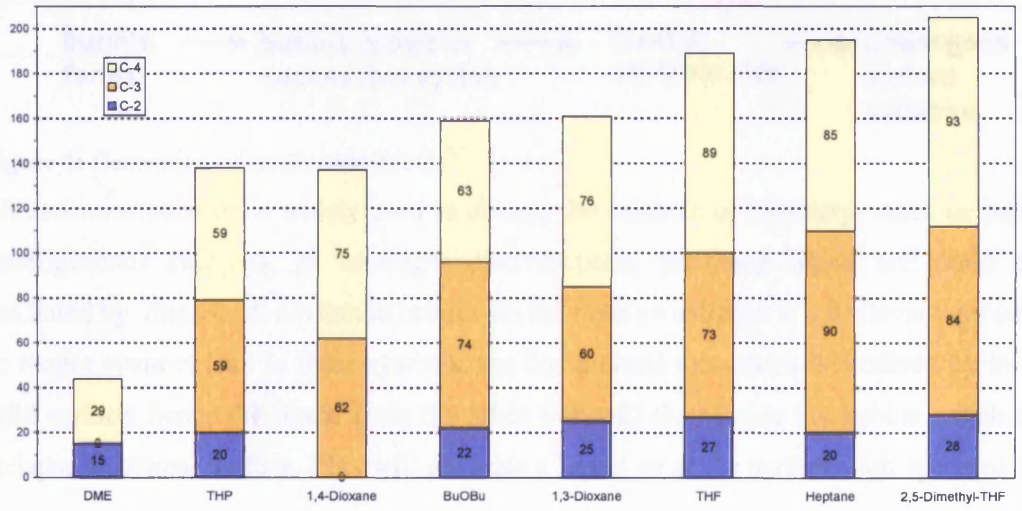
Cyclodextrins readily bind small organic molecules from aqueous solution and the binding can be measured by NMR. However this method is compromised by the presence of the ^{1}H-NMR resonances of the cyclodextrin protons. Exchange of these protons using Raney nickel, would enable a wider range of binding studies to be performed. β-Cyclodextrin **94** has a total of 28 exchangeable (7 x [H-2, H-3, H_2-6]) and previous studies have shown that complete deuteration is difficult to achieve[31,53]. It has been reported that Raney nickel catalysed hydrogen-deuterium exchange of methyl-α-D-galactopyranoside **50a** catalysed by Raney nickel is enhanced under ultrasonic irradiation.



50a **68a**

Deuterations were conducted in eight deuterium oxide/organic solvent mixtures (Graph 1). Deuterium incorporation occurred preferentially at C-4 for all solvent systems. The C-3 position was deuterated in seven solvent pairs but not in a mixture of 1,2-dimethoxyethane and D_2O. No deuterium was incorporated at C-2 position in 1,4-dioxane and D_2O, but this position was deuterated in the other seven solvents[54].

Graph 1. Raney nickel catalysed deuteration of α-methyl-D-galactopyranoside 50a. % Incorporation of deuterium at C-2, C-3, C-4, ranked left to right according to average incorporation



The use of ultrasound irradiation in organic, inorganic and organometallic synthesis (sonochemistry) has been a valuable method in the last two decades. Ultrasound has frequencies of around 20 KHz to 10 MHz with wavelengths of roughly 100 to 0.15 mm. The chemical effect of ultrasound does not result from direct interaction of sound with molecular species, instead ultrasonic irradiation causes the phenomena of acoustic cavitation in liquids.

Cavitation is the formation, growth and implosive collapse of bubbles in a liquid. When ultrasound passes through a liquid, like any sound wave it consists of expansion and compression waves, which compress and stretch the liquid, this forms bubbles that are filled with vapour (from evaporation of liquid). Once formed, small gas bubbles irradiated with ultrasound absorb energy from the sound and grow. These bubbles subsequently collapse which generates high local heating and pressure which enhances chemical reactions[55].

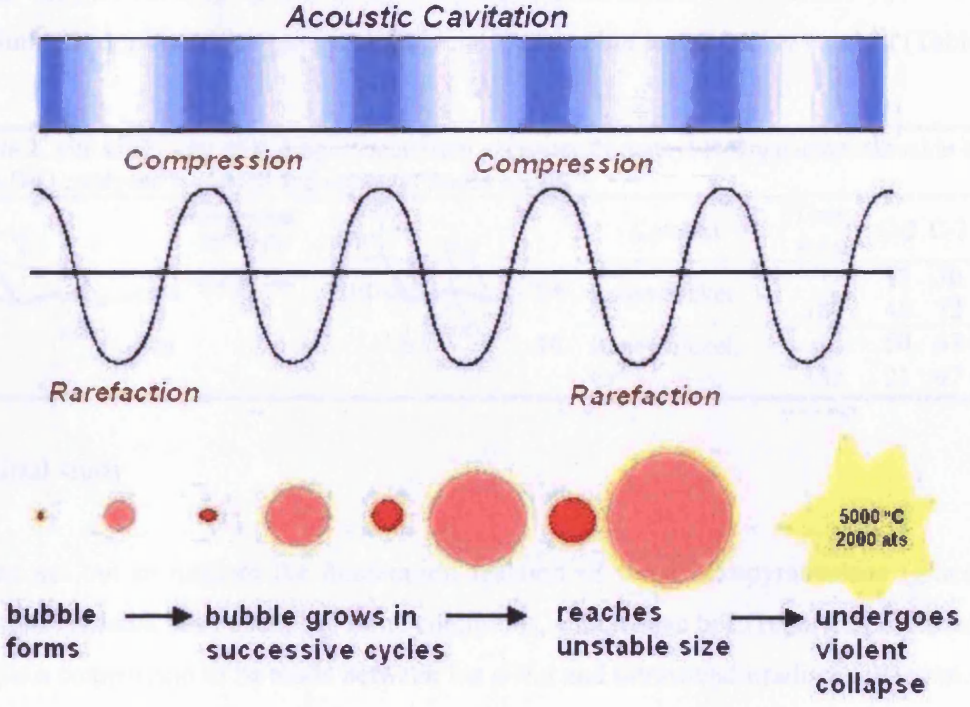


Figure 1: Generation of an acoustic bubble[56]

Ultrasound irradiation is widely used to cleanse the surfaces of jewellery, machine parts and heterogeneous catalysts. In heterogeneous reactions, involving liquid and solid phases irradiated by ultrasound, cavitation occurs but the bubbles collapse in a different way which is no longer symmetrical. In these systems, the liquid phase movement is hindered by the large solid surface, hence the liquid from the other side will flow inside the bubble which causes collapse at or near surface. This will generate a liquid jet at the surface with speed of 100 m

s^{-1} which is highly effective in removing impurities from the surface of the catalyst, and consequently increases the surface area of the catalyst available for reaction[57].

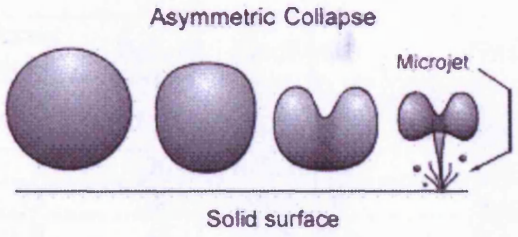
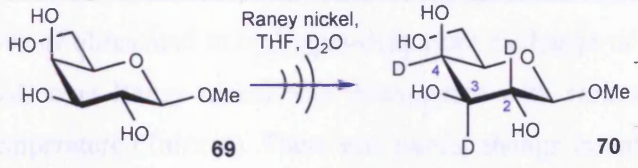


Figure 2: Bubble collapse at a solid surface[58]

Ultrasonication of Raney nickel removes superficial oxides and induces surface cracking to reveal fresh nickel surfaces[59]. Ultrasonication has been used with a range of other nickel based catalysts, for the deuteration of methyl β-D-galactopyranoside **69** with D_2O but these were all ineffective. Doping the catalyst with transition metals reduced activity, except for chromium(III) doping which gave a slightly more active but less selective catalyst (Table 2)[60].

Table 2. Site selectivity of hydrogen-deuterium exchange of methyl-β-D-galactopyranoside **69** with D_2O catalysed by doped and undoped Raney nickel[60].



Catalyst	Time, mins	C-2	C-3	C-4
Raney nickel	1	13	70	69
	180	40	72	88
Raney nickel, Cr^{3+}	1	50	67	62
	180	21	97	98

2.2. Initial study

We first set out to explore the deuteration reaction of three hexopyranosides (*gluco* **24a**, *manno* **54a**, *galacto* **50a**) under the same conditions, which have been reported previously[31,44], to enable a comparison to be made between the silent and ultrasound irradiated reaction.

Raney nickel was washed with D_2O two or three times and then glucopyranoside **24a** was added. A range of conditions were examined with either magnetic stirring or with heating to reflux alone (Table 3). Deuteration was also attempted using 10 % palladium on carbon in deuterium oxide, but this did not give the desired compound after 2 h. reflux, and instead the starting material was recovered.

Table 3. Optimisation of the deuteration of glucopyranoside **24a** by D_2O catalysed by Raney nickel.

Entry	*Gluco* **24a**, g	Raney nickel, g	D_2O, ml	Conditions	Partially deuterated protons
1.	1	10	4	3 d, r.t., 48 h, 90°C	2, 3, 4 and perhaps 5
2.	1	12	4	reflux, 18 h.	10% at C-2
3.	0.2	2	20	reflux, 2 d.	2, 3, 4, 5, 6a
4.	0.2	3	20	reflux, 3 d	2, 3, 4, 6
5.	0.2	9	11	reflux, 20 h.	2, 3, 4
6.	0.2	12	20	reflux, 21 h	2, 3 and perhaps 4

In all cases the deuteration was very slow and unselective, despite the presence of large amounts of Raney nickel; even up to 12 g/g of glucopyranoside **24a** (Entry 2). In the magnetically stirred reactions, the Raney nickel particles were attracted to the magnetic bar and dispersion of the catalyst was poor, even with rapid stirring. Consequently, in later reactions agitation by reflux alone was used.

2.3. Methodology development

Use of ultrasound in hydrogen-deuterium exchange of methyl-α-D-glucopyranoside **24a** with D_2O over Raney nickel was investigated with various reaction conditions, but all at room temperature (Table 4). There was partial change in only one case, ultrasonication for 2 hours (entry 1) resulted in circa 10 % deuteration at C-2. As a result, this approach was abandoned because the conversion was too low.

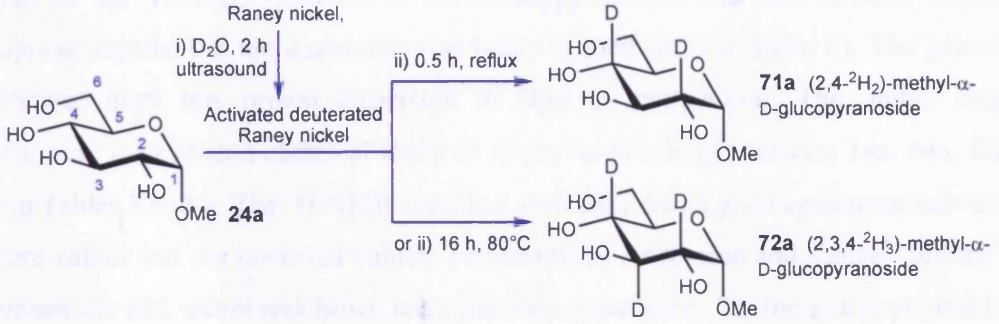
Table 4. Deuteration of glucopyranoside **24a** in D_2O (4 mls) catalysed by Raney nickel using ultrasonic irradiation, without heating.

Entry	Glucopyranoside **24a**, g	Raney Ni, g	Weight ratio[a]	Reaction time, h
1.	1	4	4	2
2.	0.2	1	5	3
3.	0.2	12	60	4

[a]Weight Raney Nickel\weight glucopyranoside **24a**

Ultrasound and Reflux: In a slightly different method for deuteration of the catalyst, Raney nickel (12 g, wet) was placed in a flask with deuterium oxide (4 ml) was added and the supernatant layer removed (5 ml). The washing was repeated twice more and every time the wet solid was left with no supernatant layer. Finally the catalyst was ultrasonicated in deuterium oxide (4 ml) for 2 h. When a mixture of the glucopyranoside **24a** (200 mg) and activated Raney nickel (12 g) were refluxed in deuterium oxide for only half an hour,

exchange occurred exclusively predominantly at C-2 and C-4 and circa 30 % at C-3 (**71a**). Whereas heating the glucopyranoside **24a** (200 mg) for 16 h. at 80°C, resulted in formation of predominantly (2,3,4-$^{2}H_3$)-methyl-α-D-glucopyranoside **72a**. The protons at C-2 and C-4 were completely exchanged and 64 % deuterium incorporation occurred at C-3 position. These two procedures were adopted as standards and are described as the *half hour* and the *sixteen hour* procedure respectively. The NMR data used in the determination of incorporation rates is discussed in the next section.



2.4. Spectroscopic characterisation

Assignment of the ^{1}H-NMR spectrum of simple glycosides is comparatively difficult because of poor signal dispersion, even at 500 MHz. The literature chemical shifts and coupling constants of methyl-α-D-hexopyranosides **24a**, **54a**, **50a** are summarised in Tables 5 and 7 and our observed values in Tables 6 and 8. Assignments of the ^{1}H and ^{13}C-NMR are based on HSQC, HMBC, and DEPT-NMR based correlations, rooted on the only two unambiguous NMR signals; the anomeric carbon and C-6 in the ^{13}C-spectra.

Table 5. Literature ^{1}H chemical shifts of methyl-α-D-hexopyranosides **24a**, **54a**, **50a** in D_2O.

Compound	1	2	3	4	5	6*R*	6*S*	OCH_3	Literature
Gluco **24a**						3.76	3.89		61
					3.65	3.75	3.85		62
	4.80	3.55	3.66	3.40	3.64	3.75[a]	3.87[a]	3.42	63
Manno **54a**						3.70[a]	3.54[a]	3.2	64
	4.77	3.94	3.77	3.67	3.61	3.78[a]	3.90[a]	3.42	65
	5.27	4.45			4.12			3.94	66[b]
- 0.52 ppm[b]	4.75	3.93			3.60				Correction[b]
	4.761	3.929	3.751	3.640	3.604	3.898[a]	3.755[a]		67
	4.76	3.93	3.76	3.64	3.62	3.90[a]	3.76[a]	3.41	63
Galacto **50a**						3.70	3.70		61
					3.90	3.69	3.69		62
	4.84	3.81	3.81	3.97	3.89	3.74[a]	3.74[a]	3.41	63

[a] *R* or *S* unknown. [b] The data reported in reference 66 apparently has a systematic error of + 0.52 ppm

Table 6. Observed ^{1}H-chemical shifts of methyl-α-D-hexopyranosides **24a**, **54a**, **50a** in D_2O and CD_3OD

Proton	Solvent	1	2	3	4	5	6a	6b	OCH_3
Gluco **24a**,	D_2O	4.72	3.47	3.58	3.31	3.55	3.78	3.66	3.33
Manno **54a**	D_2O	4.60	3.76	3.57	3.46	3.46	3.73	3.57	3.22
	CD_3OD	4.71	3.86	3.75	3.68	3.57	3.92	3.79	3.46
Galacto **50a**	D_2O	4.75	3.66	3.66	3.82	3.74	3.70	3.70	3.34
	CD_3OD	4.74	3.78	3.74	3.90	3.78	3.74	3.70	3.42

Analysis of the ^{1}H-NMR spectrum of the mannopyranoside **54a** was difficult because of overlapping signals, but the dispersion was better in methanol-d_4 (Table 6). The galactoside **50a** showed even less proton dispersion in D_2O or methanol-d_4. The proton coupling constants and their related chemical shifts of methyl-α-D-hexopyranosides **24a**, **54a**, **50a** are listed in Tables 5 and 6. The ^{1}H-NMR chemical shift data shows good agreement between the literature values and our observed values. The results of Andersson and Kenne[63] are the most comprehensive and recent and hence are used for comparisons. For the glucopyranoside **24a** there is a systematic difference (observed – literature of -0.085 ± 0.005 for all signals, except for H-6a (0.03) and H-6b (-0.21), which have a large geminal coupling constant and hence the apparent chemical shifts are more sensitive to the spectrometer frequency. For the mannopyranoside **54a** there is a systematic difference of -0.16 to -0.19 for all signals and for the galactopyranoside **50a** the differences range from -0.04 to -0.15. Overall, there is an excellent fit between the literature and our observations.

Table 7. Literature ^{1}H-^{1}H coupling constants of methyl-α-D-hexopyranosides **24a**, **54a**, **50a** in D_2O.

Compound	$^3J_{1,2}$	$^3J_{2,3}$	$^3J_{3,4}$	$^3J_{4,5}$	$^3J_{5,6a}$	$^3J_{5,6b}$	$^2J_{6a,6b}$	Literature
Gluco **24a**					5.4	2.2	12.3	62
	4.0	10.0	10.0	10.0	5.8	2.8	12.8	68
Manno **50a**					1.9[a]	5.8[a]	12.0	62
	1.6	2.9						66
	1.8	3.5	9.7	10.0	2.6	5.8	12.3	67
	1.6	3.5	10.0	10.0	1.9	5.8	12.0	68
Galacto **54a**					6.4	5.3		62
					7.8	6.0		61
	3.0	9.8	2.3	1.0	4.6	8.2	12.2	69

[a] *R* or *S* unknown

Table 8. Observed ^{1}H-^{1}H coupling constants of methyl-α-D-hexopyranosides **24a**, **54a**, **50a** in D_2O and CD_3OD.

Coupling	Solvent	$^3J_{1,2}$	$^3J_{2,3}$	$^3J_{3,4}$	$^3J_{4,5}$	$^3J_{5,6a}$	$^3J_{5,6b}$	$^2J_{6a,6b}$
Gluco **24a**	D_2O	3.7	9.8	9.4	9.6	5.5	2.0	12.2
Manno **54a**	D_2O	1.5	3.2	9.4	9.5	2.4	5.9	11.8
	CD_3OD	1.6	3.3	9.4	9.4	2.4	6.0	11.8
Galacto **50a**	D_2O	3.7	9.3	2.6	1.5	6.3	5.4	11.8
	CD_3OD	3.6	9.8	3.0	overlap	5.4	2.0	11.0

The literature and observed coupling constants are shown in Tables 7 and 8. All of the coupling constants in our work were calculated by taking the peak list and entering the data into the computer program Multiplet[70]. Briefly, this calculates coupling constants by averaging appropriate line separations in a multiplet, permutates the separations and then determines the fit of the permutation result to the data. Consequently the accuracy of the results can exceed the digital resolution of the spectra for a signal with high multiplicity. None of the signals in the data are more complex than doublets of doublets and hence the averaging has minimal effect. All of our results are the same as the literature values within the limits of digital resolution (± 0.3 Hz). The ^{13}C-NMR chemical shifts showed a systematic error of circa 1 ppm for all signals (Table 9), except for C-1 and C-3 of the glucopyranosides.

Table 9: Literature, observed and differences (observed – literature) ^{13}C chemical shifts of methyl-α-D-hexopyranosides **24a**, **54a**, **50a** in D_2O.

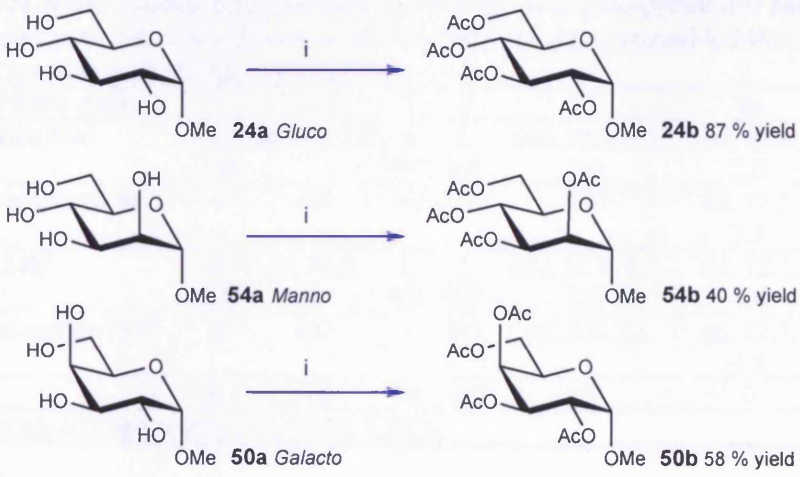
Compound	C-1	C-2	C-3	C-4	C-5	C-6	OCH_3
Gluco **24a**							
Literature[71] see also[63]	100.0	72.2	74.1	70.6	72.5	61.6	55.9
Observed	99.6	70.8	71.6	69.7	71.4	61.2	55.4
Differences	-0.4	-1.4	-2.5	-0.9	-1.1	-0.4	-0.5
Differences (observed – literature) -0.4 to -2.5 ppm (upfield)							
Manno **54a**							
Literature[71] see also[67]	101.9	71.2	71.8	68.0	73.4	62.1	55.9
Observed	100.7	69.7	70.4	66.6	72.4	60.8	54.5
Differences	-1.2	-1.5	-1.4	-1.4	-1.1	-1.3	-1.4
Differences -1.2 to -1.5 ppm (upfield).							
Galacto **50a**							
Literature[71]	100.1	69.2	70.5	70.2	71.6	62.2	56.0
Observed	99.2	68.0	69.3	69.1	70.6	61.1	54.8
Differences	-0.9	-1.2	-1.2	-1.1	-1.0	-1.1	-1.2
Differences between -0.9 to -1.2 ppm (upfield)							

Replacement of hydrogen by deuterium, obviously results in the loss of that signal from the ^{1}H-NMR spectrum and the coupling in which the hydrogen is participating. As ^{1}H-^{2}H coupling constants are 15 % of the corresponding ^{1}H-^{1}H values any coupling constant below about 7 Hz will only be observed as line broadening under most circumstances in the

deuterated compound. Substitution by deuterium also changes the chemical shift of the attached carbon (0.1 to 0.6 ppm), adjacent carbons (<0.15 ppm) and adjacent protons (0.1 to 0.2 ppm). If the ^{1}H-NMR spectrum of a compound has poor dispersion, unselective deuteration gives a mass of peaks that are exceedingly difficult to analyse. Moreover the pyranosides **24a**, **54a**, **50a** are only appreciably soluble in water, methanol and DMSO, and have similar chemical shifts in these solvents. As a consequence of this we decided to prepare derivatives for analysing the deuterated products and/or the reaction mixtures.

2.5. Acetylation of methyl-α-D-hexopyranosides 24a, 50a, 54a

The peracetates of sugars are non-polar and consequently easier to purify by chromatography; they are frequently crystalline, soluble in a range of organic solvents and the ^{1}H-NMR spectra have better dispersion. Therefore we would be able to measure multiple sets of NMR data if required in several solvents of different types (e.g. $CDCl_3$ and C_6D_6). The ^{1}H-NMR spectrum of glucopyranosides **24a**, is comparatively easy to assign, because there are few overlapping protons compared to the mannopyranoside **54a,** or the galactopyranoside **50a**, but it was also acetylated to compare with the others. The hexopyranosides **24a**, **54a**, **50a** were acetylated by a general procedure[72] which is used for carbohydrates. A large excess of pyridine and acetic anhydride (50, 20 equivalents respectively) in dichloromethane at 0°C, was added to the hexopyranosides **24a**, **54a**, **50a** with stirring for 3 h. and then kept in the refrigerator for 2 days. Water-sodium bicarbonate workup and a wash with copper sulfate to remove excess of pyridine yielded the tetra-*O*-acetyl-hexopyranosides **24b**, **54b**, **50b** in fair to good yields. ^{1}H-NMR spectroscopic analysis showed down-field proton shifts for secondary protons (2, 3, 4-H) attached to acetoxy groups, relative to the parent alcohols **24a**, **54a**, **50a**.



Scheme: Conditions i) Ac_2O, pyridine, CH_2Cl_2, 2 h., 0° C; 2 d., refrigerator.

2.5.1. Analysis of the ^{1}H-NMR spectra of tetra-*O*-acetyl-hexopyranosides and the Raney nickel catalysed deuteration of manno- and galacto-pyranosides 54a, 50a by deuterium oxide.

The preliminary study indicated that ultrasound treated Raney nickel was more active in deuteration reactions than untreated Raney nickel, and some selectivity was achieved in the deuteration of glucopyranoside **24a**. The half hour procedure resulted in incorporation of deuterium predominantly C-2 and C-4, whereas the sixteen hour procedure, resulted in deuteration predominantly at C-2, C-3 and C-4. Consequently deuteration of the manno-pyranoside **54a** and galactopyranoside **50a** were investigated under similar conditions.

In a ^{13}C-NMR spectrum of the deuterated glucopyranoside **71a**, the signals expected for C-2 and C-4, which appear at 70.8 ppm and 69.7 ppm for the undeuterated glucopyranoside **24a** were not observed due to deuteration at those positions. This was confirmed in a ^{1}H NMR spectrum in which the proton at C-1 appeared as a singlet. Deuterated glucopyranoside **71a** was acetylated and ^{1}H NMR spectrum of the tetra-*O*-acetyl derivative **71b** showed two singlet signals due to H-1 and H-3 at 4.96 and 5.49 ppm respectively. ^{13}C chemical shifts and ^{1}H-^{1}H coupling constants of *gluco* derivatives are summarised in Tables 10, 11.

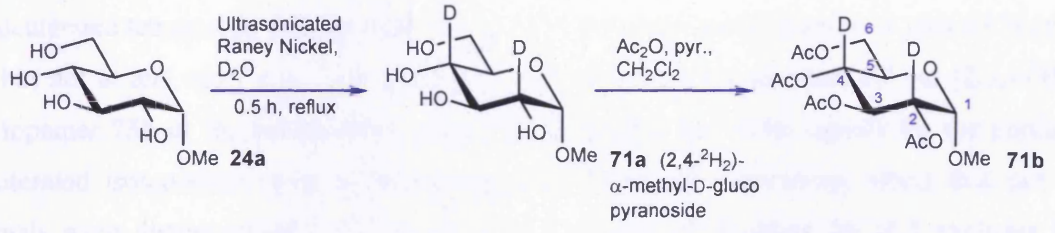


Table 10. ^{1}H-^{1}H-NMR coupling constants patterns of methyl-α-D-glucopyranoside **24a**, (2,4-$^2H_2$)-α-methyl-D-glucopyranoside **71a**, 2,3,4,6-tetra-*O*-methyl-α-D-glucopyranoside **24b**, (2,4-$^2H_2$)-2,3,4,6-tetra-*O*-methyl-α-D-glucopyranoside **71b**.

Compound / Proton position	1	2	3	4	5	6a	6b
Glucopyranoside **24a**[a]	d, 3.7	dd, 9.8, 3.7	t, 9.4	t, 9.6	ddd, 10.0, 5.5, 2.0	dd, 12.2, 2.0	dd, 12.2, 5.5
(2,4-$^2H_2$)-glucopyranoside **71a**[a]	s	*NO*	*ND*	*NO*	*ND*	dd, 12.3, 2.3	dd, 12.2, 5.5
Tetra-acetate **24b**[b]	d, 3.7	dd, 10.2, 3.7	t, 9.8	t, 9.8	ddd, 10.2, 4.5, 2.3	dd, 12.3, 4.6	dd, 12.3, 2.3
(2,4-$^2H_2$)- tetra-acetate **71b**[b]	s	*NO*	s	*NO*	dd, 4.5, 2.1	dd, 12.3, 4.5	dd, 12.3, 2.2
% Deuterium	0	100	30	100	0	0	0

NO, not observed; *ND*, not determined; [a]D_2O; [b]$CDCl_3$

Table 11. ^{13}C-NMR chemical shifts of methyl-α-D-glucopyranosides **24a**, **71a**, **24b**, **71b** (*cf* Table 9).

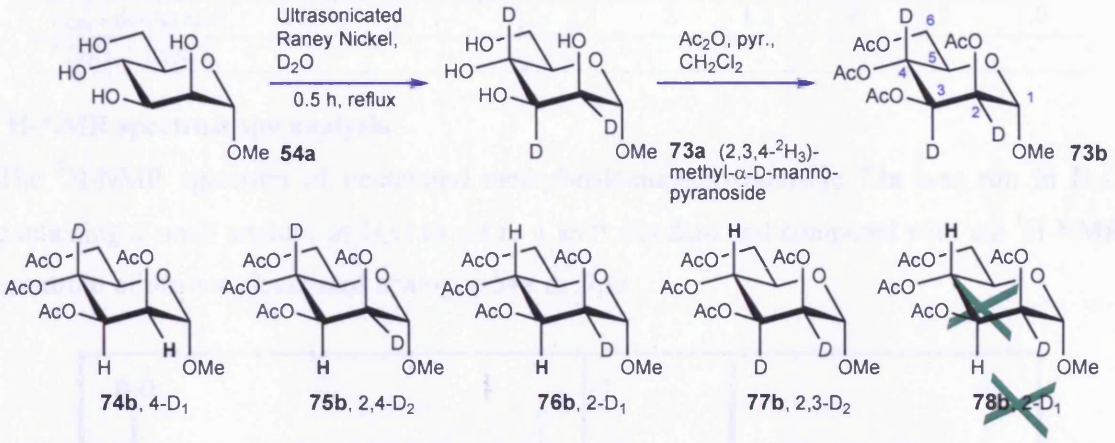
Compound / Carbon position	1	2	3	4	5	6	OCH_3
Glucopyranoside **24a**[a]	99.6	70.8	71.6	69.7	71.4	61.2	55.4
(2,4-$^2H_2$)-glucopyranoside **71a**[a]	99.5	*NO*	72.2	*NO*	71.8	60.7	55.3
$\hat{c}_{24a}$ - deuterated	0.1		- 0.6		- 0.4	0.5	0.1
Tetra-acetate **24b**[b]	97.1	71.1	70.4	68.8	67.5	62.5	55.8
(2,4-$^2H_2$)- tetra-acetate **71b**[b]	95.7	*NO*	68.9	*NO*	66.0	60.8	54.4
$\hat{c}_{24b}$ - deuterated	1.4		1.5		1.5	1.7	1.4

NO, not observed; *ND*, not determined; [a] D_2O; [b] $CDCl_3$

Unlike the deuterated glucopyranoside **71a**, the NMR spectra of the deuterated compounds formed from the mannopyranoside **54a** and galactopyranoside **50a** were comparatively difficult to interpret due to overlap of signals, consequently the use of the tetra-*O*-acetates was essential.

When the mannopyranoside **54a** was subjected to the half hour procedure, the ^{1}H-NMR spectrum of the product showed a singlet peak for H-1, and a weak doublet of 3.6 Hz for H-2 (Table 12). The coupling constants for H-6a, H-6b were identical to those observed for undeuterated mannopyranoside **54a**, which indicates there is no appreciable deuteration at C-5 or C-6. (Table 13). Comparison of the NMR spectra of the deuterated tetraacetate and the undeuterated tetraacetate **54b** indicated circa 72 % deuteration at C-2 and C-4 plus 67 % at C-3, but not at any other site. This is clearly consistent with the presence of the (2,3,4-$^2H_2$)-isotopomer **73b** as the predominant component, however the weak signals for the partially deuterated isotopomers give an interesting insight into the selectivity, albeit that not all signals were distinguishable. The absence of a doublet of doublets for H-3 excludes the possibility that any non-deuterated mannopyranoside **54b** is present, which is important for the arguments for assigning the structures of **74b** and **78b**, which could otherwise could be deuterium or hydrogen at C-4 or C-2 respectively. Similarly the absence of a doublet signal for H-2, means that C-2 of the isotopomer **77b** must bear a deuterium, albeit that chemical shift coincidence and the small value of $^3J_{1,2}$ (1.3 Hz) might cause this signal to be hidden under the observed doublet of doublets. The presence of a deuterium at C-2 is confirmed by the absence of a doublet signal for H-1, albeit as a minor component this could be easily hidden under the singlet observed. The signals and the structures which they indicate are as follows: doublet of doublets for H-2, **74b**; singlet for H-3 **75b**; doublet for H-3, **76b**; doublet for H-4, **77b**; and the triplet for H-4, **78b**, which is identical to structure **76b** deduced previously. There are eight possible regio-isotopomers for hexopyranosides containing 0 – 3

deuteriums, at positions C-2, C-3 and C-4. Of the three possible monodeuterated mannopyranosides, two are present **74b**, **76b** and the 3-[$^2H_1$]-regioisomer is absent. Similarly, of the three possible dideuterated derivatives, two are present **75b**, **77b** and the 3,4-[$^2H_1$]-regioisomer is absent. This is consistant with a preference for deuteration at C-2 and C-4, over C-3 and agrees with the degree of incorporation for each site observed for the tri-deuterated mannopyranoside **73b**.



The indicative proton is shown in bold. The italicised deuteriums in the the hexopyranosides **74b**, **78b** cannot be hydrogens, because they would then be the non-deuterated tetra-acetate **54b**, which is known to be absent because there is no doublet of doublets signal for H-3 present. Similarly D-2 in hexopyranoside **77b** cannot be hydrogen, because otherwise a doublet signal for H-2 would be present in the spectrum, albeit this might be easily hidden.

Table 12. ^{1}H-^{1}H Coupling constant patterns of mannopyranosides **54a**, **73a**, **54b**, **73b**.

Compound / Position	1	2	3	4	5	6a	6b
Mannopyranoside **54a**[a]	d, 1.6	dd, 3.2, 1.6	dd, 9.3, 3.3	t, 9.0	ddd, 9.3, 6.0, 2.4	dd, 11.8, 2.3	dd, 11.7, 5.9
($^2H_3$) **73a**[a]	s	d, 3.6	*ND*	*ND*	m	dd, 12.2, 2.2	dd, 12.1, 6.0
Tetra-acetate **54b**[b]	d, 1.5	dd, 3.1, 1.6	dd, 10.0,3.2	t, 9.8	ddd, 9.2,5.4,2.5	dd, 12.2, 5.3	dd, 12.2, 2.4
($^2H_3$) Tetra-acetate **73b**[b]	s	dd, 3.5, 1.4	s; d 9.9	d, 10.1; t 9.7	m	dd, 12.2, 5.3	dd, 12.2, 2.3
% Deuterium, **73ab**	0	~72	~67	~72	0	0	0

ND, not determined; [a]D_2O; [b]$CDCl_3$

The ^{13}C-NMR shift data (Table 13) represents a composite of major signals of the undeuterated centres in partially deuterated compounds **73a** – **77a**, **73b** – **77b**. Small chemical shift differences are present, but they do not provide any obvious interpretation.

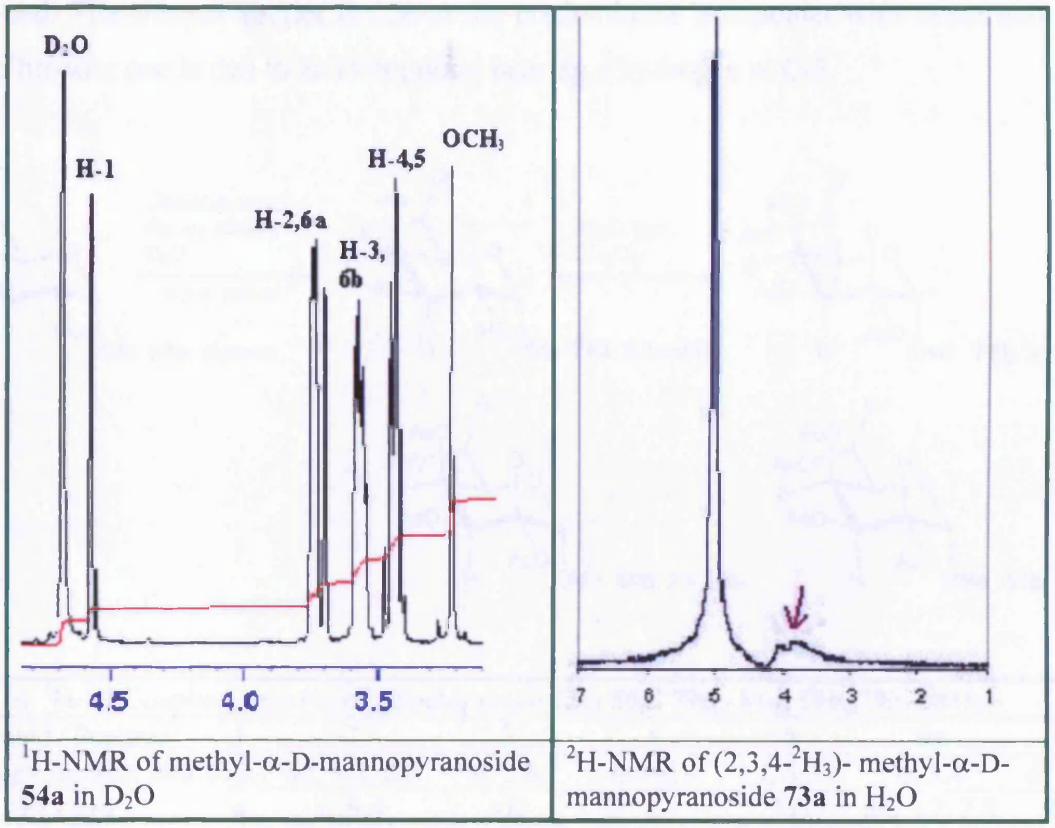
Table 13. ^{13}C chemical shifts of mannopyranosides **54a**, **54b** and deuterated derivatives **73a – 77a**, **73b – 77b**.

Compound/Carbon	1	2	3	4	5	6	OCH_3
Mannopyranoside **54a**[a]	100.7	69.7	70.4	66.6	72.4	60.8	54.5
Deuterated[a]	100.1	70.2	70.7	66.9	71.8	61.2	54.9
∂_{54a} - deuterated	0.6	-0.5	-0.3	-0.3	0.6	-0.4	-0.4
Tetra-acetate **54b**[b]	98.6	69.5	69.1	66.2	68.4	62.5	55.3
Deuterated[b]	97.5	67.2	67.1	65.1	66.5	60.8	54.3
∂_{54b} - deuterated	1.1	2.3	2	1.1	1.9	1.7	1.0

[a] D_2O; [b] $CDCl_3$;

^{2}H-NMR spectroscopy analysis

The ^{2}H-NMR spectrum of deuterated methyl-α-D-mannopyranoside **73a** was run in H_2O containing a small amount of D_2O to act as a shift standard and compared with the ^{1}H-NMR spectrum of the non-deuterated analogue **54a** in D_2O.



^{1}H-NMR of methyl-α-D-mannopyranoside **54a** in D_2O

^{2}H-NMR of (2,3,4-$^2H_3$)- methyl-α-D-mannopyranoside **73a** in H_2O

In the ^{2}H-NMR spectrum of mannopyranoside **73a**, the signals were very broad and deuterium-deuterium couplings were not observed due to non-specific deuteration and the narrow chemical shift range. Two broad signals at 4.05 and 3.86 ppm were just discernable. The chemical shift difference between these two signals was same as the differences measured between H-2 and H-4 in the ^{1}H-NMR spectrum of the non-deuterated mannopyranoside **54a**, which confirmed overlap of all protons except H-1, O-CH_3 in that area

in a similar way to mannopyranoside **54a**. Overall, the ^{2}H-NMR spectrum just confirmed the absence of selective deuteration in the ring and no deuteration in the methoxy group.

When the galactopyranoside **50a** was subjected to the half hour procedure, it was apparent from the ^{1}H-NMR spectrum that H-4 had disappeared rapidly, however analysis of incorporation at the other sites was complicated by overlapping signals (Table 14). Integration of residual peaks in the ^{1}H-NMR spectrum of the tetra-*O*-acetyl-galactopyranosides **79b** – **81b**, indicated circa 90% deuteration at C-2, 70 % at C-3, 80 % at C-4 and approximately 40 % at each proton at C-6. The degree of deuteration at each proton on C-6 (H-6*R*, H-6*S*) could not be determined due to signal overlap. As anticipated, the residual signal for H-3 appeared as a singlet due to the high rate of deuteration at C-2 and C-4 (**80b**). About 20 % of the material was undeuterated at C-2; H-2 appeared as a broad doublet of 10.5 Hz indicating that C-3 was also undeuterated (**81b**). H-1 appeared as two adjacent singlets, one of which was broadened. The sharper singlet is due to the predominant isotopomer with deuterium at C-2 and the broader one is due to an isotopomer bearing a hydrogen at C-2.

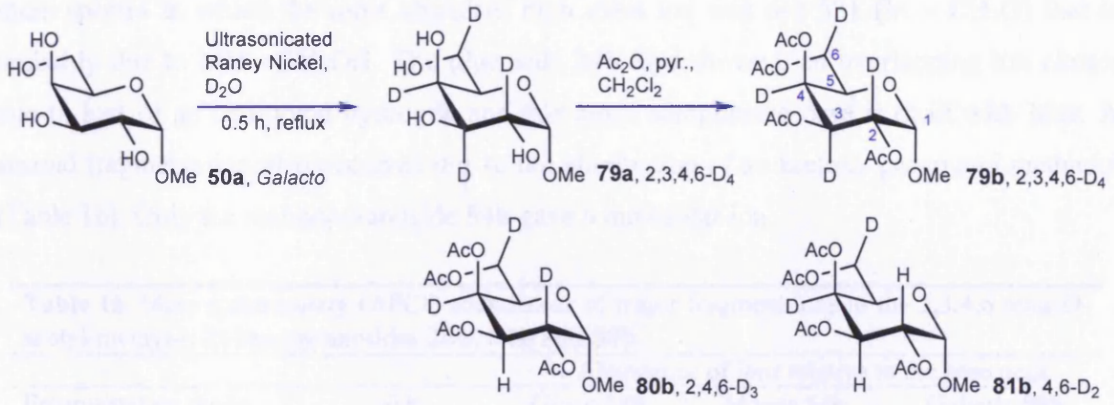


Table 14. ^{1}H-^{1}H Coupling patterns of galactopyranosides **50a**, **79a - 81a**, **50b**, **79b – 81b**.

Compound / Position	1	2	3	4	5	6a	6b
Galactopyranoside **50a**	d, 3.7	dd, 9.3, 2.9	d, 2.6	broad s	t, 6.3	d, 5.4	d, 11.8
($^{2}H_{1-5}$) **79a - 81a**	s	d, 9.4	*ND*	s	*ND*	dd, 5.2, 2.2	*ND*
Tetra-acetate **50b**	d, 3.6	dd, 10.8, 3.6	dd, 11.0, 3.4	dd, 3.2, 0.7	dd, 6.8, 6.0	m, overlap	m, overlap
($^{2}H_{1-5}$) tetra-acetates **79b - 81b**	s; br s	dd, 6.6, 3.6	s; d, 10.5	d, 8.7	d, 5.3	m	M
% Deuterium, **79b - 81b**	0	~90	~70	~80	0	~40	~40
ND, not determined							

The deuteration sites were confirmed in a ^{13}C-NMR spectrum (Table 15) that showed disappearance of the C-2 and C-4 signals and the relatively lower intensity of other carbons compared to C-1 and C-5.

Table 15. ^{13}C-NMR Chemical shifts of galactopyranosides **50a**, **79a - 81a**, **50b**, **79b – 81b**.

Compound / Carbon position	1	2	3	4	5	6	OCH_3
Galactopyranoside **50a**[a]	99.2	68.0	69.3	69.1	70.6	61.1	54.8
($^2H_{1\text{-}5}$) **79a - 81a**[a]	99.2	*NO*	69.2	*NO*	70.5	61.1	54.8
∂_{50a} - deuterated	0.0		0.1		0.1	0.0	0.0
Tetra-acetate **50b**[b]	97.5	68.4	68.5	67.9	66.5	62.2	55.9
($^2H_{1\text{-}5}$) tetra-acetate **79b – 81b**[b]	96.1	65.9	66.3	65.0	64.9	60.7	54.5
∂_{50b} - deuterated	1.4	2.5	2.2	2.9	1.6	1.5	1.4

NO, not observed; [a]D_2O; [b]$CDCl_3$

2.6. Analysis of the mass spectra of deuterated hexopyranosides

Mass spectrometry data is complementary to NMR data; it gives the total deuterium incorporation without the hazard of unfortuitous overlapping signals, but not site specific information. The 2,3,4,6-tetra-*O*-acetyl-methyl-α-D-hexopyranosides **24b**, **54b** and **50b** gave mass spectra in which the most abundant high mass ion was *m/z* 331 (M – CH_3O) that is probably due to MH - CH_3OH. The glucoside **24b** also showed, an overlapping ion cluster due to loss of an additional hydrogen and this more complicated case is dealt with later. A second fragmentation also occurred due to the elimination of an acetoxy group and methanol (Table 16). Only the mannopyranoside **54b** gave a molecular ion.

Table 16. Mass spectrometry (APCI) abundances of major fragment ions of the 2,3,4,6-tetra-*O*-acetyl-methyl-α-D-hexopyranosides **24b**, **54b** and **50b**.

		% Abundance of ions relative to the base peak		
Fragmentation mode	*m/z*	*Gluco* **24b**	*Manno* **54b**	*Galacto* **50b**
M – OCH_3	331	80	100	100
M – CH_3OH	330	31		
M – OAc	303	24	16	
M – OAc – CH_3OH	271	14	8	

Table 17. Mass spectrometry data for the MH – CH_3OH ion cluster of deuterated 2,3,4,6-tetra-*O*-acetyl-methyl-α-D-hexopyranosides **71b - 81b**.

Hexopyranoside	Major fragment ions, *m/z* and their relative abundance % (in parentheses)
Gluco **71b**	335 (7), 334 (60), 333 (35), 332 (25)
Manno **73b – 77b**	335 (13), 334 (41), 333 (38), 332 (15)
Galacto **79b – 81b**	338 (1.2), 337 (8), 336 (36), 335 (63), 334 (53), 333 (21), 332 (4)

The precise interpretation of this data posed a number different of challenges. To discuss these issues with the greatest clarity, the analysis is beneficially discussed in the reverse order to that above (*i.e galacto, manno, gluco*). The mass spectra for the deuterated glycosides **71b - 81b** can be analysed approximately by assuming that each of the peaks in the [MH - CH_3OH] ion cluster represents a different deuterated isotopomer (e.g. Table 18, lines 3 & 4). However the presence of ^{13}C-isotopomers makes this, the simple analysis inaccurate. The MH - CH_3OH ion has the elemental formula $C_{14}H_{19}O_9$; the ^{13}C- content at natural abundance gives an X + 1 ion of 15.7 % intensity and the ^{18}O content gives an X + 2 of 1.8 %, both relative to the $^{12}C^{16}O$ isotopomer. The natural deuterium content can be neglected because H_{19} only contributes 0.3 % to the X + 1 ion. Hence the calculated abundances for the $C_{14}H_{19}O_9$ ion cluster are predominantly determined by the ^{13}C-content (Table 18, line 1). The mass spectrum of the non-deuterated galactopyranoside **50b**, showed slightly lower intensities for X + 1 and X + 2 (line 2) than expected, which was probably because the spectrum was comparatively weak.

Table 18. Analysis of the MH - CH_3OH ion cluster ($C_{14}H_{19}O_9$) of tetra-*O*-acetyl-methyl-α-D-galactopyranosides **50b, 79b – 81b.**

Line	Description \ *m/z*	331	332	333	334	335	336	337	338	339
1.	Theoretical abundances for $C_{14}H_{19}O_9$	100	16.3	3.1	0.4					
2.	**50b**, observed abundances	100	10	0.5						
	Deuterated galactopyranosides **79b – 81b**									
3.	Observed abundances		4	21	53	63	36	8	1.2	0
4.	Abundances normalized to 100 max.		6.3	33.3	84.1	100	57.1	12.7	1.9	0
5.	Calculated abundances, 0.1% increm.		6.3	33.3	84.1	100	57.1	9.6	1.6	0.16
6.	Calculated errors > 0.1%, total, 3.6%		0	0	0	0	0	3.1	0.3	0.16
	Isotopomer ratios									
	Deuterated isotopomers	D_0	D_1	D_2	D_3	D_4	D_5			
7.	Ratio relative to 100	0	7.3	37.5	91.3	100	47.0			
8.	Ratio summed to 100	0	2.6	13.2	32.3	35.3	16.6			

The $C_{14}H_{19}O_9$ ion cluster for the deuterated galactopyranosides **79b – 81b** consists of seven peaks (lines 3 & 4), which are in principle composed of one undeuterated and five deuterated isotopomers (positions 2, 3, 4, 6_2), each of which consists of four ^{13}C- and ^{18}O-isotopomers (Table 18, line 1). The abundances of the ions in the ion cluster due to ^{13}C, ^{2}H and ^{18}O can be calculated using the analytical window in ChemDraw or by using a number of standard computer programs; in this case they were calculated using HiMass[73]. Determination of the ratio of deuterium isotopomers is much more difficult. The basic methodology is shown in Table 19. The observed abundance of each peak consists of the sum of the abundances of each C,O-isotopomer cluster multiplied by the abundance of the each D-isotopomer.

Table 19. Methodology for the prediction of multiple isotopomers, illustrated by the theoretical abundances of $C_{14}H_{19}O_9$ with abundances a_0-a_5 of the deuterated isotopomers, where a_0 = abundance of the D_0 isotopomer etc.

	331	332	333	334	335	336	337	338	339
a_0	$100a_0$	$16.3a_0$	$3.1a_0$	$0.4a_0$					
a_1		$100a_1$	$16.3a_1$	$3.1a_1$	$0.4a_1$				
a_2			$100a_2$	$16.3a_2$	$3.1a_2$	$0.4a_2$			
a_3				$100a_3$	$16.3a_3$	$3.1a_3$	$0.4a_3$		
a_4					$100a_4$	$16.3a_4$	$3.1a_4$	$0.4a_4$	
a_5						$100a_5$	$16.3a_5$	$3.1a_5$	$0.4a_5$
Sum each column, compare with data below, record error.									
Increment abundances, sum each column, determine error, save abundances if error is lower than recorded previously.									
Repeat until incremented permutations of a_0-a_5 are completed.									
79b – **81b** abundances	6.3	33.3	84.1	100	57.1	12.7	0	0	

The ratio of isotopomers with different deuterium contents was determined by a permutation calculation which was implemented in a Visual BASIC program with For Next loops. The problem with this form of analysis is the permutation increment. If this is 1 % then each value of the abundance (a_n) can take 100 values and as there are six variables the number of permutations is $100^6 = 1 \times 10^{12}$. Typically, each calculation takes circa 10^{-5} second and hence the total time for a calculation is 10^7 seconds, which is almost 116 days! In practice it is better to take a larger increment, such as 5 % that requires 10.7 minutes to determine rough values, and then to reduce the increment and the increment range.

Using this methodology, the abundances of the MH – CH_3OH cluster for the deuterated galactopyranoside **50b** were calculated to a final increment of 0.1 % (Table 18, line 5) with satisfactory errors (lines 7 & 8). The predominant deuterated isotopomers are D_3 and D_4, which correspond predominantly to the 2,4,6- and 2,3,4,6-deuterated-galactopyranosides isotopomers **80b**, **79b**. This accurately matches the ^{1}H-NMR data, which showed 90, 70 and 80% incorporation at C-2, C-3 and C-4 and circa 40 % replacement of each C-6 hydrogen, or equivalent to 90:70:80:80, which would give D_4 as the predominant ion.

Identical methodology was used to determine the ratios of the isotopomers of the deuterated mannopyranosides **54b** (Table 20, lines 7 & 8). In this case the predominant isotopomers were D_2 and D_3, which is consistent with the ^{1}H-NMR data which showed 72, 67, and 72 % incorporation at C-2, C-3 and C-4 respectively. Hence the assignments are D_1 **74b**, **76b**; D_2 **75b**, **77b**; D_3 **73b** and the minute amounts of D_4 and D_5 were not detected by ^{1}H-NMR.

Table 20. Analysis of the MH - CH_3OH ion clusters ($C_{14}H_{19}O_9$) of 2,3,4,6-tetra-*O*-acetyl-methyl-α-D-mannopyranosides **54b**, **73b** – **77b**.

Line	Description \ *m/z*	331	332	333	334	335	336	337	338	339
1.	Theoretical abundances for $C_{14}H_{19}O_9$	100	16.3	3.1	0.4					
2.	**54b**, measured abundances	100	12	3						
	Deuterated mannopyranoside **73b** – **77b**									
3.	Observed abundances		15	38	41	13	3	0.6		
4.	Abundances normalized to 100 max.		36.6	92.7	100	31.7	7.3	1.5	0	0
5.	Calculated abundances, 0.1% increment		36.6	92.7	100	31.7	7.3	1.1	0.12	0.08
6.	Calculated errors > 0.1%, total, 0.6%		0	0	0	0	0	0.38	0.12	0
	Isotopomer ratios									
	Deuterated isotopomers	D_0	D_1	D_2	D_3	D_4	D_5			
7.	Ratio relative to 100	0	42	100	98	17	3			
8.	Ratio summed to 100	0	16.3	38.5	37.7	6.7	0.8			

It is noteworthy for both the galactopyranosides **50b** and the mannopyranoside **54b**, the predominant losses were MH - CH_3OH rather than MH - CH_3OD. This is clearly consistant with protonation and elimination at the anomeric centre which does not become deuterated during the course of Raney nickel deuteration.

Table 21. Analysis of the MH - CH_3OH and MH - CH_3OH – H ion clusters ($C_{14}H_{19}O_9$) of 2,3,4,6-tetra-*O*-acetyl-methyl-α-D-glucopyranosides **24b**, **71b**..

Line	Description \ *m/z*	330	331	332	333	334	335	336	337	338
1.	Theoretical abundances for $C_{14}H_{19}O_9$		100	16.3	3.1	0.4				
	Glucopyranoside **24b**									
2.	Observed abundances	31	80	9	1					
3.	Abundances normalized to 100 max.	38.8	100	11.3	1.25					
4.	Calculated abundances, 1 % increm.	38.8	100	16.5	2.9					
5.	Calculated errors > 0.1%, total, 6.8%	0	0	5.2	1.7					
	Fragmentation ratios, M - X	CH_3OH	CH_3O							
6.	Ratio relative to 100	41	100							
7.	Ratio summed to 100	29	71							
	Deuterated glucopyranoside **71b**									
8.	Measured abundances			25	35	60	7	5		
9.	Abundances relative to 100 max.			41.7	58.3	100	11.7	8.3	0	0
10.	Calculated abundances, 5 % increment			8.1	58.8	100	14.0	8.2	0.8	0.09
11.	Calculated errors > 0.1%, total, 37%			33.6	0.5	0	2.3	0	0.8	0
	Isotopomer ratios									
	Deuterated isotopomers		D_0	D_1	D_2	D_3	D_4			
12.	Ratio relative to 100		0	21	100	0	0.05			
13.	Ratio summed to 100		0	17	78		5			

Analysis of the mass spectra of the glucopyranoside **24b**, **71b** is complicated by competing MH – CH_3OH and CH_3OH – H losses. Fortunately the methodology used for analysis of isotopomers can be used in exactly the same way to determine the ratio of the fragmentation pathway products. Thus the C,O-isotopomer cluster (Table 21, line 1) was permutated as two sets of ions[74] and the results compared with the observed abundances (lines 2 & 3). The fit was satisfactory, except for some minor differences in the lower abundance ions and the ratio

was 41:100 (lines 6 & 7) for the two fragmentation patterns and very close to the value deduced from the ratio of *m/z* 330:331 (38.8:100, line 3).

Calculation of the abundances of the ions for the deuterated glucopyranosides **71b** gave satisfactory fits for all the ions, except *m/z* 332, which was grossly inaccurate (Table 21, lines 10 and 11). Repeated attempts with a smaller increment for the first abundance did not improve the fit. The most likely explanation is that the deuterated compound undergoes losses of deuterium as well as hydrogen and this would have the largest effect on the lowest mass ion as observed. Although, all but one of the peaks showed satisfactory fits, no meaningful interpretation can be made unless all the peaks are accurately predicted.

2.7. Deuteration of glucopyranoside 24a with smaller amounts of catalyst

Deuteration of 200 mg of the three hexopyranosides **24a**, **54a**, **50a** with 12 g of ultrasonicated Raney nickel gave good incorporation, albeit with poor selectivity, but to obtain high yields of the product the catalyst had to be washed with large amounts of deuterium oxide. Therefore we investigated the deuteration of glucopyranoside **24a** using smaller amounts of ultrasonicated Raney nickel, to improve economy and in the hope that the reaction might be more selective.

The reaction was run with the glucopyranoside **24a** (200 mg) employing different amounts of catalyst (12 g, 1 g, 0.5 g, 0.1 g) and the reaction mixture was refluxed in D_2O, for 0.5 h or for longer in some cases when less Raney nickel was used. The standard reaction reported previously is shown in Table 22, entry 1 for comparison. When 1 gram of ultrasonicated Raney nickel was used, under conditions that were identical to those reported previously, the degree of deuterium incorporation was reduced and the C-2, C-4 *vs* C-3 selectivity was marginally lower (entry 2). Reduction of the amount of catalyst to 0.5 gram, resulted in lower conversion but apparently higher selectivity after half an hour (entry 3). However after two and a half hours, the selectivity was poorer and deuteration was still incomplete (entry 4). The reaction utilising 0.1 gram of catalyst was found to be too slow, providing circa 28% deuteration at C-2 and C-4 when the reaction was prolonged for 3.5 hr (entry 5).

In conclusion, using 0.5 gram of ultrasonicated Raney nickel gave quite good selectivity for deuteration at C-2 and C-4 of glucopyranosides **24a**, but the reaction was too slow with 0.1

gram of Raney nickel. In all cases, no deuterium incorporation was observed at C-1, C-5, or C-6.

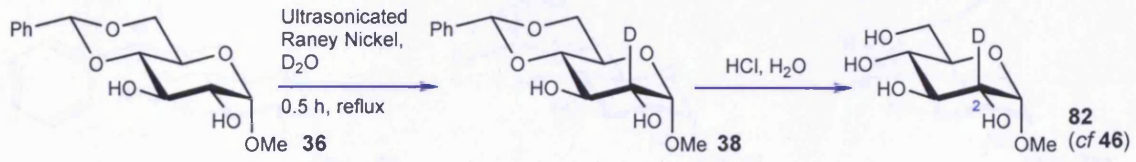
Table 22. % Deuterium incorporation into glucopyranoside **24a** (200 mg) by D_2O catalysed by different quantities of ultrasonicated Raney nickel.

Entry	Catalyst, g	Weight ratio	Reflux time, hours	Hydrogen position 2	3	4
1.	12	60	0.5	100	30	100
2.	1.0	5	0.5	66	25	50
3.	0.5	2.5	0.5	29	0	21
4.	0.5	2.5	2.5	67	40	50
5.	0.1	0.5	3.5	28	0	28

[a]Weight catalyst/weight glucopyranoside **24a**

2.8. Preparation of 2-[$^2H_1$]-methyl-α-D-glucopyranoside

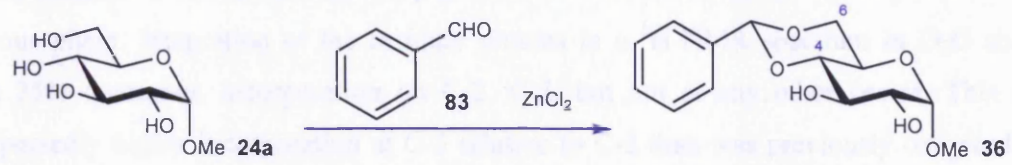
2-[$^2H_1$]-methyl-α-D-glucopyranoside **82** (cf. **46**) has been prepared by regioselective oxidation of 4,6-*O*-benzylidene-methyl-α-D-glucopyranoside **36**, reduction with sodium borodeuteride and cleavage of the benzylidene group[28] (Section 1.2.1.). We imagined that Raney nickel deuteration would enable this compound to be prepared in two rather than three synthetic steps. The deuteration of glucopyranoside **24a** by deuterium oxide catalysed by ultrasonicated Raney nickel gave quite good selectivity for deuteration at C-2 and C-4. By blocking C-4 with a benzylidene group (**38**), it appeared that, it might be possible to achieve high selectivity for deuteration at C-2. Benzylidene groups are cleaved by hydrogenation, but we assumed that as no added hydrogen is used in the deuterium exchange reaction, this should only be a minor side reaction. In the best possible case, exchange would occur at C-2 and subsequent benzylidene cleavage would exhaust the "active" hydrogen on the Raney nickel to give 2-[$^2H_1$]-α-methyl-D-glucopyranoside **82** in one pot!



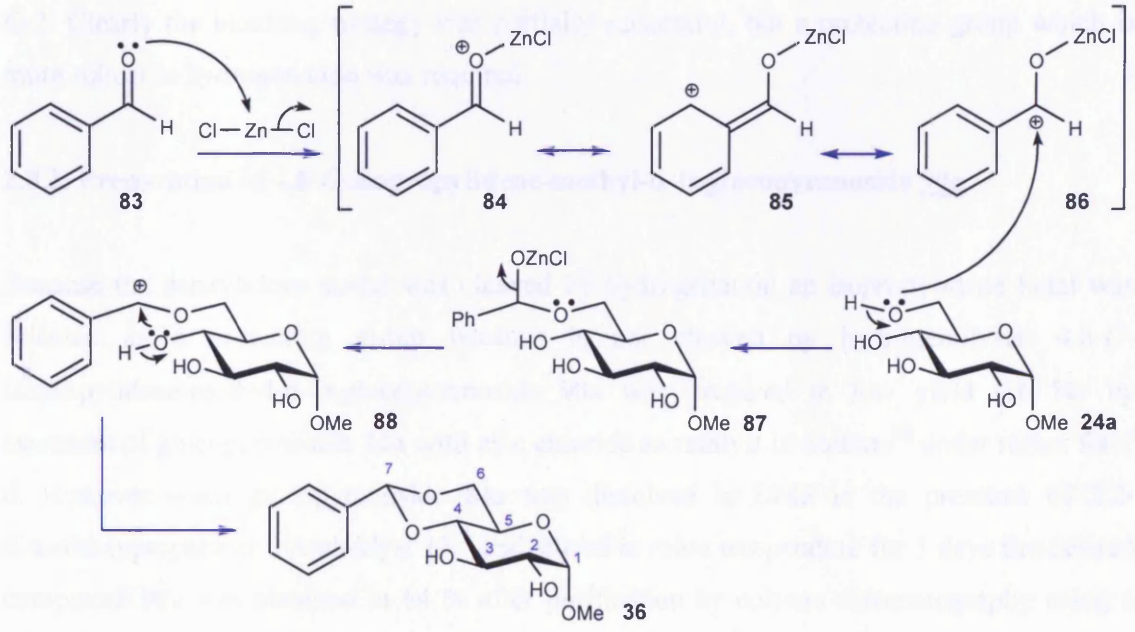
Scheme . Proposed synthesis of 2-[$^2H_1$]-methyl-α-D-glucopyranoside **82**

2.8.1. The preparation of 4,6-*O*-benzylidene-methyl-α-D-glucopyranoside 36

Stirring a mixture of glucopyranoside **24a**, zinc chloride and benzaldehyde for 3 h. at room temperature and crystallization from hot water gave 4,6-*O*-benzylidene-methyl-α-D-glucopyranoside **36** as white crystals in 51 % yield. It is essential to use good quality, dry, finely powdered zinc chloride and to use an efficient stirrer with the gelatinous reaction mixture. The product was identified by ^{1}H, ^{13}C-NMR spectroscopy, mass spectra (APCI-MS, MH *m/z* 283) and the melting point was 165 – 166 °C (lit.[75] 163 – 164 °C).



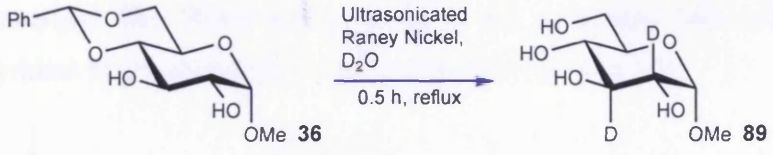
The primary hydroxyl group at C-6 of the glucopyranosides **24a** is less hindered than other hydroxyl groups in the molecule and therefore attacks the benzaldehyde metal complex **84** – **86** more easily than the other hydroxyl groups. A hemiacetal alkoxide **87** is formed which cleaves to give a benzylic oxonium ion **88**, which is attacked by the 4-hydroxyl group to give the benzylidene acetal **36**. Benzylidene groups prefer to be part of six-membered ring acetals, rather than five-membered rings. The most stable and least sterically hindered arrangement is with the phenyl substituent in an equatorial position[76,77].



Scheme . Mechanism for the formation of of 4,6-*O*-benzylidene-methyl-α-D-glucopyranoside **36**.

2.8.2. The deuteration of 4,6-*O*-benzylidene-methyl-α-D-glucopyranoside 36

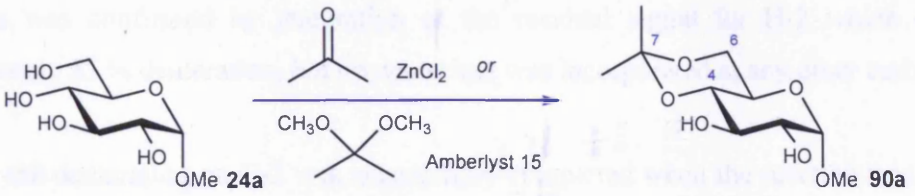
4,6-*O*-benzylidene-methyl-α-D-glucopyranoside **36** was deuterated according to the general procedure (12 g Raney nickel) and refluxed for 0.5 h. under nitrogen. TLC ($CHCl_3$: MeOH, 80:20) indicated disappearance of starting materials and formation of methyl glucopyranoside **89** plus a trace of a less polar product. The work-up was designed to separately extract organic soluble components such as remaining starting material and the water soluble hexopyranoside **89**. Evaporation of the organic soluble phase provided a trace of a sugar (10 mg) that could not be identified, and methyl glucopyranoside **89** was collected in 76 % yield from the aqueous phase. Integration of the residual protons in a ^{1}H-NMR spectrum in D_2O showed circa 75% deuterium incorporation on C-2, C-3, but not at any other centre. This is an unexpectedly higher incorporation at C-3 relative to C-2 than was previously observed with the glucopyranoside **24a,** and suggested that deuteration occurred which was then followed by hydrogenation of the benzylidene group.



The reaction was run again, with less Raney nickel (0.5 g) and refluxed for 20 minutes which resulted in cleavage of the benzylidene group and roughly 30 % deuterium incorporation at C-2. Clearly the blocking strategy was partially successful, but a protecting group which is more robust to hydrogenation was required.

2.8.3. Preparation of 4,6-*O*-isopropylidene-methyl-α-D-glucopyranoside 90a

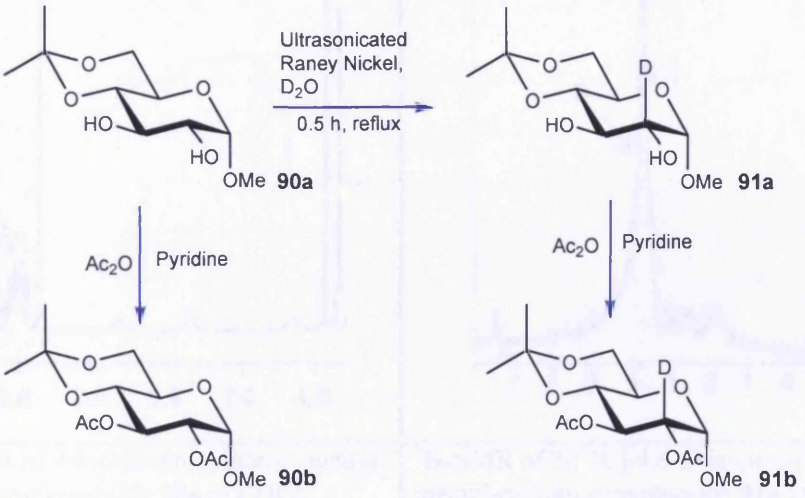
Because the benzylidene acetal was cleaved by hydrogenation an isopropylidene ketal was selected as a protecting group because is not cleaved by hydrogenolysis. 4,6-*O*-isopropylidene-methyl-α-D-glucopyranoside **90a** was prepared in low yield (16 %) by treatment of glucopyranoside **24a** with zinc chloride as catalyst in acetone[78] under reflux for 2 d. However when glucopyranoside **24a** was dissolved in DMF in the presence of 2,2-dimethoxypropane and Amberlyst 15[79] and stirred at room temperature for 5 days the desired compound **90a** was obtained in 64 % after purification by column chromatography using a solvent gradient of chloroform to ethanol.



The product **90a** was identified by ^{1}H-NMR spectroscopic analysis that showed the presence of two up-field singlet resonances at 1.60, 1.54 ppm corresponding to the methyl protons and 29.1, 19.1 ppm in the ^{13}C-NMR spectrum[80].

2.8.4. Raney nickel catalysed deuteration of 4,6-*O*-isopropylidene-methyl-α-D-gluco pyranoside 90a by deuterium oxide

Isopropylidene ketal **90a** was deuterated using the standard half hour procedure. The ^{1}H-NMR spectrum of the product showed no cleavage of the isopropylidene groups during exchange. To gain the benefit of better dispersion of the ^{1}H-NMR, the acetates of the starting material **90a** and its deuterated form **91a** were converted to the peracetates **90b**, **91b** using acetic anhydride in pyridine by the method previously described (Section 2.5).

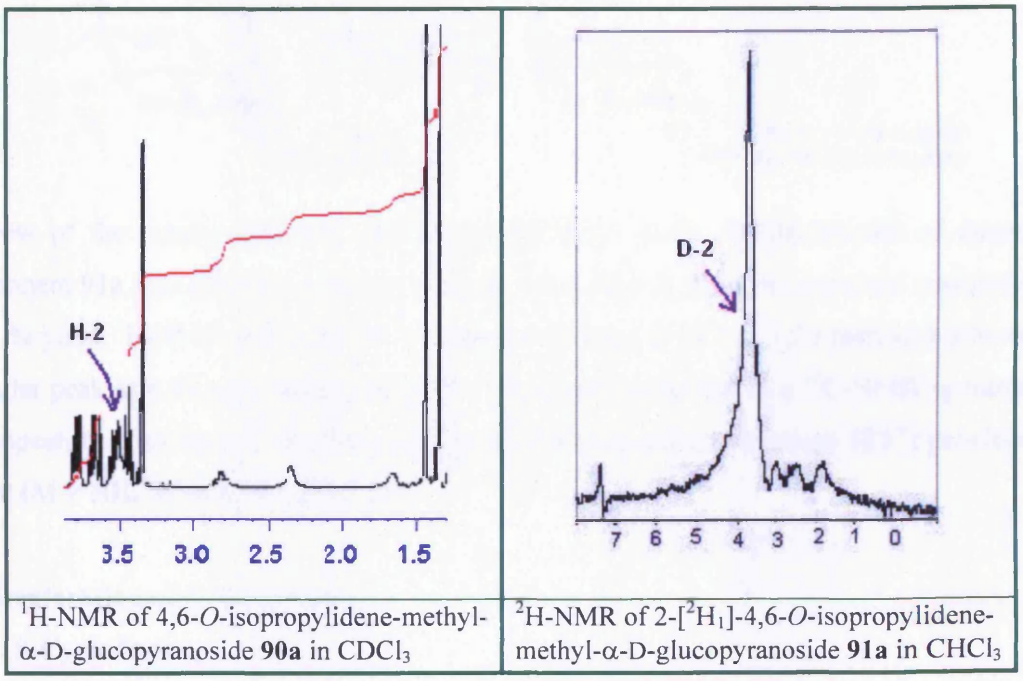


The most significant proton signals in the ^{1}H-NMR spectrum of diacetate **90b**, **91b** were H-1, H-2, and H-3. The ^{1}H-NMR spectrum of the diacetate **90b** showed the presence of a down-field triplet proton resonance at 6.07 ppm with a coupling constant of 9.9 Hz corresponding to the 3-H and a doublet of doublets resonance of 9.9 and 3.8 Hz at 5.25 ppm from 2-H, plus a doublet signal of 3.7 Hz at 4.97 ppm corresponding to 1-H. Whereas the ^{1}H-NMR spectrum of the partially deuterated diacetate **91b** showed a triplet proton resonance of 9.7 Hz at 5.92 ppm which was overlapped with a doublet of 9.5 Hz for 3-H, indicating partial deuteration at

C-2. This was confirmed by integration of the residual signal for H-2 which indicated approximately 55 % deuteration, but no deuterium was incorporated at any other carbon.

However the deuteration on C-2 was successfully completed when the reaction was repeated again using 0.9 g of catalyst and the reaction mixture was refluxed for a longer period of time (1.5 hr.) to give compound **91a** in 55 % yield. ^{1}H-NMR spectrum of that compound was comparatively easy to interpret and there was no need to acetylate. The disappearance of 2-H proton resonance gave a singlet proton resonance at 4.69 ppm corresponding to 1-H and a doublet of 9.5 Hz from 3-H at 3.68 ppm. ^{13}C-NMR spectrum showed the absence of the 2-C resonance and high resolution mass spectrometry (ES+) provided the (M + NH_4^+) ion at *m/z* 253.1505.

^{2}H-NMR spectroscopy of 2-[$^{2}H_1$]-4,6-*O*-isopropylidene-methyl-α-D-glucopyranoside 91a



^{1}H-NMR of 4,6-*O*-isopropylidene-methyl-α-D-glucopyranoside **90a** in $CDCl_3$

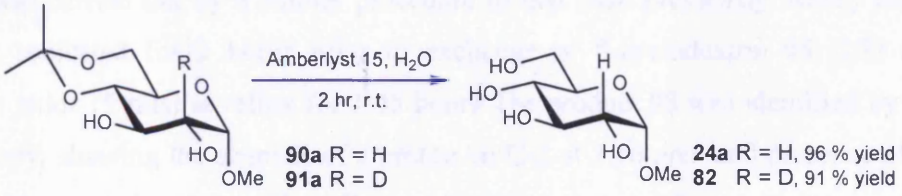
^{2}H-NMR of 2-[$^{2}H_1$]-4,6-*O*-isopropylidene-methyl-α-D-glucopyranoside **91a** in $CHCl_3$

The ^{2}H-NMR spectrum of deuterated isopropylidene glucopyranoside **91a** was run in $CHCl_3$ and compared with ^{1}H-NMR spectrum of the non-deuterated analogue **90a** in $CDCl_3$. In the ^{1}H-NMR spectrum of **90a** in $CDCl_3$, H-2 appeared at 3.60 ppm as a doublet of doublets (*J* 9.0, 3.9 Hz), whereas a broad singlet was observed at 3.62 ppm in the ^{2}H-NMR spectrum of deuterated **91a**. As ^{1}H-^{2}H couplings are 15 % of ^{1}H-^{1}H-couplings, in principle the expected

pattern could be a doublet of doublets of 1.4, 0.6 Hz. However with the normal broadening due to deuterium, the broad singlet observed is more likely.

2.8.5. Cleavage of the isopropylidene protecting group

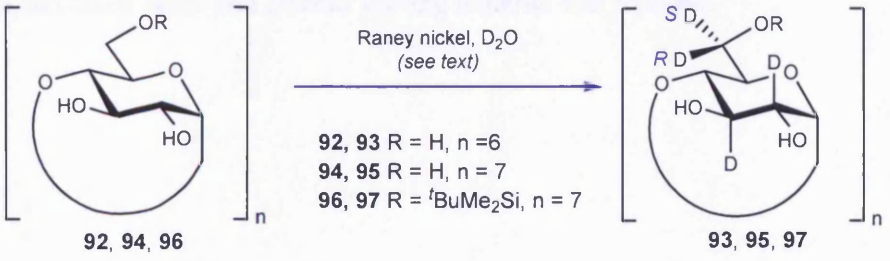
In order to prepare 2-[$^2H_1$]-methyl-α-D-glucopyranoside **82** from isopropylidene ketal **91a**, the isopropylidene group had to be removed. Initally undeuterated isopropylidene ketal **90a** was tested as a model substrate; treatment with Amberlyst 15 (a cation exchange resin) in water for 2 hr. in room temperature hydrolysed the protecting group[81]. The progress of reaction was monitored by TLC ($CHCl_3$, CH_3OH, 60:40) which revealed the absence of starting material and the presence of glucopyranoside **24a** at R_f 0.4. The yield was virtually quantitative (96 %) and the glucopyranoside **24a** produced was identical by ^1H, ^{13}C NMR spectroscopy with commercial material.



In view of the results provided from the preliminary study, the hydrolysis of deuterated component **91a** was carried out using the same procedure to form the expected compound **82** in 91 % yield. ^1H-NMR spectroscopy showed the absence of H-2 at 3.50 ppm and presence of a singlet peak at 4.73 ppm belonging to H-1. This was confirmed in a ^{13}C-NMR spectrum by "disappearance" of the 2-C resonance. High resolution mass spectroscopy (ES^+) provided the parent ($M + NH_4^+$) ion at *m/z* 213.1191.

2.9. Deuteration of Cyclodextrins

2.9.1. β-Cyclodextrin



It has been reported that Raney nickel catalysed deuteration of α-cyclodextrin **92** by deuterium oxide at reflux for 24 hours results in facile exchange of H-2, and 2H-6, but only 30 % replacement of H-3 (**93**)[31]. Similarly when β-cyclodextrin **94** was subjected to Raney nickel and deuterium oxide at reflux for 12 hours, complete exchange of H-2 occurred and there was 30%, 96% and 84% deuteration of H-3, H-6*R* and H-6*S* respectively (**95**). The degree of deuteration at C-6 was determined by using the NMR shift reagent, ASANA (sodium anthraquinone 2-sulfonate) to resolved the signals[53]. The slower rate of deuteration at C-3 mirrors the results with glucopyranoside **24a** and the selectivity is probably further enhanced by the orientation of H-3 into the hindered cavity of the cyclodextrin.

In view of the encouraging results from the initial studies of the deuteration of the hexopyranosides **24a**, **36a**, **50a**, **54a**, **90a** by ultrasonicated Raney nickel in deuterium oxide the complete deuteration of β-cyclodextrin **94** looked promising. Therefore deuterium labeling was carried out by a similar procedure to that used previously. Raney nickel (8 g, wet) was sonicated for 2 hours prior to exchange of β-cyclodextrin **94** (200 mg) with deuterium oxide (5 mls) at reflux for 1.45 hours The product **95** was identified by ^{1}H-NMR spectroscopy, showing the absence of a proton on C-2 at 3.56 ppm and presence of a singlet peak at 4.98 ppm belonging to H-1. The proton at C-3 changed from a triplet to a doublet of 9.0 Hz at 3.88 ppm due to deuteration at C-2. Approximately 30% deuteration was detected at C-3 and as expected H-4 appeared as a doublet of 9.8 Hz from coupling with proton on C-5 due to deuteration at C-3 and also a triplet of 9.5 Hz from coupling with H-3 and H-5. It was impossible to determine the exact rate of deuteration of each proton on C-6 because of signal overlapping of H-5, H-6 and H-6′. In order to examine the viability of the developed deuteration procedure to the cyclodextrin derivatives, silylated β-cyclodextrin **96** was prepared by treatment of β-cyclodextrin **94** with *tert*-butyldimethylsilyl chloride in dry pyridine (Section 4.1.3.1.). A solution of silylated β-cyclodextrin **96** in deuterium oxide was refluxed with sonicated Raney nickel for 2 h. However a ^{1}H-NMR spectrum showed no deuteration had taken place and instead starting material was isolated.

Chapter 3

Cyclodextrins

Introduction

3. Introduction

3.1. General introduction

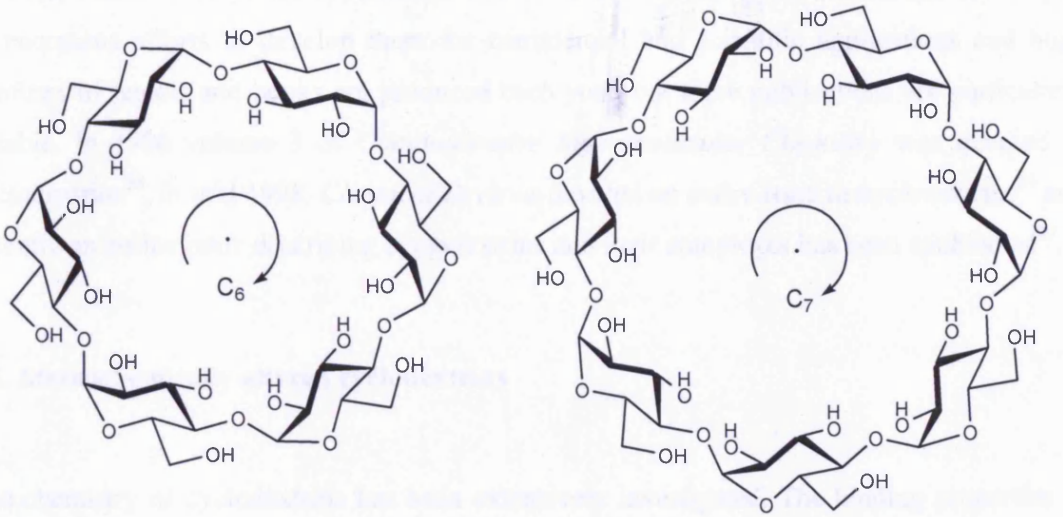
Cyclodextrins (CDs) are macrocyclic oligosaccharides consisting of six- (α-cyclodextrin **92**), seven- (β-cyclodextrin **94**), eight- (γ-cyclodextrins) or more -glucopyranoside units predominantly in the $^4C_1$ conformation linked by α-(1→4) bonds, surrounding a central cavity. They were first discovered in 1891, as water soluble products of the enzymatic degradation of starch. In 1904, Schardinger was able to isolate these compounds by the action of *Bacillus macerans* amylase upon starch and named them α-dextrin and β-dextrin. In the older literature these compounds are described as Schardinger dextrins. Freudenberg isolated γ-dextrin in 1935, but at that time the structure of those products was still uncertain[82] and was only in 1942 that the structures of α-dextrin **92** and β-dextrin **94** were determined by X-ray crystallography[83].

Scheme. A CPK depiction of a molecular model of β-cyclodextrin **94** produced using PC Model 8 and Pov-Ray. Carbon atoms are turquoise, oxygens red, and the hydrogen atoms are grey. The view is from the "top" face showing the 14 secondary hydroxyl groups.

3.1.1. Structure and physical properties

The X-ray structure of cyclodextrins, shows that the secondary hydroxyl groups on C-2, C-3, are located on the wider edge of a truncated cone and the primary hydroxyl groups on C-6 on the other edge. Apolar hydrogens on C-3, C-5 make a rigid, hydrophobic cavity and hence they are able to encapsulate non-polar compounds within their bucket-like structure (host-guest binding) and the hydrophilic exterior allows for solubilization in aqueous solution. Binding constants have been measured for hundreds of compounds[84], but the highest affinity

guest is probably lithocholic acid (a tetracyclic steroid) which binds to β-cyclodextrin **94** with a binding constant of 1.17 (± 0.10) x 10^6 Mol^{-1} in 1 % DMSO in water[85].



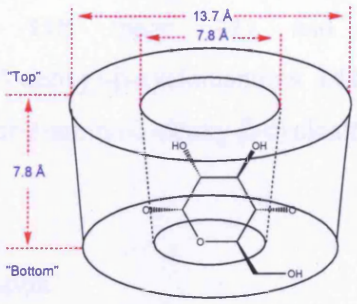
92 α-Cyclodextrin **94** β-Cyclodextrin

Scheme. The structures of α- and β-cyclodextrins.

The three dimensional form and size of cyclodextrins is the most important parameter in the formation of inclusion complexes with hydrophobic compounds (Table 23).

Table 23. Properties of cyclodextrins[86,87]

Cyclodextrin	α **92**	β **94**	γ **113**
Number of glucose units	6	7	8
Inner diameter a, Å	4.5	7.0	9.5
Outer diameter, b, Å	13.7	15.3	16.9
Height c, Å	7.9	7.9	7.9
Cavity volume, $Å^3$	174	262	427
Molecular weight, g/Mol	972	1135	1297
Solubility in H_2O, g/L	14.5	1.85	23.2



3.1.2. Applications of Cyclodextrins

As mentioned earlier, cyclodextrins are capable of accommodating organic substances within their cavity. Due to this property, cyclodextrins can be used to hold sensitive and active compounds such as flavours and fragrances and protect against heat, light or air, hence resulting in stabilization when subjected to long term storage. As cyclodextrins are soluble in water, they can greatly increase the solubility of water insoluble substances by encapsulation. The taste of foods or drugs can also be improved by masking off-flavours and unpleasant odours through cyclodextrins-complex formation. In the analytical chemistry field,

cyclodextrins are also used for the separation of enantiomers by high performance liquid chromatography (HPLC) or gas chromatography (GC). The stationary phases of these columns contain methylated cyclodextrins or other derivatives. Cyclodextrins are the subject of enormous efforts to develop them for commercial and scientific applications and huge numbers of papers and books are produced each year, but three publications are particularly notable. In 1996 volume 3 of *Comprehensive Supramolecular Chemistry* was devoted to cyclodextrins[88], in mid 1998, *Chemical Reviews* devoted an entire issue to cyclodextrins[89] and recently an online book describing cyclodextrins and their complexes has been epublished[90].

3.2. Stereochemically altered cyclodextrins

The chemistry of cyclodextrins has been extensively investigated. The binding properties of countless guests have been investigated and the structures have been modified by derivatisation or replacement of the hydroxyl groups in order to increase the compatibility between cyclodextrins as hosts, and non polar compounds as guests, e.g., mono- or disubstituted, per-alkylation, per-esterification[82], halogenation of primary hydroxyl groups[91,92], perbenzylation[93]. There is only a small group of compounds in which some modifications have been made to change the cyclodextrin stereochemistry, which includes per(2,3-anhydro)-cyclomannoepoxides **116, 117, 118** (page 44) and per-3,6-anhydrocyclodextrins **120, 122** (page 45), heptakis(3-deoxy)-β-cyclomannins **144a, 144b, 144c** (page 50), β-cycloaltrin **137a** (page 49) and per-3-amino-3-deoxy-β-cycloaltrin **137b** (page 49).

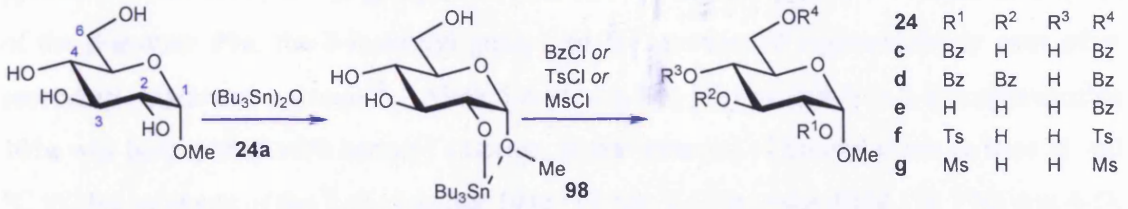
3.3. Regioselectivity of hydroxyl group functionalisation

3.3.1. Monosaccharides

3.3.1.1. Tosylation

In a unprotected monosaccharide such as methyl-α-D-glucopyranoside **24a**, there are two types of hydroxyl groups: a primary hydroxyl groups at C-6 and secondary hydroxyl groups at C-2, C-3, C-4. The primary hydroxyl group at C-6 is the most reactive one because it is the least sterically encumbered and therefore this group can be protected with high regioselectivity, and normally excellent yields are obtained. In general, the C-2 hydroxyl group in the α-anomer of D-glucopyranoside is more acidic than C-3 due to the closeness of

that group to the anomeric centre, and hence is more nucleophilic and more reactive toward electrophiles.

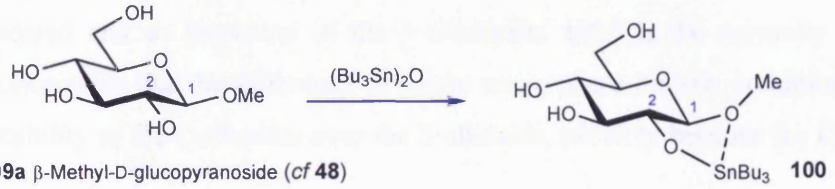


24	R^1	R^2	R^3	R^4
c	Bz	H	H	Bz
d	Bz	Bz	H	Bz
e	H	H	H	Bz
f	Ts	H	H	Ts
g	Ms	H	H	Ms

Bis(tributyltin)oxide has been used to modify and enhance the reactivity of polyols, by the formation of stannoxides, which can form complexes with adjacent oxygen substituents (**98**). The selectivity achieved depends on the ratio of bis(tributyltin)oxide and electrophile to the polyol, the conditions of the reaction with the electrophile and possibly the conditions for stannylation.

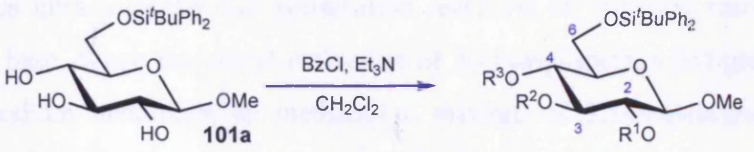
The following three examples all used the same ratio of the reagents; bis(tributyltin)oxide (3 equiv "Bu_3Sn") and subsequent reaction with 3 equivalents of benzoyl chloride. When methyl-α-D-glucopyranoside **24a** was treated with both the reagents sequentially at room temperature the product consisted of the 2,6-di-*O*-benzoate **24c** (81.4 % yield) and the 2,3,6-tri-*O*-benzoate **24d** (18.4 % yield), whereas when benzoylation was run at -10°C, the 2,6-di-*O*-benzoate **24c** was the sole product (95 % yield). But when the stannylation was performed at 140°C and the benzoylation was run at -15°C, the 6-*O*-benzoate **24e** was the major product (73 % yield) together with a small amount of the 2,6-di-*O*-benzoate **24c** (20 % yield).

In addition when the reactivity of methyl- α-D-glucopyranoside **24a** was examined with *p*-toluenesulfonyl chloride or methanesulfonyl chloride, the 2,6-di-*O*-tosylate **24f** (80.6 %) and the 2,6-di-*O*-mesylate **24g** (75.4 %) were formed with high selectivity. It has been noted from the above experiments that formation of the five membered coordination ring **98** from methyl-α-D-glucopyranoside **24a** which bears a *cis*-1,2-hydroxyether is less favorable than the formation of the corresponding coordination ring **100** from the *trans*-1,2-hydroxyether moiety of methyl-α-D-glucopyranoside **99a**[94].



99a β-Methyl-D-glucopyranoside (*cf* **48**) **100**

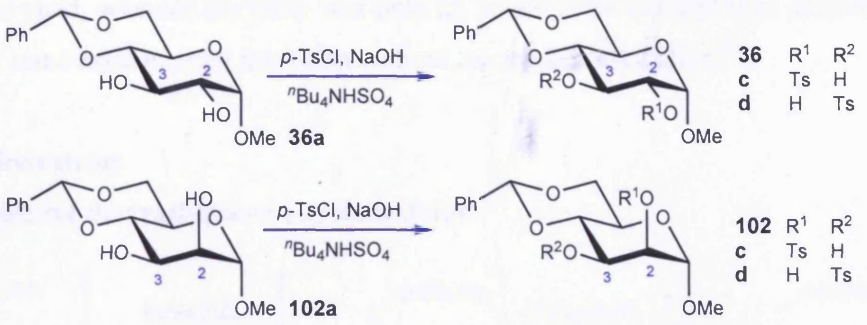
The differentiation of secondary hydroxyl groups of carbohydrates is very important in order to protect hydroxyl groups with high regioselectivity. In the α-anomer of methyl-D-gluco-pyranoside **24a**, the hydroxyl group at C-2 position is more nucleophilic whereas in the case of the β-anomer **99a**, the 3-hydroxyl group can be transformed regioselectively over other secondary hydroxyl groups[95]. Methyl-6-*O*-(*tert*-butyldiphenylsilyl)-β-D-glucopyranoside **101a** was benzoylated with benzoyl chloride, in the presence of triethylamine as base at –60 °C to give mixtures of the 2-*O*-benzoate **101c** (12 %), 3-*O*-benzoate **101d** (19.5 %) and 4-*O*-benzoate **101e** (10.5%). The dibenzoates **101f**, **101g** were also isolated as minor products (less than 5 %), and starting material **101a** was recovered at 26 % yield. However higher regioselectivity was achieved when the reaction was conducted at 0 °C, the 3-*O*-benzoate **101d** was isolated as the major product (62.5 % yield) together with the 2-*O*-benzoate **101c** and the 4-*O*-benzoate **101e** as minor products[96].



101	R^1	R^2	R^3
c	Bz	H	H
d	H	Bz	H
e	H	H	Bz
f	Bz	Bz	H
g	H	Bz	Bz

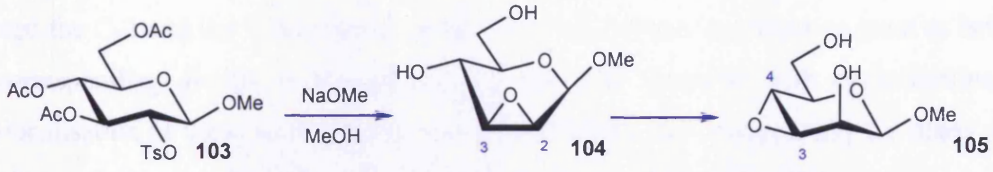
The 4,6-*O*-benzylidene-methyl-α-D-gluco- and manno-pyranosides **36a**, **102a** have unprotected hydroxyl groups at C-2 and C-3, which are regioselective monotosylated under phase-transfer conditions using tetrabutylammonium hydrogen sulphate, *para*-toluene-sulphonyl chloride and dilute sodium hydroxide. In the aqueous phase, the sodium alkoxide of the hexopyranosides **36a**, **102a** undergo cation exchange to give the tetrabutylammonium alkoxide which is then transferred into the organic phase and reacts with *para*-toluenesulphonyl chloride to form the monotosylate **36c**, **102c**, **102d**. This is more soluble in the organic phase and hence evades further deprotonation by sodium hydroxide in the aqueous phase. In the case of glucopyranoside **36a,** a higher yield of the 2-*O*-tosylate **36c** (78 %) was obtained compared with the 3-*O*-tosylate (7 %) due to the higher intrinsic reactivity of position 2. When the mannopyranoside **102a** was reacted under the same conditions the 2-*O*-tosylate **102c** was the only product whereas partial tosylation with *para*-toluenesulphonyl chloride in pyridine gave the 3-*O*-tosylate **102d** as the major product[97]. Normally equatorial hydroxyl groups are more reactive than axial hydroxyl groups because the latter are more sterically hindered and so formation of the 3-*O*-tosylate **102d** is the normally anticipated result. It is conceivable that the difference in results under phase transfer conditions is due to preferential stability of the 2-alkoxide over the 3-alkoxide, possibly because the C-O bond of

the 2-alkoxide can be stabilised by donation into the antibonding orbital of the anomeric C-O bond.

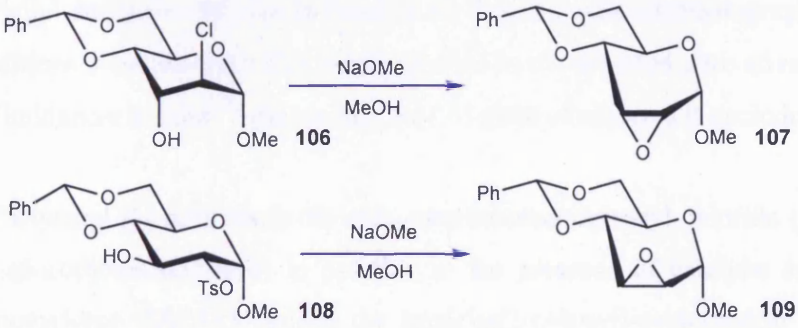


3.3.1.2. Epoxidation

2,3-Anhydro sugars (epoxides) can be readily obtained using a general method which involves intramolecular S_N2 substitution reactions of tosylated carbohydrates by treatment with a base. When the acetyl derivative of 2-*O*-tosyl-methyl-β-D-glucoside **103** was treated with sodium methoxide in methanol a mixture of 2,3-anhydromannoside **104** and 3,4-anhydroaltroside **105** were produced. The latter results from intramolecular nucleophilic attack of the 2,3-epoxide by the 4-alkoxide (**104**), which is known as a Payne rearrangement or *epoxide walk*[98,99].



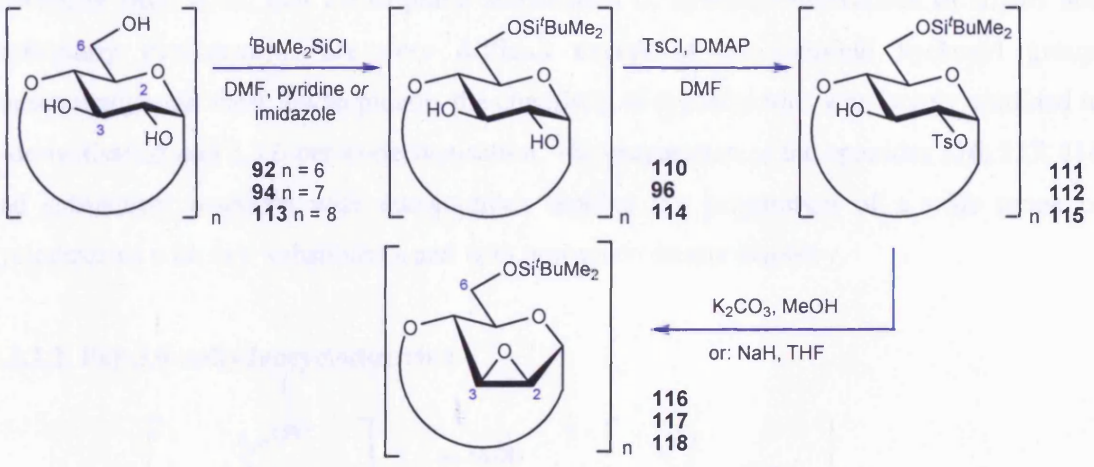
Similarly the action of sodium methoxide on 2-chloro-2-deoxy-idoside **106** gave the 2,3-anhydro-allopyranoside **107** as only product, whereas the 2-*O*-tosyl glucoside **108** yielded the 2,3-anhydro-mannoside **109** in quantitative yield[100].



Microwave irradiation (100 W) of 2-*O*-tosyl-glucopyranoside **108** at 100 °C for 6 minutes in the presence of Al_2O_3:KOH (3:1, 2 equiv.) gave the 2,3-anhydro-mannoside **109** in nearly quantitative yield, whereas the yield was only 25 % when the reaction was performed under exactly the same experimental condition without microwave irradiation[101].

3.3.2. Cyclodextrins

3.3.2.1. Selective derivatisation of cyclodextrins

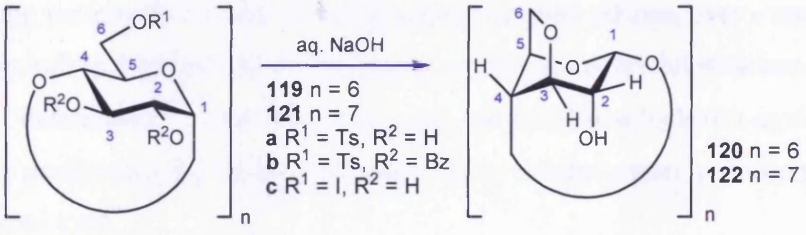


The differential reactivity (6 > 2 > 3,4) of the hydroxyl groups of α-methyl-D-gluco-pyranoside **24a** is mirrored in the reactions of cyclodextrins. The reactivity difference between the C-2 and the C-6 hydroxy groups of cyclodextrins, is at least as great as between the corresponding groups in the glucopyranoside **24a**. However with cyclodextrins, full functionalisation at these sites requires multiple reactions and consequently for many years, selective cyclodextrin chemistry was plagued by low yields and disputes over the purity of products. The break through came when it was shown that *t*-butyldimethylsilylation showed high selectivity for the C-6 hydroxy group of cyclodextrins. When β-cyclodextrin **94** was treated with *tert*-butyldimethylsilyl chloride and pyridine in DMF, the per-6-*O*-*t*-butyldimethylsilyl derivative **96** was isolated in 82 % yield after chromatography and under identical conditions γ-cyclodextrin **113** was converted to the silylated derivative **114** in 82 % yield. Use of imidazole as base[102] gave a only a 61 % yield of silylated β-cyclodextrin **96**[103].

Treatment of silylated β-cyclodextrin **96** with *para*-toluenesulphonyl chloride (3 equivalents per silylated glucopyranoside unit) in pyridine in the presence of catalytic amounts of 4-dimethylaminopyridine (DMAP) yielded the *heptakis*(2-*O*-tosyl)-cyclodextrin **112** in 50 % yield[104]. Later the procedure was refined and applied to all of the per-6-*O*-silyl ether

derivatives (**110**, **96**, **114**) to give the 2-per-*O*-tosyl-α-, β-, γ-cyclodextrins **111**, **112**, **115** in better yields (55 %, 58 %, 60 % respectively)[105]. Treatment of these tosylates with potassium carbonate in methanol gave the 3-*O*-alkoxides, which underwent intramolecular S_N2 like nucleophilic attack of the 2-*O*-tosyl substituents to give the per-(2,3-anhydro)cyclomannoepoxides **116, 117, 118** in high yield (90 - 93%). In an alternative procedure heptakis-(2-*O*-tosyl) derivative **112** was treated with sodium hydride in tetrahydrofuran, but the desired product **117** was prepared in lower yield (75 %)[105]. It has previously been noted that nucleophilic substitution of hydroxyl derivatives of sugars and particularly cyclodextrins are very difficult except at the terminal hydroxyl group, consequently until these developments the chemistry of cyclodextrins was largely confined to 6-derivatisation and 2,3,6-per-*O*-derivatisation. The preparation of the epoxides **116, 117, 118** and subsequent reactions with nucleophiles enabled the preparation of a wide range of cyclodextrins with new substituents and with non-*gluco* stereochemistry.

3.3.2.2. Per-3,6-anhydrocyclodextrins



Selective per-*O*-6-tosylation[106] of cyclodextrins and displacement by halides or other nucleophiles has been used previously for preparing 6-halo[91]- and 6-amino[107]-cyclodextrins. The displacement of 6-*O*-tosylates by a 3-hydroxyl group orientated *syn* to the 6-substituent to give 3,6-anhydro-monosaccharides was first discovered in the early years of synthetic carbohydrate chemistry[108]. Combining these ideas gave viable routes to per-3,6-anhydrocyclodextrins **120**, **122**, which were reported simultaneously by two research groups and were the first cyclodextrins with wholly non-natural conformations. Heptakis-6-*O*-tosyl-β-cyclodextrin **121a** was prepared by Stoddart's group without incident and then heated with a 1M aqueous solution of sodium hydroxide for 2 days to yield heptakis(3,6-anhydro)-β-cyclodextrin **122** in 44 % yield[109]. Tosylation of α-cyclodextrin **92** was more difficult and hexakis(6-*O*-tosyl)-α-cyclodextrin **119a** could not be isolated in a pure state. Consequently hexakis(2,3-*O*-benzoyl-6-*O*-tosyl)-α-CD **119b** was prepared in high yield (95 %)[110] and the esters hydrolysed using a mixture of triethylammonium, methanol and water (1:5:1) at reflux

for 4 days to give hexakis(6-*O*-tosyl)-α-cyclodextrin **119a**. Treatment with sodium hydroxide produced hexakis(3,6-anhydro)-β-cyclodextrin **120** in 22 % yield[109]. Alternatively, Defaye's group, treated α- or β-cyclodextrins **92, 94** with triphenylphosphine and iodine to give the 6-iodo-derivatives **119c**, **121c** (80, 88 % yield). These were cyclised with sodium hydroxide in DMSO to give the per-3,6-anhydrocyclodextrins **120**, **122**, in 75 and 88 % yield respectively[91]. When this approach was repeated by the Stoddart group using Amberlyst IRA-400 resin (hydroxide form) or potassium hydroxide, heptakis(3,6-anhydrocyclo)-β-dextrin **122** was formed in 30 % and 26 % yield respectively[111]. An important aspect of both routes involving iodide displacement, was the removal of metals ions from the product by ultra-filtration (Defaye[91]) or crystallisation (Stoddart[111]). Heptakis(3,6-anhydrocyclo)-β-dextrin **122** preferentially binds potassium over, lithium, sodium, rubidium and caesium cations[111,112] and the X-ray crystal structure of a 2KOH. $3.5H_2O$. $0.333(CH_3COCH_3)$ complex showed that the potassiums were 10- and 11-coordinate[111].

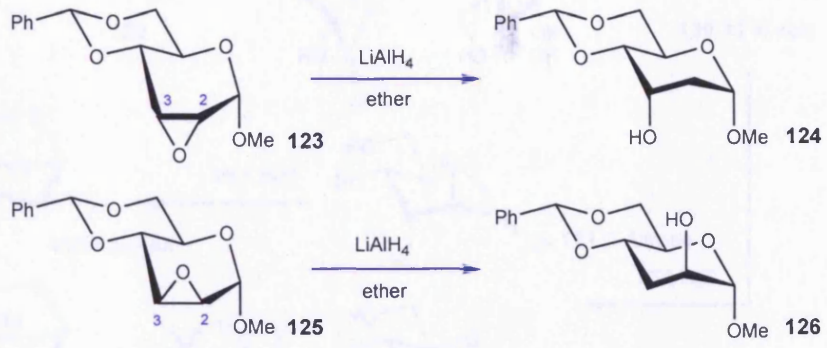
Octakis(3,6-anhydro)-γ-cyclodextrin was prepared by the tosylation route (95.2 % yield[112]) and had a strong preference for binding rubidium and caesium cations, over a total of 21 other metal ions, (including the other alkali metal ions) and the X-ray crystal structure of a caesium complex was determined[113]. Pentakis- and three tetrakis(3,6-anhydro)-α-cyclodextrins all showed strong preferences for binding rubidium and to a lesser extent potassium over all the other alkali metal ions[114].

The ^{1}H NMR spectrum of the per-3,6-anhydrocyclodextrins **120**, **122** showed signals at 3.87 and 3.94 ppm (both $^3J_{2,3}$ = 5 Hz) for H-2 and at 4.31 ($^3J_{3,4}$ = 5.5 Hz; $^3J_{4,5}$ = 2 Hz) and 4.19 ($^3J_{3,4}$ = 5 Hz; $^3J_{4,5}$ = 2.5 Hz) ppm for H-4. The values of these coupling constants are those expected for equatorial, equatorial coupling, which confirms that the 2,3-anhydro-D-glucose units are in the $^1C_4$, rather than the $^4C_1$ conformation of the parent cyclodextrins **92, 94**[115].

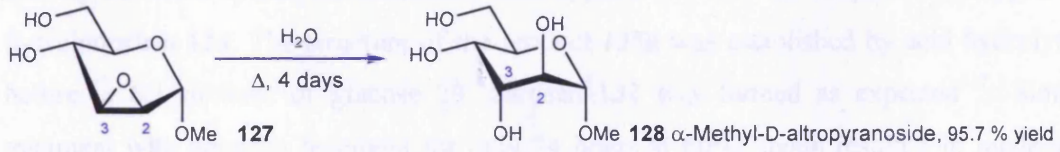
3.4. Regioselectivity of ring opening

The nucleophilic ring opening of epoxides goes via a S_N2 mechanism and hence the orientation of the former nucleophile and the hydroxyl substituent are always *trans* to each other in the product. Similarly, nucleophilic ring opening of the epoxide moiety of 2,3-anhydro-hexapyranosides with nucleophiles gives *trans*-products, but there are two possible products the *trans*-diaxial and the *trans*-diequatorial. Most frequently the products result from

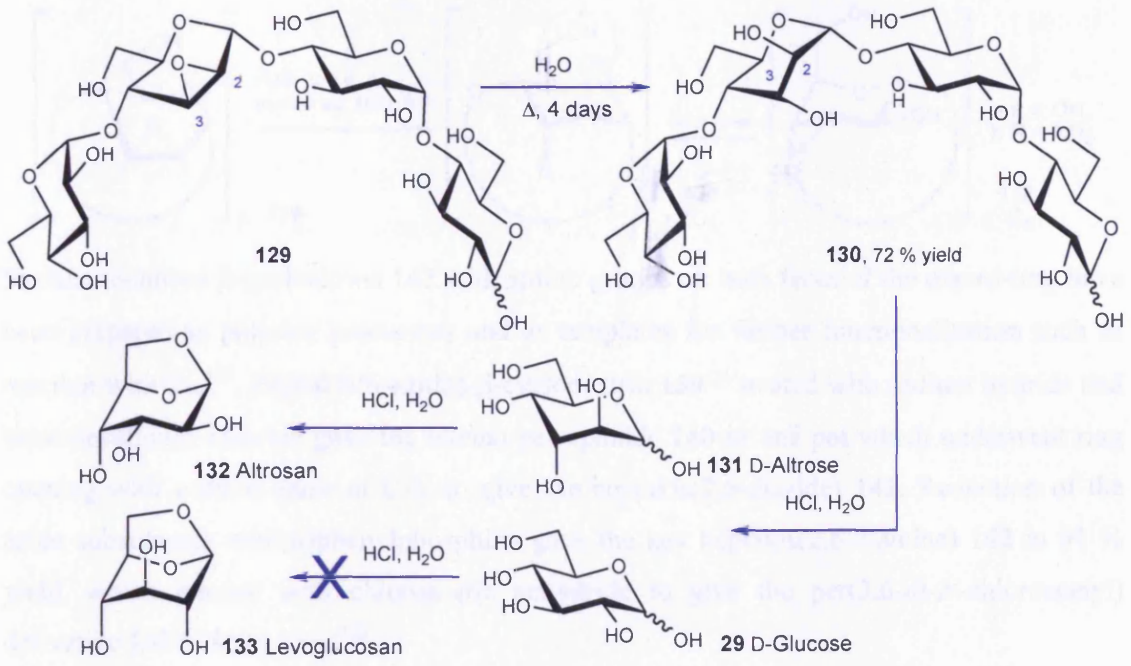
trans-diaxial ring opening and this phenomenon is often referred to as the Furst-Plattner rule. For example, reduction of methyl 2,3-anhydro-4,6-*O*-benzylidene-α-D-allopyranoside **123** with lithium aluminium hydride, gives 2-deoxy-hexopyranoside **124**, whereas 2,3-anhydro-mannopyranoside **125**, gives the 3-deoxy-hexopyranoside **126**[116,4].



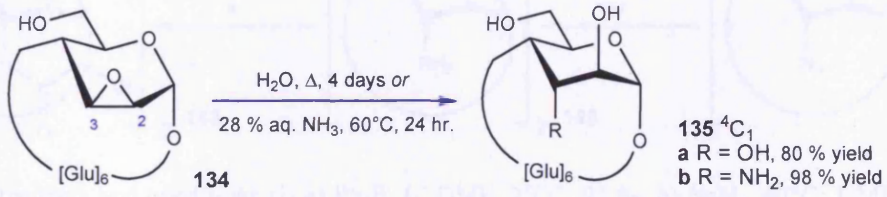
3.4.1. Per-3-amino-3-deoxycycloaltrin



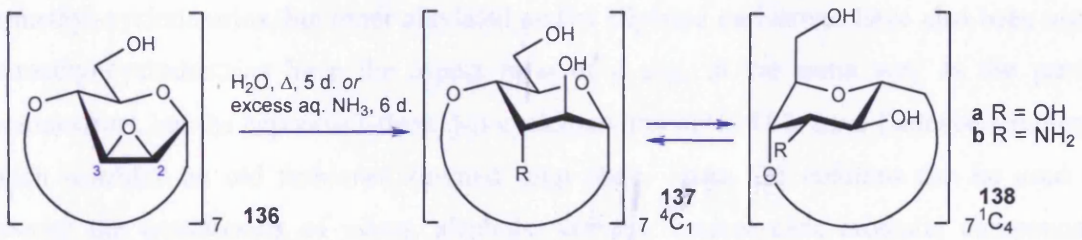
In a similar way to that described above, treatment of the 2,3-anhydro-mannopyranoside **127** with refluxing water for four days, gave altropyranoside **128** by axial attack of the epoxide by water at C-3. In a more spectacular example the tetra-saccharide mono-mannoepoxide **129** was hydrolysed under the same conditions to give Glu-Alt-Glu-Glu **130** in 72 % yield. Evidence for the assigned structure was provided by FAB-MS and acidic hydrolysis which gave a mixture of glucose **29** and altrosan **132**. Authentic altrosan **132** was prepared by the acid treatment of altrose **131** or methyl altropyranoside **128** and the products compared by HPLC. The formation of altrosan **132** without the formation of levoglucosan **133** is probably due to conformational reasons. Formation of levoglosan requires that all five of the substituents (4 x OH, CH_2OH) on the ring of **133** become axial before cyclisation can occur, whereas formation of altrosan **132** only requires three substituents to become axial.



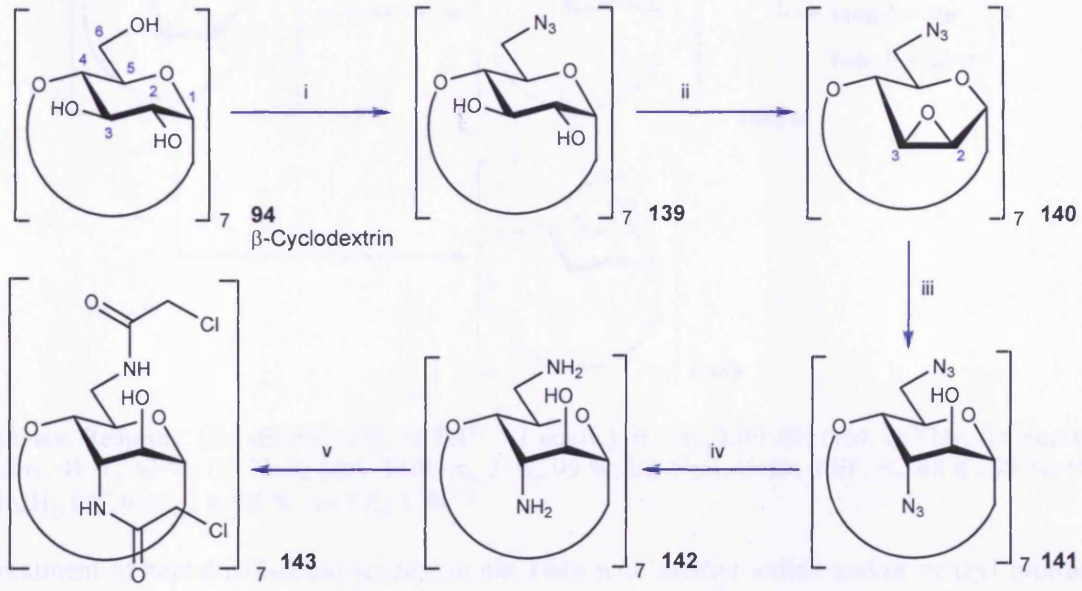
Exactly the same hydrolysis conditions were applied to mono-2,3-anhydro-mannopyranosyl-β-cyclodextrin **134**. The structure of the product **135a** was established by acid hydrolysis as before; a 6:1 mixture of glucose **29**: altrosan **132** was formed as expected[117]. Similarly treatment with aqueous ammonia for only 24 hours at 60°C, again resulted in nucleophilic attack at C-3 to give mono-(3-amino-3-deoxy-altropyranosyl)-β-cyclodextrin **135b**[118]. In the ^{1}H-NMR spectrum $^3J_{2,3}$ was 10.4 Hz for the altroside ring which is consistant with a $^1C_4$ rather than $^4C_1$ a conformation[119], whereas an X-ray crystal structure of the 5.5 hydrate, had the skew boat conformation[120].



The nucleophilic ring-opening reaction of heptakis(2,3-anhydro)-β-cyclomannin **136** with water produced β-cycloaltrin **137a**, **138a** in 72.9% yield. The coupling constant between H-1 and H-2 was 4.5 Hz which indicates either a mixture of $^4C_1$ and $^1C_4$ conformers **137a**, **138a** or a twisted boat[121]. Per-3-amino-3-deoxy-β-cycloaltrin **137b** was prepared by reaction of *manno*-epoxide **136** with ammonia, but the NMR spectra were reported to be complex which was attributed to association between the amino groups[122].



Perfunctionalised β-cycloaltrins **142** with amino groups on both faces of the macro-ring have been prepared as polymer precursors and as templates for further functionalisation such as reaction with C_{60}[123]. Heptakis(6-azido)-β-cyclodextrin **139**[124] treated with sodium hydride and benzenesulfonyl chloride gave the *manno*-per-epoxide **140** in one pot which underwent ring opening with sodium azide at C-3, to give the heptakis(2,6-diazide) **141**. Reduction of the azide substituents with triphenylphosphine gave the key heptakis(2,6-diamine) **142** in 91 % yield, which reacted with chloroacetic anhydride to give the per(3,6-di-*N*-chloroacetyl) derivative **143** in 85 % yield[125].

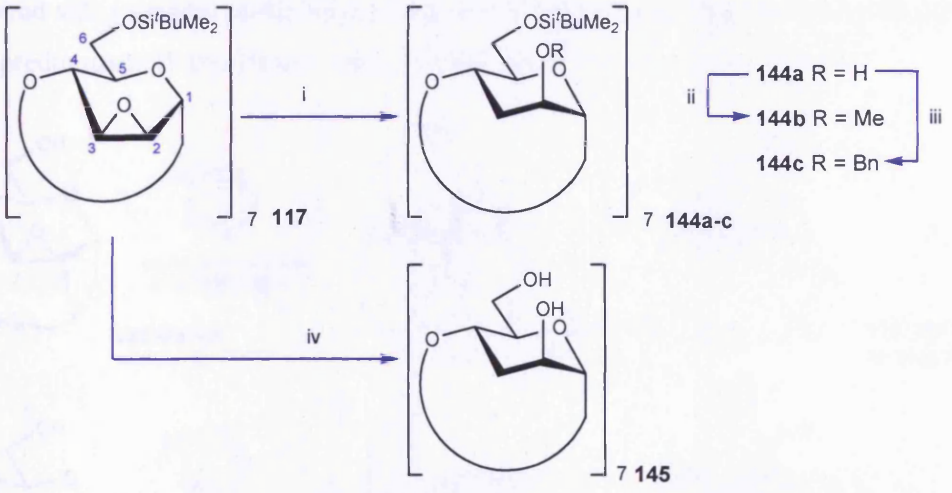


Scheme: Reagents and conditions (i) a) Ph_3P, I_2, DMF, 70°C, 92 %; b) NaN_3, 60°C; DMF, 98 %[124]; (ii) NaH, DMF, rt 5 h., $PhSO_2Cl$, rt, 1h, 84 %; (iii) NaN_3, DMF: H_2O 9:1, 120 °C, 3 d., 55 %; (iv) PPh_3, DMF, rt, 1.5 h, then NH_3 aq., rt, 24 h, 91 %; (v) $(ClCH_2CO)_2O$, MeOH, rt, 24 h, 85 %[125].

3.4.2. Per-3-deoxycyclomannins:

Heptakis(3-deoxy)-β-cyclomannin **144a** was the first cyclodextrin to be prepared which was reduced on the ring and the derivatives **144b**, **114c** are the only cyclodextrins with non-natural stereochemistry to have ever been used as stationary phases for capillary GC. In capillary GC columns, modified cyclodextrins are used as stationary phases; most commonly these are

permethyl-cyclodextrins, but other alkylated and/or silylated derivatives have also been used. Permethyl-cyclodextrins have the aspect ratio of a cup, in the same way as the parent cyclodextrins, but the heptakis(3-deoxy)-β-cyclomannins **144b**, **114c** have flattened structures which resemble an old fashioned rimmed soup plate. These GC columns can be used to separate the enantiomers of esters, alcohols, ketones, heterocycles, aromatic compounds, amino acid derivatives and others. The key step was the reduction of heptakis(2,3-anhydro, 6-*O*-TBDMS)-β-cyclomannin **117**. When excess diisobutyl aluminium hydride in toluene or lithium aluminium hydride were tried, complex mixtures were formed and in the latter case the silyl groups were also partially cleaved. However reduction with Super-Hydride (lithium triethylborohydride solution in THF) gave heptakis(3-deoxy)-β-cyclomannin **144a** in 40 % yield.



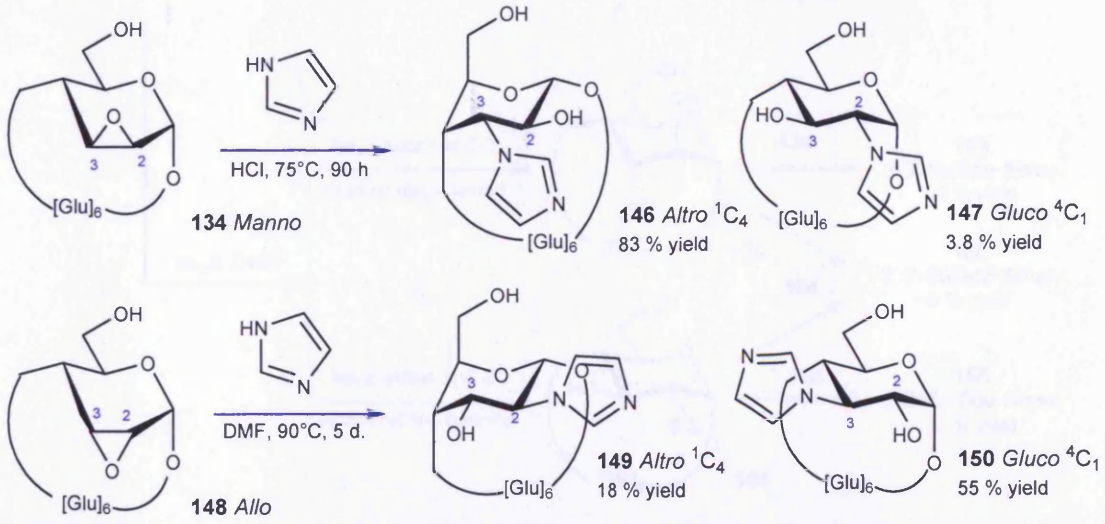
Scheme. Reagents: (i) $LiEt_3BH$ (1M, in THF, 5.1 equiv.), rt., 2h., $LiEt_3BH$ (1M, in THF, 2.6 equiv.), reflux, 48 h., 40 %; (ii) NaH, MeI, THF, rt., 12h., 90 %; iii) NaH, BnBr, THF, rt., 48 h., 59 %; (iv) $LiAlH_4$, 0°C to rt., 2 h, 65 °C for 7 d., 51%[126].

Treatment of heptakis(3-deoxy)cyclomannin **144a** with methyl iodide and/or benzyl bromide in THF using sodium hydride gave heptakis(2-*O*-methyl) **144b** and heptakis(2-*O*-benzyl) **144c** ether in 90 % and 59 % yield respectively. The ^{1}H-NMR data for these compounds gave minimal coupling constant data, because of a narrow chemical shift range and the large number of couplings. However, H-1 appeared as a signal of doublet (2.8 Hz), a broad singlet and a doublet of 2.4 Hz with chemical shifts of 4.62, 4.73, 4.84 ppm for **144a**, **144b**, **144c** respectively. These low values are consistant with equatorial-equatorial coupling and hence the $^4C_1$ conformation. As expected the H-6 protons showed large geminal couplings from 10.7-11.2 Hz but small or zero vicinal couplings to H-5, which indicates H-5 and H-6 have the *gg*-conformation[126]. It was later shown that protracted reduction of heptakis(2,3-anhydro,

6-*O*-TBDMS)-β-cyclomannin **117** with lithium aluminium hydride reduced both the epoxides and the silyl ethers to give heptakis(3-deoxy)-β-cyclomannin **145**[127]; identical methodology was used to prepare octakis(3-deoxy)-γ-cyclomannin[128].

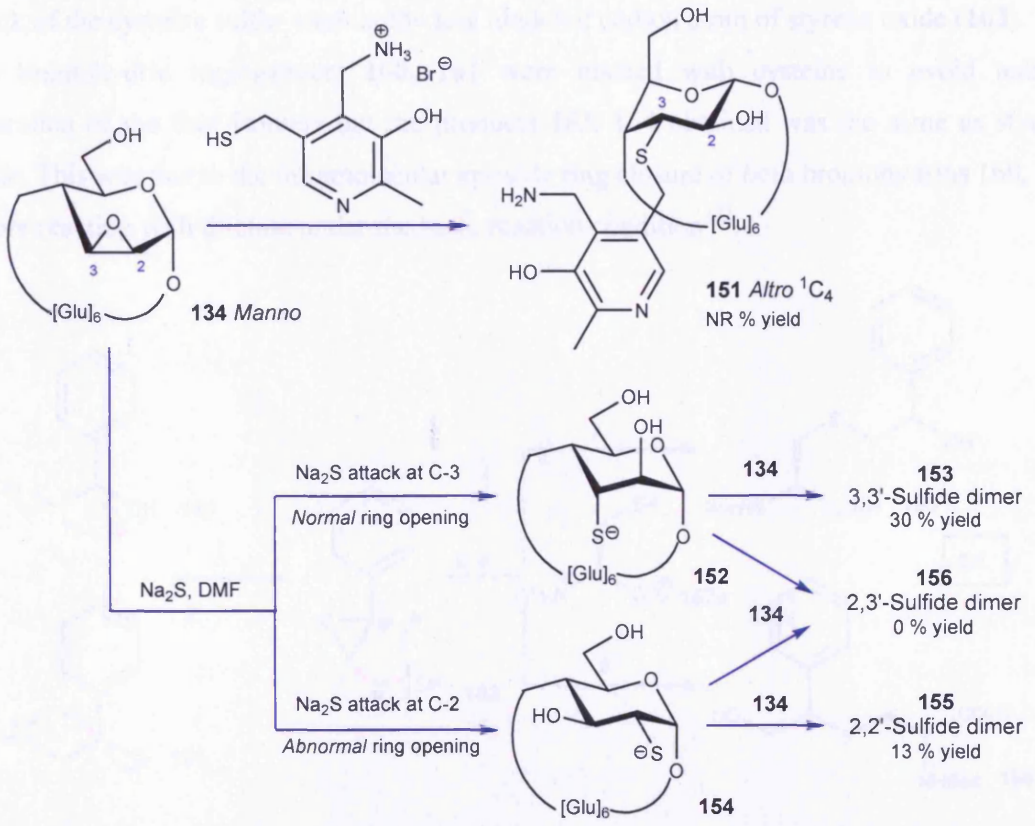
3.4.3. Abnormal ring opening of *manno* epoxide:

The mono(2,3-anhydro)-β-cyclomannins and -β-cycloallins **134**, **148** show interesting differences in reactivity with imidazole. The mono(2,3-anhydro)-β-cyclomannin **134** undergoes attack predominantly at C-3 and the ring flips to give the $^1C_4$ conformer **146** as described previously for other nucleophiles. Mono(2,3-anhydro)-β-cyclomannin **134** undergoes attack at C-2 and C-3 with little selectivity. Both modes of attack are from the unhindered side (outside) of the macro-ring and it might be expected that *trans*-diaxial attack should predominate (Furst-Plattner rule), but this yields the minor product **149**[129].



Ring opening of mono(2,3-anhydro)-β-cyclomannin **134** with sulfur nucleophiles, also gives products from ring opening at C-3 and C-2. The biomimetic pyridoxamine-cyclodextrin **151** was prepared by *normal* mode thiol attack at C-3[130] as expected, however the reaction with sodium sulfide was more complex. Treatment of mono(2,3-anhydro)-β-cyclomannin **134** with sodium sulfide (0.46 equivalents) gave the 3,3′-sulfide **153** (30 % yield) and the 2,2′-disulfide **155** (13 % yield), but none of the 2,3′-sulfide **156** was identified. The low yields do not exclude the possibility that this was formed, but not isolated[131]. The ^{1}H-NMR spectroscopic data revealed that 3,3′-sulfide **153** had the basic pattern of an altroside residue with a predominantly $^1C_4$ conformation, from the normal attack of the intermediate thiolate **152** to C-

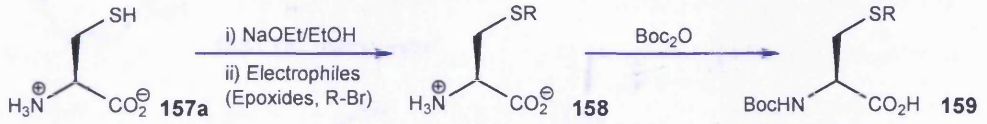
3 of the epoxide **134**. However the abnormal ring opening of epoxide **134** lead to the unexpected formation of 2.2′-disulfide **155** as a minor product, with a $^4C_1$ conformation, from the reaction of the intermediate thiolate **154** with the C-2 side of epoxide **134**. Various nucleophiles such as benzyl thiol, sodium azide and iodine have been reacted with mono(2,3-anhydro)-β-cyclomannin **134**[127] and attack generally follows the *trans*-diaxial rule[130] in which there is predominantly the formation of 3-modified altrose structures as major (*normal*) products and 2-modified glucosidic species as minor (*abnormal*) products.



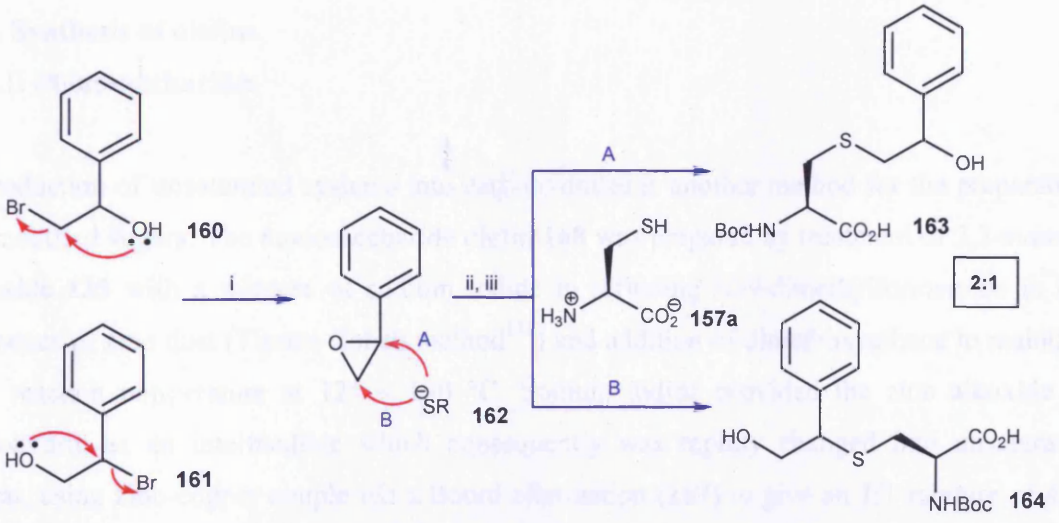
3.5. Reaction of cysteine with electrophiles

In the reactions of species bearing many nucleophilic groups, functional group modification with protecting groups is frequently used to differentiate the groups, but this is not necessarily required. Careful control of pH can give the same benefits without explicit protection-deprotection steps. For example, cysteine *S*-conjugates are important building blocks in the preparation of peptides such as modified glutathiones (GSH). L-Cysteine **157a** in sodium ethoxide solution is alkylated on sulfur by electrophiles such as epoxides and alkyl bromides

(**158**). There were no side reactions due to the protonated amino group, which was subsequently protected by *t*-butyl dicarbonate (Boc_2O) to facilitate isolation (**159**).

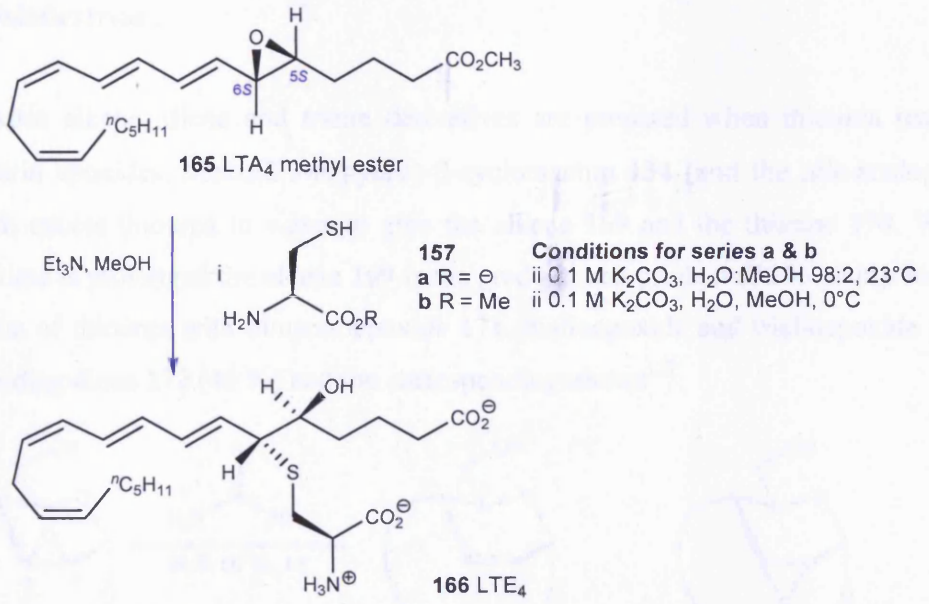


In the case of reaction between styrene oxide **162** and cysteine **157a** a mixture of regioisomers **163** and **164** (75 % yield) were recovered, of which the most abundant product was from the attack of the cysteine sulfur atom at the less hindered carbon atom of styrene oxide (**163**). The two bromohydrin regioisomers **160, 161** were reacted with cysteine to avoid tedious separation of the four isomers but the products **163**, **164** obtained was the same as styrene oxide. This was due to the intramolecular epoxide ring closure of both bromohydrins **160**, **161** before reacting with thiolate under the basic reaction condition[132].



Scheme. Reagents i) NaOEt, EtOH, ii) L-Cysteine **157a**, dry EtOH, NaOEt, rt, 2 h, then acidified with HCl (4 %), iii) NaOH, Boc_2O, water:dioxane 1:2, 0°C - rt., 4 h. (75 – 90 % yield)[132]

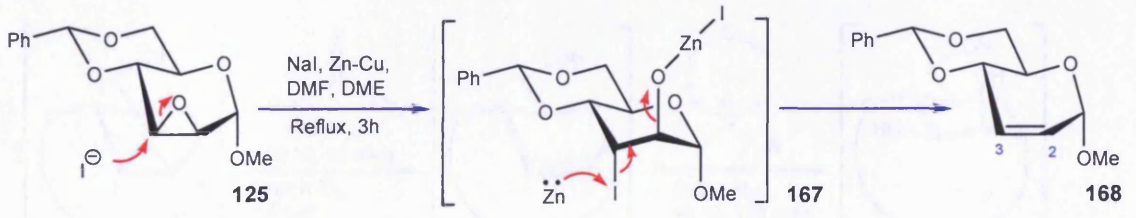
Leukotriene-E_4 (LTE_4) is one of the leukotrienes that are thought to be involved in asthmatic and allergic reactions. It is prepared synthetically and possibly biosynthetically by S_N2 attack of LTA_4 by cysteine thiolate at C-6. This centre is attacked because the protonated/solvated epoxide is able to stabilise carbocation character by resonance with the adjacent alkene bonds. It was originally reported (without experimental details) that cysteine **157a** could be used as the nucleophile with LTA_4 methyl ester **165**[133], but later cysteine methyl ester **157b** was used and gave LTE_4-methyl ester in 75 % yield, which was hydrolysed to LTE_4 **166** in 82 % yield[134].



3.6. Synthesis of olefins

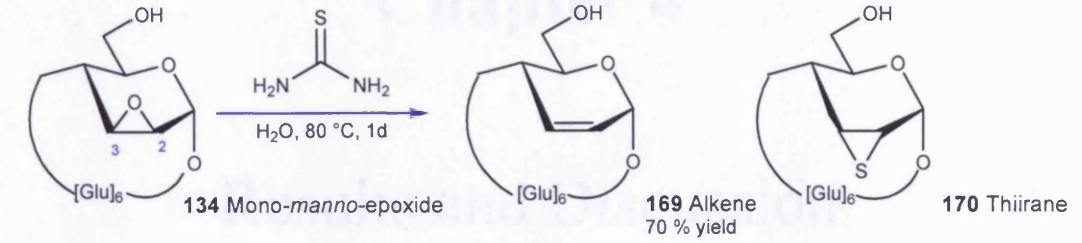
3.6.1. Monosaccharides

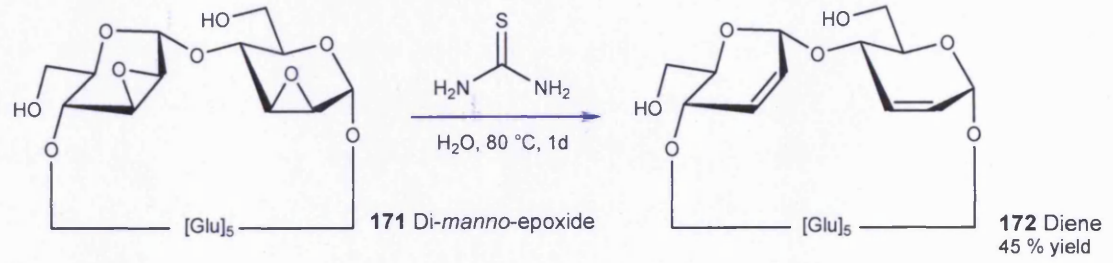
Introduction of unsaturated systems into carbohydrates is another method for the preparation of modified sugars. The monosaccharide olefin **168** was prepared by treatment of 2,3-*manno*-epoxide **125** with a mixture of sodium iodide in refluxing *N*,*N*-dimethylformamide in the presence of zinc dust (Tipson-Cohen method[135]) and addition of dimethoxyethane to maintain the reaction temperature at 125 – 130 °C. Sodium iodide provided the zinc alkoxide of iodohydrin as an intermediate which consequently was rapidly changed into unsaturated sugar, using zinc-copper couple *via* a Boord elimination (**167**) to give an 1:1 mixture of 4,6-*O*-benzylidene-2,3-dideoxy-methyl-α-D-gluco-hex-2-enopyranoside **168** and starting material **125**[136].



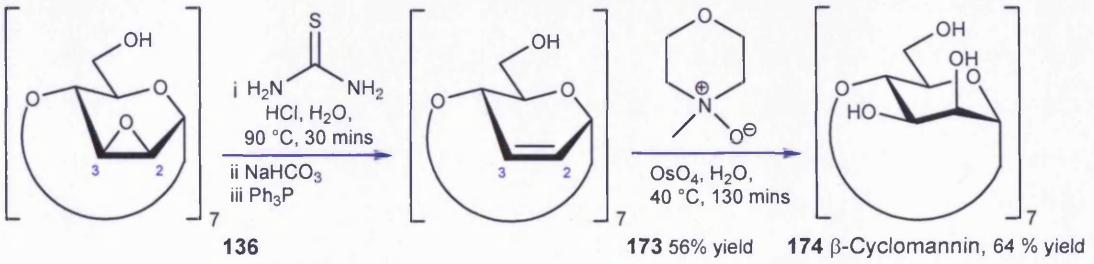
3.6.2. Cyclodextrins

Cyclodextrin alkene, diene and triene derivatives are prepared when thiourea reacts with cyclodextrin epoxides. Mono(2,3-anhydro)-β-cyclomannin **134** (and the *allo*-analogue **148**) react with excess thiourea in water to give the alkene **169** and the thiirane **170**. When the reaction time is prolonged the alkene **169** is the predominant product (70 % yield). In the case of reaction of thiourea with dimannoepoxide **171**, dialloepoxide and trialloepoxide gave the corresponding diene **172** (45 %) and the corresponding trienes[127].





In a dramatic demonstration of the power of this technique; heptakis(2,3-anhydro)-β-cyclomannin **136** was subjected to deoxygenation with thiourea, to give heptakis(2,3-dideoxy-2,3-thia)-β-cycloallin, which was reduced to give the heptakis(alkene) **173**[137]. Dihydroxylation from the outer face by catalytic amounts of osmium tetraoxide regenerated with *N*-methylmorpholine *N*-oxide gave β-cyclomannin **174**[138] in 64 % yield, in a reaction which generated 14 chiral centres in one synthetic operation!



Chapter 4

Results and Discussion

4. Synthesis of chemically modified cyclodextrins

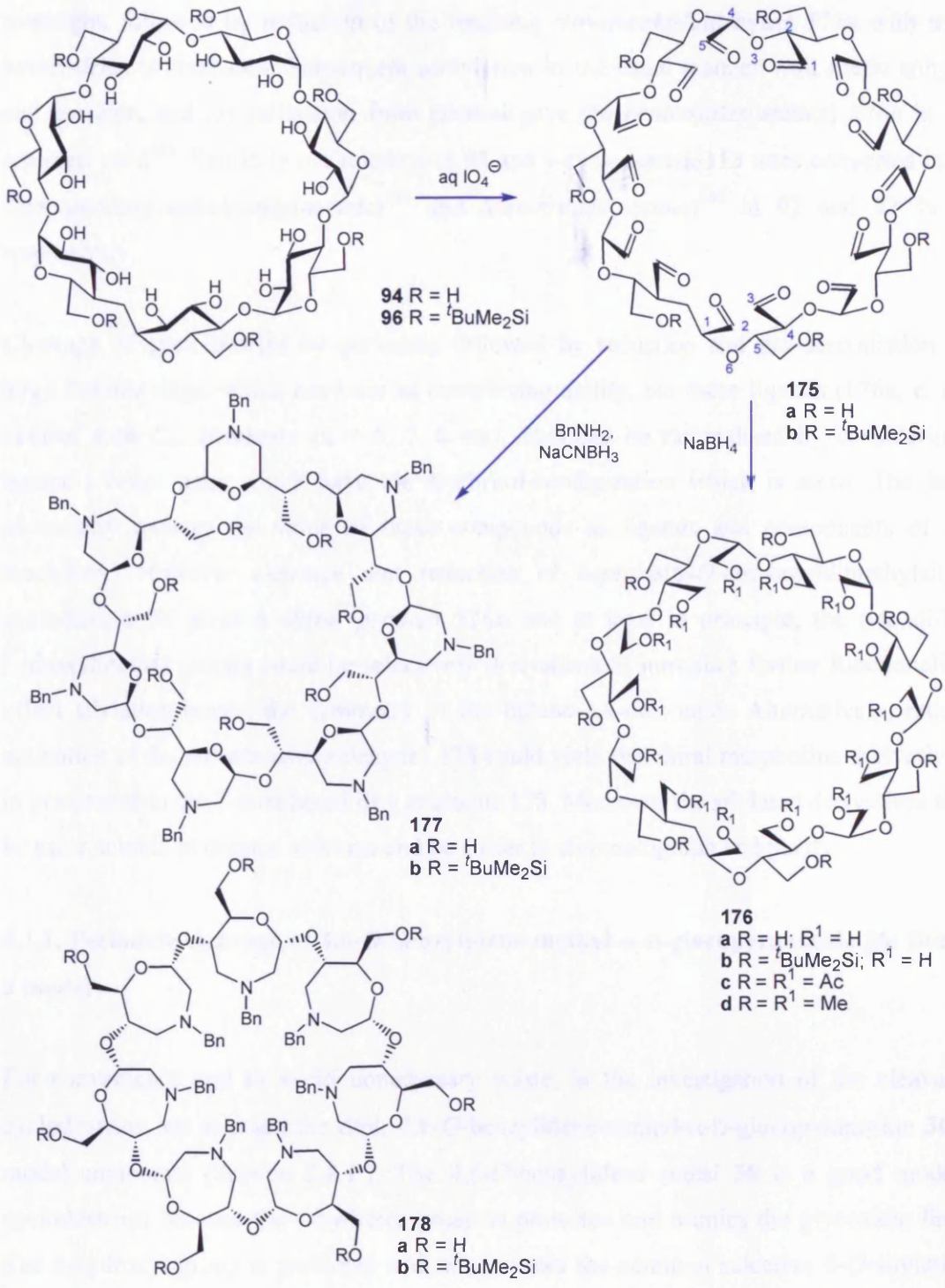
4.1. Introduction: Periodate cleavage of cyclodextrins

The potential of nanotechnology has prompted research efforts to manufacture nanoscale objects with useful physical and chemical properties. However the range of materials that can be prepared at this scale is limited by the ability to control the formation of large rings. Ring formation is essential to achieve strong and selective binding of guests, which underlies virtually all recognition and selectivity phenomenon. Macrolactonisation of natural products[139] and olefin metathesis to produce large rings[140] may extend the range of materials available, but at present most large chemical structures, such as calixarenes are devoid of functionality.

Cyclodextrins are able to form inclusion complexes with a wide range of guest molecules, and there has been much interest in the synthesis of novel cyclodextrins with increased compatibility between the cavity and guest molecules for use as supramolecular *building blocks*[88,141]. Moreover cyclodextrins are natural products, available from renewable carbohydrate sources and are ultimately biodegradable, consequently they are a sustainable source. The disadvantage of cyclodextrins is the large number of hydroxyl groups of similar reactivity and the limited range of forms available.

β-Cyclodextrin **94** can be viewed as two 35-membered rings conjoined at the anomeric oxygen atoms. Cleavage of the vicinal diol moieties in β-cyclodextrin **94** by periodate oxidation (**175**) and then reduction with sodium borohydride creates a single 35-membered ring (**176**) with additional flexibility to bind hydrogen bonding guests.

An aqueous solution of β-cyclodextrin **94** was oxidized with sodium periodate at 26 °C for 2.5 days, excess periodate and iodate were precipitated with barium chloride, the mixture (**175a**) was reduced with borohydride (**176a**) over two days and finally acetylated to give the *henicosakis*(acetate) **176c** in 11 % yield. Repetition of this procedure with α-cyclodextrin **92**, gave the corresponding *octadecakis*(acetate) in 0.6 % yield[142]. Cleavage and reduction of β-cyclodextrin **94** sequence was repeated and used to prepare the *henicosakis*(*O*-methyl)-derivative **176d**[143]. Amazingly, this work has only ever been cited once (in a review[141]) and not by any other workers, including those subsequently doing essentially identical work.



Scheme: Proposed synthesis of cyclodextrin derivatives

Alternatively cleavage of β-cyclodextrin **94** was carried out by using a three molar excess of sodium periodate and keeping the aqueous solution in a dark ice-box for 7 days. To decompose excess sodium periodate, 1,2-ethanediol was added and the reaction stirred at 0 °C

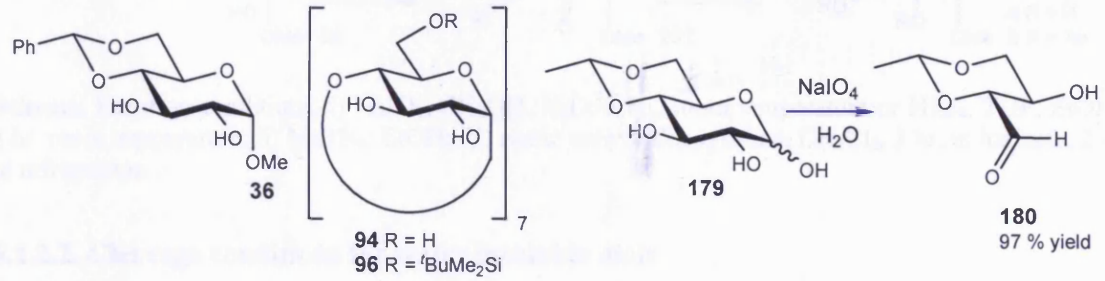
overnight, followed by reduction of the resulting *tetradecakis*(aldehyde) **175a** with sodium borohydride in methanol. Subsequent acetylation in the usual manner with acetic anhydride and pyridine, and crystallisation from ethanol gave the *henicosakis*(acetate) **176c** in 88 % reported yield[144]. Similarly α-cyclodextrin **92** and γ-cyclodextrin **113** were converted into the corresponding *octadecakis*(acetate)[145] and *tetracosakis*(acetate)[144] in 92 and 82 % yield respectively.

Cleavage of cyclodextrins by periodate followed by reduction and per-derivatistion gives large flexible rings, which have useful complexing ability, but these ligands (**176a**, **c**, **d**) are ***achiral with C_{xv} symmetry*** (x = 6, 7, 8 etc). This can be rationalised by considering the butane-1,4-diol units which have the erythritol-configuration which is *meso*. The lack of asymmetry reduces the value of these compounds as ligands and components of nano-machinery. However cleavage and reduction of *heptakis*(6-*O*-*tert*-butyldimethylsilyl)-β-cyclodextrin **96** gives a ***chiral product*** **176b** and at least in principle, the two different hydroxylmethyl groups could be selectively derivatised to introduce further functionality. In effect silylation breaks the symmetry of the butane-1,4-diol units. Alternatively, reductive amination of the *tetradecakis*(aldehyde) **175** could yield the chiral morpholine derivative **177** in preference to the 7-membered ring analogue **178**. Moreover the silylated derivatives should be more soluble in organic solvents and be easier to chromatograph and purify.

4.1.1. Periodate cleavage of 4,6-*O*-benzylidene-methyl-α-D-glucopyranoside 36: Study of a model

For convenience and to avoid unnecessary waste, in the investigation of the cleavage of cyclodextrins, we selected the diol; 4,6-*O*-benzylidene-methyl-α-D-glucopyranoside **36** as a model compound (Section 2.8.1.). The 4,6-*O*-benzylidene acetal **36** is a good model for cyclodextrins, because the 4-hydroxy group is protected and mimics the glycosidic linkage. The 6-hydroxyl group is protected and this imitates the common selective 6-*O*-silylation of cyclodextrins. Moreover the benzylidene group rigidifies the pyranoside ring, which mimics the rigidity of the macrocyclic ring. Cleavage of the benzylidene compound **36** by periodate under aqueous and non-aqueous conditions was investigated as a model for the cleavage of β-cyclodextrin **94** and heptakis(6-*O*-TBDMS)-β-cyclodextrin **96**. The polarity of 4,6-*O*-benzylidene-α-methyl-D-glucopyranoside **36** lies somewhere between these two extremes.

Therefore conditions which can be applicable to water soluble and water insoluble compounds were desired.

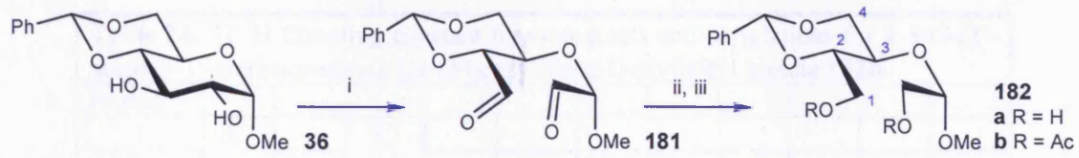


The method initially used for the cleavage of the benzylidene diol **36** with metaperiodate was based on an *Organic Syntheses* procedure intended for use on 4,6-*O*-ethylidene-D-glucose **179**[146] which is appreciably more water soluble than the benzylidene diol **36** used in this work. We additionally intended to do immediate reduction with the borohydride. Consequently the method was modified so that the aqueous metaperiodate used in the cleavage step was replaced by 20 % ethanolic aqueous periodate. Nevertheless, although about 15 % of a product was identified by ^{1}H-NMR, it could not be isolated. Therefore further efforts were taken to optimise the reaction. A suspension of the benzylidene diol **36** in water was added to a cooled suspension of sodium metaperiodate in water, maintained at approximately pH 4 by drop wise addition of 8 M sodium hydroxide for 3 hours. Reduction of the precipitate formed was directly done with sodium borohydride in ethanol, and the reaction was stored in the freezer overnight. ^{1}H-NMR spectrum of the crude product displayed signals consistant with starting material and circa 13 % of a new product, but isolation by column chromatography (silica gel, petrol to ethyl acetate gradient) yielded only starting material **36** (85 % yield).

4.1.2. Methodology development

4.1.2.1. Cleavage condition for water soluble diols

In a slightly different procedure, the diol **36** was dissolved in warm methanol and a solution of aqueous sodium periodate was added to the sugar solution, the corresponding dialdehyde **181** was formed. Reduction with borohydride gave 3-*O*-(2′-hydroxy-1′-methoxy-ethyl)-2,4-*O*-benzylidene-D-erythritol **182a** as an oil in excellent yield (90 %). A portion of the material (194 mg) was acetylated in the usual way by acetic anhydride in the presence of pyridine[72] to give diacetate **182b** (57% yield) which is less polar and crystalline. The erythritol **182a** and diacetate **182b** were successfully characterised by NMR spectroscopy.

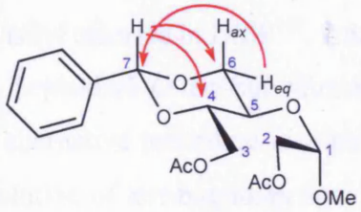


Scheme. Reaction conditions, i) $NaIO_4$, CH_3OH, H_2O; 2 hr., room temperature or HIO_4, THF, EtOH; 2 hr. room temperature; ii) $NaBH_4$; EtOH; iii) acetic anhydride, pyridine, CH_2Cl_2, 3 hr. in ice bath, 2 d. in refrigerator.

4.1.2.2. Cleavage conditions for water insoluble diols

Cleavage conditions for water insoluble diols was developed when benzylidene compound **36** was dissolved in THF and ethanol and then a solution of periodic acid in THF was added to the sugar solution and stirred for 2 hr. at r.t. Reduction by sodium borohydride monitored by TLC yielded erythritol **182a** in 80 % yield, which was characterised by NMR spectroscopy.

4.1.2.3. Spectroscopic characterisation of pyranoside 2,3-cleavage products 182



Key $^3J_{H\text{-}C}$ Correlations

Note: Assignments follow sugar numbering rather than numbering in the name

Assignment of the H-1, H-2a, H-2b spin system was comparatively easy, because of the unambiguous assignment of H-1 from the ^1J ^{1}H-^{13}C COSY NMR experiment (δ_H 4.72, δ_C 100.0). The assignment of H-2a and H-2b then follows from correlations in a ^{1}H-^{1}H COSY experiment. Assignment of the signals for the C-3 to C-6 segment of the molecule was very difficult, because H-4 overlapped with H-5 (Table 24). The side of the signal attributed to H-4, showed a possible doublet of doublets (9.5, 4.7 Hz), whereas H-5 was a complex multiplet. Moreover, the functional array of C-3 to C-6 is approximately symmetrical (erythritol like). The ends of the spin system were assigned by means of a ^3J ^{1}H-^{13}C COSY NMR experiment. This showed $^3J_{H\text{-}7,C\text{-}6}$, $^3J_{H\text{-}7,C\text{-}4}$, $^3J_{H\text{-}6a,C\text{-}7}$, and hence enabled C-6 to be distinguished from C-3. H-6a and H-6b are most likely the equatorial (δ 4.43) and axial (δ 3.67) respectively. HRMS (ES^+) provided the parent ($M+H^+$) ion at *m/z* 285.1337, and ($M+NH_4^+$) ion at *m/z* 386.1813 for the diol **182a** and the diacetate **182b** respectively.

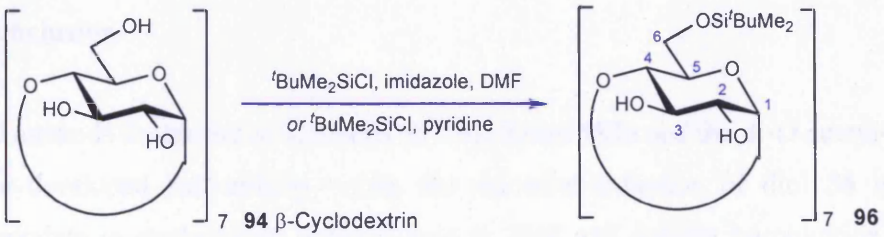
Table 24. ^{1}H-^{1}H Coupling constant measurements and correlations for 1-3-*O*-(2′-acetoxy-1′-methoxy-ethyl)-2,4-*O*-benzylidene-D-erythrityl acetate **182b**.

Proton										
1	5.7		5.2							
2a			5.0		11.7					
2b	5.9				11.8					
3a			12.2						0.8	
3b			12.1		4.7					
4	9.5				4.7				?	
5	?						?			?
6a (*eq*)			10.9				4.3			
6b (*ax*)			10.1							10.1

4.1.3. Selective modification of the primary hydroxyl face of β-CD 94

4.1.3.1. Preparation of 6-*O*-t-butyldimethylsilyl derivatives of cyclodextrins

Due to the large number of hydroxyl groups (21 in β-cyclodextrin **94**) selective modification of these groups can be difficult. Therefore protection of the primary hydroxyl groups were initially carried out by treatment of β-cyclodextrin **94** (3.0 g) with imidazole and *tert*-butyldimethylsilyl chloride in DMF[103]. After 15 days stirring at room temperature an excellent yield of the heptakis(6-*O*-*tert*-butyldimethylsilyl)-β-cyclodextrin **96** was achieved (4.6 g, 91%). In an alternative procedure a solution of β-cyclodextrin **94** (1 g) in dry pyridine was added to a solution of *tert*-butyldimethylsilyl chloride in dry pyridine[147] at 0°C, the reaction required 20 hours stirring at room temperature. The yield was lower (1.18 g, 70%), but the time scale was more practical. Therefore a large scale preparation of β-cyclodextrin (10 g) was undertaken using the dry pyridine method and a good yield achieved (11.83 g, 70%).

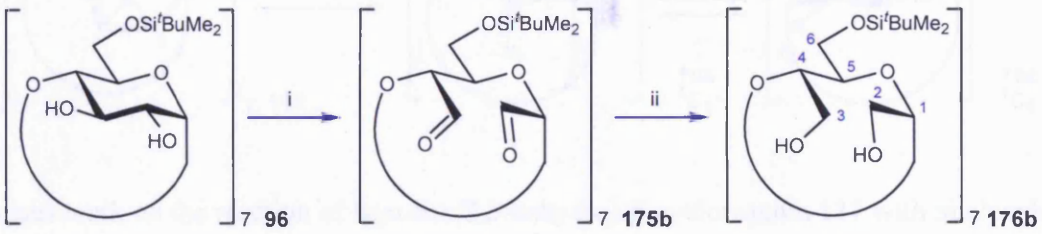


Scheme. Synthesis of heptakis(6-*O*-*tert*-butyldimethylsilyl)-β-cyclodextrin **96**.

4.1.3.2. . Periodate Cleavage of the glucose rings of β-CD

In view of the results from the preliminary study, the isolation of D-erythritol **182a** in high yield looked promising for development of new oxidation-reduction reaction to cleave the

glucose ring of 6-*O*-*tert*-butyldimethylsilyl cyclodextrins. Hence in order to examine if oxidation-reduction of silylated β-cyclodextrin **96** was practical for the synthesis of *tetradecakis*(alcohol) **176b**, a mixture of β-CD derivative **96** and sodium periodate was stirred in methanol at room temperature for 6 h. Reduction with sodium borohydride resulted in recovery of only unreacted starting material **96** in 87% yield.



Scheme: i) HIO_4, THF, EtOH or $NaIO_4$, CH_3OH, H_2O; ii) $NaBH_4$, EtOH[142].

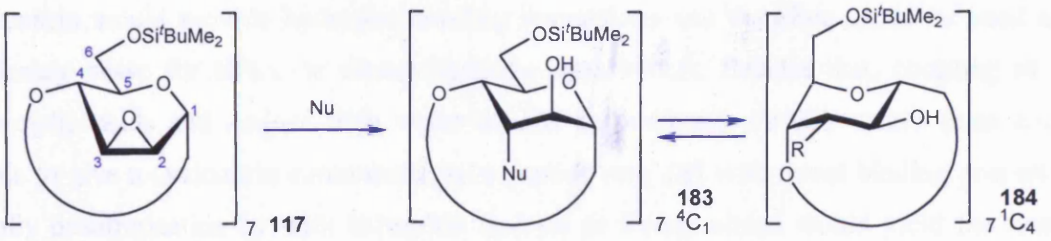
On the other hand, when the silylated cyclodextrin **96** was treated as a water insoluble compound and reacted with periodic acid, stirred at 0 °C in THF for 6 hours, the ^{1}H-NMR spectrum showed a complicated mixture which was confirmed by the presence of a large number of spots in the TLC. The periodic acid procedure was repeated again under the same reaction conditions and the progress of reaction was monitored by TLC. When TLC confirmed complete consumption of starting material after 2 hours, 1,2-ethanediol was added to the reaction mixture to decompose excess periodic acid, followed by reduction with sodium borohydride in ethanol. ^{1}H-NMR spectra indicated an ambiguous mixture which had *t*-butyl- and dimethylsilyl groups at the right chemical shifts (δ_H 0.82, 0.01 respectively) plus some overlapping complex signals in the sugar region which could not be identified.

4.1.3.3. Conclusion

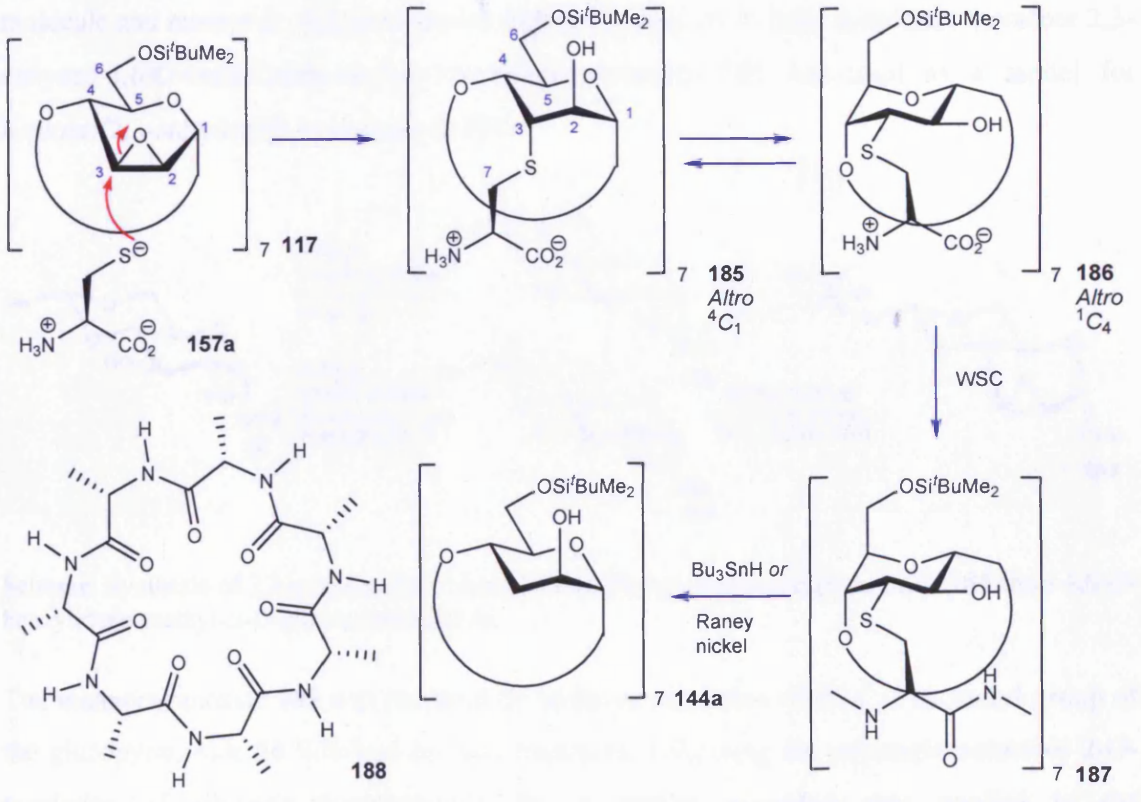
Two novel methods for the facile synthesis of D-erythritol **182a** and the di-*O*-acetyl-derivative **182b** were developed successfully, using the oxidation-reduction of diol **36** with either sodium periodate in methanol or periodic acid in THF and sodium borohydride. However oxidation-reduction of the silylated β-CD **96** under the same reaction conditions failed to give the expected *tetradecakis*(alcohol) **176b**, presumably due to the complexity of the cyclodextrins.

4.2. The synthesis of cycloaltrin-3-sulfides

4.2.1. Introduction



Previous work on the reaction of *heptakis*(2,3-anhydro)-β-cyclomannin **117** with nucleophiles indicates that attack occurs preferentially at C-3 (*trans*-diaxial ring opening[130]) to give products with *altro*-stereochemistry in the $^4C_1$ conformation (e.g. Nu = H) or more commonly the $^1C_4$ conformation. Consequently this modifies the stereochemistry of the C-2-and C-3 hydroxyl groups of cyclodextrins, and can result in extension of the length of the chiral cavity on the secondary face.



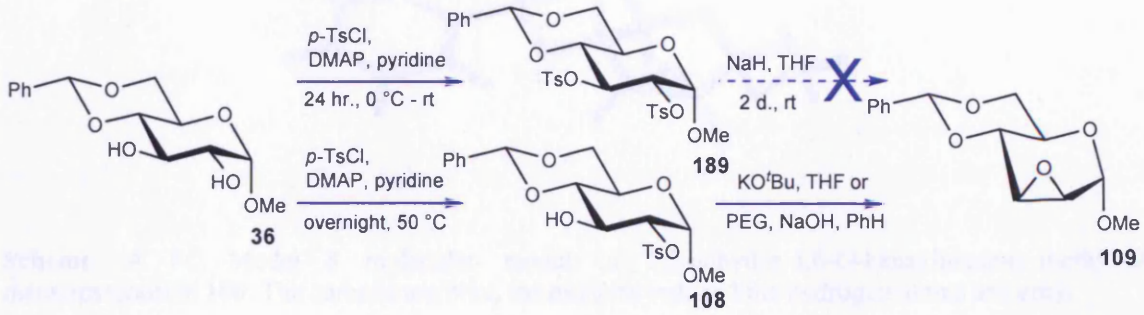
Scheme. Proposed synthesis of *cyclo*-hepta-L-alanine **188**

It was proposed that if L-cysteine **157a** acted as a nucleophile with *heptakis*(2,3-anhydro)-β-cyclomannin **117**, the epoxide ring opening process would create a cycloaltrin adorned with amino acid groups on the top face **185**, **186** (capped cyclodextrins). This amino acid cycloaltrin would provide hydrogen bonding interactions and therefore could be used as a stationary phase for HPLC to discriminate the enantiomers. Besides that, coupling of the carboxylic acids and amines with water soluble carbodiimide (WSC) would form amide bonds to give a cycloaltrin surmounted by a peptide ring **187** with novel binding properties. Finally desulfurisation by with tributyltin hydride or Raney nickel would yield the known heptakis(3-deoxy)-β-cyclomannin **144a** and also *cyclo*-heptakis-L-alanine **188**. As far as we are aware, this would be the first covalent template synthesis of a cyclic peptide.

4.2.2. Synthesis of tosyl and *manno*-derivatives

4.2.2.1. Model studies: The preparation of 2,3-anhydro-4,6-*O*-benzylidene-methyl-α-D-mannopyranoside 109

Perfunctionalisation of β-cyclodextrins requires seven sequential reactions on the same molecule and moreover cyclodextrins are difficult to prepare in large quantities, therefore 2,3-anhydro-4,6-*O*-benzylidene-methyl-α-D-mannopyranoside **109** was used as a model for *heptakis*(2,3-anhydro)-β-cyclomannin **117**.

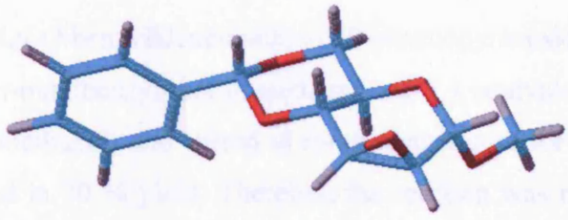


Scheme. Synthesis of 2,3-anhydro-4,6-*O*-benzylidene-methyl-α-D-mannopyranoside **109** from 4,6-*O*-benzylidene-methyl-α-D-glucopyranoside **36**

The mannopyranoside **109** was prepared by selective tosylation of the C-2 hydroxyl group of the glucopyranoside **36** followed by base treatment. Following the successful selective 2-*O*-tosylation of silylated β-cyclodextrin **96**, a similar procedure was applied to the glucopyranoside **36**, which was stirred with a 3 fold molar excess of *para*-toluenesulphonyl chloride in pyridine catalysed by DMAP for 24 hr. at 0 °C to room temperature. The product

was a mixture of mono- **108** and ditosyl- benzylidene glucosides **189** with R_f 0.3 and 0.5 respectively. Epoxidation of that mixture was tried with sodium hydride in dry THF for 2 days at room temperature, but after that time ^{1}H NMR spectrum showed no conversion. However when these processes were conducted with 1 equivalent of *para*-toluenesulphonyl chloride, a crude reaction mixture of diol **36** and monotosylate **108** were obtained after 5 h. stirring at 0 °C to room temperature and heating at 50 °C overnight.

With the anticipation that the epoxide **109** would be easier to separate from the diol **36** than the monotosyl derivative **108**, the reaction mixture was treated with potassium *tert*-butoxide in dry THF at 0 °C to room temperature for 4 days. About 5 – 10 mole % potassium *tert*-butoxide was added during that time. The epoxide **109** was easily separated from crude product by column chromatography using a solvent gradient of chloroform to ethyl acetate, but in very low yield (4 %). In order to increase the yield of epoxide **109**, the mixture was treated with polyethylene glycol in benzene and stirred under reflux with saturated sodium hydroxide[148] for 18 hours. Column chromatography, using chloroform as eluent gave 2,3-anhydro-4,6-*O*-benzylidene-methyl-α-D-mannopyranoside **109** in 75 %, which was fully characterized by ^{1}H-, DEPT-, ^1J ^{1}H-^{13}C- and ^3J ^{1}H-^{13}C- COSY NMR experiments and mass spectrometry.



Scheme. A PC Model 8 molecular model of 2,3-anhydro-4,6-*O*-benzylidene-α-methyl-D-mannopyranoside **109**. The carbons are blue, the oxygens red, and the hydrogen atoms are grey.

In the NMR spectrum C-7/H-7 and C-1/H-1 were assigned based on unambiguous shifts from a ^1J ^{1}H-^{13}C COSY NMR experiment (δ_H 5.39, δ_C 102.5), (δ_H 4.81, δ_C 96.9) respectively and similarly C-6 is the only methylene and was identified from the ^{13}C DEPT experiment. The ^1J ^{1}H-^{13}C COSY NMR then gave H-6a (δ_H 4.23, dd *J* 10.3, 4.6 Hz) and H-6b (δ_H 3.60, app. t *J* 10.3 Hz). These are assigned as equatorial and axial respectively based on the lesser chemical shift of axial protons relative to equatorial protons and the large value of $^3J_{5,6}$ for the latter which can only be due to a $^3J_{ax,ax}$ coupling. The ^3J ^{1}H-^{13}C COSY NMR experiment showed a

strong correlation from H-6a to C-4. $^{3}J\,^{1}H\text{-}^{13}C$ coupling constants follow a Karplus-like curve relationship; from molecular models the dihedral angle H-6_{eq}, C-6, C-5, C-4 is 174° whereas the corresponding angle for H-6_{ax} is 63° and hence these should give strong and weak correlations respectively. The two highest field protons signals were assigned to H-2 and H-3 (epoxides) based on shifts and coupling constants (δ_H 3.05, 3.44, $^{3}J_{2,3}$ 3.6 Hz). In a $^{3}J\,^{1}H\text{-}^{13}C$ COSY NMR experiment, H-1 showed a stronger correlation to the higher shift signal than the lower one, hence this was assigned to H-3 (δ_H 3.44, δ_C 53.87) rather than H-2 (δ_H 3.05, δ_C 50.58). The only other possible $^{3}J_{C,H}$ correlation of H-1 is to C-5.

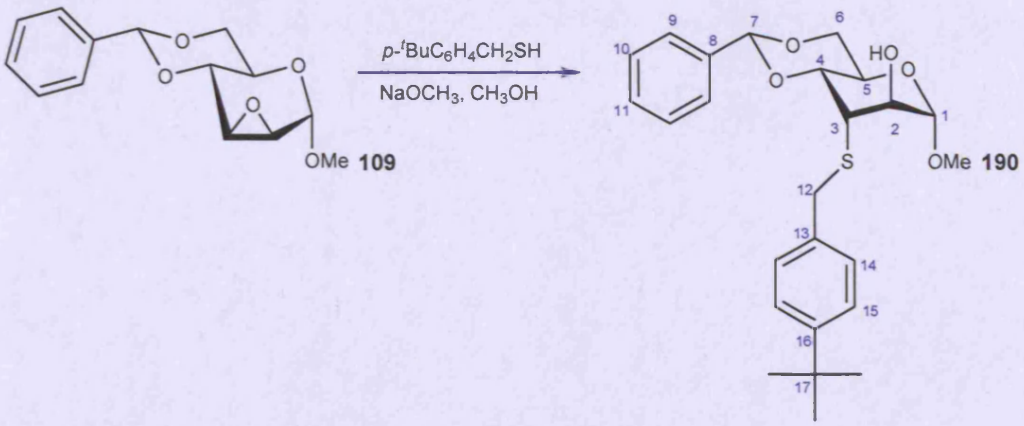
4.2.3. Nucleophilic ring opening of epoxides

The interactions between electrophiles such as epoxides with nucleophiles containing sulfur atoms was intended to include, 4-*tert*-butylbenzylthiol, thiourea and L-cysteine to synthesise *altro*-sugars from the *gluco* starting material.

4.2.3.1. 4-*tert*-Butylbenzylthiol

4.2.3.1.1. Model studies of 2,3-anhydro-4,6-*O*-benzylidene-methyl-α-D-mannopyranoside 109 with 4-*tert*-butylbenzylthiol

Initially 2,3-anhydro-4,6-*O*-benzylidene-methyl-α-D-mannopyranoside **109** was treated with 1.1 equivalent of 4-*tert*-butylbenzylthiol in methanol and 1.1 equivalent of sodium methoxide (25-30 % solution in methanol) and stirred at room temperature for 29 hr, but only starting material was recovered in 70 % yield. Therefore the reaction was repeated again using 1.2 equivalent of 4-*tert*-butylbenzylthiol and 1.1 equivalent of sodium methoxide (powder) in methanol and stirred at room temperature for 1 day and then refluxed for 3 days. The reaction mixture was monitored by TLC, and more reagent (2 equiv.) was added to promote the reaction. A base-acid workup followed by purification, provided the expected product (3-deoxy-3-(4′-*tert*-butylbenzylthiol)-4,6-*O*-benzylidene-methyl-α-D-altropyranoside) **190** in 30 % yield but unreacted starting material **109** was recovered in 46 % yield.



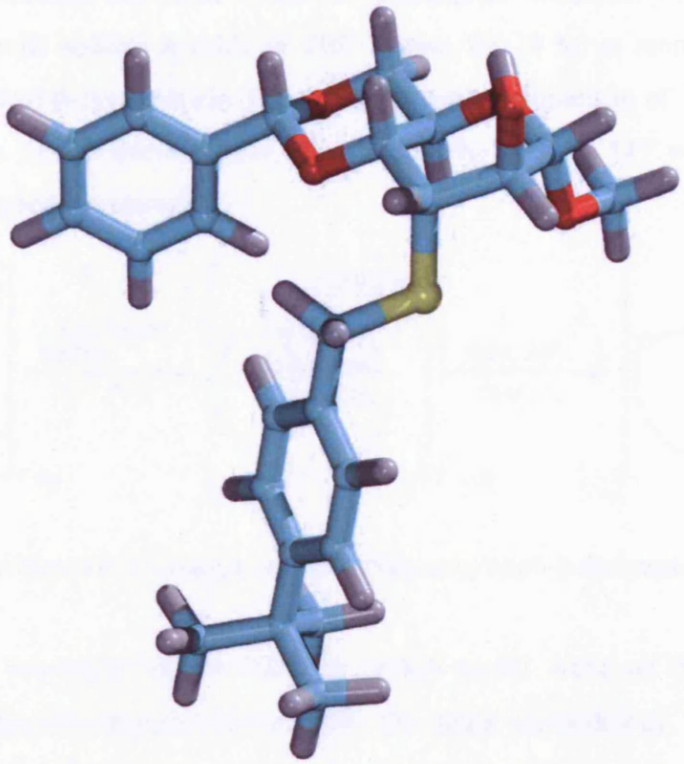
Scheme. Preparation of 3-deoxy-3-(4′-*tert*-butylbenzylthiol)-4,6-*O*-benzylidene-methyl-α-D-altro-pyranoside **190**.

NMR analysis of 3-deoxy-3-(4′-*tert*-butylbenzylthiol)-4,6-*O*-benzylidene-methyl-α-D-altropyranoside 190

The altropyranoside **190** was identified by ^{1}H, ^{13}C-DEPT, HSQC and HMBC NMR techniques rooted on the unambiguous shifts for C-1, C-7 (δ_C 100.9, 102.2 respectively) in $^{1}J_{C,H}$ correlations with H-1, and H-7 (δ_H 4.39, 5.54 respectively). The two methylene groups found in a ^{13}C-DEPT experiment were distinguished by $^{3}J_{C,H}$ correlation and the protons assigned from ^{1}J ^{1}H-^{13}C correlations. The only proton which can display a multiplet more complex than a doublet of doublets in the ^{1}H NMR spectrum is H-5 due to coupling with 2H-6 and H-4, hence C-5 was identified by ^{1}J ^{1}H-^{13}C correlation. H-1 also showed a strong correlation with C-3 which was again confirmed by the dihedral angle values calculated by molecular model (179.5). The only remaining protons and carbons are in positions 2 and 4. The ^{1}H NMR spectrum showed a doublet of doublet signal with coupling constants of 9.26, 4.01 which was as expected for H-4, but which could not be H-2, which is only involved in ^{3}J-H_{eq},H_{eq} couplings. Hence all the protons were assigned from the ^{1}J ^{1}H-^{13}C COSY NMR experiment. The position of the sulfide at C-3 and hydroxyl group at C-2 were confirmed by appearance of the H-2 signal (δ circa 3.95) at lower field than H-3 (δ 3.40) due to the more electrophilic OH group on C-2 than sulfur atom on C-3. The coupling constants for H-3 (dd, *J* 3.3, 2.4 Hz) and the absence of an appreciable ^{3}J H-1, H-2 coupling is consistant with the $^{4}C_1$ conformation which is enforced by the *trans*-ring fusion, despite the presence of three axial substituents. However it is not possible to exclude a partial skewing of the pyran oxygen and carbons -1, -2 and -3. High resolution mass spectrometry (ES^+) provided a protonated ($M+H^+$) ion at *m/z* 445.2038.

Table 25. Dihedral angles (C-C-C-H) used for estimating $^3J_{H,C}$ of altropyranoside **190** calculated using PC-Model.

θ°, *J*	C-1	C-2	C-3	C-4	C-5	C-6	C-7	C-12	C-OMe
H-1			179.5		177.4				1.7
H-2				119.4					
H-3	179.6				179.5			37.8	
H-4		66.7				22.8	16.7		
H-5	65.6		65.1						
H-6$_{ax}$				111.3			1.6		
H-6$_{eq}$				126.1			168.6		
H-7				33.9		1.7			
H-12a			19.3						
H-12b			21.4						



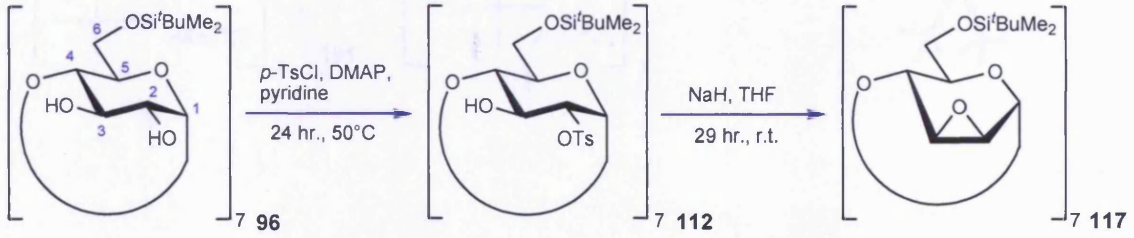
Scheme. A molecular model of 3-deoxy-3-(4′-*tert*-butylbenzylthiol)-4,6-*O*-benzylidene-methyl-α-D-altropyranoside **190** produced using PC Model 8 and rendered using Pov-Ray. Bond widths are set to the radius of hydrogen.

Table 26. Comparison of observed and calculated (PC-Model) $^3J_{H,H}$ NMR coupling constants for altropyranoside **190**.

Coupling Constant (Hz)	1,2	2,3	3,4	4,5	5,6$_{eq}$	5,6$_{ax}$
Calculated	2.4	2.3	5.3	9.4	6.1	10.6
Observed	0	2.4	4.0	9.3	5.2	10.3
Difference	2.4	0.1	1.7	0.1	0.9	0.3

4.2.3.1.2. Heptakis(2,3-anhydro-6-*O*-*t*-butyldimethylsilyl)-β-cyclomannin 117 and 4-*tert*-butylbenzylthiol

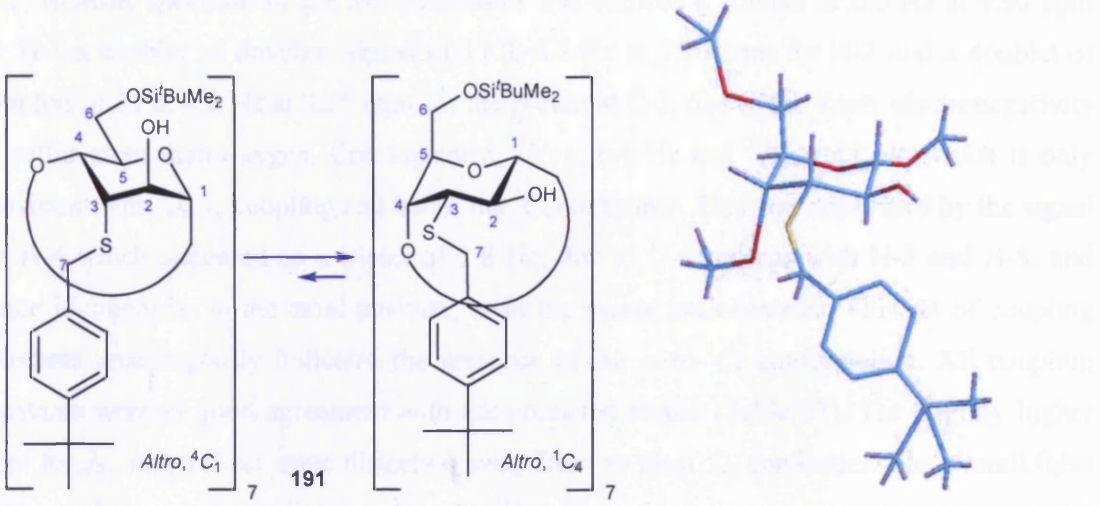
Heptakis(6-*O*-*t*-butyldimethylsilyl)-β-cyclodextrin **96** was tosylated selectively at the 2-*O* positions according to the procedure of Coleman[104] using *para*-toluenesulfonyl chloride (3 equivalents per silylated glucopyranose unit) in pyridine in the presence of 4-dimethylaminopyridine (DMAP) as a catalyst for 24 hr at 50 °C. Heptakis(2-*O*-tosyl-6-*O*-tertbutyldimethylsilyl)-β-cyclodextrin **112** was purified by column chromatography using a solvent gradient of chloroform to ethanol but in low yield (4.7% yield). Further material was recovered from the column and recolumned using a less polar eluent; chloroform and ethyl acetate, which increased the yield to 62 %. Subsequent treatment of the tosylated β-cyclodextrin **112** with sodium hydride in THF stirring for 29 hr. at room temprature gave heptakis(2,3-anhydro)-β-cyclomannin **117** in 82 % yield. Comparison of ^{1}H- and ^{13}C-NMR spectroscopic data of the intermediates **96**, **112** and the product **117** with literature data confirmed the assigned structures[105].



Scheme: Synthesis of heptakis(2,3-anhydro-6-*O*-*t*-butyldimethylsilyl)-β-cyclomannin **117**

Having achieved successful results from the initial model work on 2,3-anhydro-4,6-*O*-benzylidene-α-methyl-D-mannopyranoside **109**, the same methodology was applied to a cyclodextrin epoxide. Heptakis(2,3-anhydro)-β-cyclomannin **117** was refluxed for 7 days with a 35 fold molar excess of 4-*tert*-butylbenzylthiol (5 equiv. per glucose ring), and sodium methoxide (31.5 equiv.) in methanol. The reaction mixture was monitored by TLC which indicated a complex mixture and this was confirmed by the ^{1}H-NMR spectrum of the crude reaction mixture. Purification of those materials resulted in recovery of epoxide **117** and 4-*tert*-butylbenzylthiol in a ratio of 1.0:1.5 as major components and a minor compound which could not be identified.

As a consequence of these results, the reaction was run with a lower concentration of epoxide **117** but a higher concentration of reagent and base. Therefore double equivalents of the reagent, as well as base were used in methanol and the sulfide **191** was successfully formed in good yield after only 2 days reflux. Purification of crude reaction mixture by column chromatography using a slow gradient of chloroform to ethyl acetate gave heptakis(3-deoxy-3-(4′-*tert*-butylbenzylthiol)-6-*O*-*tert*-butyldimethylsilyl)-β-cycloaltrin **191** in 75% yield.



Scheme. The equilibrium between the $^4C_1$ and $^1C_4$ conformers of heptakis(3-deoxy-3-(4′-*tert*-butylbenzylthiol)-6-*O*-*tert*-butyldimethylsilyl)-β-cycloaltrin **191** and a molecular model of a 4-*O*-methyl-methyl-α-D-altropyranoside analogue of the $^1C_4$ conformer created using PC Model 8 and Pov-Ray.

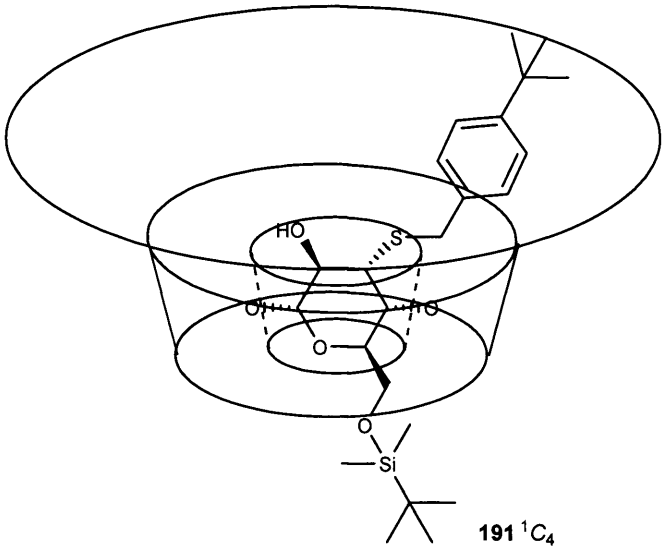
NMR analysis of heptakis(3-deoxy-3-(4′-*tert*-butylbenzylthiol)-6-*O*-*tert*-butyldimethylsilyl)-β-cycloaltrin 191

Altro-cyclodextrins undergo equilibrium interconversion between $^4C_1$ and $^1C_4$ conformers, Therefore an intermediate coupling constant of approximately 4-5 Hz between H-1 and H-2 was expected. Monosaccharide analogues of both conformers were modeled using PC-Model and the coupling constants measured using the modified Karplus routine in the program (Table 27).

Table 27. Comparison of the observed $^3J_{H,H}$ NMR coupling constants of heptakis(3-deoxy-3-(4'-*tert*-butylbenzylthiol)-6-*O*-*tert*-butyldimethylylsilyl)-β-cycloaltrin **191** with those for the $^4C_1$ and $^1C_4$ conformers of a monosaccharide model calculated using PC-Model.

$^3J_{x,y}$	Calculated		Found	Difference	
	$^4C_1$	$^1C_4$		$^4C_1$	$^1C_4$
$J_{1,2}$	2.6	7.4	6.7	+4.1	-0.7
$J_{2,3}$	2.5	11.5	11.0	+8.5	-0.5
$J_{3,4}$	5.6	1.7	3.8	-1.8	+2.1

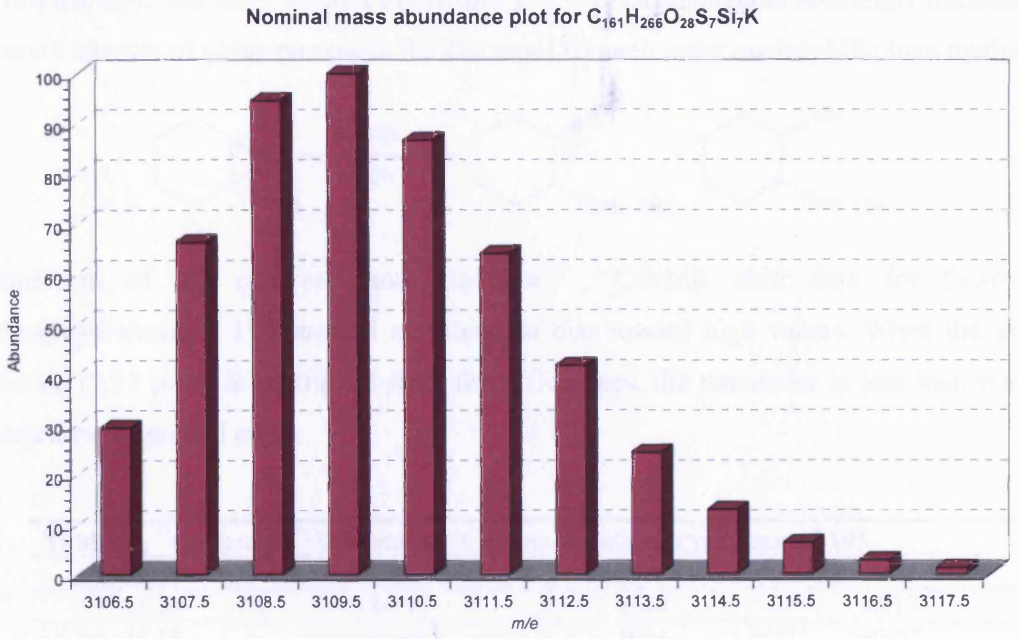
The ^{1}H-NMR spectrum of the altropyranoside **191** showed a doublet of 6.6 Hz at 4.90 ppm for H-1 a doublet of doublets signal of 11.0, 6.7 Hz at 3.70 ppm for H-2 and a doublet of doublets of 11.0, 4.2 Hz at 3.25 ppm for the proton at C-3, due to the lower electronegativity of sulfur atom than oxygen. Consequently, $^3J_{1,2}$ = 6.6 Hz and $^3J_{2,3}$ = 11 Hz which is only consistent with $^3J_{ax,ax}$ coupling and hence the $^1C_4$ conformer. This was confirmed by the signal for H-4 which appeared as a triplet of 3.8 Hz, due to ^3J-couplings with H-3 and H-5, and hence it cannot be in the axial position, as in the parent cyclodextrins. This set of coupling constants unambigously indicates the presence of the *altro*-$^1C_4$ conformation. All coupling constants were in good agreement with the predicted values (Table 27). The slightly higher error for $J_{3,4}$ may reflect some distortion away from an ideal $^1C_4$ conformer. The overall form of the cyclodextrin is predicted to be a broader cone with a ring of aromatics groups on the "top" face.



191 $^1C_4$

Scheme. Heptakis(3-deoxy-3-[4-*t*-butyl-benzyl]-6-*O*-*t*-butyldimethylsilyl)-β-cycloaltrin **191** viewed at 45° from the top face.

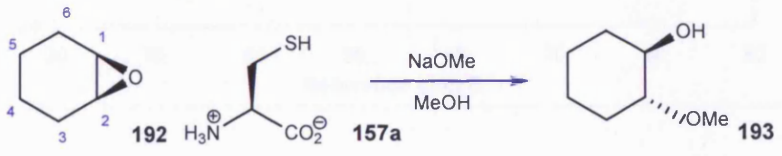
A MALDI mass spectrum provided the [M +K^+] ion at *m/z* 3109.5 for $C_{161}H_{266}O_{28}S_7Si_7K$. Simulation of the ion cluster shows perfect agreement with the measured value. Interestingly, the highest abundance peak at high resolution was calculated to be 3107.5492.



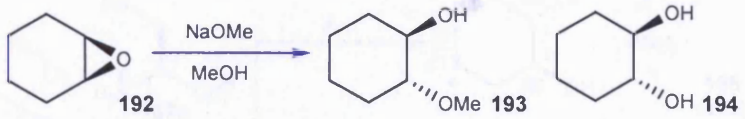
4.2.3.2. L-Cysteine

4.2.3.2.1. Model studies of cyclohexane oxide with L-cysteine

A Beilstein search for the reactions of epoxides with cysteines revealed 152 examples in which activated epoxides (mostly allyl epoxides) which had been opened by cysteine but only one report of an non-activated epoxide; cyclohexene oxide[149]. Therefore we repeated this reaction and applied the same methodology to 2,3-anhydro-4,6-*O*-benzylidene-methyl-α-D-mannopyranoside **109** as a model of heptakis(2,3-anhydro)-β-cyclomannin **117** as before. Firstly, cyclohexane oxide **192** was treated with L-cysteine **157a** in methanol at room temperature in the presence of sodium methoxide (25 – 30% solution in methanol) for 24 h. An acid-base wash and extraction with ethyl acetate collected (±)-*trans*-2-methoxycyclohexanol **193** in 38 % yield. Unreacted L-cysteine **157a** (in 60 % yield) was collected from the aqueous phase.



Formation of (±)-*trans*-2-methoxycyclohexanol **193** during reaction was confirmed when cyclohexane oxide **192** was stirred at r.t. for 24 h with sodium methoxide in methanol alone. Purification of the crude product using a solvent gradient of petroleum ether to ethyl acetate gave (±)-*trans*-2-methoxycyclohexanol **193** and (±)-*trans*-1,2-dicyclohexanol **194** in a ratio of 1:2 respectively. The large amount of the diol **194** was surprising and seemingly indicates that the small amount of water present in the methanol is much more nucleophilic than methanol.

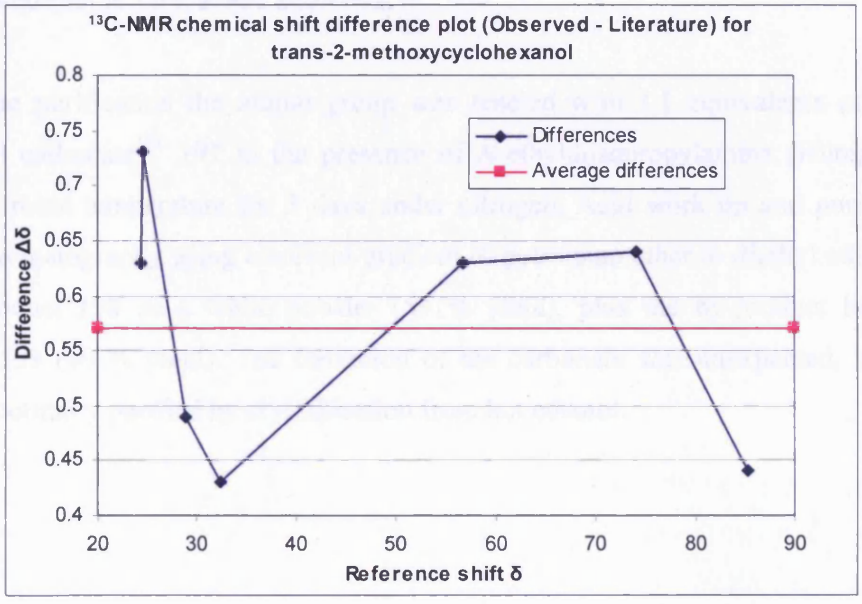


192 **193** **194**

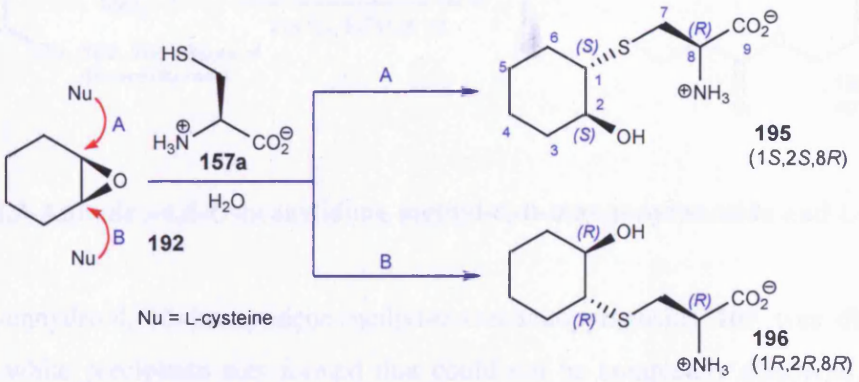
Comparison of the observed and literature[150] ^{13}C-NMR shift data for (±)-*trans*-2-methoxycyclohexanol **193** showed a systematic bias toward high values. When the average deviation (0.57 ppm) is subtracted from the differences, the remainder is less than normally expected experimental error.

Table 28. ^{13}C-chemical shift data for (±)-*trans*-2-methoxycyclohexanol **193**.

Observed	Literature[150]	Difference	Difference - 0.57
85.39	84.95	0.44	-0.13
74.15	73.51	0.64	+0.07
56.73	56.1	0.63	+0.06
32.43	32.0	0.43	-0.14
28.71	28.22	0.49	-0.08
24.53	23.8	0.73	+0.16
24.35	23.72	0.63	+0.06
Absolute averages		0.57	0.1

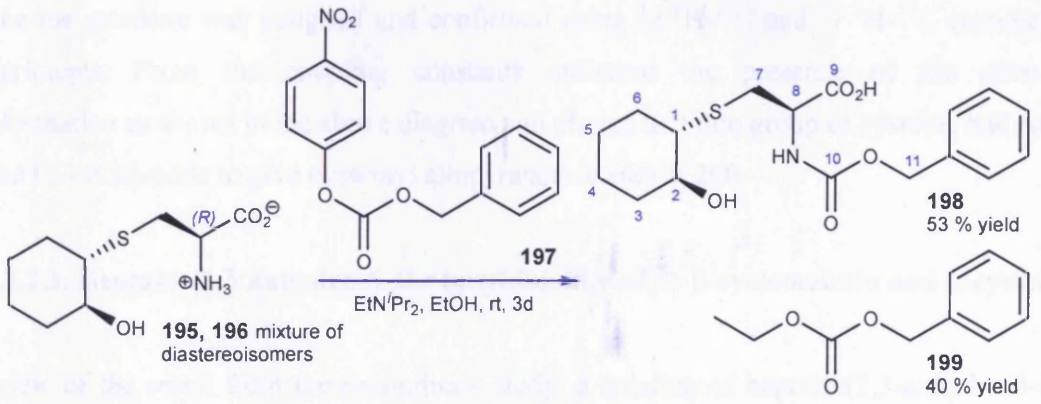


From information gleaned from the literature study, L-cysteine **157a** and an excess of cyclohexene oxide were reacted in water at room temperature over 4 days. The solution was extracted with ether and the aqueous evaporated to give the diastereomeric adducts **195**, **196** in 52 % yield. These result from attack of prochiral cyclohexane oxide **192** at the two possible sites.



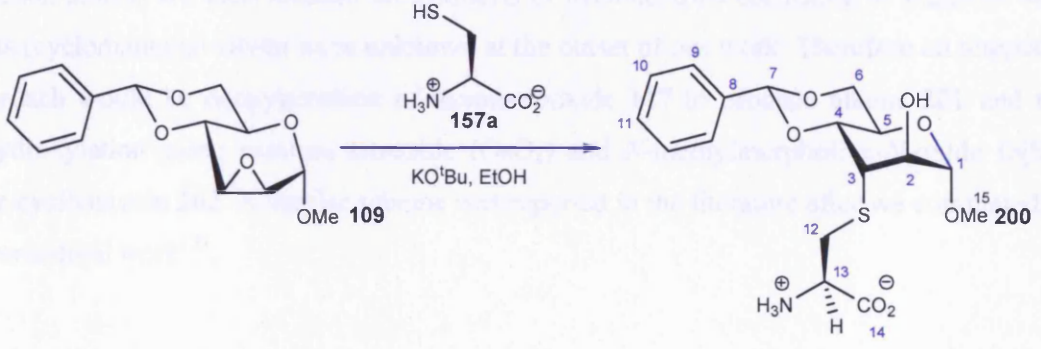
The ^{1}H-NMR spectrum of *trans*-1-(2-amino-2-carbonylethylthio)-2-cyclohexanol **195**, **196** showed a characteristic signal for each proton but assignment of the spectrum was very difficult because of overlapping signals, thus making it difficult to calculate coupling constants. The cysteinyl moiety had clearly been incorporated into the product; ^{1}H-NMR signals were found for H-8 (δ 3.25, dd, *J* 7.0, 4.9 Hz) which was overlapped with H-2, H-7a (δ 2.77, ddd, *J* 13.2, 8.1, 5.1 Hz,) and H-7b (δ 2.65, td, *J* 13.7, 7.0 Hz). In the ^{13}C-NMR spectrum each signal appeared as a pair. IR spectroscopy indicated the presence of a medium, broad NH_3^+ stretch at 2081 cm^{-1} and carboxylate anion at 1608 cm^{-1}. HRMS (ES^+) provided the protonated ($M+H^+$) ion at *m/z* 220.1002.

To facilitate purification the amino group was reacted with 1.1 equivalents of benzyl 4-nitrophenyl carbonate[151] **197** in the presence of *N*-ethyldiisopropylamine (Hünigs base) in ethanol, at room temperature for 3 days under nitrogen. Acid work up and purification by column chromatography using a solvent gradient of petroleum ether to diethyl ether gave the desired product **198** as a white powder (53 % yield), plus the by-product benzyl ethyl carbonate **199** (40 % yield). The formation of the carbonate was unexpected, because the reagent is normally purified by crystallisation from hot ethanol.



4.2.3.2.2. 2,3-Anhydro-4,6-*O*-benzylidene-methyl-α-D-mannopyranoside and L-cysteine

When 2,3-anhydro-4,6-*O*-benzylidene-methyl-α-D-mannopyranoside **109** was dissolved in ethanol, a white precipitate was formed that could not be completely dissolved even with warming or sonicating. Addition of a solution of cysteine (5 equivalents) in 4 ml H_2O, and reflux for 3 d., yielded only starting material and cysteine. Therefore the reaction was repeated again using potassium *tert*-butoxide (4 equivalents) as base and refluxed for 18 hr. A ^{1}H-NMR spectrum of the precipitates showed formation of the cysteinyl allopyranoside adduct **200** in 42 % yield with no further purification needed. The spectrum showed a singlet at 4.60 ppm for H-1, and a small doublet of 1.5 Hz at 4.10 ppm for H-2 indicating an equatorial-equatorial coupling with H-1. At 3.32 ppm, a doublet of doublets signal appeared with coupling constants of 3.7 and 2.5 Hz for H-3. The proton at C-4 appeared at 4.51 ppm as a doublet of doublets of 9.6, 4.2 Hz. The two geminal protons at C-12 position were displayed as two sets of doublet of doublets at 2.98, 2.85 ppm.



The two sets of methylene groups at C-6 and C-12 were simply distinguished in a ^{13}C-DEPT NMR spectrum by appearance of an upfield signal at 40.3 ppm belongs to C-12 due to its attachment to the sulfur atom. The carboxylate carbon (C-14) showed a signal at 180.8 ppm,

hence the structure was assigned and confirmed using ^{1}J ^{1}H-^{13}C and ^{3}J ^{1}H-^{13}C correlations experiments. From the coupling constants indicated the presence of the *altro*-$^{4}C_{1}$ conformation as shown in the above diagram and proved that thio group of cysteine had added to the C-3 of epoxide to give cysteinyl allopyranoside adduct **200**.

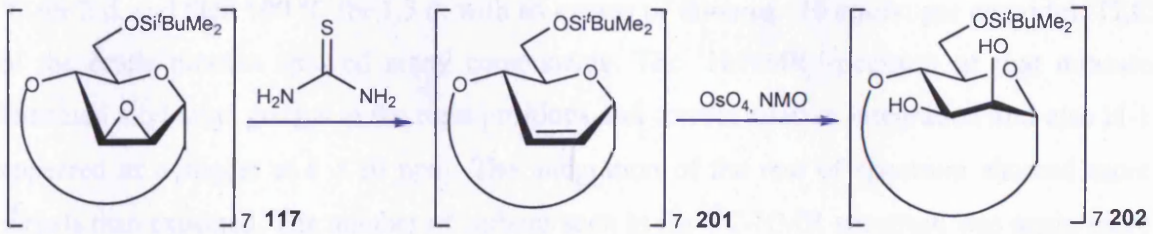
4.2.3.2.3. Heptakis(2,3-anhydro-6-*O*-*t*-butyldimethylsilyl)-β-cyclomannin and L-cysteine

In view of the result from the preliminary study, a solution of heptakis(2,3-anhydro-6-*O*-*t*-butyldimethylsilyl)-β-cyclomannin **117** in ethanol was refluxed with cysteine and potassium *tert*-butoxide, for three days under nitrogen. But only starting material was recovered from the organic layer. A ^{1}H-NMR spectrum of the crude mixture from the aqueous phase indicated cleavage of silyl groups due to the presence of water in the reaction mixture and a complicated mixture that could not be identified. A conclusion of these studies is that this methodology was not applicable to heptakis(2,3-anhydro)-β-cyclomannin **117**, as had been hoped at the outset.

4.3. The synthesis of cyclomannins by epoxide elimination-dihydroxylations

4.3.1. Introduction

The earlier work has described some methods for the synthesis of novel cyclodextrins including *manno*-epoxide ring opening with nucleophiles to give cyclodextrins with *altro*-stereochemistry rather than *gluco* (Section **5.2.4.1.2**) and also attempts to cleave the glucose ring. In this regard, two new methods were developed and worked well with the model (Section **5.3.1**). We then focused on synthesis of cyclodextrins consisting of mannose sugar units (cyclomannins) which were unknown at the outset of our work. Therefore an alternative approach would be deoxygenation of *manno*-epoxide **117** to produce alkene **201** and then dihydroxylation using osmium tetroxide (OsO_4) and *N*-methylmorpholine-*N*-oxide (NMO) give cyclomannin **202**. A similar scheme was reported in the literature after we completed our experimental work[138].

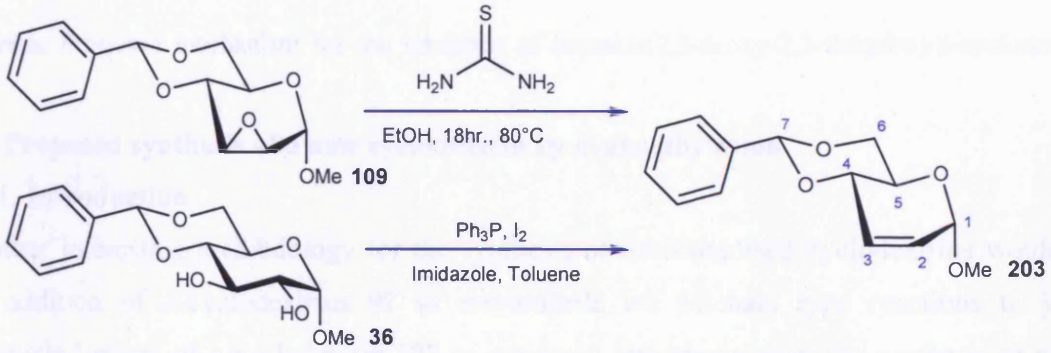


Scheme. Proposed approach for the synthesis of cyclomannin **202**

4.3.2. Model studies

4.3.2.1. Elimination reactions of model epoxides with thiourea

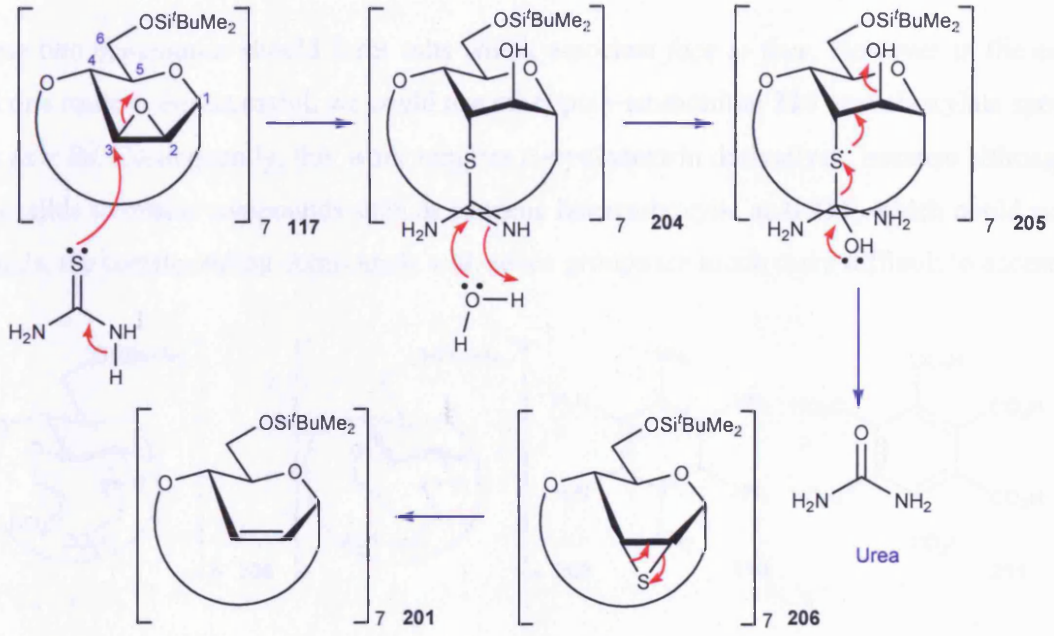
Both 2,3-anhydro-β-cyclomanin **136** and -allin were reported to react with excess thiourea in DMF to form the alkene **173** as the predominant product[127]. Hence the utility of this procedure was first tested with the monosaccharide model by refluxing a solution of 2,3-anhydro-4,6-*O*-benzylidene-α-methyl-D-mannopyranoside **109** in ethanol with a 10 fold molar excess of thiourea for 18 h. under nitrogen. After that time, ^{1}H NMR spectroscopy of the crude product showed two signals with chemical shifts 6.08 of 5.67 ppm which appeared as doublets with a coupling constant ($^3J_{2,3}$ 10.5 Hz), expected for alkene protons. Purification by column chromatography gave 4,6-*O*-benzylidene-2,3-dideoxy-methyl-α-D-gluco-hex-2-enopyranoside **203** in moderate yield (51 %). Comparison of the ^{1}H- and ^{13}C-NMR data for the alkene **203** with the data for the alkene obtained by reaction of 4,6-*O*-benzylidene-methyl-α-D-glucopyranoside **36** with triphenylphosphine, imidazole and iodine in refluxing toluene[152], confirmed the structure.



4.3.2.2. The reaction of heptakis(2,3-anhydro-6-*O*-*t*-butyldimethysilyl)-β-cyclomannin with thiourea

The highly facile one-pot synthesis of mono alkene **203** using thiourea was successful, thus heptakis(2,3-anhydro-6-*O*-*t*-butyldimethysilyl)-β-cyclomannin **117** was heated in DMF at 80

°C for 2 d. and then 100 °C for 1.5 d. with an excess of thiourea (10 equiv. per epoxide). TLC of the crude product showed many components. The ^{1}H-NMR spectrum of that mixture indicated silyl alkyl groups in the right positions and correct relative integration and also H-1 appeared as a singlet at δ 5.10 ppm. The integration of the rest of spectrum showed more signals than expected. The number of carbons seen in the ^{13}C-NMR spectrum was again more than expected, presumably due to incomplete reaction. The reaction was repeated under the same reaction conditions except with the addition of 10% w/w H_2O/DMF in order to remove urea from the reaction, the reaction time was prolonged to 10 d., unfortunately a very complicated reaction mixture was formed again that could not be identified.

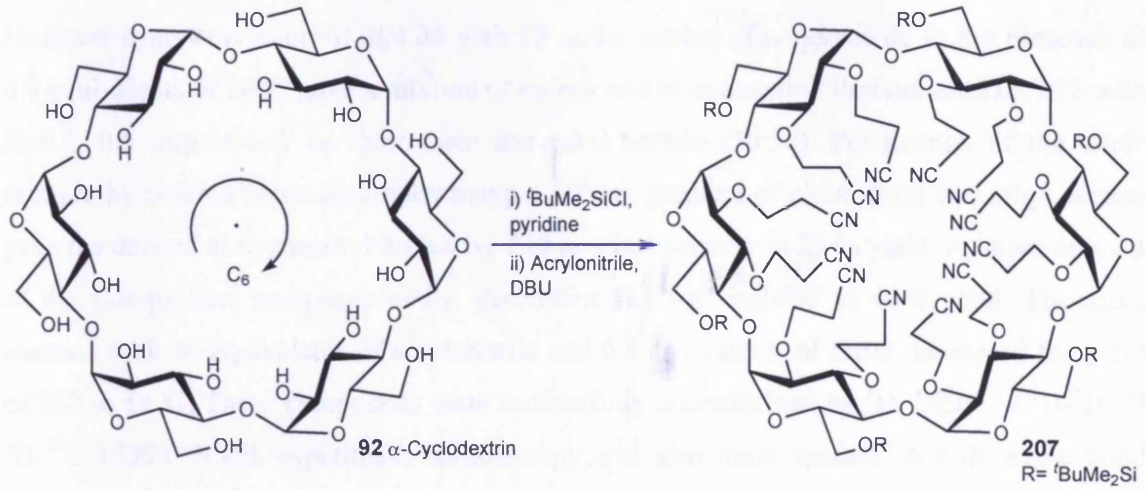


Scheme. Proposed mechanism for the synthesis of heptakis(2,3-deoxy-2,3-dehydro)-β-cyclodextrin **201**.

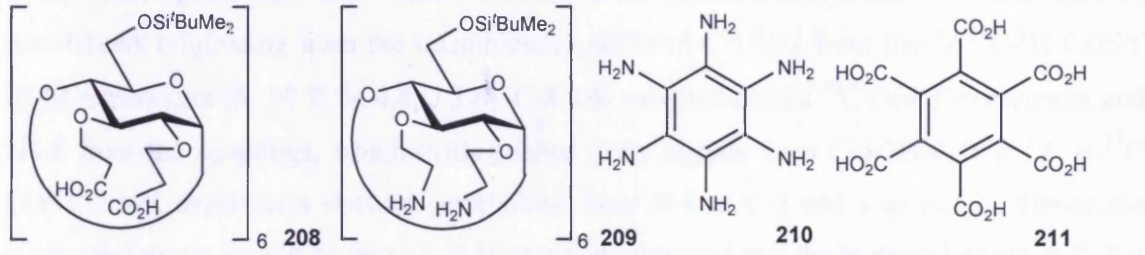
4.4. Proposed synthesis of a new cyclodextrin by cyanoethylation

4.4.1. Introduction

Another interesting methodology for the synthesis of functionalised cyclodextrins would be the addition of α-cyclodextrins **92** to acrylonitrile *via* Michael type reactions to yield cyanoethyl ethers of α-cyclodextrin **207** as important intermediates in the synthesis of novel cyclodextrin derivatives. Hydrolysis of the nitrile **207** would give the corresponding carboxylic acid derivative **208** whereas reduction would yield the amine **209**. From mixing the carboxylic acid **208** and the amine **209** an interesting salt will result.

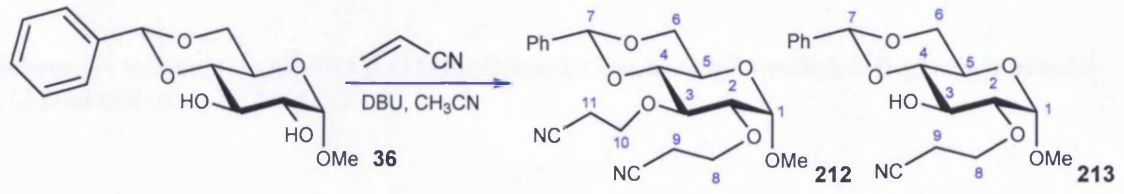


These two compounds should form salts which associate *face to face*. However in the event that one route is unsuccessful, we could use other poly-ammonium **210** or carboxylate species **211** as a *lid*. Consequently, this work requires α-cyclodextrin derivatives, because although it is possible to obtain compounds such as benzene hexacarboxylic acid **211**, which could act as ligands, the corresponding compounds with seven groups are much more difficult to access.



4.4.2. Cyanoethylation of the model diol; 4,6-*O*-benzylidene-α-methyl-D-glucopyranoside

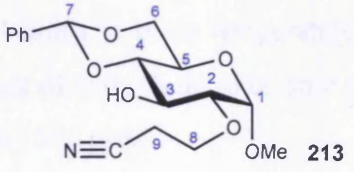
As a model for cyclodextrins, the diol **36** was used to prepare methyl 4,6-*O*-benzylidene-2,3-di-*O*-(cyanoethyl)-methyl-α-D-glucopyranoside **212**, using McGeary's methodology for the synthesis of glucose- and galactose-based monosaccharides[153]. The diol **36** was initially treated with acrylonitrile and 1,8-diazabicyclo[5.4.0]undec-7-ene (DBU) as a base in acetonitrile and stirred at room temperature for 2 d. to give the di-(cyanoethyl ether) glucoside **212**.



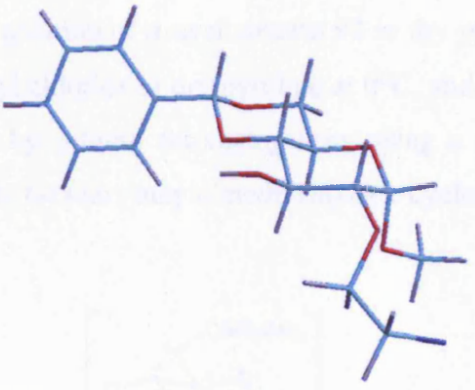
However cyanoethylation of diol **36** with 10 molar excess of acrylonitrile in the presence of 0.4 equivalents of DBU gave a mixture of mono- and di-cyanoethyl derivatives **213**, **212** with R_f 0.3, 0.5 respectively in chloroform and ethyl acetate (50:50). Purification of the crude mixture by column chromatography using a solvent gradient of chloroform and ethyl acetate gave the desired di-cyanoethyl derivative **212** as white powder in 28 % yield. A larger amount of the side-product mono-cyanoethyl derivative **213** was isolated in 45% yield. The same reaction with 20 equivalents of acrylonitrile and 0.8 equivalents of DBU, increased the yield of **212** to 54 %. These compounds were successfully characterized by ^{1}H, DEPT, J ^{1}H-^{1}H, ^3J ^{1}H-^{13}C COSY NMR experiment spectroscopy and also mass spectra. A C-N triple bond stretch at 2250 cm^{-1} was observed in the IR spectrum of the dinitrile **212** and the mononitrile **213.**

NMR analysis of 4,6-*O*-benzylidene-2-*O*-(cyanoethyl)-methyl-α-D-glucopyranoside 213

Assignment of the cyanoethyl group to the 2-*O*- or 3-*O*-positions required careful assignment of the NMR signals for C-2/C-3 and H-2/H-3. The assignments of H-2 and H-3 were based on correlations originating from the unambiguous shifts of C-1/H-1 from the ^1J ^{13}C-^{1}H COSY NMR experiment (δ_C 97.8, δ_H 4.8, J 3.6). C-8 was assigned from a ^{13}C DEPT experiment and 2H-8 from the couplings, which distinguished these signals from C-6/2H-6. The ^3J ^{1}H-^{13}C COSY NMR experiment showed correlations from H-8 to C-2 and also to CN. Hence the cyanoethyl group should be on C-2. It is generally observed that the hydroxyl group at C-2 is more reactive than that at C-3, because it is slightly more acidic.



213



Scheme. A molecular model of 4,6-*O*-benzylidene-2-*O*-(cyanoethyl)-methyl-α-D-glucopyranoside **213** produced using PC Model 8.

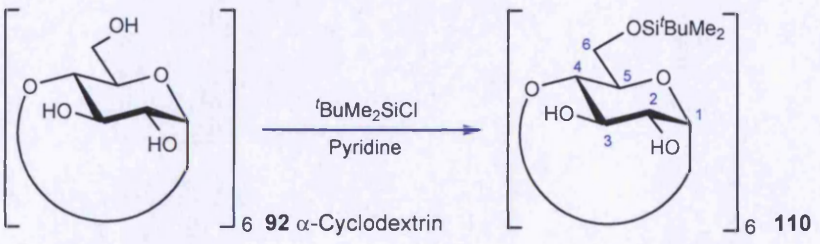
In order to optimise the reaction condition of the cyanoethylation, the reaction was repeated again by using more acrylonitrile and DBU (20, 8 equivalents respectively) and the reaction mixture was left to stir at room temperature for 3 d. At that time, only di-cyano derivative **212** was produced in 54 % yield. However when the reaction was conducted in THF and stirred at room temperature for 2 or 5 days in the presence of 4 equivalents potassium *tert*-butoxide, starting material **36** and mono-cyanoether derivative **213** were produced. When the more polar solvent, DMF was used and the reaction heated at 50 °C for 2 days, a mixture of mono- and di-derivatives **213**, **212** was formed in a ratio of 1:1 and surprisingly only starting material **36** was recovered when DMSO was used as solvent. The results are summarised in Table 29.

Table 29. Optimization of the cyanoethylation of methyl 4,6-*O*-benzylidene-methyl-α-D-glucopyranoside **36**.

Entry	Base	Equiv. of base	Solvent	Reaction time	product			Ratio
1.	DBU	0.4	CH_3CN	2d., r.t.	SM **36**	mono **144**	di **145**	0.5: 2.5, 7
2.	DBU	8	CH_3CN	3d., r.t.		mono **144**	di **145**	1:9
3.	KO^{t}Bu	4	THF	2d., r.t.	SM **36**			
4.	KO^{t}Bu	4	THF	5d., r.t.		mono **144**		
5.	KO^{t}Bu	8	DMF	2d., 50°C		mono **144**	di **145**	1:1
6.	KO^{t}Bu	8	DMSO	2d., 50°C	SM **36**			

4.4.3. Cyanoethylation of hexakis(6-*O*-*tert*-butyldimethylsilyl)-α-cyclodextrin

Due to the large number of hydroxyl groups (eighteen on α-cyclodextrin **92**) selective modification of these groups can be difficult. Therefore protection of the primary hydroxyl groups of α-cyclodextrin **92** were initially carried out using same method which was applied to the β-cyclodextrin **94** (Section 4.1.3.1.)[103]. A solution of α-cyclodextrin **92** in dry pyridine was added to a solution of *tert*-butyldimethylsilyl chloride in dry pyridine at 0°C, and stirred for 24 hours at room temperature. Purification by column chromatography using a solvent gradient of chloroform to ethanol yielded hexakis(6-*O*-*tert*-butyldimethylsilyl)-α-cyclodextrin **110** in 36 % yield.



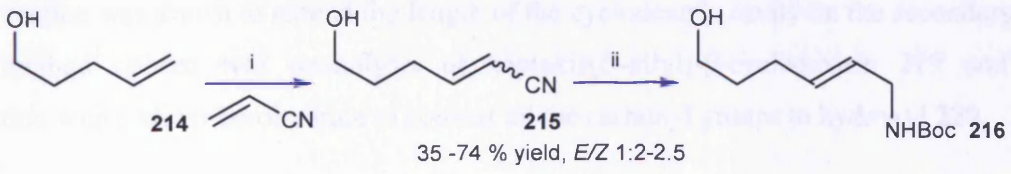
Application of the acrylonitrile method to the silyl ether **110** in the presence of DBU in acetonitrile was investigated. To a solution of the silyl ether **110** in acetonitrile, was added DBU (1.0 equiv. per glucose ring) and acrylonitrile (30 equiv. per glucose ring) which were stirred at r.t. for 2 d. and then refluxed for 1 d. (Entry 2). At that time ^{1}H-NMR spectra of the crude reaction mixture showed a mixture which was confirmed by TLC (chloroform:ethanol, 80:20) as having many components. Purification of the crude mixture was attempted first using a solvent gradient of dichloromethane to ethanol and then for a second time petroleum ether as a less polar solvent was used. But ^{1}H-NMR spectra of the fractions indicated an ambiguous mixture presumably due to an uncompleted reaction. Hence this reaction was repeated with different amounts of base and reagent and various reaction times, nevertheless a complex mixture was formed each time (Table 30).

Table 30. Optimization of the cyanoethylation of hexakis(6-*O*-*t*-butyldimethylsilyl)-α-cyclodextrin **110**.

Entry	DBU, equiv.	Acrylonitrile, equiv.	Solvent	Reaction time
1.	0.9	20	CH_3CN	3d., r.t.
2.	1	30	CH_3CN	2d., r.t.; 1d., reflux
3.	0.4	100	CH_3CN	5d., r.t
4.	0.4	100	THF	4d., r.t.; 4d., reflux
5.	18	83	neat	1d., 45°C

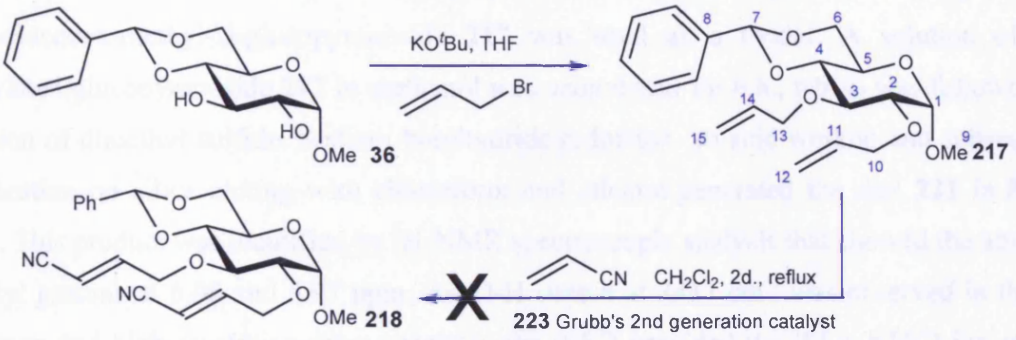
4.4.4. Allylation, and olefin metathesis or ozonolysis of glucopyranosides

It has been reported that selective cross-metathesis reactions between pent-4-en-1-ol and a range of terminal alkenes bearing cyano-groups proceeds in good yield. Acrylonitrile has a low tendency to undergo self-metathesis and hence cross-metathesis with alkenes such as pent-4-en-1-ol is a useful way to achieve homologation (**215**) under mild conditions. Reduction of the nitrile with diisobutyl aluminum hydride (DIBAH) and protection gave the amine **216**[154].



Scheme. Reaction conditions: i) Ru complex **223** (5 mol%), CH_2Cl_2, N_2, reflux, 2h., ii) DIBAH, Boc_2O, $NaBH_4$ in MeOH, $NaHCO_3$, rt., 18h[154].

Application of this procedure to per(*O*-allyl-ethers) of cyclodextrin would give a product similar to that of cyanoethylation. Accordingly, we set out to investigate a new cross metathesis process for the synthesis of a nitrile cyclodextrin that avoided use of a base catalyst. To examine if the one-step three-component reaction was possible with allyl compounds a model was studied. 2,3-Di-*O*-allyl-4,6-*O*-benzylidene-methyl-α-D-gluco-pyranoside **217** was prepared via an S_N2 replacement by treatment of diol **36** with allyl bromide and potassium *tert*-butoxide in THF, under nitrogen for 28 h at room temperature, in 90 % yield as the sole product. The product was confirmed by ^{1}H NMR spectroscopy showing the presence of the 11-H and 14-H acyclic vinylic protons resonance as ddt, at 5.97, 6.08 ppm respectively. IR spectroscopy indicated the presence of an alkene stretch at 1646 cm^{-1}.

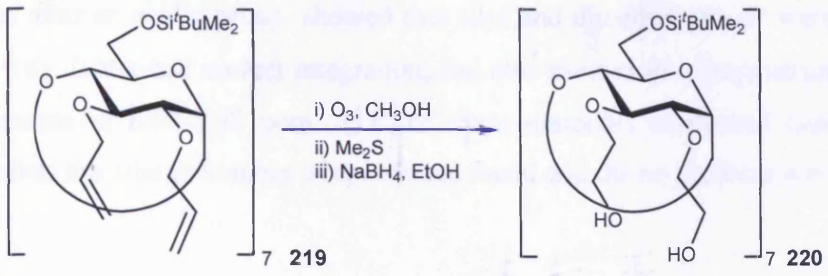


A mixture of Grubbs catalyst **223** (5 mol%), acrylonitrile (4 equiv.) and diallyl glucopyranoside **217** (0.07 M) were refluxed in distilled dichloromethane under nitrogen and the reaction progress was monitored by TLC. After 2 days starting material was identified from the crude reaction mixture.

4.5. Extension of the height of β-cyclodextrin

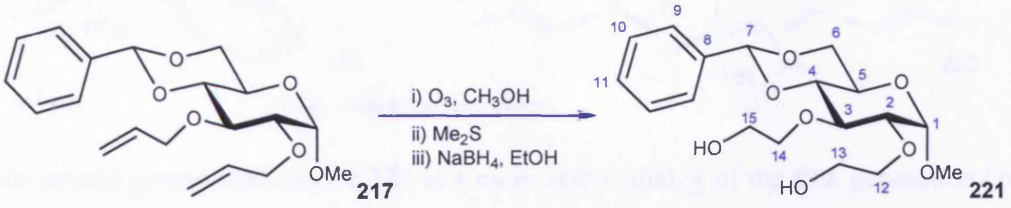
4.5.1. Introduction

In order to increase the compatibility between host (CD's) and guest (non-polar compounds), our intention was drawn to extend the length of the cyclodextrin cavity on the secondary face. The method chosen was ozonolysis of heptakis(*O*-allyl)-β-cyclodextrin **219** and then reduction with sodium borohydride to convert all the carbonyl groups to hydroxyl **220**.



4.5.1.1. Ozonolysis of the model alkene; 2,3-di-*O*-allyl-4,6-*O*-benzylidene-α-methyl-D-glucopyranoside

In order to examine the viability of the proposed ozonolysis method, 2,3-di-*O*-allyl-4,6-*O*-benzylidene-α-methyl-D-glucopyranoside **217** was used as a model. A solution of the diallylated glucopyranoside **217** in methanol was ozonolysed for 6 h., which was followed by addition of dimethyl sulfide. Sodium borohydride reduction, an acid workup and subsequent purification on silica eluting with chloroform and ethanol generated the diol **221** in 85 % yield. This product was identified by ^{1}H-NMR spectroscopic analysis that showed the absence of allyl protons at 6.08 and 5.97 ppm. An O-H stretch at 3417 cm^{-1} was observed in the IR spectrum and high resolution mass spectroscopy (ES^+) provided the ($M + NH_4^+$) ion at *m/z* 388.1966.



4.5.1.2. Ozonolysis of heptakis(2,3-di-*O*-allyl-6-*O*-*t*-butyldimethylsilyl)-β-cyclodextrin

Having developed the ozonolysis methodology for the preparation of diol **221** from diallylated glucopyranoside **217**, the synthetic approach towards tetradecakis(hydroxyl)-β-cyclodextrin **220** from the tetradecakis(*O*-allyl)-derivative **219** began. The allylated-β-cyclodextrin **219** was prepared in 36 % yield by using allyl bromide and sodium hydride in DMF (Section 4.6.3.2.1.).

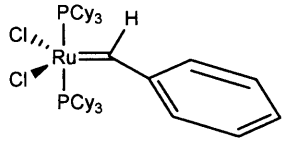
A solution of **219** in methanol was then treated with ozone for 8 h., dimethyl sulfide added and subsequently reduced with sodium borohydride overnight. ^{1}H-NMR spectrum of the

crude product after an acid workup, showed that silyl and dimethyl groups were in the right position (δ_H 0.85, 0.00), had correct integration, and also showed the disappearance of vinylic proton resonances at 6.03-5.92 ppm. TLC of those materials confirmed consumption of starting materials but a large number of spots were found and the no products were isolable.

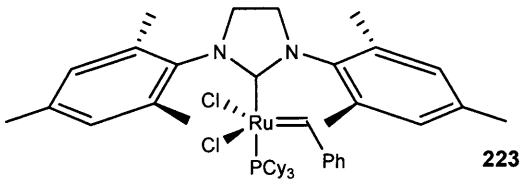
4.6. Ring Closing Metathesis (RCM)

4.6.1. Introduction

The ring closing olefin metathesis (RCM) employing the Grubb's catalyst has become an efficient tool in organic synthesis for constructing various sizes of cyclic compounds over the past decade[140]. RCM has been applied to a wide range of molecules containing ether, ester, amide and/or amine functionalities employing the first- and second-generation olefin metathesis catalysts **222**, **223** respectively[155]. There has been a single example of the application of RCM to cyclodextrins, which was reported after I finished my experimental work. This report described the formation of a single ring from a 6^A,6^D-di-*O*-allyl-α-cyclodextrin[156].



222



223

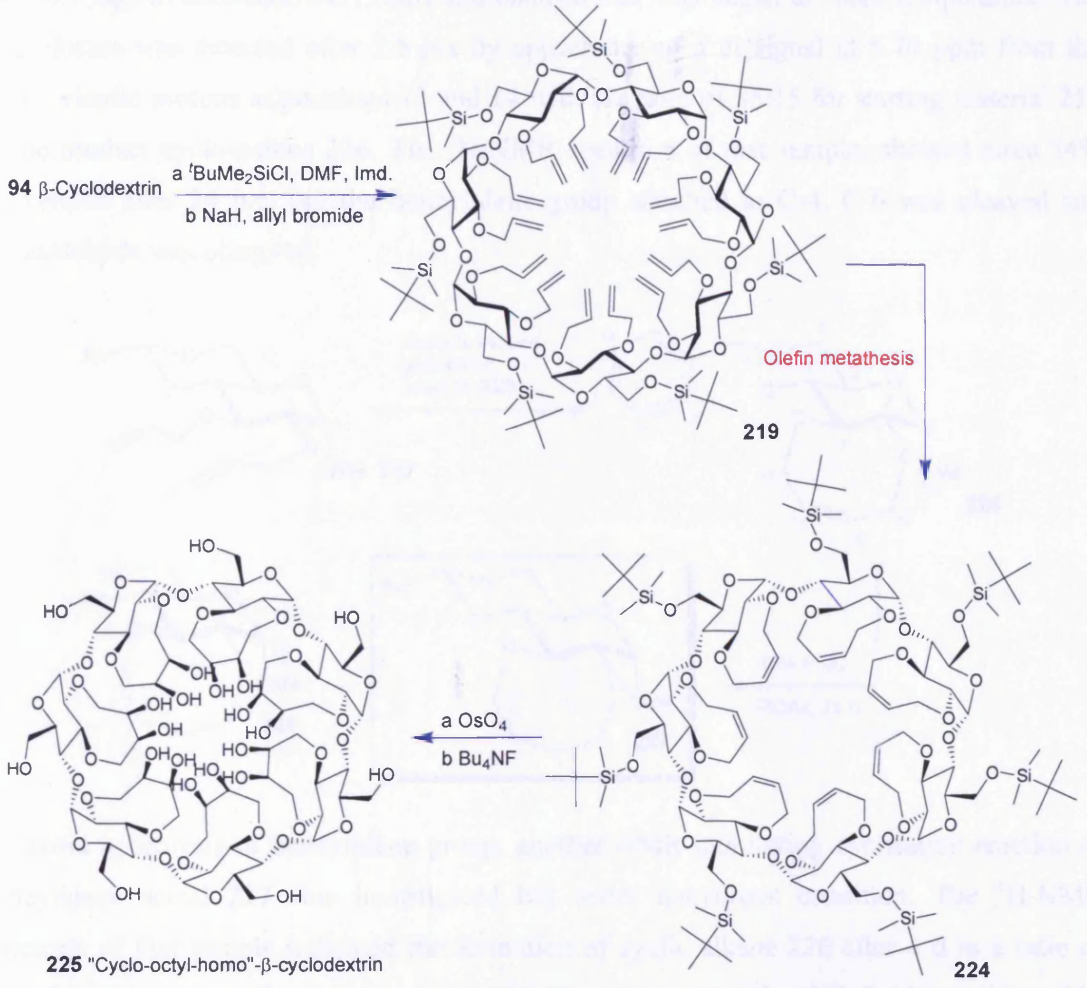
PCy_3 : tricyclohexylphosphine

Grubbs second generation catalyst **223** is a more active analog of the first generation Grubbs catalyst for ring closing metathesis. It ring closes olefins with excellent functional group tolerance and selectivity. Hence we have chosen this catalyst in our work.

4.6.2. Proposed synthesis of "cyclo-octyl-homo"-β-cyclodextrin 225

The per-silylated derivatives of CD can be prepared in only one step as explained in section (4.1.3.1.) rather than the multi-step procedure required for the synthesis of per-epoxidised cyclodextrins **117** (Section 4.2.2.). The epoxy-cyclodextrins and the thiiranes described previously are the only cyclodextrin derivatives known with new *fused* rings. Therefore our attention was drawn to apply the ring closing metathesis reactions of the β-cyclodextrin **94**. The RCM precursor is heptakis(2,3-di-*O*-allyl-6-*O*-*tert*-butyldimethylsilyl)-β-cyclodextrin

219, which upon olefin metathesis would produce the cyclic olefin derivatives **224** within the cyclodextrin bucket. *Syn*-dihydroxylation with osmium tetroxide followed by desilylation using tetrabutylammonium fluoride would give cyclo-octyl-homo-β-cyclodextrin **225** as an interesting novel CD with seven 8-membered rings on the top face.



Scheme. Proposed synthesis of "cyclo-octyl-homo"-β-cyclodextrin **225**.

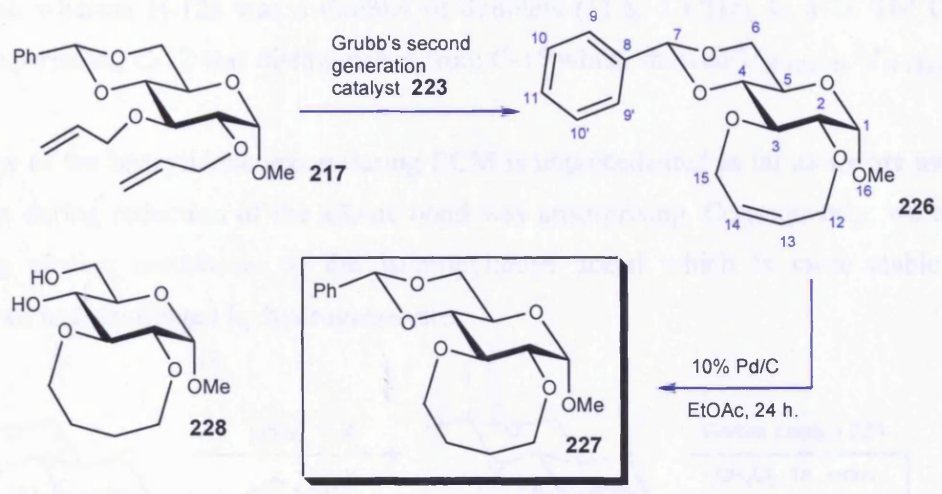
4.6.3. Ring closing metathesis

4.6.3.1. Model studies: Ring closing metathesis of 2,3-di-*O*-allyl-methyl-α-D-gluco-pyranosides 217, 229

RCM was first carried out on the 2,3-di-*O*-allyl-4,6-*O*-benzylidene-methyl-α-D-gluco-pyranoside **217** in dichloromethane with 20 mol % of the second generation Grubbs catalyst **223** under nitrogen at room temperature for 3 days, but after that time no expected product was formed. ^{1}H-NMR spectrum of reaction of di-allyl-pyranoside **217** with 13 mol % and

0.15 mol % of the same catalyst in dichloromethane and refluxing for six days showed minor conversion and low amount of materials were recovered.

Therefore an NMR monitoring reaction using a lower concentration of di-*O*-allyl-pyranoside **217** (0.7 mg) in chloroform-*d* (1 ml) and catalyst **223** was began at room temperature. The ring closure was detected after 2.5 hrs by appearance of a dt signal at 5.70 ppm from the cyclic vinylic protons at positions 13 and 14 with the ratio of 85:15 for starting material **217** to the product cyclo-octene **226**. The ^{1}H-NMR spectrum of that sample, showed circa 54% conversion after 24 hrs, but the benzylidene group attached to C-4, C-6 was cleaved and benzaldehyde was observed.

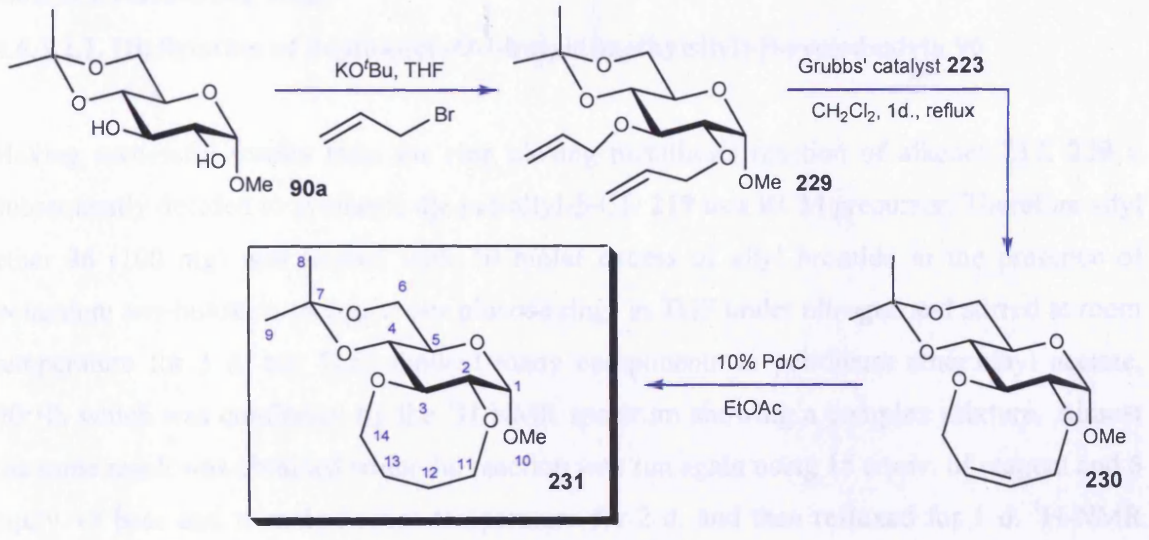


To avoid hydrolysis of benzylidene group, another NMR monitoring cyclisation reaction of benzylidene acetal **217** was investigated but under anhydrous condition. The ^{1}H-NMR spectrum of that sample indicated the formation of cyclic alkene **226** after 4 d in a ratio of circa 85:15 to unreacted starting material **217**. Therefore a sample of diallyl benzylidene **217** was placed under high vacuum for 1.5 hr. and distilled dichloromethane was added under nitrogen. Cyclisation with 5 mol % Grubbs catalyst **223** in dichloromethane gave cyclooctene **226** in 56 % yield after 24 hr. reflux following purification by column chromatography on silica. Hydrogenation of cyclooctene **226** for 24 hr. in ethyl acetate over 10 % palladium on carbon, gave the tricyclic benzylidene cyclooctane **227** in 72% yield following purification on silica, and also the bicyclic cyclooctane **228** formed by hydrogenolysis of the benzylidene group was isolated in 25 % yield. The structures of the products **227**, **228** were confirmed by ^{1}H, DEPT, *J* ^{1}H-^{1}H, ^{1}J ^{1}H-^{13}C and ^{3}J ^{1}H-^{13}C COSY NMR data, IR spectrum and mass spectroscopy.

NMR analysis of tricyclic benzylidene 1,4-dioxa-cyclooctane 227

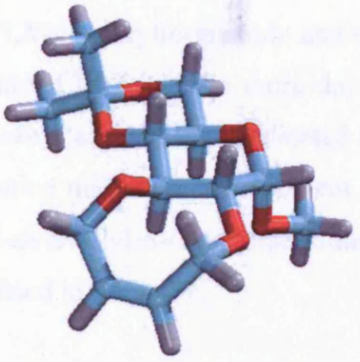
The ^{1}H-NMR spectrum of **227** showed unambiguous shifts for H-7, H-1 in a ^{1}J ^{1}H-^{13}C correlations with C-7, C-1 (δ_H 5.50, 4.78; δ_C 101.9, 100.5 respectively). From correlations in a ^{1}H-^{1}H COSY45 experiment, H-2, 3, 4 were assigned. The two sets of methylene groups at C-13, C-14 were distinguished from the other methylene groups at C-12, C-15, C-6 by appearance of the up-field multiplet signals in proton and carbon spectrum. Assignment of the signals for the H-6a, H-6b were based in a ^{1}J ^{1}H-^{13}C COSY NMR experiment (δ_H 4.30, 3.65, δ_C 69.2). The assignment of the signals for the methylene groups at C-12, C-15 was difficult because of signal overlapping. The side of the signal attributed to H-5 showed a complex multiplet, whereas H-12a was a doublet of doublets (11.8, 5.1 Hz). In a ^{3}J ^{1}H-^{13}C COSY NMR experiment, C-12 was distinguished from C-15 which showed $^{3}J_{\text{H-12,C-2}}$, $^{3}J_{\text{H-15,C-3}}$.

Cleavage of the benzylidene group during RCM is unprecedented as far as we are aware, but cleavage during reduction of the alkene bond was unsurprising. Consequently, we turned to the ring closing metathesis of the isopropylidene acetal which is more stable toward hydrolysis and unaffected by hydrogenolysis.



Therefore using a similar procedure the isopropylidene acetal **90a** reacted with allyl bromide in the presence of potassium *tert*-butoxide in THF to give the diene **229** in 85 % yield and without the need for column chromatography in this latter case. Successful cyclisation of diene **229** using 5 mol % Grubbs catalyst **223** in dichloromethane refluxed overnight, resulted in preparation of **230** in 75 % yield after purification using slow gradient of petroleum ether, ethyl acetate. Catalytic hydrogenation of the alkene **230** was carried out using 10 % palladium

on carbon in ethyl acetate resulting in the preparation of cyclooctane **231** after 24 hr. stirring at room temperature. The product was identified by ^{1}H NMR spectroscopy showing the absence of the 12-H, 13-H vinylic protons resonance at 5.64 ppm. Analysis of the high resolution mass spectrum (ES^+) showed the protonated MH^+ at *m/z* 289.1647. The structures of the cyclooctene **230** and cyclooctane **231** were confirmed by HSQC and HMBC NMR techniques.

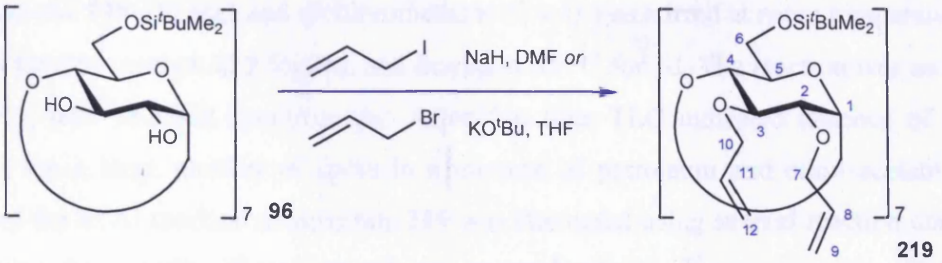


Scheme. A molecular model of cyclooctane **231** produced using PC Model 8 and Pov-Ray. The carbon atoms are blue, the oxygen's red, and the hydrogen atoms are grey.

4.6.3.2. Cyclodextrin study

4.6.3.2.1. Diallylation of heptakis(6-*O*-*t*-butyldimethylsilyl)-β-cyclodextrin 96

Having successful results from the ring closing metathesis reaction of alkenes **217**, **229** it subsequently decided to synthesis the per-allyl-β-CD **219** as a RCM precursor. Therefore silyl ether **96** (100 mg) was treated with 10 molar excess of allyl bromide in the presence of potassium *tert*-butoxide (4 equiv. per glucose ring) in THF under nitrogen and stirred at room temperature for 3 d. but TLC showed many components in petroleum ether:ethyl acetate, 90:10, which was confirmed by the ^{1}H-NMR spectrum showing a complex mixture. Almost the same result was obtained when the reaction was run again using 15 equiv. of reagent and 6 equiv. of base and stirred at room temperature for 2 d. and then refluxed for 1 d. ^{1}H-NMR spectrum revealed an unambiguous multiplet at δ 6.02 belonging to one of the allyl protons at position 8 or 11, but the material was a mixture. Allyl iodide was used as more reactive reagent with potassium *tert*-butoxide, but after 3 days stirring in room temperature, no product could be isolated. However, when the more powerful base; sodium hydride was used, with allyl iodide and the reaction was conducted in *N*,*N*-dimethylformamide[157] a 7% yield of the desired product **219** was obtained.



An improved yield of 36% was observed when sodium hydride was added to a chilled (0°C) solution of silyl β-CD **96** in dry *N,N*-dimethylformamide and stirred over night, and then to that solution allyl bromide was added[158]. After one more day stirring at room temperature TLC eluted with petroleum ether:ethyl acetate, 95:5 indicated the product **219** as the fastest spot. Purification of the crude reaction mixture using a solvent gradient of petroleum ether to ethyl acetate yielded heptakis(2,3-di-*O*-allyl-6-*O*-*tert*-butyldimethylsilyl)-β-cyclodextrin **219** as a syrup. The results are summarised in Table 31.

Table 31. Different reaction condition for the synthesis of per-ally β-CD **219**.

Entry	Solvent	Reagent and equiv.	Base and equiv.	Reaction time	Product
1	THF	Allyl bromide, 10	KO^{t}Bu, 4	3d., r.t.	mixture
`2	THF	Allyl bromide, 15	KO^{t}Bu, 6	2d., r.t., 1d.,68°C	mixture
3	THF	Allyl iodide, 10	KO^{t}Bu, 4	3d., r.t.	mixture
4	DMF	Allyl iodide, 5	NaH, 5	2d., r.t.	7%
5	DMF	Allyl bromide , 18	NaH, 11	1d., 0°C, 1d., r.t	36%

Allylation at position 2 and 3 was verified by data from ^{1}H, DEPT, J ^{1}H-^{1}H, ^3J ^{1}H-^{13}C COSY NMR experiment spectroscopy. In the ^{1}H-NMR spectrum two sets of multiplets were observed at 6.03, 5.92 related to allyl protons at position 11, 8 respectively. From J ^{1}H-^{1}H COSY NMR spectrum the correlation of H-8 with protons at C-7, C-9 observed and also H-11 with protons on C-10, 12. The methylene protons at position 10, 7 showed a ^3J correlation with C-2, C-3 respectively in a ^3J ^{1}H-^{13}C COSY NMR experiment. A MALDI mass spectroscopy analysis revealed a significant peak at *m/z* 2534.3 ($M + K^+$) corresponding to the expected molecular mass of per-allyl-β-cyclodextrin **219**.

4.6.3.2.2. Ring closing metathesis of heptakis(2,3-di-*O*-allyl-6-*O*-*t*-butyldimethylsilyl)-β-cyclodextrin 219

Following the preliminary study of RCM reactions which resulted in successful formation of eight-membered ring products **226, 230** from dienes **217** and **229**, a solution of the allylated

β-cyclodextrin **219** (20 mg) and dichloromethane (2 ml) was stirred at room temperature over 5 mol % Grubbs catalyst **223** for 2 d. and heated at 35 °C for 1d. The reaction was monitored using TLC and ^{1}H-NMR spectroscopy. After that time TLC indicated absence of starting material, but a large number of spots in a mixture of petroleum and ethyl acetate (95:5). Therefore the RCM reaction of substrate **219** was attempted using several reaction conditions and varying the quantity of catalyst, solvent, temperature and also reaction time (Table 32). To avoid the formation of products by intermolecular reaction (cross metathesis, CM)[155], a solution of alkene β-CD **219** in THF at 1 mM or 0.58 mM concentration was heated at reflux for 4 or 10 days in the presence of 2-5 mol% of catalyst **223**, but again a complicated mixture was obtained confirmed by ^{1}H-NMR spectroscopy even after purification by column chromatography.

The synthesis of radicicol, a macrocycle natural product which is an antifungal antibiotic, used Grubbs catalyst **223** in the ring closing metathesis reaction. When the reactions were performed in toluene at 110°C for a short period of time and also at lower substrate concentration the monomer macrocycles were present as predominant products over the dimers[159]. Therefore in a further attempt, the reaction was conducted in refluxing toluene using a Dean-Stark apparatus in the presence of 10 mol% of the Grubbs catalyst **223**, but again TLC indicated many spots which were impossible to separate.

Table **32.** Ring closing metathesis reaction of substrate **219** under different reaction conditions

Entry	Condition	Concentration, mM	Catalyst, mol%
1.	CH_2Cl_2, r.t., 2 d., 35°C, 1 d.	10	5
2.	THF, reflux, 4 d.	1	2
3.	THF, reflux, 10 d.	0.58	2
4.	Toluene, reflux, 2 d.	1	10

4.7. Conclusion and future studies:

In summary, the purpose of the work in this thesis was to investigate the potential for the synthesis of cyclodextrins with novel stereochemistry. In this regards, the study was exclusively focused on the reactions to modify the secondary face of the cyclodextrins, which is wider and are able to accommodate a wide range of non-polar compounds within their cavities, for use as supramolecular building blocks.

Therefore, cleavage of the vicinal diol moieties in β-cyclodextrin (Section 4.1), allylation of the silylated β-cyclodextrin and subsequently ring closing metathesis (RCM) reaction (Section 4.6), ozonolysis reaction (Section 4.5), cyanoethylation reaction (Section 4.4), and finally epoxide-ring opening reaction with thiourea to form alkenes (Section 4.3), and also with a range of nucleophiles (Section 4.2) were investigated. The *heptakis*(2,3-anhydro)-β-cyclomannin reacted with 4-*tert*-butylbenzylthiol and gave the *altro*-$^{1}C_{4}$ conformation.

There are several lines of research arising from this work which is, to investigate the guests for the thiol-cyclodextrin **191**, using functional thiol-CD's, and also to extend the cyclodextrins ring on both sides (primary and secondary).

Chapter 5

Experimentals

5. Experimental

5.1. General

Commercially available reagents were used as received without further purification. Reactions were carried out under an atmosphere of dry nitrogen. Flash chromatography was performed using Matrex silica 60 35-70 micron. Analytical thin layer chromatography (TLC) was carried out using aluminium-backed plates coated with Merck Kiesegel 60 GF_{254} that were visualized under UV light (at 254 and/or 360 nm) and/or ethanolic phosphomolybdic acid (5%) and heat as developing agents. Melting points were recorded on a Kofler hot stage apparatus and are uncorrected.

Infra-red spectra were recorded in the range 4000-600 cm^{-1} on a Perkin-Elmer 1600 series FT-IR spectrometer either as a thin film between NaCl plates, in solution in dichloromethane or as a KBr disk, as indicated. Low-resolution mass spectra were recorded using a Fisons VG platform II spectrometer and on a Micromass Q-TOF Micro spectrometer using electrospray ionization (ES) unless otherwise stated. High-resolution mass spectra were obtained from EPSRC Mass Spectrometry Service at University College of Wales, Swansea using the ionization method indicated. NMR spectra were recorded using Bruker DPX 400 instruments operating at 400 MHz for ^{1}H spectra and 100 MHz for ^{13}C spectra at 25°C, or on a Bruker Avance 500 spectrometer operating at 500 MHz for ^{1}H and 125 MHz for ^{13}C at 25°C. All chemical shifts are reported in ppm downfield from TMS. Coupling constants (*J*) values were reported in Hz. Chiral compounds were analysed using a AA-1000 Polarimeter apparatus using the sodium D line at the indicated temperature for [α] and are given in degree.

5.2. Analysis of mass spectrometry abundance data for multiple isotopomers

There are numerous programs available for calculating the ratio of isotopomers derived from a single molecular formulae, such the analytical data window in ChemDraw and HiMass (written by D. R. Kelly). However as far as I am aware there are no programs available for the calculating the ion cluster pattern for *mixtures* of isotopomers.

The current program (8_Iso_15P) is based on an earlier program developed for the analysis of mixtures of the three protonated/deuterated isotopomers of dichloromethane[160]. The program is written in Microsoft Visual Basic 3.0 using the *Skeleton environment*, which consists of a standard form, subroutines, functions, error code files and display utilities, suitable for

scientific programming. In practice this means that a complete program can be quickly written by changing the code invoked by the calculate button.

The program was designed to accommodate isotopomers consisting of eight ions with eight different amounts of incorporation of another isotope. Eight ions is sufficient to cover most elemental compositions including the majority of tin species to adequate accuracy. The eight degrees of incorporation is sufficient to cover the ring protons of a monosaccharide (D_0 to D_7). The specification; eight ions and eight degrees of incorporation requires the abundances of 15 peaks to be determined. In general for *n* ions and *m* degrees of incorporation the number of peaks to be determined is $n + m - 1$.

The central loops of the program are shown below, with some of the display, monitoring and counting code removed for the purposes of clarity. Comment statements are shown in an alternative font and multiple lines of code with an underscore.

All numerical variables and arrays are double precision and strict variable declaration is used. Operators starting with "FN" are functions and "DP" in a function name indicates that the function returns a double precision number.

```
Calc_Diff_Save = 50: ' This is the maximum acceptable error and later on in the program the lowest error found so far during the loops. The value is arbitrary and the value chosen only affects the number of nonsense results displayed in the early stages of the running of the program.
Start_Loop = 0: End_loop = 1: Step_Loop = .1: ' Loop parameters set
For n1 = Start_Loop To End_loop Step Step_Loop
For n2 = Start_Loop To End_loop Step Step_Loop
For n3 = Start_Loop To End_loop Step Step_Loop
For n4 = Start_Loop To End_loop Step Step_Loop
For n5 = Start_Loop To End_loop Step Step_Loop
For n6 = Start_Loop To End_loop Step Step_Loop
For n6 = Start_Loop To End_loop Step Step_Loop
For n7 = Start_Loop To End_loop Step Step_Loop
For n8 = Start_Loop To End_loop Step Step_Loop
' CalcAbun(n) is the calculated abundance of each peak. SAb(n) is the abundance of individual isotopomer in a single sub-cluster.
    CalcAbun(1) = (SAb(1)*n1)
    CalcAbun(2) = (SAb(2)*n1) + (SAb(1)*n2)
    CalcAbun(3) = (SAb(3)*n1) + (SAb(2)*n2) + (SAb(1)*n3)
    CalcAbun(4) = (SAb(4)*n1) + (SAb(3)*n2) + (SAb(2)*n3) + (SAb(1)*n4)
    CalcAbun(5) = (SAb(5)*n1) + (SAb(4)*n2) + (SAb(3)*n3) + (SAb(2)*n4) +
_(SAb(1)*n5)
    CalcAbun(6) = (SAb(6)*n1) + (SAb(5)*n2) + (SAb(4)*n3) + (SAb(3)*n4) +
_(SAb(2)*n5) + (SAb(1)*n6)
    CalcAbun(7) = (SAb(7)*n1) + (SAb(6)*n2) + (SAb(5)*n3) + (SAb(4)*n4) +
_(SAb(3)*n5) + (SAb(2)*n6) + (SAb(1)*n7)
    CalcAbun(8) = (SAb(8)*n1) + (SAb(7)*n2) + (SAb(6)*n3) + (SAb(5)*n4) +
_(SAb(4)*n5) + (SAb(3)*n6) + (SAb(2)*n7) + (SAb(1)*n8)
```

```
    CalcAbun(9) = (SAb(8)*n2) + (SAb(7)*n3) + (SAb(6)*n4) + (SAb(5)*n5) +
_(SAb(4)*n6) + (SAb(3)*n7) + (SAb(2)*n8)
    CalcAbun(10) = (SAb(8)*n3) + (SAb(7)*n4) + (SAb(6)*n5) + (SAb(5)*n6) +
_(SAb(4)*n7) + (SAb(3)*n8)
    CalcAbun(11) = (SAb(8)*n4) + (SAb(7)*n5) + (SAb(6)*n6) + (SAb(5)*n7) +
_(SAb(4)*n8)
    CalcAbun(12) = (SAb(8)*n5) + (SAb(7)*n6) + (SAb(6)*n7) + (SAb(5)*n8)
    CalcAbun(13) = (SAb(8)*n6) + (SAb(7)*n7) + (SAb(6)*n8)
    CalcAbun(14) = (SAb(8)*n7) + (SAb(7)*n8)
    CalcAbun(15) = (SAb(8)*n8)
    MaxCalcAbun# = FNMaxDP(CalcAbun(), 1, 15) / 100: 'Find the maximum abundance
    Call NormaliseDP(CalcAbun(), 1, 15, MaxCalcAbun#): 'Normalise abundances to
```
maximum abundance value. In this case, elements 1 to 15 of the double precision array CalcAbun() are normalized to MaxCalcAbun#.

```
' FoundAbun(n) contains the abundances of the experimental data. The following loop calculates the sum of the
```
absolute differences, between the data and calculated abundances.

```
    Calc_diff = 0
    For n101 = 1 To 15 Step 1
        Calc_diff = Calc_diff + Abs(FoundAbun(n101) - CalcAbun(n101))
    Next n101
```

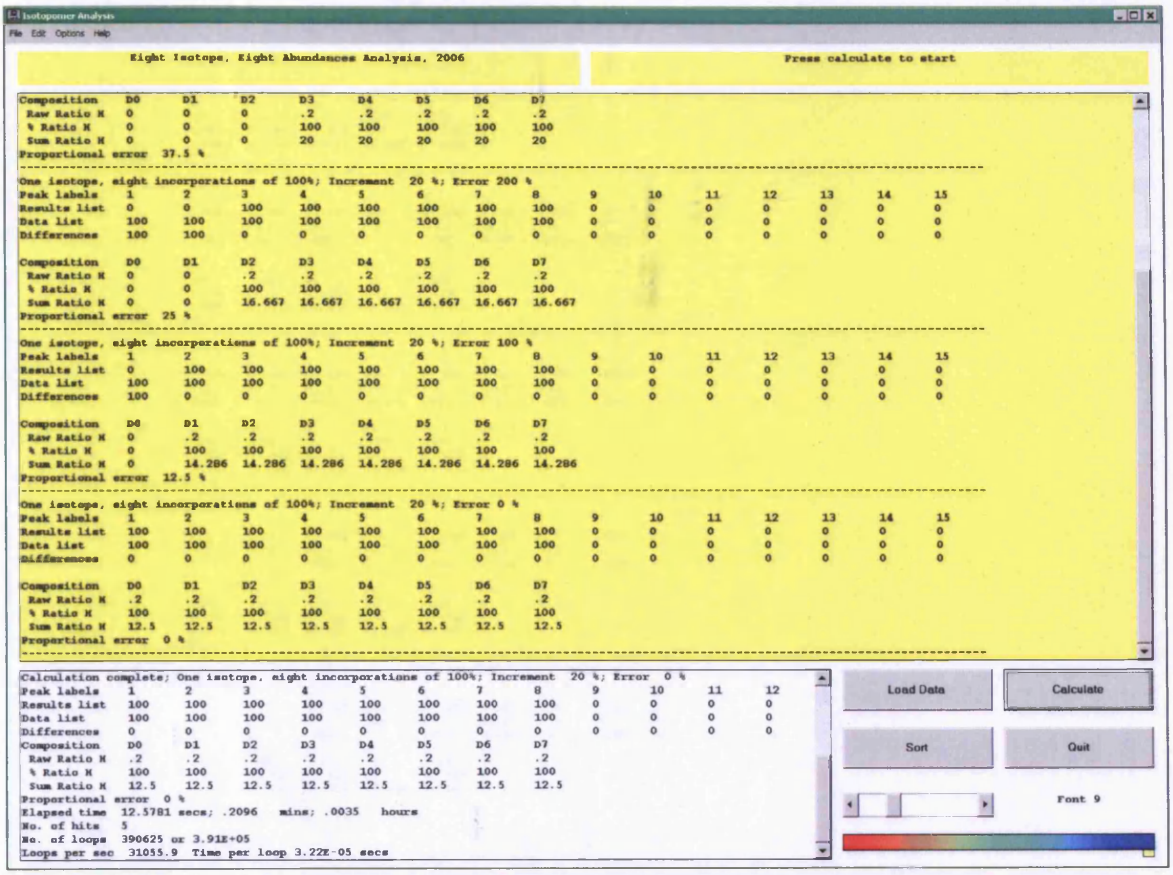
' The following statement determines if the sum of the absolute differences between found abundances and the data abundances ("the error") are less than the maximum acceptable error. If they are the results are displayed and the maximum acceptable error is set to the current error value. Therefore as the loops progress the error of the data displayed is progressively reduced and the fit between calculated and found abundances increases.

```
    If Calc_diff < Calc_Diff_Save Then
       Calc_Diff_Save = Calc_diff
       <<Display results>>
    End If
Next n8, n7, n6, n5, n4, n3, n2, n1
```

The majority of the time spent executing this loop is expended in the FNMaxDP function and the NormaliseDP subroutine, which find the maximum value in an array and normalize an array respectively; both work on double precision (DP) variables.

The results are sieved on the basis of total error, but this methodology is disproportionately affected by higher abundance peaks, because they have a higher numerical range and hence a larger potential effect on the sum of errors. However this is mitigated by the normalization routine, because the largest peak in the calculated abundances becomes 100% and if it matches the position of the largest peak in the data it can never show an error. As a comparison the proportional error is also reported, which is the absolute sum, of the errors of the differences between the calculated and found abundances, divided by the found abundances. Consequently for this measure, the difference between 80 and 60 has the same contribution as the difference between 4 and 3 for calculated and found abundances respectively.

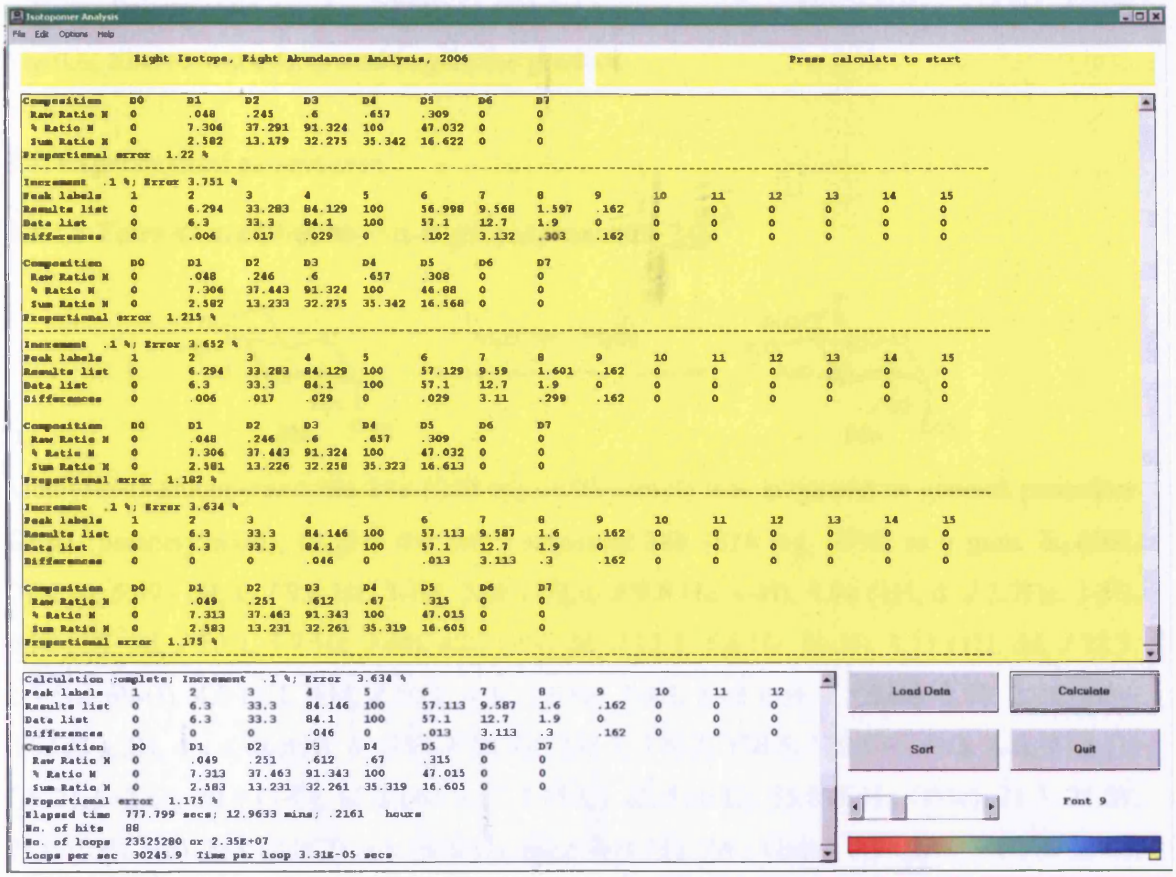
Screen capture of test data



A partial screen capture of the same run showing the top of the bottom left-hand side list box is shown below. This shows the raw data (8 x 30%), which is normalized to give the data set used in the loops (8 x 100) and the isotopes list, which is the list of abundances in the subcluster.

```
Started at 23:24:55 on Tuesday, 21 November 2006
Opened C:\Program Files\Isotopomers_8_peaks_15\Error.txt for input OK
Data read correctly from C:\Program Files\Isotopomers_8_peaks_15\Error.txt using the format; error reading
Current data set is One isotope, eight incorporations of 100%;
Peak labels     1       2       3       4       5       6       7       8       9       10      11      12
Raw Data list   30      30      30      30      30      30      30      30      0       0       0       0
Data list       100     100     100     100     100     100     100     100     0       0       0       0
Composition     D0      D1      D2      D3      D4      D5      D6      D7
Isotopes list   100     0       0       0       0       0       0       0
```

Screen capture of the final analysis of the tetra-*O*-acetyl-galactopyranosides **79b – 81b**.



5.3. General experimental procedures

5.3.1. General procedure for the deuteration of hexanopyranosides using Raney nickel (GP01)

Raney nickel (12 g, wet) was placed in a flask, deuterium oxide (4 ml) was added, the flask shaken and the supernatant (5 ml) decanted. The washing was repeated and deuterium oxide (4 ml, 221 mmol) added to the wet solid. The reaction was sonicated for 2 hr to "activate" the Raney Nickel. The hexopyranoside (200 mg) was added and the reaction refluxed. Filtered samples were analysed by ^{1}H-NMR.

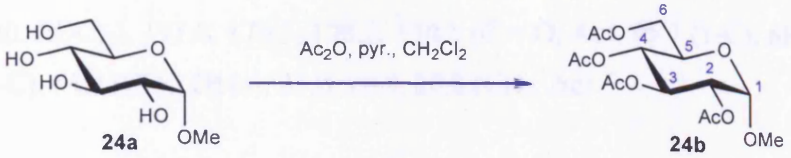
5.3.2. General procedure for the acetylation of hexanopyranosides[72] (GP02)

A mixture of pyridine (4.2 ml, 51.5 mmol, 50 equiv.) in dichloromethane (10 ml) and acetic anhydride (1.94 ml, 20.6 mmol, 20 equiv.) were placed in a flask in an ice bath. The sugar (200 mg) was added to the flask, which was kept in the ice bath for 3 hrs and in a refrigerator for two days. The solution was poured into a litre beaker filled with crushed ice. Sodium bicarbonate solution was added to the ice which melted to give a solution of pH 9. The

bicarbonate–water solution was extracted with ethyl acetate (3 x 100 ml) and washed with copper sulphate solution (3 x 60 ml) to remove excess pyridine. The solution was dried with $MgSO_4$, filtered and evaporated to give the product.

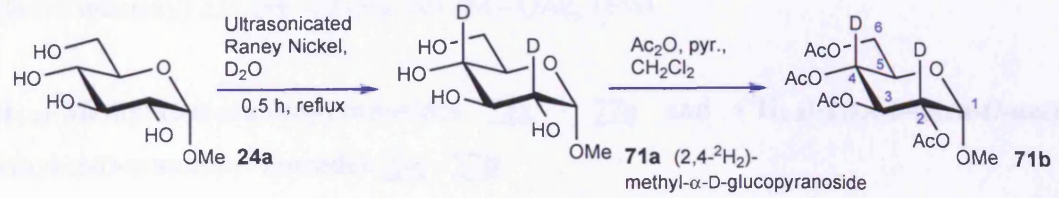
5.4. Experimental procedures

2,3,4,6-Tetra-*O*-acetyl-methyl-α-D-glucopyranoside 24b[72]



Methyl-α-D-glucopyranoside **24a** (200 mg, 1.03 mmol) was subjected to general procedure **GP02** (peracetylation), to give the *title compound* **24b** (326 mg, 87%) as a gum. δ_H (400, $CDCl_3$), 5.49 (1H, t, *J* 9.8 Hz, 3-H), 5.08 (1H, t, *J* 9.8 Hz, 4-H), 4.96 (1H, d, *J* 3.7Hz, 1-H), 4.90 (1H, dd, *J* 10.2, 3.7 Hz, 2-H), 4.27 (1H, dd, *J* 12.3, 4.6 Hz, 6a-H), 4.11 (1H, dd, *J* 12.3, 2.3 Hz, 6b-H), 4.0 (1H, ddd, *J* 10.2, 4.5, 2.3 Hz, 5-H), 3.42 (3H, s, OMe), 2.10, 2.08, 2.04, 2.02 (4 x 3H, 4 x s, acetyl); δ_C (100, $CDCl_3$), 171.1, 170.5, 170.5, 170.0 (C = O, Ac), 97.1 (1-C), 71.1 (2-C), 70.4 (3-C), 68.8 (4-C), 67.5 (5-C), 62.5 (6-C), 55.8 (CH_3, OMe), 21.1, 21.09, 21.0 (CH_3, Ac). MS (APCI) *m/z* (relative intensity) 331 (M - OMe), 330 (M - MeOH, 30%), 303 (M - OAc, 24%).

(2,4-$^2H_2$)-methyl-α-D-glucopyranoside 71a and (2,4-$^2H_2$)-2,3,4,6-tetra-*O*-acetyl-methyl-α-D-glucopyranoside 71b

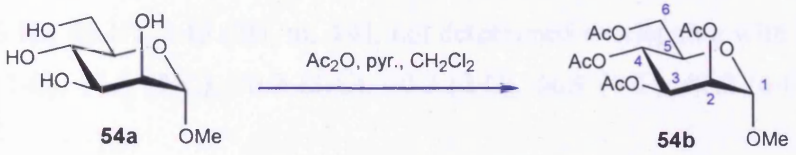


Methyl-α-D-glucopyranoside **24a** (200 mg, 1.03 mmol) was treated with Raney nickel (12 g, wet) according to general procedure **GP01**, refluxed for 0.5 hr. The catalyst was removed by filtration and the reaction was azeotroped with toluene (3 x 10 ml) to give dry product, (2,4-$^2H_2$)-methyl-α-D-glucopyranoside **71a** (110 mg), which was acetylated using general procedure **GP02** to give (2,4-$^2H_2$)-2,3,4,6-tetra-*O*-acetyl-methyl-α-D-glucopyranoside **71b** (160 mg, 80%).

(2,4-$^2H_2$)-Methyl-α-D-glucopyranoside 71a: δ_H (400, D_2O), 4.70 (1H, s, 1-H), 3.77 (1H, dd, *J* 12.3, 2.3 Hz, 6a-H), 3.64 (1H, dd, *J* 12.2, 5.5 Hz, 6b-H), 3.56 (2H, m, overlapping, 3-H, 5-H), 3.32 (3H, s, OMe); δ_C (100, D_2O), 99.5 (1-C), 72.2 (3-C), 71.8 (5-C), 60.7 (6-C), 55.3 (CH_3, OMe).

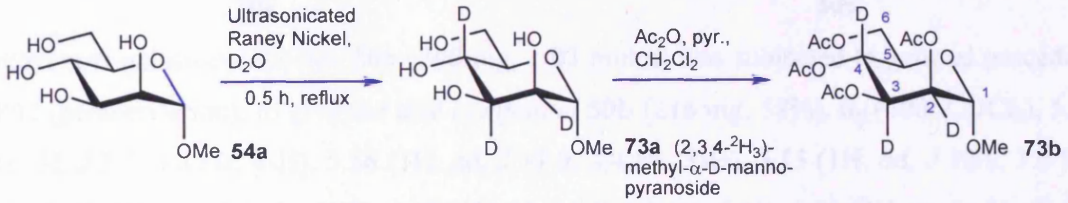
(2,4-$^2H_2$)-2,3,4,6-Tetra-*O*-methyl-α-D-glucopyranoside 71b: δ_H (400, $CDCl_3$), 5.49 (1H, s, 3-H), 4.96 (1H, s, 1-H), 4.27 (1H, dd, *J* 12.3, 4.5 Hz, 6a-H), 4.10 (1H, dd, *J* 12.3, 2.2 Hz, 6b-H), 3.98 (1H, dd, *J* 4.5, 2.1, 5-H), 3.40 (3H, s, OMe), 2.09, 2.06, 2.04, 2.0 (4 x 3H, 4 x s, acetyl); δ_C (100, $CDCl_3$), 171.0, 170.3, 170.2, 170.0 (C = O, Ac), 95.7 (1-C), 68.9 (3-C), 66.0 (5-C), 60.8 (6-C), 54.4 (CH_3, OMe), 21.0, 20.9, 20.8 (CH_3, Ac).

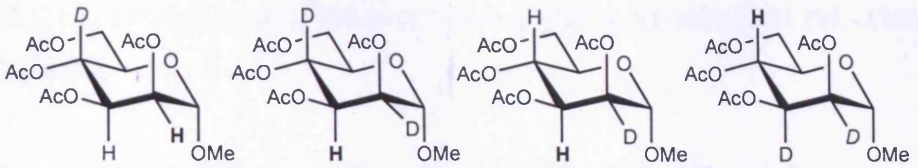
2,3,4,6-Tetra-*O*-acetyl-methyl-α-D-mannopyranoside 54b



Methyl-α-D-mannopyranoside **54a** (200 mg, 1.03 mmole) was subjected to general procedure **GP02** (peracetylation), to give the *title compound* **54b** (151 mg, 40%) as a gum. δ_H (400, $CDCl_3$), 5.33 (1H, dd, *J* 10.0, 3.2 Hz, 3-H), 5.28 (1H, t, *J* 9.8 Hz, 4-H), 5.21 (1H, dd, *J* 3.1, 1.6 Hz, 2-H), 4.72 (1H, d, *J* 1.3 Hz, 1-H), 4.30 (1H, dd, *J* 12.2, 5.3 Hz, 6a-H), 4.11 (1H, dd, *J* 12.2, 2.4 Hz, 6b-H), 3.96 (1H, m, 5-H), 3.41 (3H, s, OMe), 2.16, 2.11, 2.03, 2.0 (4 x 3H, 4 x s, acetyl); δ_C, 172.2, 171.8, 171.6, 171.6 (C = O), 98.6 (C-1), 69.5 (C-2), 69.1 (C-3), 68.4 (C-5), 66.2 (C-4), 62.5 (C-6), 55.3 (CH_3, OMe), 20.9, 20.8, 20.7 (CH3, Ac). MS (APCI) *m/z* (relative intensity) 331 (M - OMe), 303 (M - OAc, 16%).

($^2H_{1-3}$)-Methyl-α-D-mannopyranosides 73a - 77a and ($^2H_{1-3}$)-2,3,4,6-tetra-*O*-acetyl-methyl-α-D-mannopyranosides 73b - 77b





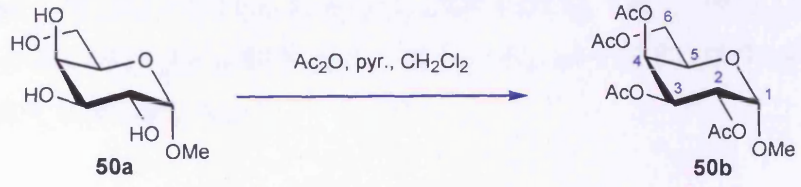
74b, 4-D_1 **75b**, 2,4-D_2 **76b**, 2-D_1 **77b**, 2,3-D_2

Methyl-α-D-mannopyranoside **54a** (200 mg) was refluxed for 0.5 h. with Raney nickel according to general procedure **GP01**. The catalyst was removed by filtration and the reaction was azeotroped with toluene (3 x 10 ml) to give dry product, ($^2H_{1-3}$)-methyl-α-D-mannopyranosides **73a - 77a** (100 mg), which were acetylated using general procedure **GP02** to give ($^2H_{1-3}$)-2,3,4,6-tetra-*O*-acetyl-methyl-α-D-mannopyranosides **73b - 77b** (80 mg).

($^2H_{1-3}$)-Methyl-α-D-mannopyranosides 73a - 77a: δ_H (400, D_2O), 4.60 (H, s, 1-H), 3.75 (H, d, *J* 3.6 Hz, 2-H), 3.72 (H, dd, *J* 12.2, 2.2 Hz, 6a-H), 3.56 (H, not determined, 3-H), 3.56 (H, dd, *J* 12.1, 6.0 Hz, 6b-H), 3.45 (2H, m, 4-H, not determined overlapping with 5-H); δ_C (100, D_2O), 100.1 (1-C), 71.8 (5-C), 70.7 (3-C), 70.2 (2-C), 66.9 (4-C), 61.2 (6-C), 54.9 (CH_3, OMe).

($^2H_{1-3}$)-2,3,4,6-Tetra-*O*-acetyl-methyl-α-D-mannopyranosides 73b - 77b: δ_H (400, $CDCl_3$), 5.32 (1H, s, d, *J* 9.9 Hz, 3-H), 5.26 (1H, d, *J* 10.1 Hz, t, *J* 9.7 Hz, 4-H), 5.20 (1H, dd, *J* 3.5, 1.4 Hz, 2-H), 4.70 (1H, s, 1-H), 4.30 (1H, dd, *J* 12.2, 5.3 Hz, 6a-H), 4.11 (1H, dd, *J* 12.2, 2.3 Hz, 6b-H), 3.94 (1H, m, 5-H), 3.40 (3H, s, OMe), 2.15, 2.11, 2.01, 2.0 (4 x 3H, 4 x s, acetyl); δ_C, 171.2, 170.7, 170.4, 170.3 (C = O), 97.5 (C-1), 67.2 (C-2), 67.1 (C-3), 66.5 (C-5), 65.1 (C-4), 60.8 (C-6), 54.3 (CH_3, OMe), 19.9, 19.7, 19.5 (CH_3, Ac).

2,3,4,6-Tetra-*O*-acetyl-methyl-α-D-galactopyranoside 50b

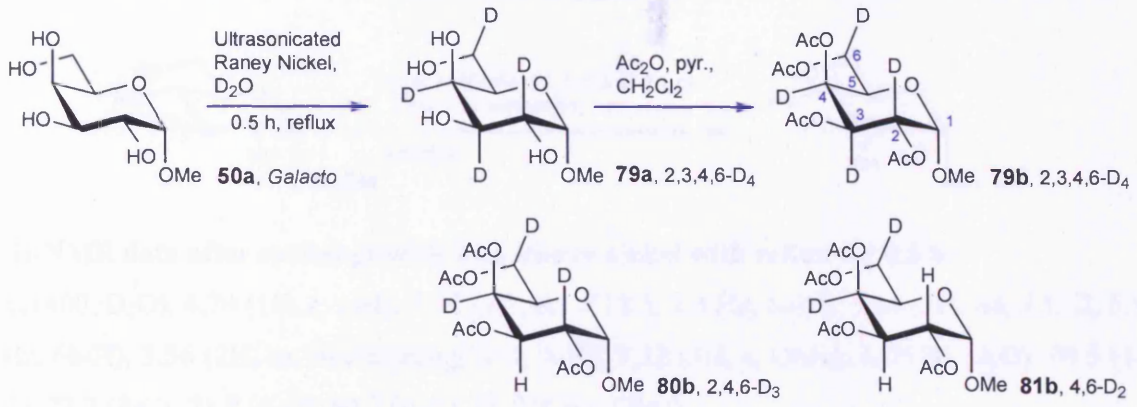


50a **50b**

Methyl-α-D-galactopyranoside **50a** (200 mg, 1.03 mmol) was subjected to general procedure **GP02** (peracetylation), to give the *title compound* **50b** (216 mg, 58%). δ_H (400, $CDCl_3$), 5.47 (1H, dd, *J* 3.2, 0.8 Hz, 4-H), 5.36 (1H, dd, *J* 11.0, 3.4 Hz, 3-H), 5.15 (1H, dd, *J* 10.8, 3.6 Hz, 2-H), 4.99 (1H, d, *J* 3.6 Hz, 1-H), 4.18 (1H,dd, *J* 6.8, 6.0 Hz, 5-H), 4.10 (2H, m, 6a-H, 6b-H), 3.40 (3H, s, OMe), 2.16, 2.08, 2.03, 1.97 (4 x 3H, 4 x s, acetyl); δ_C: 170.8, 170.8, 170.6, 170.3 (C = O), 97.5 (1-C), 68.5 (3-C), 68.4 (2-C), 67.9 (4-C), 66.5 (5-C), 62.2 (6-C), 55.9 (CH_3,

OMe), 21.2, 21.1, 21.0 (CH_3, Ac); MS (APCI) *m/z* (relative intensity) 331 (M - OMe), 332 (H + M - OMe, 10%).

($^2H_{1\text{-}5}$)-Methyl-α-D-galactopyranosides 79a - 81a and ($^2H_{1\text{-}5}$)-2,3,4,6-tetra-*O*-acetyl-methyl-α-D-galactopyranosides 79b – 81b

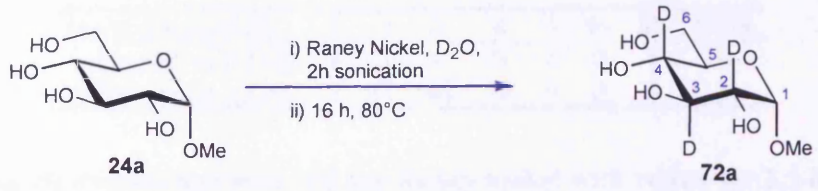


Deuterated and peracetylated galactopyranoside were prepared by general procedures **GP01** to give ($^2H_{1\text{-}5}$)-methyl-α-D-galactopyranosides **79a - 81a** (80 mg), and **GP02** to give ($^2H_{1\text{-}5}$)-2,3,4,6-tetra-*O*-acetyl-methyl-α-D-galactopyranosides **79b - 81b** (75 mg).

($^2H_{1\text{-}5}$)-Methyl-α-D-galactopyranosides 79a - 81a: δ_H (400, D_2O), 4.75 (1H, s, 1-H), 3.81 (1H, s, 4-H), 3.74 (N.D*, not determined), 3.70 (1H, dd, *J* 5.2, 2.2 Hz, 6a-H), 3.70 (N.D), 3.65 (1H, d, *J* 9.4 Hz, 2-H), 3.65 (N.D); δ_C (100, D_2O), 99.2 (1-C), 70.5 (5-C), 69.2 (3-C), 61.1 (6-C), 54.8 (CH_3, OMe).

($^2H_{1\text{-}5}$)-2,3,4,6-Tetra-*O*-acetyl-methyl-α-D-galactopyranoside 79b – 81b: δ_H (400, $CDCl_3$), 5.46 (H, d, *J* 8.7 Hz, 4-H), 5.36 (H, s, d, *J* 10.5 Hz, 3-H), 5.13 (H, dd, *J* 6.6, 3.6 Hz, 2-H), 4.98 (H, s, br s, 1-H), 4.17 (H, d, *J* 5.3 Hz, 5-H), 4.10 (2H, m, 6a-H, 6b-H), 3.40 (3H, s, OMe), 2.15, 2.06, 2.04, 1.96 (4 x 3H, 4 x s, acetyl); δ_C (100, $CDCl_3$), 170.7, 170.7, 170.6, 170.3 (C = O, Ac), 96.1 (1-C), 66.3 (3-C), 65.9 (2-C), 65.0 (4-C), 64.9 (5-C), 60.7 (6-C), 54.5 (CH_3, OMe), 21.0, 20.9, 20.8 (CH_3, Ac).

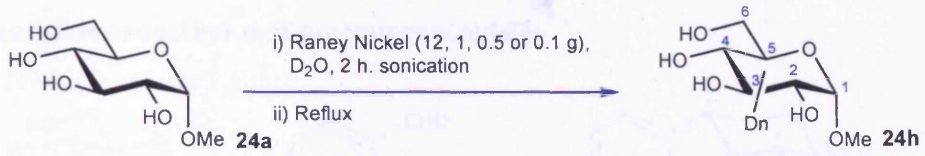
2,3,4-($^2H_3$)-methyl-α-D-glucopyranoside 72a



Methyl-α-D-glucopyranoside **24a** (200 mg, 1.03 mmol) was deuterated according to general procedure **GP01** heated at 80 °C for 16 h. δ_H (400 MHz, D_2O) **72a**: 4.76 (hidden by residual

H_2O, 1H, 1-H), 3.74 (1H, dd, *J* 12.4, 2.2 Hz, 6a-H), 3.63 (1H, dd, *J* 12.3, 5.5 Hz, 6b-H), 3.52 (1H, m, 5-H), 3.27 (3H, s, OMe); δ_C (100 MHz, D_2O), 99.8 (1-C), 72.0 (5-C), 60.9 (6-C), 55.6 (CH_3, OMe).

Deuteration of methyl-α-D-glucopyranoside 24a with different amounts of Raney nickel:



^{1}H-NMR data after exchange with 12 g Raney nickel with reflux for 0.5 h:

δ_H (400, D_2O), 4.70 (1H, s, 1-H), 3.77 (1H, dd, *J* 12.3, 2.3 Hz, 6a-H), 3.64 (1H, dd, *J* 12.2, 5.5 Hz, 6b-H), 3.56 (2H, m, overlapping, 3-H, 5-H), 3.32 (3H, s, OMe); δ_C (100, D_2O), 99.5 (1-C), 72.2 (3-C), 71.8 (5-C), 60.7 (6-C), 55.3 (CH_3, OMe).

Hydrogen number	1	2	3	4	5	6a	6b	OCH_3
% Deuterium	0	100	30	100	0	0	0	0

^{1}H-NMR data after exchange with 1 g Raney nickel with reflux for 0.5 h:

δ_H (400, D_2O), 4.76 (1H, d, *J* 3.6 Hz, 1-H), 3.77 (1H, dd, *J* 12.3, 2.2 Hz, 6a-H), 3.67 (1H, dd, *J* 12.2, 5.5 Hz, 6b-H), 3.57 (1H, t, *J* 9.4 Hz, 3-H), 3.55 (1H, m, 5-H, overlaps with 3-H), 3.47 (1H, dd, *J* 9.7, 3.8 Hz, 2-H), 3.32 (3H, s, OMe), 3.30 (1H, t, *J* 9.7 Hz, third line hidden under OMe, 4-H).

Hydrogen number	1	2	3	4	5	6a	6b	OCH_3
% Deuterium	0	66	25	50	0	0	0	0

^{1}H-NMR data after exchange with 0.5 g Raney nickel with reflux for 0.5 h and 2.5 h:

δ_H (400, D_2O), 4.76 (1H, d, *J* 3.7 Hz, 1-H), 3.79 (1H, dd, *J* 12.3, 2.0 Hz, 6a-H), 3.68 (1H, dd, *J* 12.3, 5.4 Hz, 6b-H), 3.59 (1H, t, *J* 9.3 Hz, 3-H), 3.58 (1H, m, 5-H), 3.48 (1H, dd, *J* 9.8, 3.7 Hz, 2-H), 3.36 (3H, s, OMe), 3.32 (1H, t, *J* 9.4 Hz, 4-H).

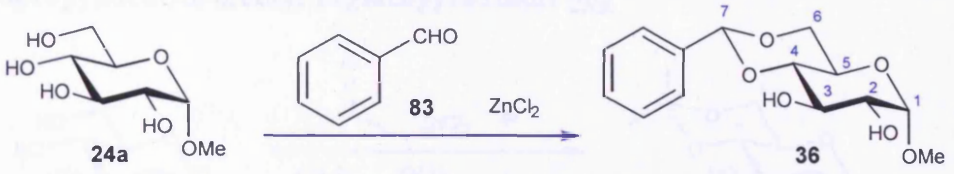
Hydrogen number	1	2	3	4	5	6a	6b	OCH_3
% Deuterium 0.5 hrs	0	29	0	21	0	0	0	0
% Deuterium 2.5 hrs	0	67	40	50	0	0	0	0

^{1}H-NMR data after exchange with 100 mg Raney nickel with reflux for 3.5 h:

δ_H (400, D_2O), 4.76 (1H, d, *J* 3.7 Hz, 1-H), 3.79 (1H, dd, *J* 12.3, 2.0 Hz, 6a-H), 3.68 (1H, dd, *J* 12.3, 5.4 Hz, 6b-H), 3.59 (1H, t, *J* 9.3 Hz, 3-H), 3.58 (1H, m, 5-H), 3.48 (1H, dd, *J* 9.8, 3.7 Hz, 2-H), 3.36 (3H, s, OMe), 3.32 (1H, t, *J* 9.4 Hz, 4-H).

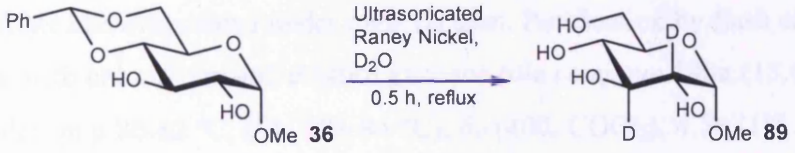
Hydrogen number	1	2	3	4	5	6a	6b	OCH_3
% Deuterium	0	28	0	28	0	0	0	0

4,6-*O*-Benzylidene-methyl-α-D-glucopyranoside 36[75]



Methyl-α-D-glucopyranoside **24a** (30 g, 154.6 mmol), zinc chloride (22.5 g, 165.4 mmol, 1.07 eq.) and benzaldehyde **83** (70 ml, 680 mmol) were stirred for 3 h. The mixture was poured into a mixture of ice:water (1:1, 1L), and the solid was filtered off. The white precipitates were washed with petroleum ether (100 ml) and left to be dried. Recrystallization from hot water gave the *title compound* **36** (22.5 g, 51 %) as a coloreless crystalline solid, m.p 165-166°C, (lit.[75] 163-164°C); δ_H (400 MHz, $CDCl_3$), 7.50 (2H, m, Ph-H), 7.42 (3H, m, Ph-H), 5.55 (1H, s, 7-H), 4.81 (1H, d, *J* 3.8 Hz, 1-H), 4.32 (1H, dd, *J* 9.6, 4.2 Hz, 6a-H), 3.96 (1H, t, *J* 9.3, Hz, 3-H), 3.83 (1H, dd, *J* 9.8, 4.7 Hz, 6b-H), 3.77 (1H, m, 5-H), 3.63 (1H, dd, *J* 8.9, 3.6 Hz 2-H), 3.52 (1H, t, *J* 9.2, Hz, 4-H), 3.48 (3H, s, OMe), 2.95 (1H, br. s, OH), 2.43 (1H, br d, *J* 8.7 Hz, OH). δ_C (DEPT, δ_H ^1H-^{13}C ^1J-COSY), 129.7 (C, Ph-C), 129.0 (CH,7.42, Ph-C), 126.8 (CH, 7.50, Ph-C), 102.2 (CH, 5.55, 7-C), 99.8 (CH, 4.81, 1-C), 81.0 (CH, 3.52, 4-C), 72.3 (CH, 3.63, 2-C), 71.2 (CH, 3.96, 3-C), 68.4 (CH_2, 4.32, 3.83, 6-C), 62.0 (CH, 3.77, 5-C), 55.8 (CH_3, 3.48, OMe-C); MS (APCI) *m/z* 283 (MH^+).

Deuteration of 4,6-*O*-benzylidene-methyl-α- D-glucopyranoside 36

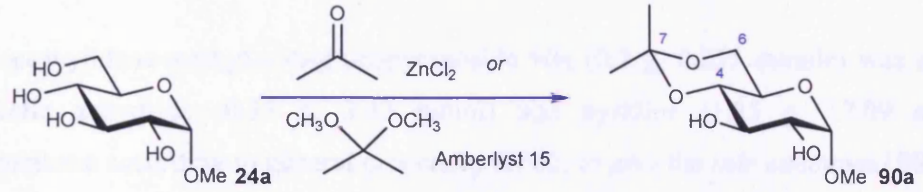


4,6-*O*-benzylidene-methyl-α-D-glucopyranoside **36** (200 mg, 0.708 mmol) was deuterated according to the **GP01** (12 g and 0.5 g Raney nickel) and refluxed for 0.5 h. Ethyl acetate (10 ml) was added to the reaction mixture. The Raney nickel was filtered off, and washed with ethyl acetate and water. The two phases were separated and evaporated. The organic soluble

phase was dried over magnesium sulphate, and evaporated to give a trace of a sugar (10 mg) that could not be identified. The aqueous phase was evaporated under high vacuum to give the compound **89** (106 mg, 76%). ^{1}H NMR spectrum of that compound in D_2O showed circa 75% deuterium incorporation on C-2, C-3, but not at any other center

Hydrogen number	1	2	3	4	5	6a	6b	OCH_3
% Deuterium **89**	0	75	75	0	0	0	0	0

4,6-*O*-Isopropylidene-α-methyl-D-glucopyranoside 90a

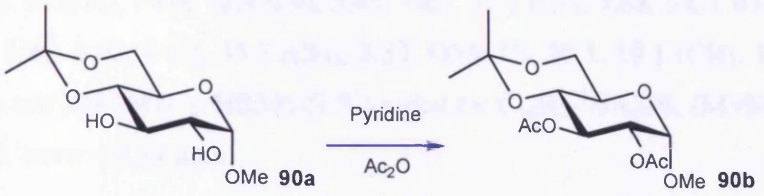


I. Preparation from acetone and zinc chloride[78]**:** Methyl-α-D-glucopyranoside **24a** (10 g, 51.54 mmol) was placed in a stoppered flask. Acetone (200 ml) was added with stirring and zinc chloride (30 g, 220.13 mmol). The mixture was refluxed for 2 days. The cooled solution was poured into a beaker containing a mixture of ice and saturated sodium hydrogen carbonate. The precipitate was collected and the filtrate was extracted with chloroform (3 x 80 ml), evaporated and columned using a solvent gradient of chloroform to ethanol gave the *title compound* **90a** (2 g, 16%) as white powder.

II. Preparation from 2,2-dimethoxypropane[79]**:** DMF (100 ml) was added with stirring to methyl-α-D-glucopyranoside **24a** (20 g, 0.103 mol) and 2,2-dimethoxypropane (31.6 ml, 26.77 g, 0.257 mol). Amberlyst 15 (100 mg) was added and the reaction left to stir in a stoppered flask with TLC monitoring. the reaction mixture was concentrated under reduced pressure and 2,2-dimethoxypropane (31.6 ml) was added again. This was done twice at intervals of three days. The reaction mixture was extracted with chloroform (3 x 80 ml), dried over sodium sulfate and evaporated under high vacuum. Purification by flash chromatography on silica eluting with chloroform and ethanol gave the *title compound* **90a** (15.43 g, 64%) as a coloreless powder. m.p 80-82 °C, (lit.[79] 84-86 °C); δ_H (400, $CDCl_3$), 4.74 (1H, d, *J* 3.9 Hz, 1-H), 3.82 (1H, dd, *J* 10.5, 5.2 Hz, 6a-H), 3.73 (1H, t, *J* 9.1, 3-H), 3.70 (1H, dd, *J* 10.5, 9.9 Hz, 6b-H), 3.62 (1H, m, 5-H), 3.60 (1H, dd, *J* 9.0, 3.9 Hz, 2-H), 3.52 (1H, t, *J* 9.1 Hz, 4-H), 3.36 (3H, s, OMe), 1.60, 1.54 (6H, 2 x s, $C(CH_3)_2$), δ_C (DEPT, δ_H ^{1}H-^{13}C ^{1}*J*-COSY), 99.76 (CH, 4.74, 1-C), 99.74 (C, 7-C), 73.4 (CH, 3.52, 4-C), 73.0 (CH, 3.60, 2-C), 72.1 (CH, 3.73, 3-C),

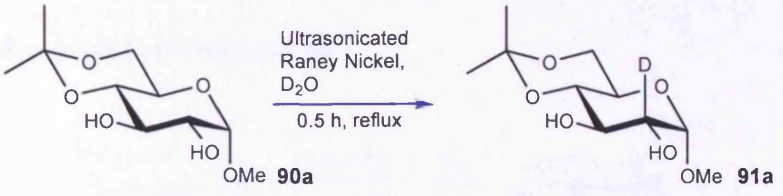
63.2 (CH, 3.62, 5-H), 62.3 (CH_2, 3.82, 3.70, 6-H), 55.5 (CH_3, 3.36, OMe-C), 29.1, 19.1 (CH_3, 1.60, 1.54, CH_3-C); MS (APCI) *m/z* 235 (MH^+).

4,6-*O*-Isopropylidene-2,3-di-*O*-acetyl-methyl-α-D-glucopyranoside 90b



4,6-*O*-Isopropylidene-methyl-α-D-glucopyranoside **90a** (0.2 g, 0.855 mmole) was acetylated using acetic anhydride (0.35 g, 3.42 mmol) and pyridine (1.35 g, 17.09 mmol) in dichloromethane according to general procedure **GP02**, to give the *title compound* **90b** in 89% yield as oil after purification by column chromatography using a slow gradient of chloroform : ethanol; R_f 0.5 in chloroform : ethanol; 90:10; IR (film), 2995, 2942, 1751, 1442, 1371, 1331, 1241, 1143, 1058, 943, 912, 851, 754, 649, 602 cm^{-1}; δ_H (400 MHz, C_6D_6), 5.78 (1H, t, *J* 9.4 Hz, 3-H), 4.97 (1H, dd, *J* 9.9, 3.6 Hz, 2-H), 4.71 (1H, d, *J* 3.6 Hz, 1-H), 3.68 (1H, m, 5-H), 3.63 (1H, dd, *J* 10.4, 6.0 Hz, 6a-H), 3.53 (1H, t, *J* 9.6 Hz, 4-H), 3.48 (1H, t, *J* 10.1 Hz, 6b-H), 2.80 (3H, s, OMe), 1.59, 1.53 (2 x 3H, 2 x s, acetyl), 1.27, 1.08 (2 x 3H, 2 x s, 8-H's, 9-H's). δ_C (DEPT, δ_H ^1H-^{13}C ^1J-COSY), 169.9, 169.2 (C = O, Ac), 99.7 (C, 7-C), 97.8 (CH, 4.71, 1-C), 72.3 (CH, 3.53, 4-C), 72.1 (CH, 4.97, 2-C), 69.5 (CH, 5.78, 3-C), 63.6 (CH, 3.68, 5-C), 62.2 (CH_2, 3.63, 3.48, 6-C), 54.5 (CH_3, 2.80, OMe-C), 29.1 (CH_3, 1.27, 8-C or 9-C), 20.4, 20.1 (CH_3, 1.59, 1.53, OAc-C), 18.8 (CH_3, 1.08, 8-C or 9-C); MS (APCI) *m/z* (relative intensity) 287 (M - OMe), 319 (M + H, 41%), 261 (M + H – OAc, 28%).

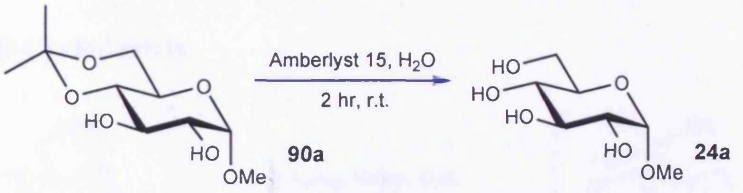
2-[$^2H_1$]-4,6-*O*-Isopropylidene-methyl-α-D-glucopyranoside 91a



Raney nickel (0.9 g, wet) was washed with deuterium oxide (2 x 3 ml; 2.5ml, 138 mmol) and sonicated according to **GP01**. 4,6-*O*-Isopropylidene-methyl-α-D-glucopyranoside **90a** (200 mg, 0.855 mmol) was added and the reaction refluxed for 1.5 hr, left to cool and filtered to give the *title compound* **91a** (110 mg, 55%) as a powder, mp 81-83 °C: $[\alpha]_D^{23}$ +110.3 (*c* 0.08,

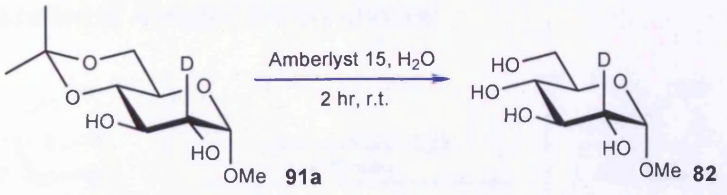
$CHCl_3$); IR (CH_2Cl_2), 3430, 2357, 1646, 1379, 1270, 1199, 1170, 1108, 1064, 1029, 857 cm^{-1}; δ_H (400 MHz, $CDCl_3$), 4.69 (1H, s, 1-H), 3.82 (1H, dd, *J* 10.5, 5.2 Hz, 6_a-H), 3.68 (2H, t, *J* 10.4 Hz,overlap 3-H, 6_b-H), 3.55 (1H, m, 5-H), 3.45 (1H, t, *J* 9.3 Hz, 4-H), 3.37 (3H, s, OMe), 2.62, 2.18 (2 x OH, s), 1.50, 1.38 (6H, $C(CH_3)_2$). δ_C (DEPT, δ_H ^1H-^{13}C ^1J-COSY), 99.7 (CH, 4.69, 1-C), 99.6 (C, 7-C), 73.4 (CH, 3.45, 4-C), 72.2 (CH, 3.68, 3-C), 63.3 (CH, 3.55, 5-C), 62.3 (CH_2, 3.82, 3.68, 6-C), 55.5 (CH_3, 3.37, OMe-C), 29.1, 19.1 (CH_3, 1.50, 1.38, CH_3-C); MS (APCI) *m/z* 236 (MH^+); HRMS (ES^+) *calcd* for $C_{10}H_{21}{}^2H_1O_6N_1$ ($M+NH_4^+$), 253.1504, found 253.1505, error – 0.39 ppm.

Methyl-α-D-glucopyranoside 24a[81]



Amberlyst 15 (10 mg) was added to 4,6-*O*-isopropylidene-methyl-α-D-glucopyranoside **90a** (100 mg, 0.427 mmol) in water (2 ml), and the reaction left to stir in a stoppered flask for 2 h. at r.t. The Amberlyst was filtered off, and the filtrate was evaporated under reduced pressure to gave the *title compound* **24a** (80 mg, 96%) as white crystalline solid. R_f 0.4 in chloroform:methanol; 60:40; δ_H (400 MHz, D_2O), 4.63 (1H, d, *J* 2.6 Hz, 1-H), 3.65 (1H, d, *J* 12.1 Hz, 6a-H), 3.54 (1H, dd, *J* 12.1, 5.0 Hz, 6b-H), 3.41 (2H, t, *J* 9.4, 3-H, 5-H overlaps with 3-H), 3.33 (1H, dd, *J* 9.6, 2.9 Hz, 2-H), 3.20 (3H, s, OMe), 3.17 (1H, t, 9.7 Hz, 4-H, third line hidden under OMe). δ_C (DEPT, δ_H ^1H-^{13}C ^1J-COSY), 99.1 (CH, 4.63, 1-C), 72.9 (CH, 3.41, 3-C), 71.4 (CH, 3.33, 2-C), 71.1 (CH, 3.41, 5-C), 69.4 (CH, 3.17, 4-C), 60.4 (CH_2, 3.65, 3.54, 6-C), 54.8 (CH_3, 3.20, OMe-C).

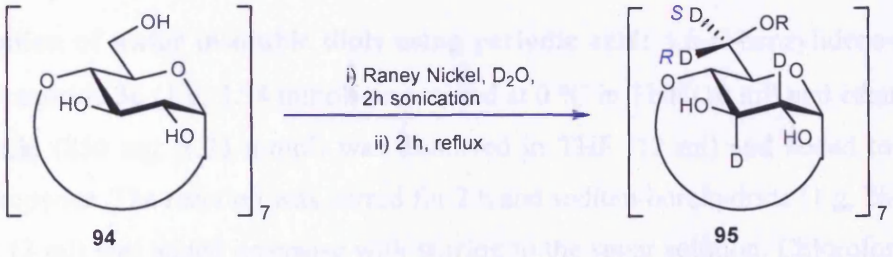
2-($^2H_1$)-Methyl-α-D-glucopyranoside 82



Amberlyst 15 (5 mg) was added to 2-[$^2H_1$]-4,6-*O*-Isopropylidene-methyl-α-D-glucopyranoside **91a** (50 mg, 0.212 mmol) in water (1 ml), and the reaction left to stir in a stoppered

flask for 2.5 h. at r.t. The Amberlyst was filtered off, and the filtrate was evaporated under reduced pressure to gave the *title compound* **82** (38 mg, 90%) as white crystalline solid, m.p 155-158°C; R_f 0.4 in chloroform:methanol; 60:40; $[\alpha]_D^{23}$ + 22 (*c* 1.0, H_2O); IR (film), 3385, 2926, 2358, 1644, 1366, 1191, 1094, 1032, 854 cm^{-1}; δ_H (400 MHz, D_2O), 4.73 (1H, s, 1-H), 3.82 (1H, dd, *J* 12.3, 2.2 Hz, 6a-H), 3.68 (1H, dd, *J* 12.3, 5.5 Hz, 6b-H), 3.60 (1H, d, *J* 9.2 Hz, 3-H), 3.57 (1H, m, 5-H), 3.37 (3H, s, OMe), 3.34 (1H, t, *J* 9.5 Hz, 4-H, third line hidden under OMe). δ_C (DEPT, δ_H ^1H-^{13}C ^1J-COSY), 99.3 (CH, 4.73, 1-C), 73.0 (CH, 3.60, 3-C), 71.6 (CH, 3.57, 5-C), 69.6 (CH, 3.34, 4-C), 60.6 (CH_2, 3.82, 3.68, 6-C), 55.0 (CH_3, 3.37, OMe); MS (ES^+) *m/z* 196 (MH^+); HRMS (ES^+) *calcd* for $C_7H_{17}{}^2H_1O_6N_1$ ($M+NH_4^+$), 213.1191, found 213.1191, error 0.0 ppm.

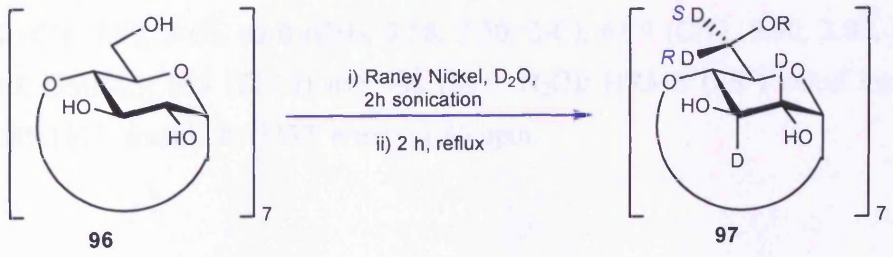
Deuteration of β-Cyclodextrin



94 **95**

Raney nickel (8 g, wet) was washed with deuterium oxide (2 x 5 ml; 5ml, 276 mmol) and sonicated according to **GP01**. β-cyclodextrin **94** (200 mg, 0.173 mmol) was placed in a flask, deuterium oxide (3 ml) was added and dried under reduced pressure and then added to the Raney nickel. The reaction mixture refluxed for 1.45 hr, left to cool and filtered to give the deuterated compound **95** (125 mg) as a white powder. δ_H (500 MHz, D_2O), 4.98 (1H, s, 1-H), 3.88 (1H, d, *J* 9.03 Hz, 3-H), 3.75 (2 or 3 H, d, *J* 11.5 Hz, 5, 6, 6′-H, overlapping), 3.56 (1H, t, *J* 9.5 Hz, d, *J* 9.8 Hz, 4-H). δ_C (DEPT, δ_H ^1H-^{13}C ^1J-COSY), 101.8 (CH, 4.98, 1-C), 81.1 (CH, 3.56, 4-C), 73.0 (CH, 3.88, 3-C), 71.7 (CH, 3.75, 5-C), 60.3 (CH_2, 3.75, 6-C).

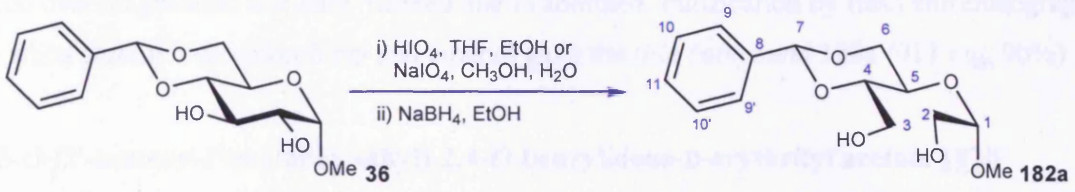
Attempted deuteration of silylated β-Cyclodextrin



96 **97**

Raney nickel (8 g, wet) was washed with deuterium oxide (2 x 5 ml; 5ml, 276 mmol) and sonicated according to **GP01**. Heptakis (6-*O*-*tert*-butyldimethylsilyl)β-cyclodextrin **96** (200 mg, 0.1 mmol) was placed in a flask, chloroform and deuterium oxide (3 + 2 ml) was added and dried under reduced pressure and then added to the Raney nickel. The reaction mixture was refluxed for 2 hr, left to cool and filtered and evaporated to give a white powder (110 mg, 55 %). The ^{1}H-NMR spectrum was identical to starting material.

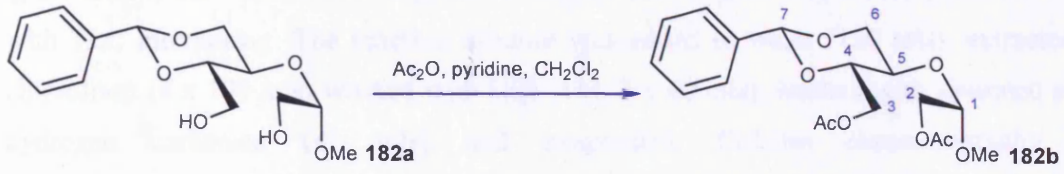
3-*O*-(2′-Hydroxy-1′-methoxy-ethyl)-2,4-*O*-benzylidene-D-erythritol 182a



I. Preparation of water insoluble diols using periodic acid: 4,6-*O*-benzylidene-methyl-α-D-glucopyranoside **36** (1 g, 3.54 mmol) was stirred at 0 °C in THF (10 ml) and ethanol (5 ml). Periodic acid (850 mg, 3.73 mmol) was dissolved in THF (12 ml) and added to the sugar solution dropwise. The reaction was stirred for 2 h and sodium borohydride (1 g, 26.45 mmol) in ethanol (3 ml) was added dropwise with stirring to the sugar solution. Chloroform (50 ml) and water (100 ml) were added to separate the phases. The chloroform extract was dried over sodium sulphate, filtered and evaporated. Purification by flash chromatography on silica eluting with chloroform and ethanol gave the *title compound* **182a** (804 mg, 80 %) as an oil. R_f 0.32 in chloroform:methanol; 90:10; $[\alpha]_D^{23}$ - 63 (*c* 1.5, $CHCl_3$); IR (film), cm^{-1} 3386, 1643, 1452, 1392, 1215, 1068, 752; δ_H (400, $CDCl_3$), 7.40, (2H, 2 x d, *J* 3.7 Hz, 9, 9′-H), 7.30 (3H, m, 10, 10', 11-H), 5.47 (1H, s, 7-H), 4.53(1H, dd, *J* 6.1, 4.8 Hz, 1-H), 4.35 (1H, dd, *J* 10.6, 5.3 Hz, 3_{eq}-H), 3.98 (1H, m, 5-H), 3.90 (1H, dd, *J* 12.4, 3.6 Hz, 6a-H), 3.86 (1H, dd, *J* 12.4, 2.6 Hz, 6b-H), 3.70 (1H, m, 4-H), 3.63 (1H, t, *J* 10.5 Hz, 3_{ax}-H), 3.58 (1H, dd, *J* 12.4, 6.1 Hz, 2_{ax}-H), 3.50 (1H, dd, *J* 12.2, 4.7 Hz, 2_{eq}-H), 3.40 (3H, s, OMe-H). δ_C (DEPT, δ_H ^{1}H-^{13}C ^{1}*J*-COSY), 137.3 (C, 8-C), 129.2 (CH, 7.30, 11-C), 128.4 (CH, 7.30, 10-C), 126.1 (CH, 7.40, 9-H), 103.2 (CH, 5.47, 7-C), 101.2 (CH, 4.53, 1-C), 80.4 (CH, 3.70, 4-C), 69.9 (CH_2, 4.35, 3.63, 3-C), 64.7 (CH, 3.98, 5-C), 62.0 (CH_2, 3.58, 3.50, 2-C), 61.9 (CH2, 3.90, 3.86, 6-C), 55.9 (CH3, 3.40, OMe-C); MS (EI/CI) *m/z* 302 (M + H_2O); HRMS (ES^+) *calcd* for $C_{14}H_{20}O_6$ ($M+H^+$), 285.1333, found 285.1337, error – 1.40 ppm.

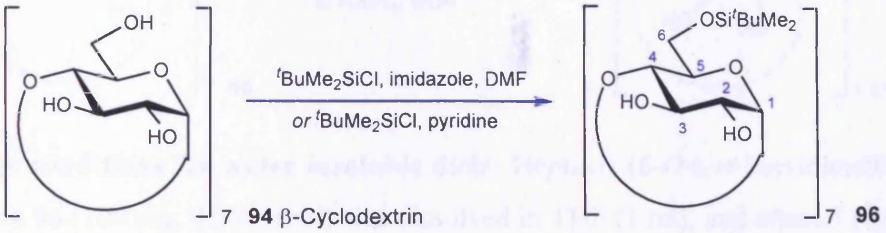
II. Preparation of water soluble diols using sodium periodate 4,6-*O*-benzylidene-methyl-α-D-glucopyranoside **36** (1 g, 3.54 mmol) was dissolved in methanol (12 ml) with magnetic stirring and warming in a stoppered flask (100 ml). Sodium periodate (1.5 g, 1.844 mmol) was dissolved in water (5 ml) with warming. The periodate solution was added to the sugar solution dropwise. A precipitate formed which did not dissolve on warming or addition of methanol (5 ml). Sodium hydroxide (8 M, 2 ml) was added, and the reaction left to stir for 2 h. Sodium borohydride (1 g, 26.45 mmol) was added slowly and the reaction stirred for 30 minutes. Water (50 ml) was added and the mixture was extracted with chloroform (3 x 30 ml) dried over magnesium sulphate, filtered and evaporated. Purification by flash chromatography on silica eluting with chloroform and ethanol gave the *title compound* **182a** (911 mg, 90%)

1-3-*O*-(2′-acetoxy-1′-methoxy-ethyl)-2,4-*O*-benzylidene-D-erythrityl acetate 182b



A mixture of pyridine (1.01 ml, 13.6 mmol, 20 equiv.) in dichloromethane (3 ml) and acetic anhydride (0.3 ml, 2.72 mmol, 4 equiv.) were placed in the flask in an ice bath. 3-*O*-(2′-hydroxy-1′-methoxy-ethyl)-2,4-*O*-benzylidene-D-erythritol **182a** (194 mg, 0.68 mmol) was added to the flask and acetylation was carried out using the general procedure **GP02**. Purification by flash chromatography on silica eluting with chloroform and methanol gave the *title compound* **182b** (144 mg, 57%) as a yellow oil. R_f 0.42 in chloroform:methanol; 95:05; $[\alpha]_D^{23}$ – 48.6 (*c* 1.0, $CHCl_3$); IR (film), cm^{-1} 3488, 2966, 2915, 2855, 1740, 1599, 1448, 1373, 1318, 1258, 1232, 1067, 1022, 981, 916, 791, 760, 710; δ_H (400, $CDCl_3$), 7.47, 7.36 (5H, 2 x d, *J* 1.4 Hz, Ph-H, overlapping), 5.52 (1H, s, 7-H), 4.72 (1H, dd, *J* 5.7, 5.2 Hz, 1-H), 4.53 (1H, dd, *J* 12.2, 0.8 Hz, 3a-H), 4.43 (1H, dd, *J* 10.9, 4.3 Hz, 6a-H), 4.29 (1H, dd, *J* 12.1, 4.7 Hz, 3b-H), 4.13 (1H, dd, *J* 11.7, 5.0, H-2a), 4.03 (1H, dd, *J* 11.8, 5.9 Hz, 2b-H), 3.91 (2H, m, 5-H overlapping 4-H, dd, *J* 9.5, 4.7 Hz), 3.67 (1H, m, 6b-H), 3.42 (3H, s, OMe), 2.12, 2.09 (2 x 3H, 2 x s, acetyl). δ_C (DEPT, δ_H ^1H-^{13}C ^1J-COSY), 170.0 (C, C=O), 130.2 (C, Ph-C), 128.1, 127.2 (2 x CH, 7.47, 7.36 , Ph-C), 101.8 (CH, 5.52, 7-C), 100.0 (CH, 4.72, 1-C), 98.8 (CH, 3.91, 4-C), 69.0 (CH_2, 4.43, 3.67, 6-C), 65.8 (CH, 3.91, 5-C), 63.4 (2 x CH_2, 4.53, 4.29, 4.13, 4.03, 2, 3-C), 55.2 (CH_3, 3.42, OMe-C), 21.1 (CH_3, 2.12, 2.09, Ac-C); MS (APCI) *m/z* 337 (M - OMe); HRMS (ES^+) *calcd* for $C_{18}H_{24}O_8$ ($M+NH_4^+$), 386.1809, found 386.1813, error – 1.03 ppm.

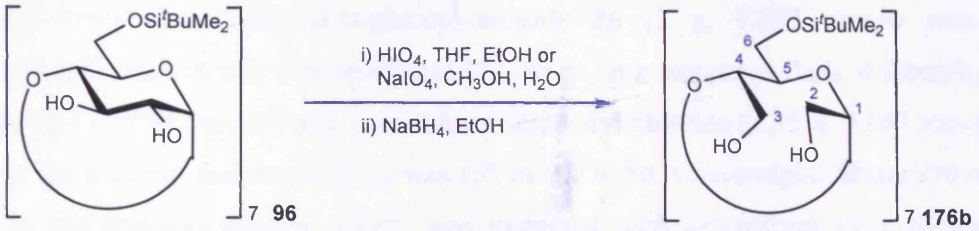
Heptakis(6-*O*-*tert*-butyldimethylsilyl)-β-cyclodextrin 96



I. Preparation by treatment of β-cyclodextrin 94 with imidazole and *tert*-butyldimethylsilyl chloride in *N,N*-dimethylformamide: A solution of *tert*-butyldimethylsilyl chloride (3.1 g, 21 mmol) in anhydrous *N,N*-dimethylformamide (25 ml) was added dropwise with stirring over 30 min at r.t. to a mixture of dried β-cyclodextrin **94** (3 g, 2.6 mmol) and imidazole (2.9 g, 0.043 mmol). The mixture was stirred at r.t. for 15 days with TLC monitoring. The reaction mixture was added to water (150 mls), extracted with chloroform (4 x 100 mls) washed with HCl (1M, 2 x 60 mls), washed with saturated sodium hydrogen carbonate (40 mls) and evaporated. Column chromatography using (chloroform:ethanol, 10:0 to 8:2) gave the *title compound* **96** (4.66 g, 91%) as white solid. R_f 0.3 in chloroform:ethanol; 85:15. δ_H (400, $CDCl_3$), 6.73 (OH, s, broad), 5.22 (OH, s, broad), 4.84 (1H, d, *J* 3.3 Hz, 1-H), 4.00 (1H, t, *J* 9.1 Hz, 4-H), 3.84 (1H, d broad, 6a-H), 3.66 (1H, m, 5-H), 3.60 (1H, d, *J* 3.4 Hz, 6b-H), 3.56 (1H, m, 2-H), 3.52 (1H, m, 3-H), 0.83 (9H, $C(CH_3)_3$), 0.00 (6H, $Si(CH_3)_2$). δ_C (DEPT, δ_H ^1H-^{13}C ^1J-COSY), 102.0 (CH, 4.84, 1-C), 81.7 (CH, 4.00, 4-C), 73.6 (CH, 3.56, 2-C), 73.4 (CH, 3.52, 3-C), 72.5 (CH, 3.66, 5-C), 61.6 (CH_2, 3.84, 3.60, 6-C), 25.9 (CH_3, 0.83, $C(CH_3)_3$), 18.3 (C, $C(CH_3)_3$), -5.1, - 5.2 (CH3, $Si(CH_3)_2$).

II. Preparation from *tert*-butyldimethylsilyl chloride in dry pyridine: A solution of β-cyclodextrin **94** (1 g, 0.88 mmol) in dry pyridine (15 ml) was added to a solution of *tert*-butyldimethylsilyl chloride (1 g, 6.63 mmol) in dry pyridine (6.5 ml) at 0°C, and stirred at r.t. for 20 hrs. Water was added (100 ml), then extracted with chloroform (4 x 50 mls), the organic phase was washed with HCl (1M, 2 x 20 mls), washed with saturated sodium hydrogen carbonate (20 mls) and evaporated. Column chromatography (chloroform:ethanol, 10:0 to 8:2) gave the *title compound* **96** (1.18 g, 70%) as white solid.

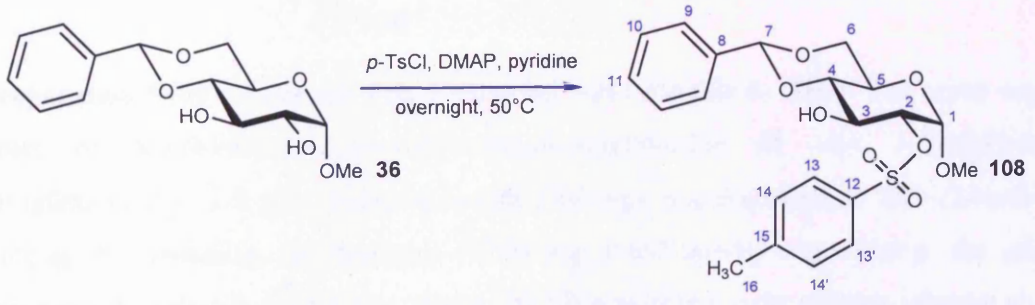
Attempted periodate cleavage of heptakis(6-*O*-*tert*-butyldimethylsilyl)-β-cyclodextrin 176b



I. Cleavage conditions for water insoluble diols: Heptakis (6-*O*-*tert*-butyldimethylsilyl)-β-cyclodextrin **96** (100 mg, 0.05 mmol) was dissolved in THF (1 ml), and ethanol (0.5 ml) with stirring at 0° C. Periodic acid (85.5 mg, 0.37 mmol) in THF (1 ml) was added to the solution mixture dropwise. The reaction mixture was left to stir for 1 hour. Ethylene glycol was added to decompose excess periodic acid and stirred for 20 min. Sodium borohydride (93.76 mg, 2.478 mmol) in ethanol (3 ml) was added dropwise with stirring to the solution. Chloroform (50 ml) and water (100 ml) were added and the aqueous layer extracted with chloroform, (3 x 30 ml). The chloroform extracts were combined, dried over magnesium sulphate, filtered and evaporated to give a black oil (127 mg).

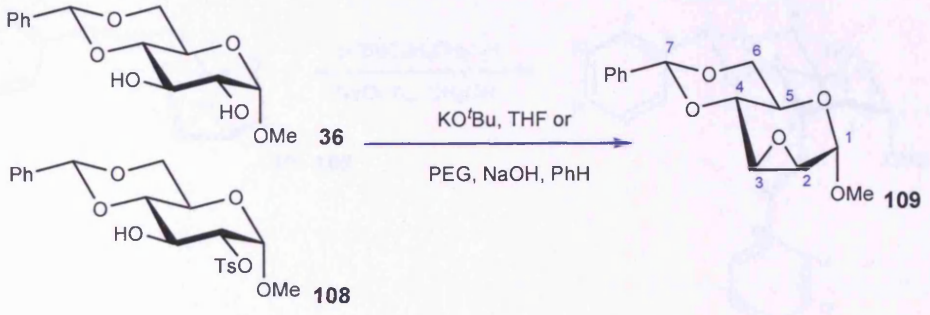
II. Cleavage conditions for water soluble diols: Heptakis (6-*O*-*tert*-butyldimethylsilyl)β-cyclodextrin **96** (100 mg, 0.05 mmol) was dissolved in methanol (2.5 ml) with stirring and warming in a stoppered flask (100 ml). Sodium periodate (148.2 mg, 0.693 mmol) was dissolved in water (0.5 ml) with warming. The periodate solution was added to the sugar solution dropwise. A white precipitate formed which did not dissolve on warming or addition of methanol (1 ml). Sodium hydroxide (8 M, 1 ml) was added, and the reaction left to stir for 2h. Sodium borohydride (99.14 mg, 2.62 mmol) in ethanol (3 ml) was added slowly and the reaction stirred for 30 minutes. Water (50 ml) was added and the mixture was extracted with chloroform (3 x 50 ml), dried over magnesium sulphate, filtered and evaporated to give a yellow powder (87 mg). The ^{1}H-NMR spectrum was identical to starting material.

2-*O*-*p*-Toluenesulfonyl-4,6-*O*-benzylidene-methyl-α-D-glucopyranoside 108



To 4,6-*O*-benzylidene-methyl-α-D-glucopyranoside **36** (2 g, 7.087 mmol) was added anhydrous pyridine (10 ml) with stirring at 0° C to r.t. in a stoppered flask. 4-Dimethylamin-pyridine (1.3 g, 7.087 mmol) and *para*-toluenesulphonyl chloride (1.35 g, 7.087 mmol) were added to the solution and the mixture was left to stir at 50 °C overnight. Water (70 ml) was added to the reaction mixture which was extracted with chloroform (3 x 60 ml). The combined organic extracts were washed with hydrochloric acid (2M, 2 x 50 ml), saturated sodium hydrogen carbonate (60 ml) and copper sulphate (60 ml), dried over Na_2SO_4 and evaporated to give a mixture of 2-*O*-tosylate **108** and starting material **36** as an oil (1.6 g). Purification of a portion (100 mg) by flash chromatography on silica using chloroform to ethanol gradient gave the *title compound* (50 mg, 46 %) as yellow pale solid. R_f 0.3 in chloroform:ethanol; 97:3; δ_H (400, $CDCl_3$), 7.77 (2H, d, *J* 8.3 Hz, 13, 13'-H), 7.38 (2H, d, *J* 3.65 Hz, 9, 9'-H) 7.29 (5H, overlapping; 14, 14'-H, 10, 10'-H, 11-H), 5.42 (1H, s, 7-H), 4.76 (1H, d, *J* 3.7 Hz, 1-H), 4.30 (1H, dd, *J* 9.37, 3.7 Hz, 2-H), 4.19 (1H, dd, *J* 10.1, 4.7 Hz, 6a-H), 4.08 (1H, t, *J* 9.33 Hz, 3-H), 3.75 (1H, m, 5-H), 3.64 (1H, t, *J* 10.2 Hz, 6b-H), 3.39 (1H, t, *J* 9.4 Hz, 4-H), 3.28 (3H, s, OMe), 2.37 (3H, s,16-H). δ_C (DEPT, δ_H ^1H-^{13}C ^1J-COSY), 145.2 (C, 15-C), 136.8 (C, 8-C), 133.2 (C, 12-C), 129.8 (CH, 7.29, 14-C), 129.3 (CH, 7.29, 11-C), 128.3 (CH, 7.29, 10-C), 128.1 (CH,7.77, 13-C), 126.2 (CH, 7.38, 9-C), 101.9 (CH, 5.42, 7-C), 98.2 (CH, 4.76, 1-C), 80.9 (CH, 3.39, 4-H), 79.5 (CH, 4.30, 2-C), 68.7 (CH_2, 4.19, 3.64, 6-C), 68.3 (CH, 4.08, 3-C), 61.9 (CH, 3.75, 5-C), 55.7 (CH_3, 3.28, C-OMe), 21.7 (CH3, 2.37, 16-C);

2,3-Anhydro-4,6-*O*-benzylidene-α-methyl-D-mannopyranoside 109

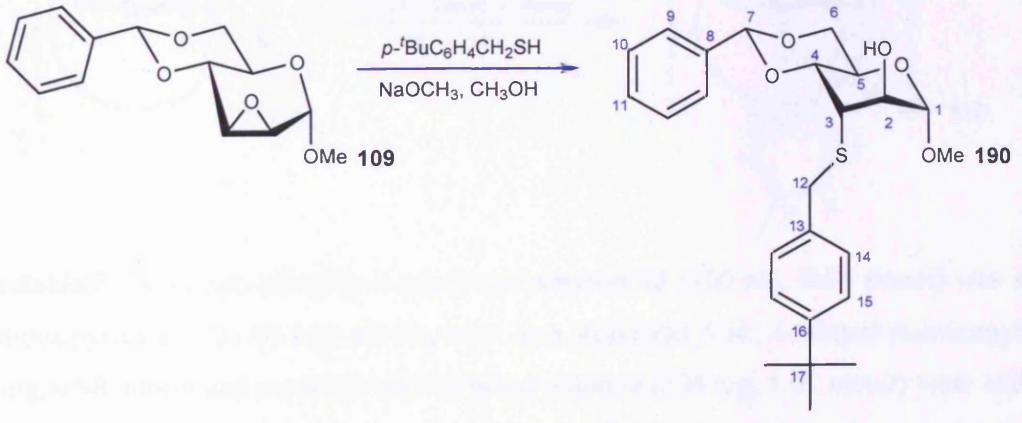


I. Preparation from treatment with potassium *tert*-butoxide in THF: The crude reaction mixture of 4,6-*O*-benzylidene-methyl-α-D-glucopyranoside **36** and 2-*O*-tosyl-4,6-*O*-benzylidene-methyl-α-D-glucopyranoside **108** (300 mg) was dissolved in THF (24 ml) with stirring at 0°C. Potassium *tert*-butoxide (77.08 mg, 0.687 mmol) was added to the solution which was left to stir for 4 days at rt. Water (20 ml) was added to the reaction mixture washed

with hydrochloric acid (2 M, 10 ml), extracted with ethyl acetate (3 x 10 ml), dried over magnesium sulphate and evaporated. Purification by flash chromatography on silica eluting with chloroform and ethyl acetate gave the *title compound* **109** (7 mg, 4%) as a powder. R_f 0.5 in chloroform; IR (CH_2Cl_2), cm^{-1} 3026, 2925, 1452, 1376, 1240, 1124, 1084, 1009, 968, 956, 920, 849, 814, 746, 692; δ_H (400, $CDCl_3$), 7.43 (2H, d, *J* 1.84 Hz, Ph-H), 7.30 (3H, m, Ph-H, overlapping) 5.39 (1H, s, 7-H), 4.81 (1H, s, 1-H), 4.23 (1H, dd, *J* 10.3, 4.6 Hz, 6a-H), 3.93 (1H, m, 5-H), 3.74 (1H, d, *J* 9.6 Hz, 4-H), 3.60 (1H, t, *J* 10.3 Hz, 6b-H), 3.44 (1H, d, *J* 3.5 Hz, 3-H), 3.19 (3H, s, OMe), 3.05 (1H, d, *J* 3.6 Hz, 2-H); δ_C (DEPT, δ_H ^1H-^{13}C ^1J-COSY), 129.3 (C, Ph), 128.4, 126.2 (CH, Ph), 102.5(CH, 5.39, 7-C), 96.9 (CH, 4.81, 1-C), 74.9 (CH, 3.74, 4-C), 69.4 (CH_2, 4.23, 3.60, 6-C), 61.7 (CH, 3,93, 5-C), 55.8 (CH_3, OMe-C), 53.9 (CH, 3.44, 3-C), 50.6 (CH, 3.05, 2-C); MS (APCI) *m/z* 265 (MH^+).

II. Preparation from treatment with polyethylene glycol and sodium hydroxide in benzene: To a solution of polyethylene glycol (26 g) and benzene (4 ml) was added the crude reaction mixture of 4,6-*O*-benzylidene-methyl-α-D-glucopyranoside **36** and 2-*O*-tosyl-4,6-*O*-benzylidene-methyl-α-D-glucopyranoside **108** (500 mg). Saturated sodium hydroxide (0.5 ml) was added to the mixture and refluxed for 18 hr. Water (30 ml) was added to the reaction mixture, washed with hydrochloric acid (1 M, 10 ml), extracted with chloroform (3 x 20 ml), dried over magnesium sulphate and evaporated. Purification by flash chromatography on silica eluting with chloroform and ethyl acetate gave the *title compound* **109** (200 mg, 75%)

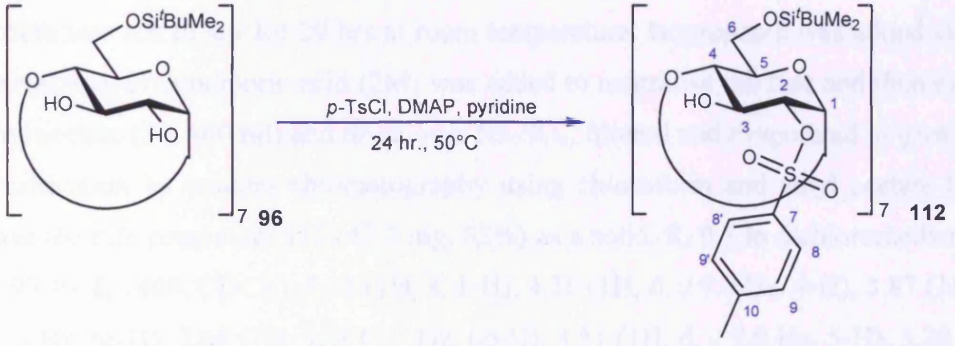
3-(4′-*tert*-Butylbenzylthiol)-4,6-*O*-benzylidene-methyl-α-D-altropyranoside 190



A solution of 4-*tert*-butylbenzyl thiol (0.085 ml, 0.454 mmol) and sodium methoxide (0.022 g, 0.416 mmol) in methanol (5 ml) was added dropwise to the epoxide **109** (100 mg, 0.378 mmol) and left to stir at r.t. for 1 day and then refluxed for 2 d. Aqueous sodium hydrogen

carbonate (3 ml) and dichloromethane (10 ml) was added and the organic phase extracted with dichloromethane (3 x 20 ml), washed with water (10 ml), dried over anhydrous sodium sulphate and evaporated. Purification of the crude reaction mixture using a slow gradient of petroleum ether and ethyl acetate 80:0 to 50:30 gave the *title compound* **190** (31 mg, 30%) as a white powder; R_f 0.4 in petroleum ether and ethyl acetate, 70:30; mp 175-177 °C; $[\alpha]_D^{23}$ -86.6 (*c* 1.0, $CHCl_3$); IR (CH_2Cl_2), cm^{-1} 3420, 2963, 1640, 1456, 1364, 1217, 1108, 1032, 754, 698; δ_H (400 MHz, C_6D_6) 7.80(2H, 2 x d, *J* 1.7 Hz, 9, 9'-H), 7.45 (3H, d, *J* 7.3 Hz, overlapping, 11, 14, 14'-H), 7.32 (2H, m, 10, 10'-H), 7.20 (2H, d, *J* 8.3 Hz, 15, 15'-H), 5.54 (1H, s, 7-H), 4.77 (1H, m, 5-H), 4.43 (1H, dd, *J* 9.3, 4.0 Hz, 4-H), 4.39 (1H, s, 1-H), 4.35 (1H, dd, *J* 10.3, 5.2 Hz, 6a-H), 4.17 (1H, d, *J* 12.8 Hz, 12a-H), 3.95 (2H, dd, *J* 12.9, 4.42 Hz, overlapping of 2-H, 12b-H), 3.80 (1H, t, *J* 10.3 Hz, 6b-H), 3.40 (1H, dd, *J* 3.3, 2.4 Hz, 3-H), 3.13 (3H, s, OMe), 1.28 (9H, $C(CH_3)_3$). δ_C (DEPT, δ_H ^1H-^{13}C ^1J-COSY), 137.5 (C, 13 or 16-C), 134.8 (C, 13 or 16-C), 131.7 (C, 8-C), 129.4 (C, 11-C), 128.9 (CH, 7.20, 15-C), 128.1 (CH, 7.45, 14-C), 125.6 (CH, 7.80, 9-C), 125.3 (CH, 7.32, 10-C), 102.2 (C, 5.54, 7-C), 100.9 (CH, 4.39, 1-C), 76.7 (CH, 4.43, 4-C), 71.6 (CH, 3.95, 2-C), 67.9 (CH_2, 4.35, 3.80, 6-C), 58.7 (CH, 4.77, 5-C), 53.0 (CH_3, 3.13, OMe-C), 44.9 (CH, 3.40, 3-C), 36.9 (CH_2, 4.17, 3.95, 12-C), 33.0 (CH_3, 1.28, $(CH_3)_3$), 27.6 (C, 17-C); MS (APCI) *m/z* 445 (MH^+); HRMS (ES^+) *calcd* for $C_{25}H_{32}O_5S_1$ ($M + H^+$) 445.2043, found 445.2038, error 1.12 ppm.

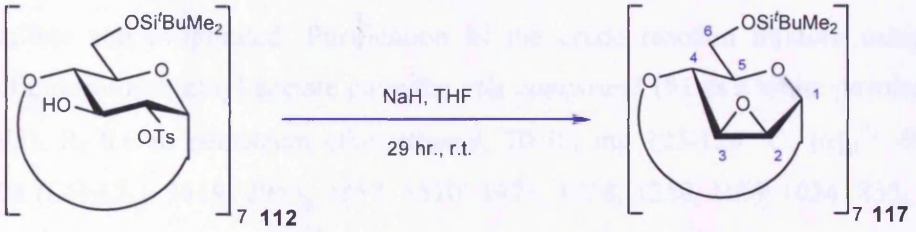
Heptakis (2-*O*-*p*-toluenesulfonyl-6-*O*-*tert*-butyldimethylsilyl)-β-cyclodextrin 112[104]



To heptakis(6-*O*-*tert*-butyldimethylsilyl)-β-cyclodextrin **96** (100 mg, 0.05 mmol) was added anhydrous pyridine (2.5 ml) with stirring at r.t. in a stoppered flask. 4-Dimethylaminopyridine (120 mg, 0.98 mmol) and *para*-toluenesulphonyl chloride (194 mg, 1.01 mmol) were added to the solution and the mixture was stirred for 24 hr at 50 °C. Water (10 ml) was added to the reaction mixture which was extracted with chloroform (3 x 20 ml). The organic extracts were washed with hydrochloric acid (2M, 2 x 30 ml), saturated sodium hydrogen carbonate (30 mls

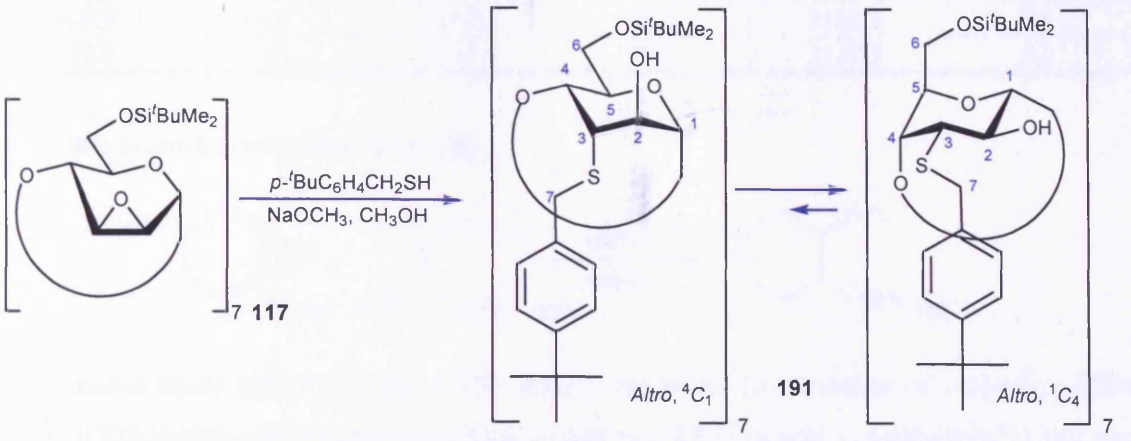
and saturated copper sulfate (10 ml), dried over Na_2SO_4 and evaporated. Purification by flash chromatography on silica eluting with chloroform and ethyl acetate gave the *title compound* **112** (93.4 mg, 62 %) as white solid. δ_H (400, $CDCl_3$), 7.80 (2H, d, *J* 8.7 Hz, tosyl, 8, 8'-H), 7.36 (2H, d, 8.7 Hz, tosyl, 9, 9'-H), 5.20 (1H, d, *J* 3.7 Hz, 1-H), 4.25 (1H, dd, *J* 9.9, 3.7 Hz, 2-H), 3.86 (1H, dd, *J* 11.6, 2.8 Hz, 6a-H), 3.82 (1H, t, *J* 9.3 Hz, 3-H), 3.65-3.62 (2H, dt, *J* 10.2, 9.4 Hz , 4-H, 6b-H), 3.48 (1H, m, 5-H), 2.45 (3H, s, tosyl), 0.84 (9H, $C(CH_3)_3$), 0.00 (6H, $Si(CH_3)_2$). δ_C (DEPT, δ_H ^1H-^{13}C ^1J-COSY) 145.1 (C, tosyl, 10-C), 133.0 (C, tosyl, 7-C), 129.6 (CH, 7.36, tosyl, 9, 9'-C), 128.3 (CH, 7.80, tosyl, 8, 8'-C), 98. 9 (CH, 5.20, 1-C), 80.0 (CH, 3.65, 4-C), 79.8 (CH, 4.25, 2-C), 72.5 (CH, 3.48, 5-C), 69.9 (CH, 3.82, 3-C), 61.6 (CH_2, 3.86, 3.62, 6-C), 25.8 (CH_3, 0.84, $(CH_3)_3$), 21.8 (CH_3, 2.45, tosyl, CH_3), 18.3 (C, $C(CH_3)_3$), -5.1, -5.3 (CH3, $Si(CH_3)_2$).

Heptakis (2,3-anhydro)-β-cyclomannin 117[105]



To a solution of heptakis(2-*O*-tosyl-6-*O*-*tert*-butyldimethylsilyl)-β-cyclodextrin **112** (100 mg, 0.033 mmol) in THF (5.5 ml) was added NaH (994 mg, 41.41 mmol) slowly with stirring. The mixture was left to stir for 29 hrs at room temperature. Isopropanol was added slowly to stop the reaction. Hydrochloric acid (2M) was added to neutralise mixture and then extracted with ethyl acetate (2 x 100 ml) and dried over Na_2SO_4, filtered and evaporated to give a white solid. Purification by column chromatography using chloroform and ethyl acetate (10:0 to 0:10) gave the *title compound* **117** (47.7 mg, 82%) as a solid. R_f 0.3 in dichloromethane:ethyl acetate; 90:10. δ_H (400, $CDCl_3$), 5.13 (1H, s, 1-H), 4.10 (1H, d, *J* 9.0 Hz, 4-H), 3.87 (1H, dd, *J* 11.6, 3.13 Hz, 6a-H), 3.64 (1H, d, *J* 11.1 Hz, 6b-H), 3.51 (1H, d, *J* 7.9 Hz, 5-H), 3.29 (1H, d, *J* 3.6 Hz, 2-H), 3.09 (1H, d, *J* 3.54, 3-H), 0.85 (9H, $C(CH_3)_3$), 0.00 (6H, $Si(CH_3)_2$). δ_C (DEPT, δ_H ^1H-^{13}C ^1J-COSY) 96.2 (CH, 5.13, 1-C), 70.8 (CH, 3.51, 5-C), 70.7 (CH_2, 3.87, 3.64, 6-C), 69.8 (CH, 4.10, 4-C), 68.0 (CH, 3.29, 2-C), 49.1 (CH, 3.09, 3-C), 25.9 (CH_3, 0.85, $(CH_3)_3$), 17.3 (C, $C(CH_3)_3$), -4.1, - 4.7 (CH3, $Si(CH_3)_2$).

Heptakis(3-deoxy-3-(4'-*tert*-butylbenzylthiol)-6-*O*-*tert*-butyldimethylsilyl)-β-cycloaltrin 191



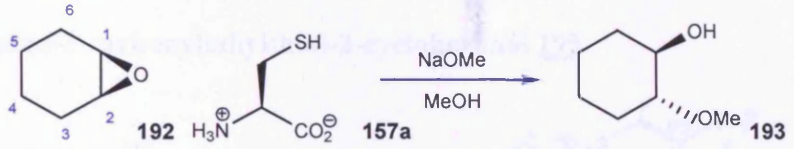
A solution of 4-*tert*-butylbenzyl thiol (397.6 mg, 0.41 ml, 2.205 mmol) and sodium methoxide (107 mg, 1.984 mmol) in methanol (4 ml) was added dropwise to a solution of epoxide **117** (50 mg, 0.027 mmol) in methanol (4 ml) and refluxed for 2 days. Aqueous sodium hydrogen carbonate (10 ml) and chloroform (20 ml) added and the organic phase was extracted with chloroform (3 x 20 ml), washed with water (10 ml), dried over anhydrous sodium sulfate and evaporated. Purification of the crude reaction mixture using a slow gradient of chloroform: ethyl acetate gave the *title compound* **191** as a white powder (60 mg, 75 % yield). R_f 0.6 in petroleum ether:ethanol, 70:30; mp 125-126 °C; $[\alpha]_D^{23}$ -97 (*c* 1.0, $CHCl_3$); IR (CH_2Cl_2), 3419, 2955, 2857, 1510, 1471, 1358, 1258, 1099 1024, 835, 775, 650 cm^{-1}; δ_H (500 MHz, C_6D_6), 7.38, (2H, d, *J* 8.3 Hz, 9a-H, 9b-H), 7.20 (2H, d, *J* 8.3 Hz, 10a-H, 10b-H), 4.90 (1H, d, *J* 6.6 Hz, 1-H), 4.44 (1H, dd, *J* 11.0, 6.7 Hz, 2-H), 4.35 (1H, ddd, *J* 5.0, 4.1, 3.8 Hz, 5-H), 4.20 (1H, d, *J* 13.2 Hz, 7a-H), 4.15 (1H, t, *J* 3.8 Hz, 4-H), 3.90 (1H, d, *J* 13.2 Hz, 7b-H), 3.80, (1H, dd, *J* 10.7, 4.0 Hz, 6a-H), 3.70 (1H, dd, *J* 10.8, 5.0 Hz, 6b-H), 3.57 (OH-2, s), 3.25 (1H, dd, *J* 11.0, 4.2 Hz, 3-H), 1.20 (9H, s, $C(CH_3)_3$), 0.92 (9H, s, $Si(C(CH_3)_3)$, 0.0 (6H, s, $Si(CH_3)_2$). δ_C (DEPT, δ_H ^{1}H-^{13}C ^{1}*J*-COSY), 149.4 (C, 12-C), 136.0 (C, 8-C), 129.2 (CH, 7.38, 9-C), 125.4 (CH, 7.20, 10-C), 106.8 (CH, 4.90, 1-C), 80.0 (CH, 4.15, 4-C), 76.9 (CH, 4.35, 5-C), 74.0 (CH, 4.44, 2-C), 63.3 (CH_2, 3.80, 3.70, 6-C), 48.2 (CH, 3.25, 3-C), 36.9 (CH_2, 4.20, 3.90, 7-C), 34.2 (C, 12-C), 31.3 (CH_3, 1.20, $C(CH_3)_3$), 26.0 (CH_3, 0.92, $Si(CH_3)_3$), 18.3 (C, $Si(C(CH_3)_3$), 1.15 (CH_3, 0.0, $Si(CH_3)_2$); MS *m/z* MALDI [M+K^+] $C_{161}H_{266}O_{28}S_7Si_7K$ found 3109.5, which is at the centre of the ion cluster (Table 34, cf chart in section **4.2.3.1.2**, page 72).

Table 34. Nominal mass abundance data for $C_{161}H_{266}O_{28}S_7Si_7K$

Mass	Abundance	Mass	Abundance	Mass	Abundance
3106.5	29.3	3110.5	86.6	3114.5	12.9
3107.5	66.4	3111.5	64.1	3115.5	6.3

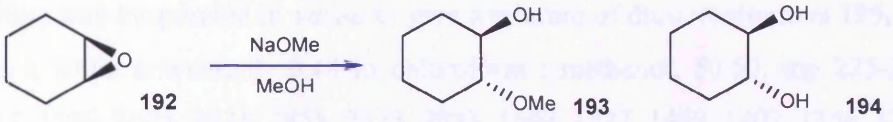
3108.5	94.5	3112.5	41.7	3116.5	2.8
3109.5	100	3113.5	24.3	3117.5	1.2

(±)-*Trans*-2-methoxycyclohexanol 193



Cyclohexene oxide **192** (0.081 mg, 0.825 mmol) was added to a solution of L-cysteine **157a** (0.1 g, 0.825 mmol) and sodium methoxide (0.044 mg, 0.825 mmol) in methanol (1.5 ml) and left to stir at r.t. for 24 h. under nitrogen. The reaction mixture was acidified with hydrochloric acid (1M), and then neutralised with saturated sodium hydrogen carbonate. The reaction mixture was extracted with ethyl acetate (3 x 10 ml), dried over magnesium sulphate and evaporated to collect the *title compound* **193** in 38 % yield. Unreacted L-cysteine **157a** (in 60 % yield) was collected from the aqueous phase.

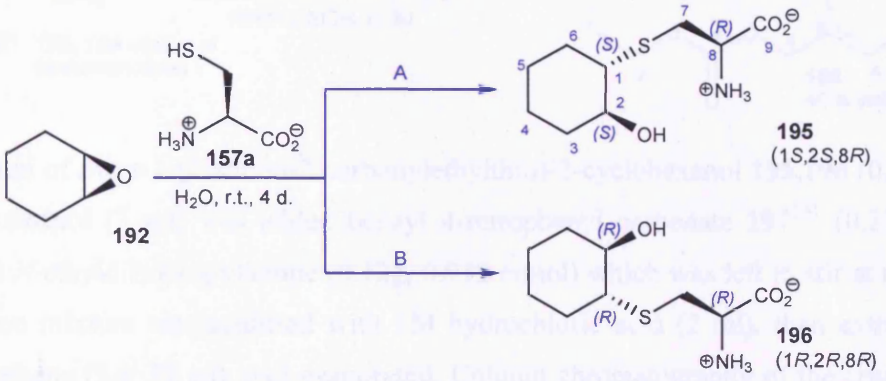
(±)-*Trans*-2-methoxycyclohexanol 193 and (±)-*trans*-1,2-dicyclohexanol 194



Cyclohexene oxide **192** (0.081 mg, 0.825 mmol) was added to a solution of sodium methoxide (0.044 mg, 0.825 mmol) in methanol (1.5 ml), and left to stir at r.t. for 24 h. The reaction mixture was acidified with hydrochloric acid (1M), and then neutralised with saturated sodium hydrogen carbonate. The reaction mixture was extracted with ethyl acetate (3 x 10 ml), dried over magnesium sulphate and evaporated. Purification by column chromatography using a slow gradient of petroleum ether : ethyl acetate gave *trans*-2-methoxycyclohexanol **193** (33 mg, 25 %) as a yellow oil and also *trans*-1,2-dicyclohexanol **194** (61 mg, 52 %) as powder in a ratio of 1:2 respectively; R_f 0.6, 0.4 in petroleum ether:ethyl acetate, 70:30 respectively; **NMR data for *trans*-2-methoxycyclohexanol 193**[150], δ_H (400 MHz, $CDCl_3$), 3.33 (3H, s, OMe), 2.85 (1H, m, 1-H), 2.60 (OH, s), 2.05 (1H, m, 2-H), 1.93 (1H, m, 3a-H), 1.69-1.58 (2H, m, 3b-H, 6a-H), 1.26-1.12 (5H, m, 4a, 4b, 5a, 5b, 6b-H). δ_C (100 MHz, $CDCl_3$), 85.4 (CH, 2.85, 1-C), 74.1 (CH, 2.05, 2-C), 56.7 (CH_3, 3.33, OMe), 32.4 (CH_2, 1.69, 1.26, 6-C), 28.7 (CH_2, 1.93 1.69, 3-C), 24.5 (CH_2, 1.26, 4-C), 24.3 (CH_2, 1.26, 5-C); IR (film),cm^{-1} 2915, 2855, 2353, 1734, 1709, 1458, 1378, 1257, 1087, 1031, 800. **NMR data for *trans*-1,2-dicyclohexanol 194**, δ_H (400 MHz, $CDCl_3$), 3.30 (OH, s), 2.70

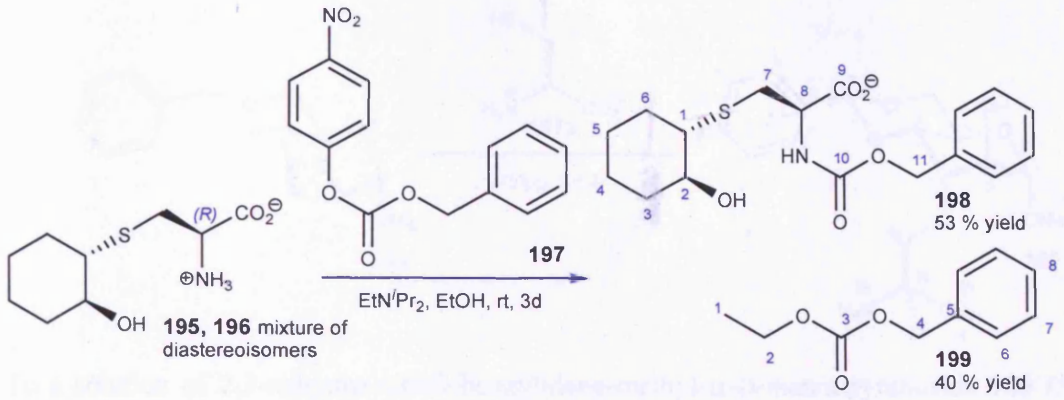
(2H, overlapping, 1-H, 6-H), 1.92 (2H, overlapping, 2a-H, 5a-H), 1.64 (2H, overlapping, 2b-H, 5b-H), 1.22 (4H, overlapping, 3a, 3b, 4a, 4b-H). δ_C (100 MHz, $CDCl_3$), 75.3 (2 x CH, 2.70, 1-C, 6-C), 31.5 (2 x CH_2, 1.92, 1.64, 2-C, 5-C), 23.0 (2 x CH_2, 1.22, 3-C, 4-C).

***Trans*-1-(2-amino-2-carbonylethylthio)-2-cyclohexanol 195**



Cyclohexene oxide **192** (0.2 g, 2.037 mmol) was added to a solution of L-cysteine **157a** (0.2 g, 1.65 mmol) in H_2O (3.5 ml) and left to stir at r.t. for 4 days under nitrogen. Water (20 ml) was added to the reaction mixture which was extracted with diethyl ether (3 x 30 ml). The aqueous phase was evaporated in *vacuo* to give a mixture of diastereoisomers **195, 196** (0.22 g, 52%) as a white powder. R_f 0.48 in chloroform : methanol, 50:50. mp 225-227°C; IR (KBr), 3317, 3196, 3005, 2925, 2855, 2573, 2081, 1608, 1527, 1449, 1402, 1356, 1262, 1229, 1158, 1118, 1070, 1039, 964, 917 cm^{-1}; δ_H (500 MHz, $CDCl_3$), 3.27-3.21 (2H, dd, *J* 7.0, 4.9 Hz, 2-H, 8-H overlap), 2.76 (1H, ddd, *J* 13.2, 8.1, 5.1 Hz, 7a-H), 2.67 (1H, td, *J* 13.7, 7.0 Hz, 7b-H), 2.39 (1H, ddd, *J* 11.5, 9.3, 4.4 Hz, 1-H), 1.92 (1H, dd, *J* 9.3, 3.7 Hz, 6a-H), 1.83 (1H, t, *J* 2.4 Hz, 3a-H), 1.58-1.48 (2H, m, overlap, 4a-H, 5a-H), 1.23 (1H, dd, *J* 15.5, 3.3 Hz, 6b-H), 1.19 (1H, dd, *J* 12.3, 8.7 Hz, 3b-H), 1.12 (1H, dd, *J* 10.6, 8.4 Hz, 5b-H), 1.08 (1H, dd, *J* 12.6, 2.0 Hz, 4b-H). δ_C (DEPT, δ_H ^1H-^{13}C ^1J-COSY), 180.9 (C = O, 9-C), 73.0 (CH, 3.27, 2-C), 55.4 (CH, 3.27, 8-C), 51.2 (CH, 2.39, 1-C), 35.1 (CH_2, 2.76, 2.67, 7-C), 34.2 (CH_2, 1.83, 1.19, 3-C), 32.4 (CH_2, 1.92, 1.23, 6-C), 25.2 (CH_2, 1.48, 1.08, 4-C), 23.8 (CH_2, 1.48, 1.12, 5-C); MS (ES^+) *m/z* 220.1 ($M+H^+$); HRMS (ES^+) *calcd* for $C_9H_{17}SO_3N$ ($M+H^+$), 220.1002, found 220.1002, error 0.0 ppm.

2-Benzyloxycarbonyl amino-3-(2-hydroxy-cyclohexylsulfanyl)-propionic acid 198

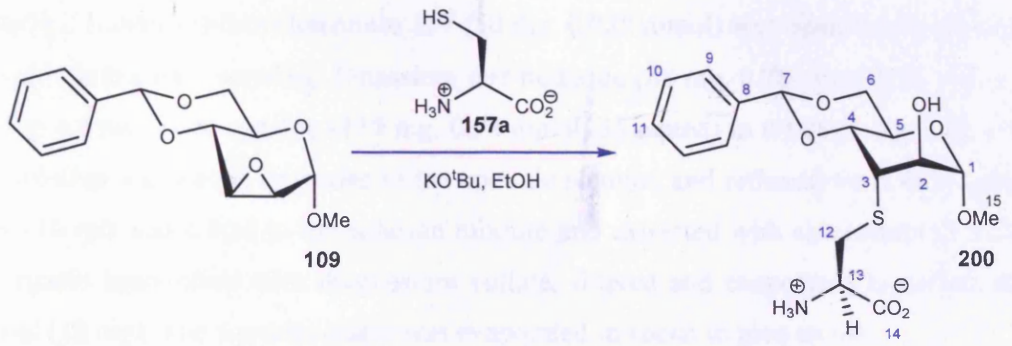


To a solution of *trans*-1-(2-amino-2-carbonylethylthio)-2-cyclohexanol **195,196** (0.2 g, 0.912 mmol) in ethanol (3 ml) was added benzyl 4-nitrophenyl carbonate **197**[151] (0.27 g, 1.003 mmol) and *N*-ethyldiisopropylamine (0.12g, 0.912 mmol) which was left to stir at r.t. for 3 d. The reaction mixture was acidified with 1M hydrochloric acid (2 ml), then extracted with dichloromethane (3 x 30 ml), and evaporated. Column chromatography of the crude mixture using petroleum ether and diethyl ether gave the *title compound* **198** (173 mg, 53 %) as a white powder, plus the benzyl ethyl carbonate **199** as an oil (72 mg, 40 %).

2-Benzyloxycarbonyl amino-3-(2-hydroxy-cyclohexylsulfanyl)-propionic acid 198: R_f 0.3 in petroleum ether:ethanol, 85:15. mp 95-96°C; IR (CH_2Cl_2), 3397, 1628, 1452, 1383, 1320, 1088, 824 cm^{-1}; δ_H (500 MHz, CD_3OD), 7.30-7.15 (5H, m, Ph-H), 5.03 (2H, 2 x d, 12.6, 12a, 12b-H), 4.90 (1H, d, *J* 6.4 Hz 10-H), 4.25 (1H, m, 8-H), 3.07 (1H, dd, *J* 13.8, 4.0, Hz, 7a-H), 2.93 (1H, dd, *J* 13.7, 9.0 Hz, 7b-H), 2.42 (1H, dd, *J* 9.5, 4.5 Hz, 2-H), 1.95 (2H, m, overlapping, 1-H, 4a-H), 1.60 (2H, m, overlapping, 4b, 5a-H), 1.20 (5H, m, overlapping, 3a, 3b, 5b, 6a, 6b-H,). δ_C (DEPT, δ_H ^1H-^{13}C ^1J-COSY), 173.0 (C, 9-C), 157.0 (C, 11-C), 137.3 (C, 13-C), 128.2, 127.1 (CH, 7.30-7.15, Ph-C), 73.5 (CH, 2.42, 2-C), 66.2 (CH_2, 5.03, 12-C), 52.7 (CH, 4.25, 8-C), 34.4 (CH_2, 1.20, 3-C), 34.2 (CH_2, 3.07, 2.93, 7-C), 32.9 (CH_2, 1.20, 6-C), 25.2 (CH_2, 1.60, 1.20, 5-C), 23.9 (CH_2, 1.95, 1.60, 4-C); MS (APCI) *m/z* 336 (M – H_2O); HRMS (ES^+) *calcd* for $C_{17}H_{23}SO_5N$ ($M+H^+$), 354.1370, found 354.1372, error - 0.6 ppm.

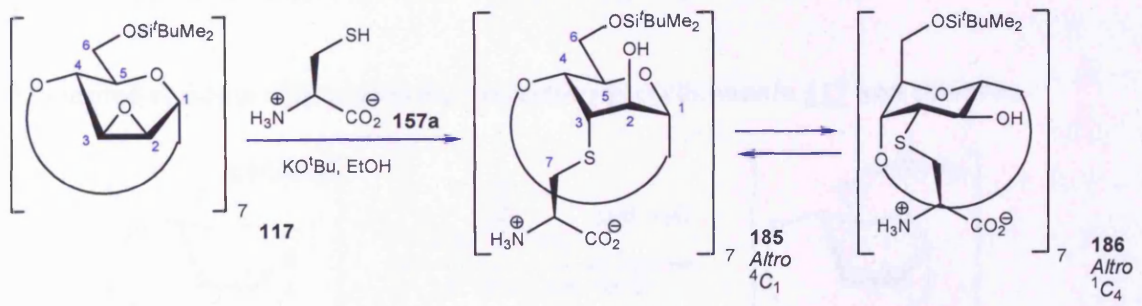
Benzyl ether carbonate 199: R_f 0.6 in chloroform:ethanol, 90:10; δ_H (400 MHz, $CDCl_3$), 7.30 (5H, m, Ph-H, overlapping), 5.08 (2H, s, 4-H), 4.12 (2H, q, 7.1 Hz, 2-H), 1.23 (3H, t, 7.1 Hz, 1-H); δ_C (DEPT, δ_H ^1H-^{13}C ^1J-COSY), 155.3 (C=O, 3-C), 136.2 (C-5), 128.4, 128.1, 127.3 (CH, 7.30, Ph-C), 67.6 (CH_2, 5.08, 4-C), 63.2 (CH_2, 4.12, 2-C), 13.9 (CH_3, 1.23, 1-C); MS (APCI) *m/z* 181 ($M + H^+$).

3-(2′-Amino-2′-carbonylethylthio)-4,6-*O*-benzylidene-α-methyl-D-altropyranoside 200:



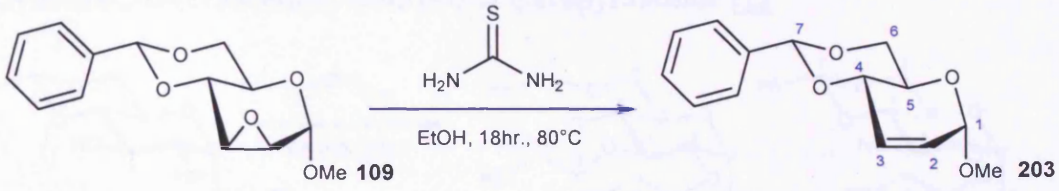
To a solution of 2,3-anhydro-4,6-*O*-benzylidene-methyl-α-D-mannopyranoside **109** (50 mg, 0.189 mmol) in ethanol (3 ml), was added a solution of cysteine **157a** (114.5 mg, 0.945 mmol, 5 equiv.) in H_2O (2 ml) dropwise with stirring. Potassium *tert*-butoxide (84.8 mg, 0.756 mmol, 4 equiv.), was added and the reaction mixture refluxed at 120°C under nitrogen for 18 h. The precipitates were collected using sinter filter funnel and evaporated in *vacuo* to give the *title compound* **200** (30 mg, 42 %) as white powder, mp 180-185°C; R_f 0.37 in petroleum ether : ethyl acetate, 70:30; $[\alpha]_D^{23}$ +30 (*c* 1.0, H_2O); IR (nujol), 2972, 2893, 2843, 1630, 1599, 1298, 1129, 1022, 976, 845, 722; δ_H (500 MHz, D_2O), 7.60 (2H, d, *J* 2.8 Hz, 9, 9'-H), 7.52 (3H, m, 10, 10', 11-H), 5.90 (1H, s, 7-H), 4.60 (1H, s, 1-H), 4.51 (1H, dd, *J* 9.6, 4.3 Hz, 4-H), 4.33 (1H, dd, *J* 10.3, 5.1 Hz, 6a-H), 4.20 (1H, m, 5-H), 4.10 (1H, d, *J* 1.5 Hz, 2-H), 3.98 (1H, t, *J* 10.3 Hz, 6b-H), 3.40 (3H, s, OMe overlap with 13-H), 3.32 (1H, dd, *J* 3.7, 2.5 Hz, 3-H), 2.98 (1H, dd, *J* 13.3, 4.5 Hz, 12a-H), 2.85 (1H, dd, *J* 13.3, 7.9 Hz, 12b-H); δ_C (DEPT, δ_H ^{1}H-^{13}C ^{1}*J*-COSY), 180.8 (COO, 14-C) 136.5 (C, 8-C), 129.7 (CH, 7.52, 11-C), 128.7 (CH, 7.52, 10-C), 126.2 (CH, 7.60, 9-C), 103.1 (CH, 5.90, 7-C), 101.9 (CH, 4.60, 1-C), 76.4 (CH, 4.51, 4-C), 73.5 (CH, 4.20, 5-C), 68.4 (CH_2, 4.33, 3.98, 6-C), 59.5 (CH, 4.10, 2-C), 55.8 (CH, 3.40, 13-H), 54.5 (CH_3, 3.40, OMe), 49.6 (CH, 3.32, 3-C), 40.3 (CH_2, 2.98, 2.85, 12-C).

Attempted reaction of heptakis(2,3-*manno*-epoxide)-β-cyclodextrin <u>117</u> and cysteine <u>157a</u>



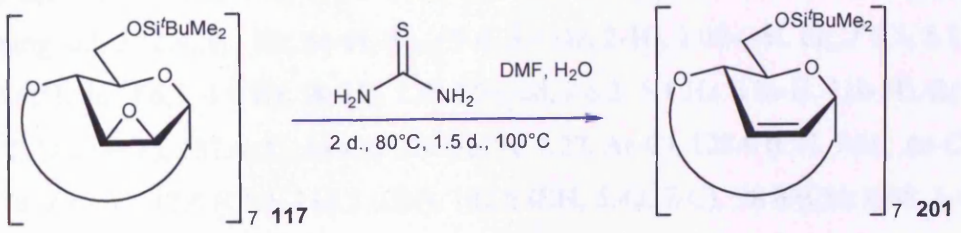
Heptakis(2,3-anhydo)-β-cyclomannin **117** (50 mg, 0.028 mmol) was dissolved in ethanol (0.5 ml) with stirring and warming. Potassium *tert*-butoxide (88 mg, 0.784 mmol, 28 equiv.) was added to a solution of cysteine (119 mg, 0.98 mmol, 35 equiv.) in water (2 ml). The cysteine **157a** solution was added dropwise to the epoxide solution and refluxed for 3 days under N_2. Water (10 ml) was added to the solution mixture and extracted with chloroform (3 x 20 ml). The organic layer dried over magnesium sulfate, filtered and evaporated to collect starting material (10 mg). The aqueous phase was evaporated in *vacuo* to give an oil.

4,6-*O*-benzylidene-2,3-dideoxy-α-D-gluco-hex-2-enopyranoside 203[152]



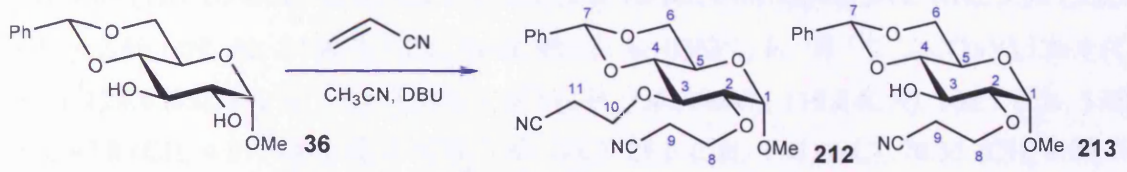
To a solution of 2,3-anhydro-4,6-*O*-benzylidene-α-methyl-D-mannopyranoside **109** (83 mg, 0.315 mmol) in ethanol (2 ml) was added a solution of thiourea (240 mg, 3.153 mmol) in ethanol (4 ml), which was heated at 80° C under nitrogen for 18 h. Water (10 ml) was added and then the reaction mixture extracted with dichloromethane (3 x 30 ml). The organic extractions were collected, dried over sodium sulfate, filtered and evaporated. Purification by flash chromatography on silica eluting with petroleum ether and ethyl acetate gave the *title compound* **203** (40 mg, 51 %) as a white solid; R_f 0.42 in petroleum ether and ethyl acetate, 80:20, δ_H (400, $CDCl_3$), 7.34, 7.29 (5H, m, Ph-H), 6.08 (1H, d, *J* 10.7 Hz, 3-H), 5.67 (1H, dt, *J* 10.4, 2.5 Hz, 2-H), 5.52 (1H, s, 7-H), 4.83 (1H, s, 1-H), 4.24 (1H, dd, *J* 9.4, 3.6 Hz, 4-H), 4.10 (1H, m, 5-H), 3.80-3.75 (2H, m, 6a-H, 6b-H), 3.39 (3H, s, OMe). δ_C (DEPT, δ_H ^{1}H-^{13}C ^{1}*J*-COSY), 136.3 (C, Ph-C), 129.8 (CH, 6.08, 3-C), 128.2, 127.3, 125.6 (CH, Ph-C), 125.2 (CH, 5.67, 2-C), 101.2 (CH, 5.52, 7-C), 96.0 (CH, 4.83, 1-H), 74.2 (CH, 4.24, 4-C), 68.4 (CH_2, 3.80, 3.75, 6-C), 63.0 (CH, 4.10, 5-C), 55.0 (CH_3, 3.39, OMe-C).

Attempted reaction of heptakis-(2,3-anhydro)-β-cyclomannin 117 and thiourea



To a solution of heptakis(2,3-anhydro)-β-cyclomannin **117** (100 mg , 0.055 mmol) in DMF (3 ml) was added a solution of thiourea (210 mg, 2.76 mmol) in 10 wt % H_2O/DMF (0.8 / 7.7 ml) which was refluxed at 80 °C for 2 d. and 100 °C for 1.5 d. Water (30 ml) was added to the reaction mixture which was extracted with chloroform (3 x 40 ml). The organic extracts were washed with water (30 ml), dried over magnesium sulfate, filtered and evaporated to give a dark brown oil (77 mg). TLC of the crude product showed many components that could not be identified.

4,6-*O*-benzylidene-2,3-di-*O*-(cyanoethyl)-Methyl α-D-glucopyranoside 212, 4,6-*O*-benzylidene-2-*O*-(cyanoethyl)-methyl-α-D-glucopyranoside 213



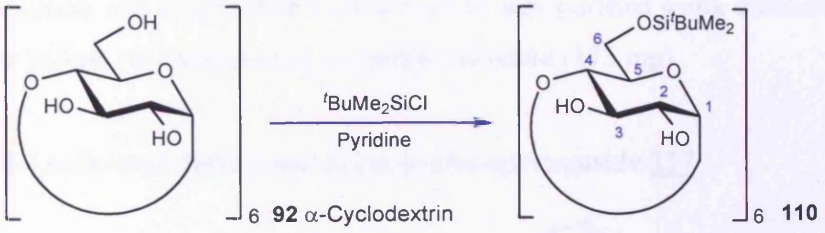
To a solution of 4,6-*O*-benzylidene-methyl-α-D-glucopyranoside **36** (2 g, 7.085 mmol) in acetonitrile (50 mls) in a round bottom flask was added DBU (0.86 g, 5.6 mmol, 0.8 ml, 0.8 equiv.) and the reaction was left to stir for 0.5 hr. Acrylonitrile (7.52 g, 141.7 mmol, 9.4 ml, 20 equiv.) was added dropwise to give a clear pale yellow solution which was left to stir at r.t. for 2d. The solution was evaporated and the residue was purified using chloroform and ethyl acetate to give the dicyanoethyl derivative **212** (1.15 g, 54 %) and the monocyanoethyl derivative **213** (0.54 g, 20 %) as white powders. R_f 0.5, 0.3 respectively in chloroform : ethyl acetate, 50:50.

4,6-*O*-Benzylidene-2,3-di-*O*-(cyanoethyl)-methyl-α-D-glucopyranoside 212: mp 109-111°C; $[\alpha]_D^{23}$ +91.5 (*c* 1.0, $CHCl_3$); IR (CH_2Cl_2), 3428, 2937, 2435, 2245, 1634, 1450, 1368, 1091, 996, 747, 696 cm^{-1}; δ_H (500 MHz, C_6D_6),7.78 (2H, d, *J* 7.29 Hz, overlapping, Ar-H), 7.38 (2H, t, *J* 7.64 Hz, overlapping, Ar-H), 7.27 (1H, d, *J* 2.4 Hz, Ar-H), 5.42 (1H, s, 7-H), 4.65 (1H, d, *J* 3.7 Hz, 1-H), 4.22 (1H, dd, *J* 10.2. 5.0 Hz, 6a-H), 3.90 (1H, m, 5-H), 3.80 (1H, t, *J* 9.2 Hz, 3-H), 3.57 (3H, t, *J* 10.3 Hz, 6b-H, overlapping with 10a-H, 8a-H), 3.43, (2H, overlapping, t, *J* 9.3 Hz, 4-H, dd, *J* 11.8, 9.9 Hz, 10b-H), 3.18 (3H, s, OMe), 3.15 (2H, overlapping, dd, *J* 12.4, 9.3 Hz, 8b-H, dd, *J* 9.0, 3.4 Hz, 2-H), 2.00 (1H, dq, *J* 7.5, 5.1 Hz, 9a-H), 1.83 (1H, dq, *J* 6.5, 4.9 Hz, 9b-H), 1.73 (2H, dd, *J* 6.2, 5.4 Hz, 11a-H, 11b-H). δ_C (DEPT, δ_H ^1H-^{13}C ^1J-COSY), 137.0 (C, Ar-C), 129.0 (CH, 7.27, Ar-C), 128.0 (CH, 7.38, Ar-C), 126.5 (CH, 7.78, Ar-C), 117.4 (CN), 116.5 (CN), 102.6 (CH, 5.42, 7-C), 98.8 (CH, 4.65, 1-C), 82.1

(CH, 3.43, 4-C), 80.1 (CH, 3.80, 3-C), 78.6 (CH, 3.15, 2-C), 69.0 (CH_2, 4.22, 3.57, 6-C), 67.2 (CH_2, 3.57, 3.43, 10-C), 66.4 (CH_2, 3.57, 3.15, 8-C), 62.4 (CH, 3.90, 5-C), 55.0 (CH_3, 3.18, OMe-C), 18.7 (CH_2, 2.00,1.83, 1.73, 9-C or 11-C), 18.6 (CH_2, 2.00, 183, 1.73, 9-C or 11-C); MS (EI/CI) *m/z* 389.2 (M + H); HRMS (ES^+) *calcd* for $C_{20}H_{24}O_6N_2$ ($M+H^+$) 389.1707, found 389.1708, error – 0.25 ppm.

4,6-*O*-Benzylidene-2-*O*-(cyanoethyl)-methyl-α-D-glucopyranoside 213: mp 123 - 124°C; $[\alpha]_D^{23}$ +60.8 (*c* 1.0, $CHCl_3$); IR (CH_2Cl_2), 3455, 2916, 2250, 1452, 1089, 989, 919, 752, 700 cm^{-1}; δ_H (500 MHz, $CDCl_3$), 7.42 (2H,d, *J* 2.7 Hz, d, *J* 1.8 Hz,Ar-H), 7.30 (3H, m, Ar-H), 5.40 (1H, s, 7-H), 4.81 (1H, d, *J* 3.6 Hz, 1-H), 4.25 (1H,dd, *J* 10.1, 4,7 Hz, 6a-H), 4.09 (1H, t, *J* 9.2 Hz, 3-H), 3.94 (1H, dd, *J* 16.0, 6.2 Hz, 8a-H), 3.87 (1H, t, *J* 6.7 Hz, 8b-H), 3.78 (1H, m, 5-H), 3.69 (1H, t, *J* 10.2 Hz, 6b-H), 3.41 (2H, t, *J* 9.0 Hz, overlapping 2-H, 4-H), 3.38 (3H, s, OCH_3), 2.60 (2H, dd, *J* 6.8, 6.3 Hz, 9a-H, 9b-H). δ_C (DEPT, δ_H ^1H-^{13}C ^1J-COSY),136.9 (C, Ar-C), 128.4, 128.3 (CH, 7.30, Ar-C), 126.3 (CH, 7.42, Ar-C), 119.8 (CN), 102.1 (CH, 5.40, 7-C), 97.8 (CH, 4.81, 1-C), 81.4 (CH, 3.41, 4-C), 81.0 (CH, 3.41, 2-C), 70.55 (CH, 4.09, 3-C), 70.52 (CH_2, 4.25, 3.69, 6-C), 67.2 (CH_2, 3.94, 3.87, 8-C), 61.9 (CH, 3.78, 5-C), 55.5 (CH_3, 3.38, OCH_3), 19.8 (CH_2, 2.60, 9-C); MS (EI/CI) *m/z* 336.2 (M + H); HRMS (ES^+) *calcd* for $C_{17}H_{21}O_6N$ ($M+H^+$) 336.1442, found 336.1444, error – 0.6 ppm.

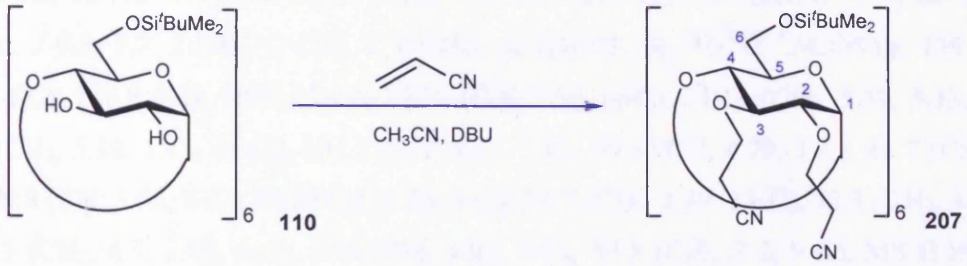
Hexakis (6-*O*-*tert*-butyldimethylsilyl)-α-cyclodextrin 110



A solution of α-cyclodextrin **92** (5 g, 5.14 mmol) in dry pyridine (40 ml) was added to a stirred, chilled (0° C) solution of *tert*-butyldimethylsilyl chloride (5 g, 33.4 mmol) in anhydrous pyridine (30 ml) over 30 minutes. The stirring was continued for 24 hrs. Chloroform (100 ml) and water (100 ml) was added to the reaction mixture and extracted with chloroform (3 x 70 ml). The organic extracts were then washed with 1M hydrochloric acid, and aqueous sodium hydrogen carbonate, dried over magnesium sulphate, filtered, and evaporated. Purification of the crude reaction mixture using a solvent gradient of chloroform to ethanol gave the *title compound* **110** (3.05 g, 36%) as crystals. R_f 0.46 in chloroform:ethanol; 80:20. δ_H (500 MHz, $CDCl_3$), 4.85 (1H,d, *J* 3.2 Hz, 1-H), 3.98 (1H, t, *J*

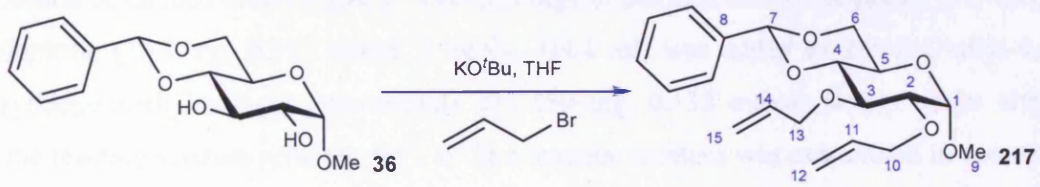
9.0 Hz, 3-H), 3.88 (1H, dd, *J* 11.4, 3.4 Hz, 6a-H), 3.81 (1H, d broad, *J* 9.8 Hz, 5-H), 3.72 (1H, d, *J* 10.5, Hz, 6b-H), 3.61 (1H, dd, *J* 9.6, 3.2 Hz, 2-H), 3.54 (1H, t, *J* 9.0 Hz, 4-H), 0.85 (9H, s, $Si(C(CH_3)_3)$, 0.0 (6H, s, $Si(CH_3)_2$). δ_C (DEPT, δ_H ^1H-^{13}C ^1J-COSY),101.3 (CH, 4.85, 1-C), 81.2 (CH, 3.54, 4-C), 74.5 (CH, 3.98, 3-C), 73.0 (CH, 3.61, 2-C), 72.1 (CH, 3.81, 5-C), 62.0 (CH, 3.88, 3.72, 6-C), 26.0 (CH_3, 0.85, $C(CH_3)_3$), 18.4 (C, $C(CH_3)_3$), -5.0, - 5.1 (CH3, $Si(CH_3)_2$).

Attempted cyanoethylation of hexakis (6-*O*-*tert*-butyldimethylsilyl)-α-cyclodextrin 110



To a solution of hexakis (6-*O*-*tert*-butyldimethylsilyl)-α-cyclodextrin **110** (200 mg, 0.122 mmol) in acetonitrile (30 ml) in a round bottom flask was added DBU (0.04 g, 0.3 mmol, 0.4 ml) and the reaction was left to stir for 0.5 hr. Acrylonitrile (0.4 g, 7.32 mmol, 0.5 ml) was added dropwise to give an orange-brown solution which was left to stir at r.t. for 2d. After that time, more DBU (0.05 ml) and acrylonitrile (1 ml) was added and the solution refluxed for 1 d. The solution was evaporated and the residue was purified using dichloromethane and ethanol to give yellow oil composed of a complex mixture (175 mg)

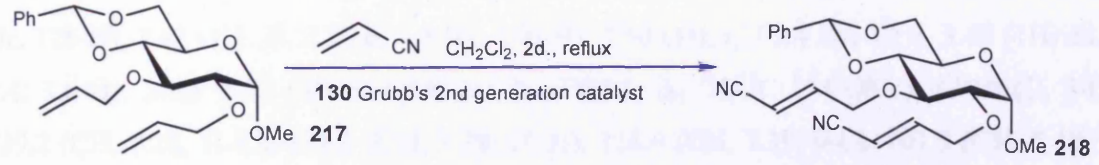
2,3-Di-*O*-allyl-4,6-*O*-benzylidene-methyl α-D-glucopyranoside 217



4,6-*O*-benzylidene-methyl-α-D-glucopyranoside **36** (2 g, 7.085 mmol) was dissolved in distilled THF (40 ml) with stirring in a 100 ml stoppered flask. Potassium *tert*-butoxide (3.2 g, 28.34 mmol) was added to the solution, which became yellow solution immediately. To that solution, allyl bromide (8.6 g, 70.85, 6.2 mls) was added dropwise and left stirring at r.t. under nitrogen for 24 hr. Water (50 ml) was added to the solution mixture which was extracted with dichloromethane (3 x 100 ml). The organic layer was dried over magnesium

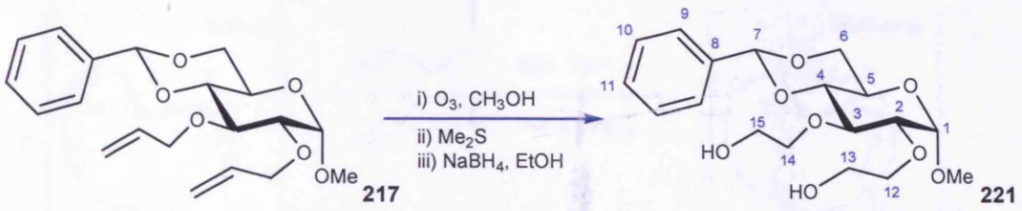
sulphate, filtered and evaporated to give the *title compound* **217**, (2.3 g, 90%) as a white powder. ^{1}H-NMR showed pure compound and consequently no further purification was required. R_f 0.38 in petroleum ether:ethylacetate, 90:10; mp 60 - 61°C; $[\alpha]_D^{23}$ +78 (*c* 1.0, $CHCl_3$); IR (film), 2915, 2863, 1646, 1454, 1374, 1087, 1053, 995, 924, 750, 699, 650 cm^{-1}; δ_H (500 MHz, C_6D_6), 7.56 (5H, m, ar-H), 6.08 (1H, ddt, *J* 17.2, 10.2, 5.3 Hz, 14-H), 5.97 (1H, ddt, *J* 17.2, 10.6, 5.3 Hz, 11-H), 5.47 (1H, s, 7-H), 5.39 (2H, dq, *J* 17.3, 1.8 Hz, 15-H, 12-H), 5.13 (2H, dq, *J* 10.3, 1.8 Hz, 15-H, 12-H), 4.79 (1H, d, *J* 3.7 Hz, 1-H), 4.49 (2H, ddt, *J* 13.2, 5.3, 1.6 Hz, 13-H), 4.3 (1H, m, 6a-H), 4.23 (1H, t, *J* 9.2 Hz, 3-H), 4.21 (2H, ddt, *J* 13.0, 5.7, 1.5 Hz, 10-H), 4.04 (1H, ddd, *J* 10.0, 10.0, 5.0 Hz, 5-H), 3.65 (2H, m, 4-H, 6b-H), 3.55 (1H, dd, *J* 9.3, 3.7, 2-H), 3.2 (3H, s, OCH_3). δ_C (DEPT, δ_H ^{1}H-^{13}C ^{1}*J*-COSY), 135.9 (CH, 6.08, 14-C), 135.5 (CH, 5.97, 11-C), 127.8 (CH, 7.56, ar-C), 116.1 (CH_2, 5.39, 5.13, 12-C), 115.5 (CH_2, 5.39, 5.13, 15-C), 101.5 (CH, 5.47, 7-C), 99.4 (CH, 4.79, 1-C), 82.7 (CH, 3.65, 4-C), 79.8 (CH, 3.55, 2-C), 78.0 (CH, 4.23, 3-C), 73.7 (CH_2, 4.49, 13-C), 72.3 (CH_2, 4.21, 10-C), 69.1 (CH_2, 4.3, 3.65, 6-C), 62.6 (CH, 4.04, 5-C), 54.8 (CH_3, 3.2, 9-C); MS (EI/CI) *m/z* 363.3 (M + H); HRMS (ES^+) *calcd* for $C_{20}H_{26}O_6$ ($M+H^+$) 363.1802, found 363.1803, error – 0.27 ppm.

Attempted synthesis of 4,6-*O*-benzylidene-2,3-di-*O*-(but-2-enenitrile)-α-methyl-D-glucopyranoside 218



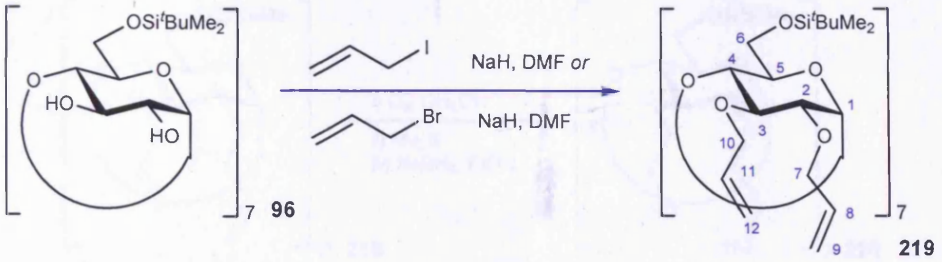
A solution of Grubbs catalyst **130** (5 mol%, 6 mg) in distilled dichloromethane (0.7 ml), and acrylonitrile (29.4 mg, 0.552 mmol, 4 equiv., 0.04 ml) was added to 2,3-di-*O*-allyl-4,6-*O*-benzylidene-methyl-α-D-glucopyranoside **217** (50 mg, 0.138 mmol) slowly under nitrogen and the reaction mixture refluxed for 2 d. The reaction mixture was evaporated in *vacuo*. The ^{1}H-NMR spectrum of the crude reaction mixture only allowed staring material to be identified (38 mg).

4,6-*O*-Benzylidene-2,3-di-*O*-2-ethanol-α-methyl-D-glucopyranoside 221



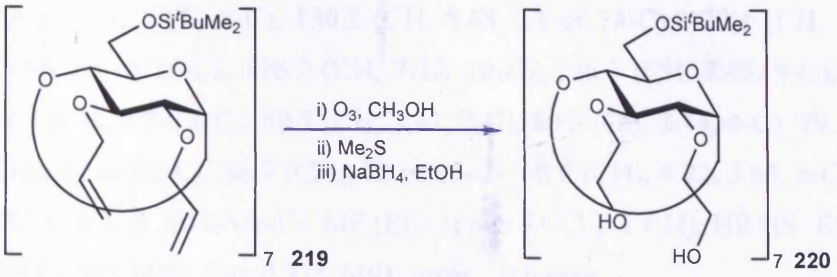
A solution of 2,3-di-*O*-allyl-4,6-*O*-benzylidene-α-methyl-D-glucopyranoside **217** (300 mg, 0.83 mmol) in methanol (20 ml) was ozonolysed for 6 hr. Dimethyl sulfide (0.5 ml) was added and left to stirring for 10 min. A solution of sodium borohydride (250 mg, 6.64 mmol) in ethanol (2 ml) was added slowly and the reaction stirred for overnight. Water (15 ml) and hydrochloric acid (1M, 10 ml) was added. The mixture was extracted with chloroform (3 x 50 ml), dried over sodium sulfate, filtered and evaporated. Purification by flash chromatography on silica eluting with chloroform and ethanol gave the *title compound* **221** (260 mg, 85 %) as yellowish solid. R_f 0.4 in chloroform:ethanol; 97:03; mp 103-105°C; $[\alpha]_D^{23}$ + 53.6 (*c* 1.0, $CHCl_3$); IR (CH_2Cl_2), 3417, 2925, 2875, 2343, 1649, 1453, 1368, 1087, 1052, 991, 921, 871, 750, 692 cm^{-1}; δ_H (500, $CDCl_3$), 7.39 (2H, 2 x d, *J* 2.2, 1.5 Hz, 9, 9'-H), 7.28 (3H, m, overlapping 10, 10', 11-H), 5.48 (1H, s, 7-H), 4.80 (1H, d, *J* 3.6 Hz, 1-H), 4.20 (1H, dd, *J* 10.0, 4.6 Hz, 6a-H), 3.88 (1H, dt, *J* 11.5, 4.2 Hz, 14a-H), 3.80 (1H, t, *J* 9.3 Hz, 3-H), 3.78 (1H, m, overlapping with H-3, 5-H), 3.76 ((1H, dt, *J* 4.9, 2.1 Hz, 14b-H), 3.75 (1H, dd, *J* 4.6, 2.0 Hz, 15a-H), 3.74 (1H, dt, *J* 11.5, 4.9 Hz, 12a-H), 3.71 (1H, dt, *J* 7.6, 2.7 Hz, 13a-H), 3. 68 (1H, dd, *J* 10.0, 3.2 Hz, 6b-H), 3.67 (1H, dd, *J* 12.9, 2.7 Hz, 13b-H), 3.64 (1H, dt, *J* 4.8, 2.6 Hz, 12b-H), 3.62 (1H, dt, *J* 15.6, 4.5 Hz, 15b-H), 3.50 (1H, t, *J* 9.4 Hz, 4-H), 3.45 (1H, dd, *J* 9.4, 3.6 Hz, 2-H), 3.37 (3H, s, OMe-H). δ_C (DEPT, δ_H ^1H-^{13}C ^1J-COSY), 137.0 (C, 8-C), 129.2 (CH, 7.28, 11-C), 128.3 (CH, 7.28, 10-H), 126.0 (CH, 7.39, 9-C), 101.5 (CH, 5.48, 7-C), 98.4 (CH, 4.80, 1-H), 81.9 (CH, 3.50, 4-C), 80.4 (CH, 3.45, 2-C), 78.0 (CH, 3.80, 3-C), 74.5 (CH_2, 3.88, 3.76, 14-C), 72.9 (CH_2, 3.74, 3.64, 12-C), 69.0 (CH_2, 4.20, 3.68, 6-C), 62.4 (CH, 3.78, 5-C), 61.9 (CH2, 3.71, 3.67, 13-C), 61.7 (CH2, 3.75, 3.62, 15-C), 55.3 (CH3, 3.37, OMe-C); MS (ES^+) *m/z* 393.1 (M + Na^+), HRMS (ES^+) *calcd* for $C_{18}H_{26}O_8$ (M + NH_4^+), 388.1966, found 388.1967, error – 0.25 ppm.

Heptakis(2,3-di-*O*-allyl-6-*O*-*tert*-butyldimethylsilyl)-β-cyclodextrin 219



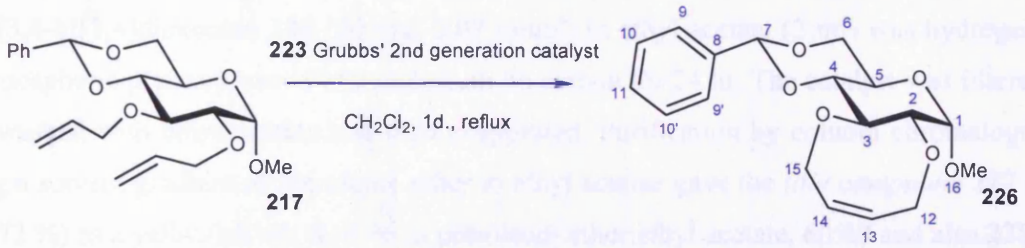
To a stirred, chilled (0°C) solution of heptakis(6-*O*-*tert*-butyldimethylsilyl)-β-cyclodextrin **96** (200 mg, 0.1 mmol) in dry *N,N*-dimethylformamide (6.1 ml) was added sodium hydride (0.2 g of a 60% oil immersion, rinsed 3 times with diethyl ether) and the mixture was stirred under nitrogen for 2 hr. at 0 °C and overnight at room temperature. Allyl bromide (1.1 ml) was then added dropwise and the reaction was left to stir for 1 day. Water (50 ml) and chloroform (50 ml) added. Extracted with chloroform (3 x 50 ml), dried over sodium sulphate, filtered and evaporated. Purification of the crude reaction mixture by column chromatography using a solvent gradient of petroleum ether to ethyl acetate gave the *title compound* **219** (90 mg, 36%) as a syrup. R_f 0.5 in petroleum ether:ethyl acetate, 95:5; $[\alpha]_D^{23}$ +80 (*c* 1.0, $CHCl_3$); IR (CH_2Cl_2), 2928, 2855, 1644, 1463, 1403, 1383, 1348, 1252, 1147, 1092, 1026, 926, 881, 825, 780, 650 cm^{-1}; δ_H (500 MHz, $CDCl_3$) 6.05 (1H,m, 8-H), 5.92 (1H, m, 11-H), 5.25 (2H, t, *J* 17.1 Hz, overlapping 9a-H, 12a-H), 5.20 (1H, d, *J* 3.3 Hz 1-H), 5.10 (2H, t, *J* 8.9 Hz, 9b-H, 12b-H), 4.50 (1H, dd, *J* 11.8, 5.5 Hz, 7a-H), 4.25 (1H, dd, *J* 11.8, 5.9 Hz, 7b-H), 4.17 (3H, m, overlapping, 10-H, 10-H, 6a-H), 3.83 (1H, t, *J* 9.1 Hz 4-H), 3.72 (1H, t, *J* 9.2 Hz, 3-H), 3.62 (1H, d, *J* 11.4 Hz, 6b-H), 3.55 (1H, d, *J* 9.4 Hz, 5-H), 3.21 (1H, dd, *J* 9.7, 3.3 Hz, 2-H), 0.88 (9H, s, $Si(C(CH_3)_3)$), 0.0 (6H, s, $Si(CH_3)_2$). δ_C (DEPT, δ_H ^1H-^{13}C ^1J-COSY) 136.4 (CH, 6.05, 8-C), 135.4 (CH, 5.92, 11-C), 116.5 (CH_2, 5.25, 5.10, 9-C), 115.8 (CH_2, 5.25, 5.10, 12-C), 98.2 (CH, 5.20, 1-C) 80.2 (CH, 3.21, 2-C), 79.4 (CH, 3.72, 3-C), 77.9 (CH, 3.83, 4-H), 74.7 (CH_2, 4.50, 4.25, 7-C), 72.2 (CH, 3.55, 5-C), 72.1 (CH_2, 4.17, 10-C), 62.3 (CH_2, 4.17, 3.62, 6-C), 29.7 (CH_3, 0.88), 18.3 (C, 0.88), -5.0 (CH_3, 0.0); MS (MALDI) *m/z* 2534.3 ($M + K^+$), *calcd* for $C_{126}H_{224}O_{35}Si_7$ ($M + K^+$) 2534.4, found 2534.3, error 39.4 ppm.

Attempted ozonolysis of heptakis(2,3-di-*O*-allyl-6-*O*-*tert*-butyldimethylsilyl)-β-cyclodextrin 219



A solution of heptakis(2,3-di-*O*-allyl-6-*O*-*tert*-butyldimethylsilyl)-β-cyclodextrin **219** (270 mg, 0.108 mmol) in methanol (20 ml) was ozonolysed for 8 hr. Dimethyl sulfide (1 ml) was added and the reaction left to stir for 10 min. A solution of sodium borohydride (230 mg, 6.048 mmol) in ethanol (2 ml) was added slowly and the reaction mixture left to stir overnight. Water (20 ml) and hydrochloric acid (1M, 15 ml) were added. The mixture was extracted with chloroform (3 x 60 ml), dried over sodium sulphate, filtered and evaporated to give a complex mixture (260 mg) that could not be separated to give pure products.

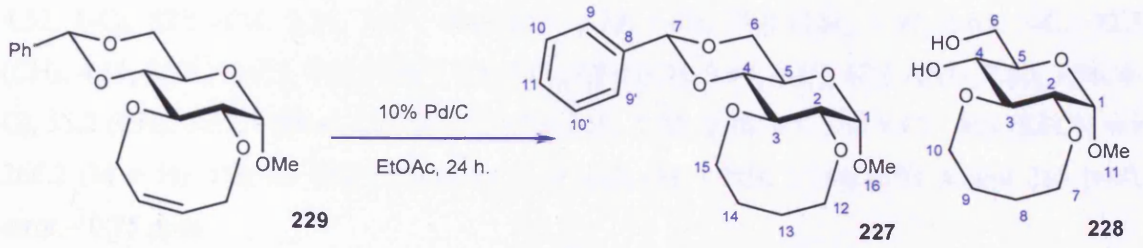
6-Methoxy-2-phenyl-4,4a,6,6a,8,11,12a,12b-octahydro[1,3]dioxine[4',5':5,6]pyrano[3,4-b][1,4]dioxocine 226



Grubbs catalyst **223** (5 mol%, 12 mg) was added to a solution of 2,3-di-*O*-allyl-4,6-*O*-benzylidene-methyl-α-D-glucopyranoside **217** (100 mg, 0.276 mmol) in distilled dichloromethane (10 ml), under nitrogen and refluxed for one day. The Grubbs catalyst was filtered off through silica gel and washed with dichloromethane. The organics were then evaporated. The crude reaction mixture purified by column chromatography using a solvent gradient of petroleum ether, ethyl acetate to give the *title compound* **226** (50 mg, 56%) as an oil. R_f 0.5 in petroleum ether:ethyl acetate, 60:40; $[\alpha]_D^{23}$ +47.8 (*c* 1.0, $CHCl_3$); IR (film), 3441, 2363, 2097, 1636, 1453, 1373, 1257, 1087, 941, 790, 695 cm^{-1}; δ_H (500 MHz, $CDCl_3$), 7.45 (2H, 2 x d, *J* 2.2, 1.6 Hz, 9, 9'-H), 7.30 (3H, m, 10, 10', 11-H), 5.68 (2H, m, 13, 14-H), 5.45 (1H, s, 7-H), 4.77 (1H, d, *J* 3.8 Hz, 1-H), 4.40-4.35, (3H,overlapping, dd, *J* 15.1, 5.0 Hz, dd, *J* 10.1, 3.2 Hz observed, 12a, 15a, 15b-H), 4.28 (1H, dd, *J* 14.5, 4.3 Hz, 12b-H), 4.22 (1H, *J* 10.1, 4.8 Hz, 6a-H), 3.81 (1H, m, 5-H), 3.77 (1H, t, *J* 10.0 Hz, 3-H), 3.66 (1H, t, *J* 10.3 Hz, 6b-H), 3.44 (1H, t, *J* 9.6 Hz, 4-H), 3.42 (1H, dd, *J* 9.0, 3.8 Hz, 2-H), 3.38 (3H, s, OMe-H), δ_C (DEPT, δ_H

^{1}H-^{13}C ^{1}J-COSY), 137.3 (C, 8-C), 130.2 (CH, 5.68, 13 or 14-C), 129.1 (CH, 7.30, 11-C), 128.8 (CH, 5.68, 13 or 14-C), 128.2 (CH, 7.30, 10-C), 126.3 (CH, 7.45, 9-C), 101.8 (CH, 5.45, 7-C), 99.3 (CH, 4.77, 1-C), 80.7 (CH, 3.42, 2-C), 80.5 (CH, 3.44, 4-C), 79.1 (CH, 3.81, 5-C), 69.1 (CH_2, 12 or 15-C), 68.9 (CH_2, 12 or 15-C), 68.6 (CH_2, 4.22, 3.66, 6-C), 62.2 (CH, 3.77, 3-C), 55.3 (CH3, 3.38, OMe-C). MS (EI/CI) *m/z* 335.2 (M + H); HRMS (ES^+) *calcd* for $C_{18}H_{22}O_6$ ($M+H^+$) 335.1489, found 335.1491, error – 0.6 ppm.

(6*S*,6a*R*,12a*S*)-2-phenylperhydro[1,3]dioxino[4',5':5,6]pyrano[3,4-b][1,4]dioxocin-6-yl methylether 227 and 6a*S*, 7*S*, 10*R*, 10a*S*)-octahydro-9-(hydroxymethyl)-7-methoxy-2H-pyrano[3,4-b][1,4]dioxocin-10-ol 228



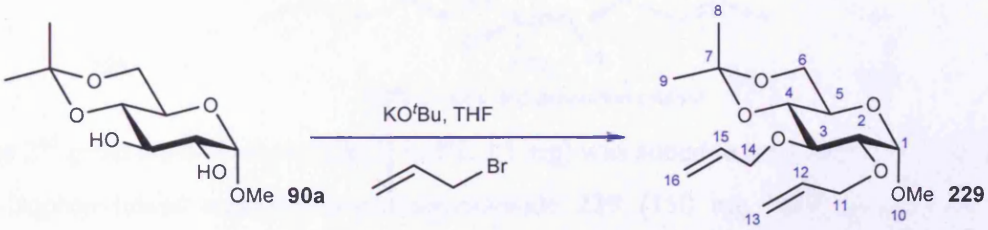
A solution of 6-methoxy-2-phenyl-4,4a,6,6a,8,11,12a,12b-octahydro[1,3]dioxine[4′,5′:5,6]pyrano[3,4-b][1,4]dioxocine **226** (20 mg, 0.07 mmol) in ethyl acetate (2 ml) was hydrogenated at atmospheric pressure over 10 % palladium on carbon for 24 hr. The catalyst was filtered off and washed with ethyl acetate and then evaporated. Purification by column chromatography using a solvent gradient of petroleum ether to ethyl acetate gave the *title compound* **227** (16.6 mg, 72 %) as a yellowish oil. R_f 0.46 in petroleum ether:ethyl acetate, 60:40 and also **228** (4.3 mg, 25 %) as an oil. R_f 0.4 in dichloromethane:methanol, 90:10.

(6*S*,6a*R*,12a*S*)-2-phenylperhydro[1,3]dioxino[4',5':5,6]pyrano[3,4-b][1,4]dioxocin-6-yl methylether 227: $[\alpha]_D^{23}$ + 36 (*c* 1.0, $CHCl_3$); IR (film), 2955, 2922, 2852, 1462, 1377, 1093, 986, 740, 695 cm^{-1}; δ_H (500 MHz, C_6D_6), 7.80 (2H, d, *J* 7.2 Hz, 9, 9'-H), 7.30 (3H,m, 10, 10', 11-H), 5.50 (1H, s, 7-H), 4.78 (1H, d, *J* 3.5 Hz, 1-H), 4.30 (1H, dd, *J* 10.1, 5.0 Hz, 6a-H), 4.19 (1H, t, *J* 9.3Hz, 3-H), 4.10 (1H, dd, *J* 11.8, 5.1 Hz, 12-H) 4.07 (1H, m, 5-H), 4.03 (1H, m, 15-H), 3.74 (1H, m, 15-H), 3.72 (1H, dd, *J* 11.4, 3.9 Hz, overlapping with 3.74, 12-H), 3.65 (1H, t, *J* 10.3 Hz, 6b-H), 3.58 (1H, t, *J* 9.5 Hz, 4-H), 3.40 (1H, dd, *J* 9.2, 3.6 Hz, 2-H), 3.20, (3H, s, OMe-H), 1.70, 1.63, 1.52, 1.40 (4H, 4 x m, 13 and 14-H). δ_C (DEPT, δ_H ^{1}H-^{13}C ^{1}J-COSY), 138.4 (C, 8-C), 128.8 (CH, 7.30, 11-C), 127.9 (CH, 7.30, 10-C), 126.8 (CH, 7.80, 9-C), 101.9 (CH, 5.50, 7-C), 100.5 (CH, 4.78, 1-C), 82.2 (CH, 3.40, 2-C), 80.7 (CH, 3.58, 4-C), 78.7 (CH, 4.19, 3-C), 72.1 (CH_2, 12-C), 71.9 (CH_2, 15-C), 69.2 (CH_2, 4.30, 3.65, 6-C), 63.3 (CH, 4.07,

5-C), 54.8 (CH_3, 3.20, OMe-C), 27.9, 26.4 (CH_2, 13 and 14-C). MS (EI/CI) *m/z* 337.2 (M + H); HRMS (ES^+) *calcd* for $C_{18}H_{24}O_6$ (M + H^+) 337.1646, found 337.1645, error 0.3 ppm.

(6a*S*, 7*S*, 10*R*, 10a*S*)-octahydro-9-(hydroxymethyl)-7-methoxy-2H-pyrano[3,4-b][1,4]dioxocin-10-ol 228: $[\alpha]_D^{23}$ + 64 (*c* 1.0, $CHCl_3$); IR (film), 2925, 2855, 1458, 1373, 1092, 1052, 986, 750, 695, 650 cm^{-1}; δ_H (500 MHz, $CDCl_3$), 4.52 (1H, d, *J* 3.6 Hz, 1-H), 4.11 (1H, dt, *J* 12.0, 5.1 Hz, 10-H), 4.07 (1H, dt, *J* 12.0, 5.5 Hz, 7-H, overlapping with 10-H), 3.80 (1H, dd, *J* 11.7, 3.4 Hz, 6a-H), 3.76 (1H, dd, *J* 11.6, 4.3 Hz, 6b-H), 3.74 (1H, m, 10'-H, overlapping with 6b-H), 3.63 (1H, dt, *J* 11.0, 4.3 Hz, 7'-H , overlapping 5-H), 3.58 (1H, m, 5-H), 3.56 (1H, t, *J* 9.3 Hz, 3-H), 3.44 (1H, t, *J* 9.4 Hz, 4-H), 3.35 (3H, s, OMe), 3.28 (1H, dd, *J* 9.5, 3.6 Hz, 2-H), 1.75, 1.70 (2 x 2H, 2 x m, 8-H, 9-H). δ_C (DEPT, δ_H ^1H-^{13}C ^1J-COSY), 99.4 (CH, 4.52, 1-C), 82.5 (CH, 3.56, 3-C), 80.5 (CH, 3.28, 2-C), 73.0 (CH_2, 4.07, 3.63, 7-C), 72.5 (CH_2, 4.11, 3.74, 10-C), 70.9 (CH, 3.58, 5-C), 69.9 (CH, 3.44, 4-C), 62.5 (CH_2, 3.80, 3.76, 6-C), 55.2 (CH_3, 3.35, OMe-C), 27.5, 26.3 (2 x CH_2, 1.75, 1.70, 8-C and 9-C). MS (EI/CI) *m/z* 266.2 (M + H); HRMS (ES^+) *calcd* for $C_{11}H_{20}O_6$ (M + NH_4^+) 266.1598, found 266.1600, error – 0.75 ppm.

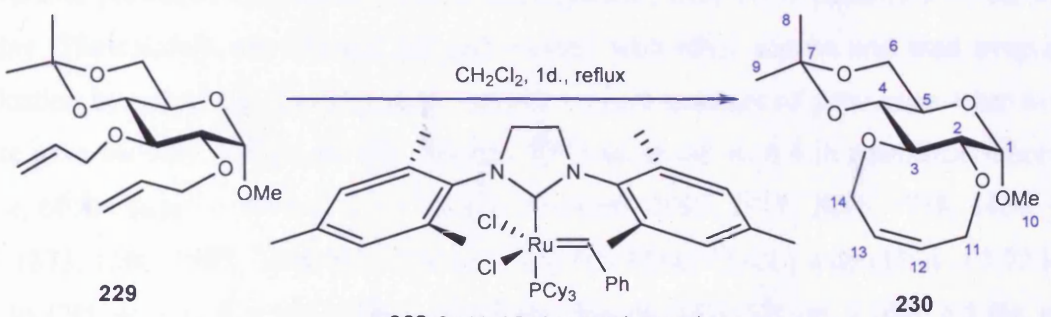
2,3-Di-*O*-allyl-4,6-*O*-isopropylidene-methyl-α-D-glucopyranoside 229



4,6-*O*-Isopropylidene-methyl-α-D-glucopyranoside **90a** (3 g, 12.82 mmol) was dissolved in distilled THF (30 ml) with stirring in a 100 mls stoppered flask. Potassium *tert*-butoxide (5.75 g, 51.28 mmol) was added to the sugar solution. A yellow solution formed immediately. To that solution allyl bromide (15.5 g, 128.2 mmol, 11 ml) was added dropwise and left stirring at r.t. under nitrogen for 24 hr. Water (50 mls) was added to the solution mixture which was extracted with dichloromethane (3 x 100 ml). The organic layer was dried over magnesium sulphate, filtered and concentrated to give the *title compound* **229** (3.3 g, 85 %) as a white powder. ^1H-NMR showed pure compound and no further purification was required. R_f 0.3 in petroleum ether:ethylacetate, 90:10; mp 46-48°C; $[\alpha]_D^{23}$ +84.2 (*c* 1.0, $CHCl_3$); IR (CH_2Cl_2), 2992, 2915, 1730, 1646, 1459, 1423, 1382, 1266, 1197, 1078, 997, 925, 852, 753 cm^{-1}; δ_H (500 MHz, C_6D_6) 6.08 (1H, ddt, *J* 17.2, 10.6, 5.3 Hz, 12-H), 5.97 (1H, ddt, *J* 17.2, 10.2, 5.3 Hz, 15-H), 5.4 (2H, dq, *J* 17.13, 1.8 Hz, 13- H, 16-H), 5.15 (2H, dq, *J* 10.5, 1.6 Hz, 13-H, 16-

H), 4.77 (1H, d, *J* 3.7 Hz, 1-H), 4.49 (2H, ddt, *J* 13.2, 5.2, 1.7 Hz, 14-H), 4.21 (2H, ddt, *J* 13.0, 5.1, 1.6 Hz, 11-H), 4.12 (1H, t, *J* 9.1 Hz, 3-H), 3.97 (1H, dd, *J* 9.7, 5.0 Hz, 6a-H), 3.93 (1H, dd, *J* 9.9, 5.0 Hz, 6b-H), 3.8 (2H, m, 4-H, 5-H), 3.55 (1H, dd, *J* 9.3, 3.7 Hz, 2-H) , 3.2 (3H, s, OMe), 1.6 (3H, s, CH_3-9), 1.42 (3H, s, CH_3-8). δ_C (DEPT, δ_H ^{1}H-^{13}C ^{1}*J*-COSY) 136.1 (CH, 5.97, 15-C), 135.5 (CH, 6.08,12-C), 116.0 (CH_2, 5.4, 5.15, 13-C), 115.3 (CH_2, 5.4, 5.15, 16-C), 99.2 (CH, 4.77, 1-C), 99.1 (C, 7-C), 80.0 (CH, 3.55, 2-C), 78.4 (CH, 4.12, 3-C), 75.3 (CH, 3.8, 4-C), 73.5 (CH_2, 4.21, 11-C), 72.2 (CH_2, 4.49, 14-C), 63.6 (CH, 3.8, 5-C), 62.8 (CH_2, 3.97, 3.93, 6-C), 54.7 (CH_3, 3.2, 10-C), 29.3 (CH_3, 1.6, 9-C), 18.8 (CH_3, 1.42 , 8-C); MS (EI/CI) *m/z* 315.2 (M + H); HRMS (ES^+) *calcd* for $C_{16}H_{26}O_6$ ($M+H^+$) 315.1802, found 315.1803, error – 0.31 ppm.

6-Methoxy-2,2-dimethyl-4,4a,6,6a,8,11,12a,12b-octahydro-1,3,5,7,12-pentaoxacycloocta [a]-naphthalene 230

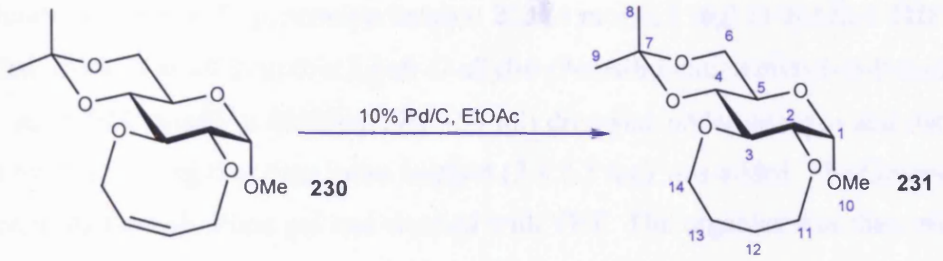


223 Grubbs' 2nd generation catalyst

Grubbs 2nd generation catalyst **223** (5 mol%, 15 mg) was added to a solution of 2,3-di-*O*-allyl-4,6-*O*-isopropylidene-methyl-α-D-glucopyranoside **229** (150 mg, 0.47 mmol) in distilled dichloromethane (15 ml), under nitrogen at reflux for one day. The Grubbs catalyst was filtered off through silica gel and washed with dichloromethane. The organics were then evaporated. The crude reaction mixture purified by column chromatography using a solvent gradient of petroleum ether, ethyl acetate to give the *title compound* **230** (100 mg, 75 %) as an oil. R_f 0.3 in petroleum ether:ethyl acetate, 50:50; $[\alpha]_D^{23}$ + 15.3 (*c* 1.0, $CHCl_3$); IR (film), 3417, 2955, 2925, 2855, 2363, 1634, 1252, 1097, 1036, 795 cm^{-1}; δ_H (500 MHz, $CDCl_3$) 5.64 (2H, dt, *J* 11.8, 4.4 Hz, 13-H, 12-H), 4.72 (1H, d, *J* 3.7 Hz, 1-H), 4.46 (1H, m, 11a-H), 4.37 (2H, d, *J* 5.54 Hz, 14-H), 4.19 (1H, ddd, *J* 16.1, 4.3, 1.3 Hz, 11b-H), 3.81 (1H, dd, *J* 11.7, 3.56 Hz, 6a-H), 3.78 (1H, dd, *J* 11.7, 4.6 Hz, 6b-H), 3.65 (1H, m, 5-H), 3.59 (1H, t, *J* 9.0 Hz, 3-H), 3.48 (1H, t, *J* 9.3 Hz, 4-H), 3.36 (3H, s, OCH_3), 3.29 (1H, dd, *J* 9.3, 3.7 Hz, 2-H), 1.45 (3H, s, CH_3), 1.38 (3H, s, CH_3). δ_C (DEPT, δ_H ^{1}H-^{13}C ^{1}*J*-COSY) 129.8 (CH, 5.64, 13-C), 128.7 (CH, 5.64, 12-C), 98.9 (CH, 4.72, 1-C),98.7 (C, 7-C), 81.8 (CH, 3.59, 3-C), 80.7 (CH,

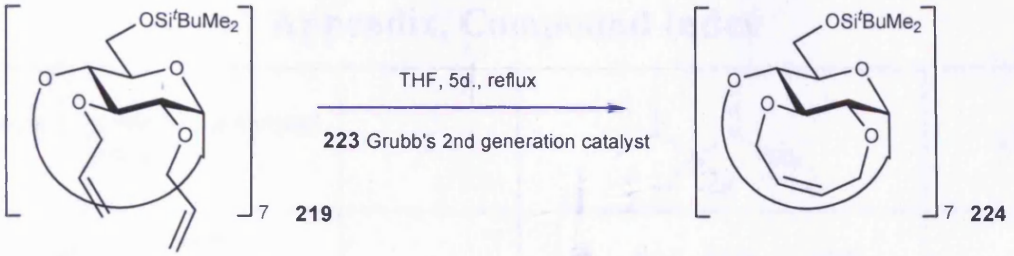
3.29, 2-C), 69.4 (CH, 3.65, 5-C), 69.3 (CH, 3.48, 4-C), 68.7 (CH_2, 4.46, 4.19, 11-C), 67.0 (CH_2, 4.37, 14-C), 61.6 (CH_2, 3.81, 3.78, 6-C), 54.2 (CH_3, 1.38, 10-C); MS (EI/CI) *m/z* 287.2 (M + H); HRMS (ES^+) *calcd* for $C_{14}H_{22}O_6$ ($M+H^+$) 287.1489, found 287.1487, error 0.7 ppm.

6-Methoxy-2,2-dimethyl-decahydro-1,3,5,7,12-pentaoxa-cycloocta[a]naphthalene 231



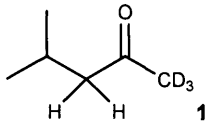
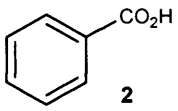
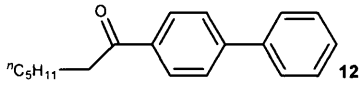
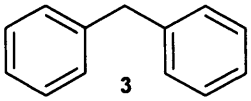
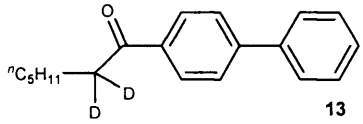
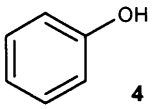
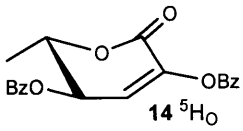
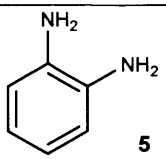
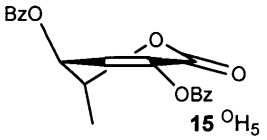
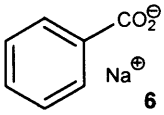
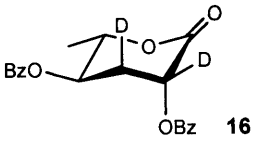
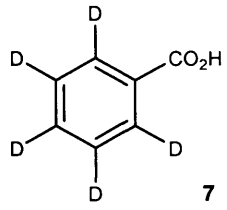
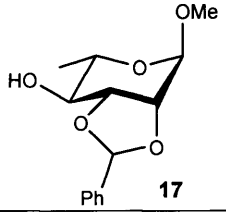
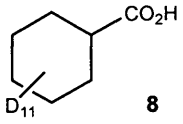
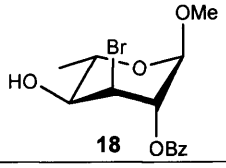
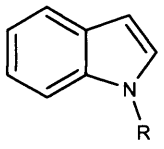
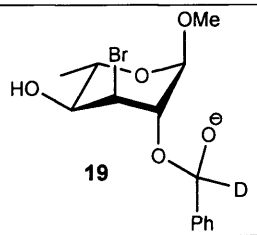
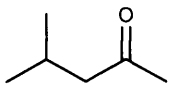
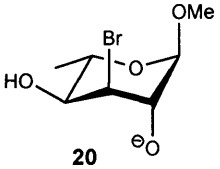
A solution of 6-methoxy-2,2-dimethyl-4,4a,6,6a,8,11,12a,12b-octahydro-1,3,5,7,12-pentaoxa-cycloocta[a]-naphthalene **230** (40 mg, 0.14 mmol) in ethyl acetate (4 ml) was hydrogenated at atmospheric pressure (water displacement hydrogenator) over 10 % palladium on carbon for one day. The catalyst was filtered off and washed with ethyl acetate and then evaporated. Purification by column chromatography using a solvent gradient of petroleum ether to ethyl acetate gave the *title compound* **231** (30 mg, 75%) as an oil. R_f 0.4 in petroleum ether:ethyl acetate, 60:40; $[\alpha]_D^{23}$ + 12.6 (*c* 1.0, $CHCl_3$); IR (film), 2962, 2915, 2845, 1738, 1669, 1446, 1408, 1373, 1260, 1097, 1018, 861, 799 cm^{-1}; δ_H (500 MHz, $CDCl_3$) 4.68 (1H,d, *J* 3.73 Hz, 1-H), 4.95 (2H, dt, *J* 11.5, 4.8 Hz observed, 11a-H, 14a-H), 3.80 (1H, dd, *J* 10.4, 4.8 Hz, 6a-H), 3.72 (1H, m, 11b-H), 3.65 (2H, m, 6b-H, 14b-H,), 3.62 (1H, t, *J* 9.0 Hz, 3-H), 3.58 (1H, m, 5-H), 3.48 (1H, t, *J* 9.4 Hz, 4-H), 3.35 (1H, m, 2-H), 3.35 (3H, s, $0CH_3$), 1.70 (4H, m, 12H, 13H), 1.42 (3H, s, CH_3), 1.38 (3H, s, CH_3). δ_C (DEPT, δ_H ^1H-^{13}C ^1J-COSY) 99.8 (CH, 4.68, 1-C), 77.2 (CH, 3.35, 2-C), 78.0 (CH, 3.62, 3-C), 76.6 (CH_2, 4.95, 3.72, 3.65, 11-C or 14-C), 78.8 (CH_2, 4.95, 3.72, 3.65, 11-C or 14-C) 72.6 (CH, 3.48, 4-C), 63.7 (CH, 3.58, 5-C), 62.5 (CH_2, 3.80, 3.65, 6-H), 55.1 (CH_3, 3.35, 10-C), 29.2 (CH_3, 1.42, 8-C), 19.1 (CH_3, 1.38, 9-C); MS (EI/CI) *m/z* 289.2 (M + H); HRMS (ES^+) *calcd* for $C_{14}H_{24}O_6$ (M + H^+) 289.1646, found 289.1647, error – 0.34 ppm.

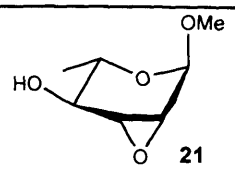
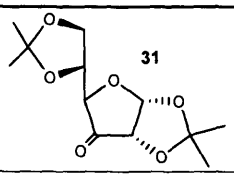
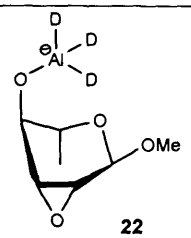
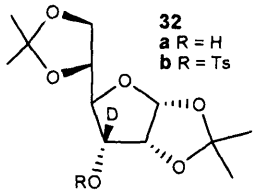
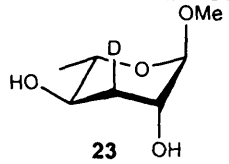
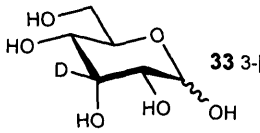
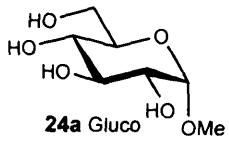
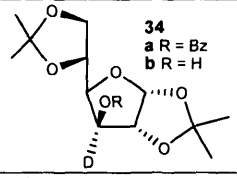
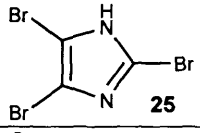
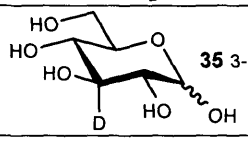
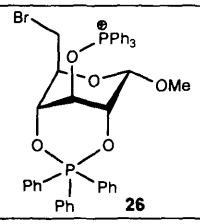
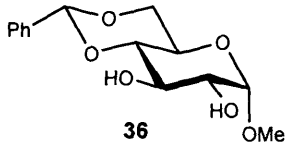
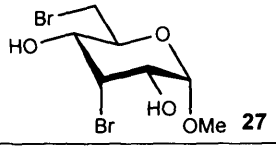
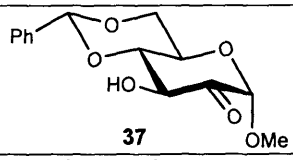
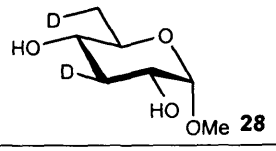
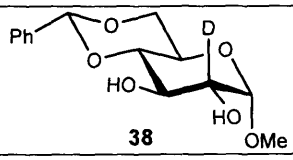
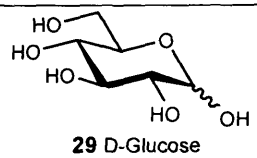
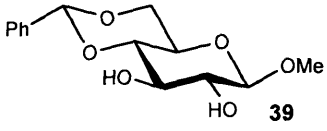
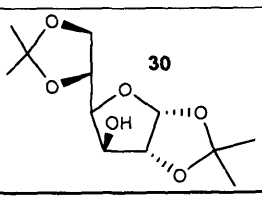
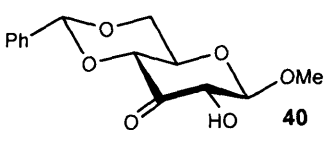
Attempted ring closing metathesis reaction of heptakis(2,3-di-*O*-allyl-6-*O*-*tert*-butyldimethylsilyl)-β-cyclodextrin 219

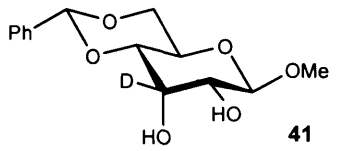
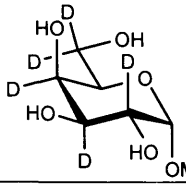
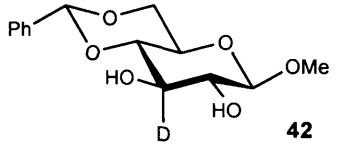
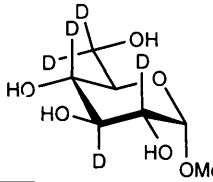
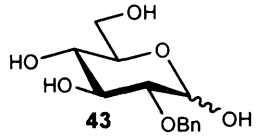
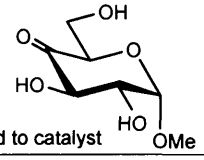
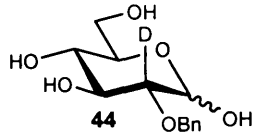
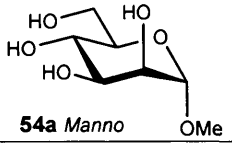
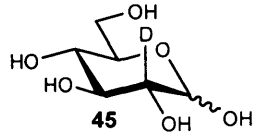
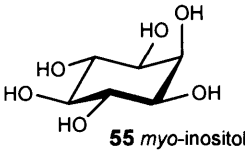
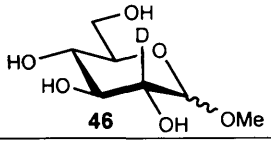
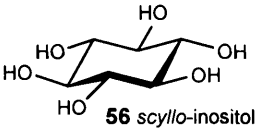
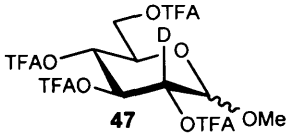
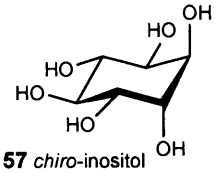
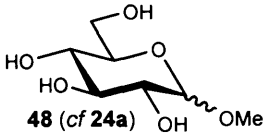
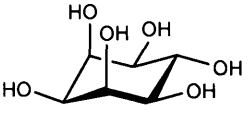
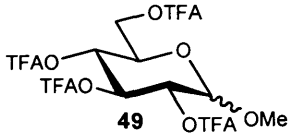
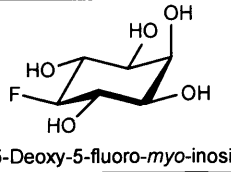
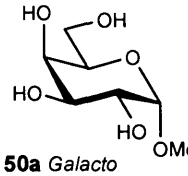
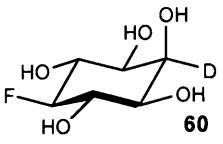


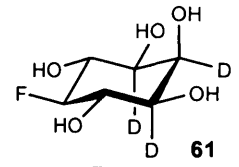
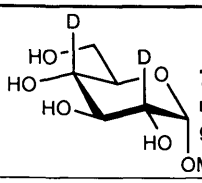
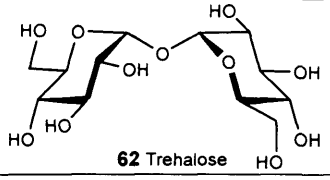
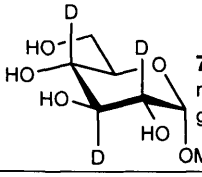
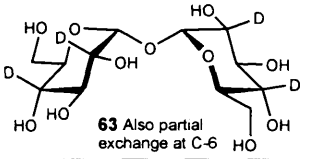
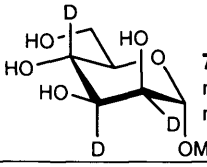
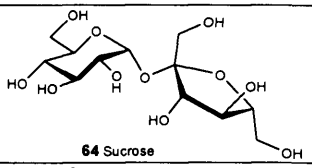
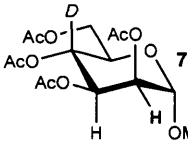
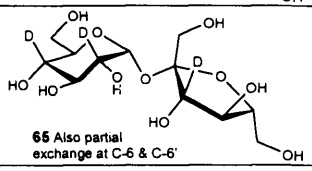
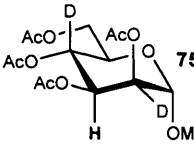
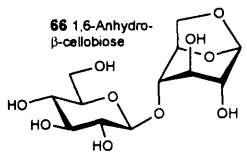
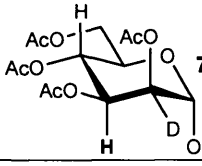
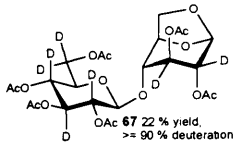
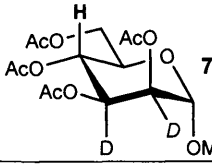
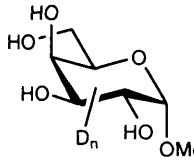
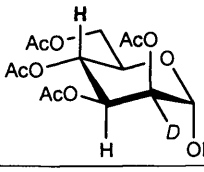
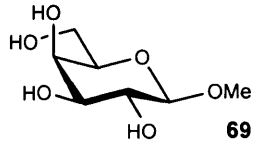
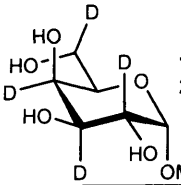
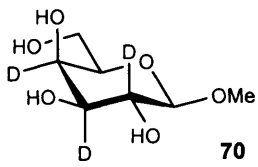
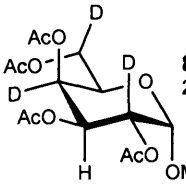
To a solution of Grubbs 2nd generation catalyst **223** (4 mol%, 1 mg) in distilled THF (70 ml), was added a solution of heptakis(2,3-di-*O*-allyl-6-*O*-*tert*-butyldimethylsilyl)-β-cyclodextrin **219** (70 mg, 0.028 mmol) in distilled THF (50 ml) dropwise under nitrogen and the reaction refluxed for 5 d. During that time more catalyst (2 x 0.3 mg) was added. The Grubbs catalyst was filtered off through silica gel and washed with THF. The organics was then evaporated. An attempt was made to purify the crude reaction mixture (54 mg) by column chromatography using a solvent gradient of hexane, diethyl ether but the complicated mixture could not be separated into pure fractions.

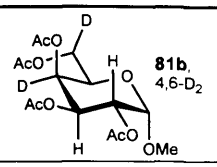
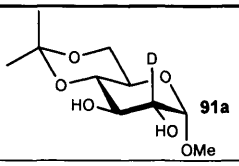
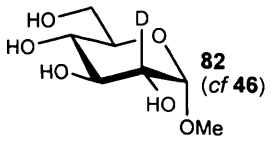
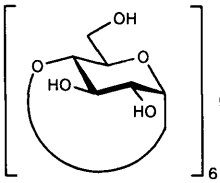
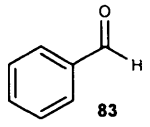
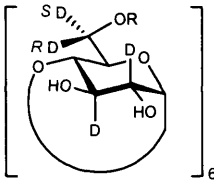
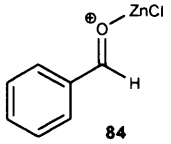
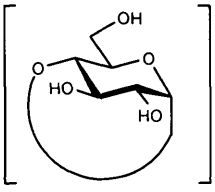
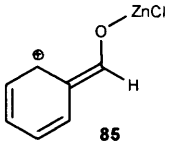
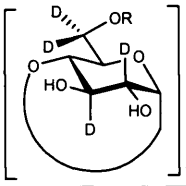
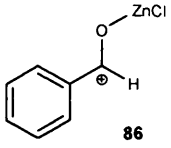
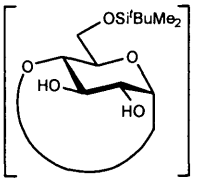
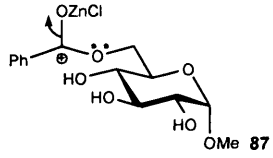
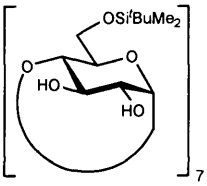
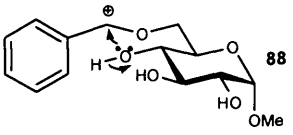
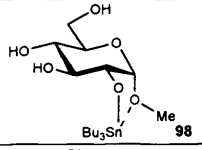
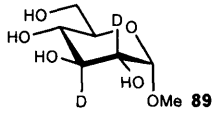
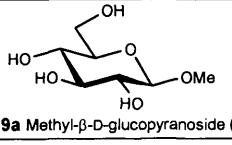
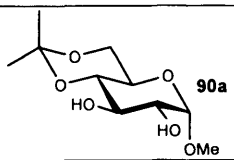
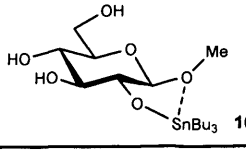
Appendix, Compound Index

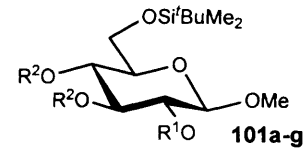
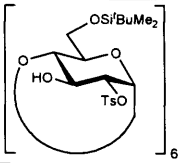
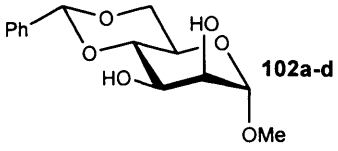
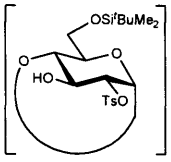
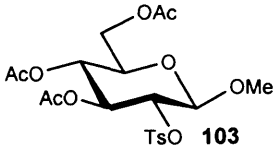
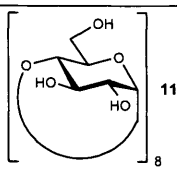
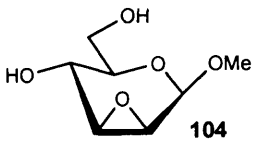
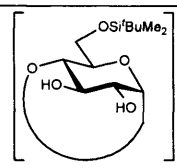
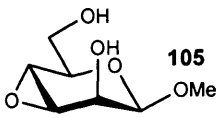
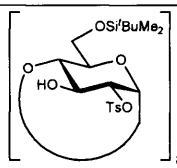
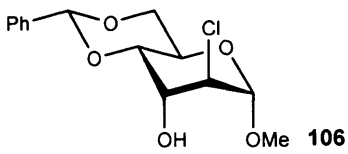
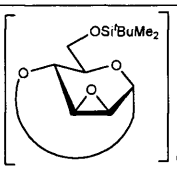
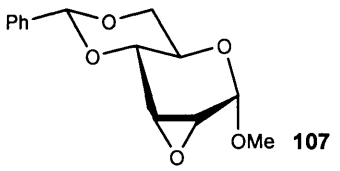
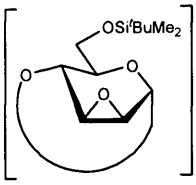
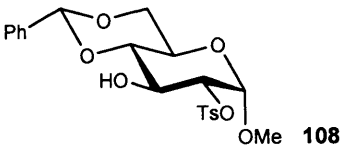
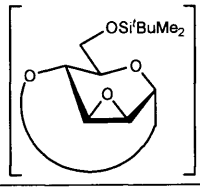
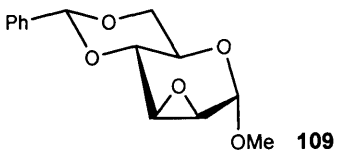
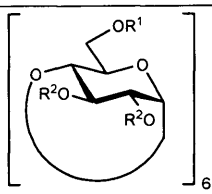
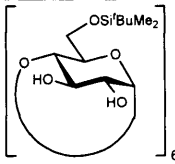
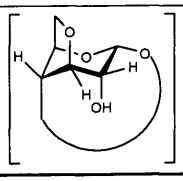
Scheme 1, partially deuterated RNA **1**	**1**	 **11**	**11**
 **2**	**2**	 **12**	**12**
 **3**	**3**	 **13**	**13**
 **4**	**4**	 **14** $^{5}H_{O}$	**14**
 **5**	**5**	 **15** $^{O}H_{5}$	**15**
 **6**	**6**	 **16**	**16**
 **7**	**7**	 **17**	**17**
 **8**	**8**	 **18**	**18**
 **9** **a** R = H **b** R = CH_3	**9**	 **19**	**19**
 **10**	**10**	 **20**	**20**

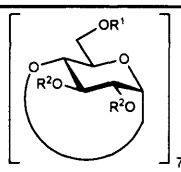
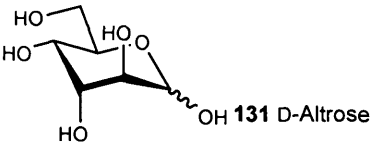
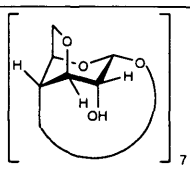
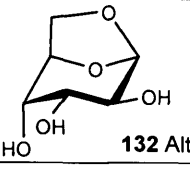
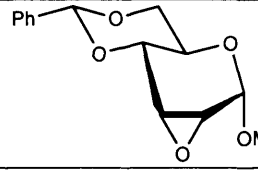
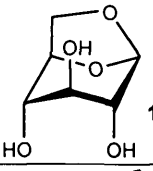
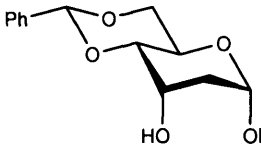
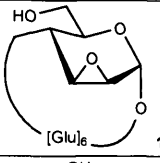
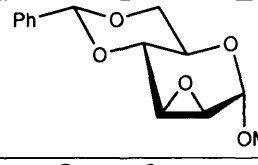
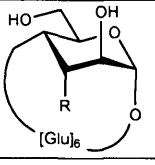
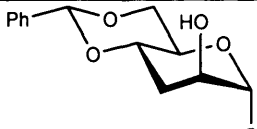
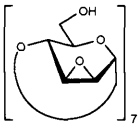
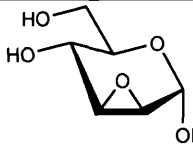
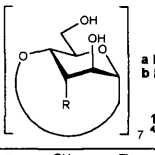
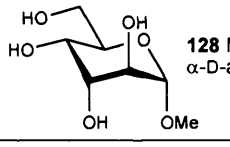
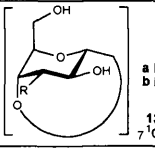
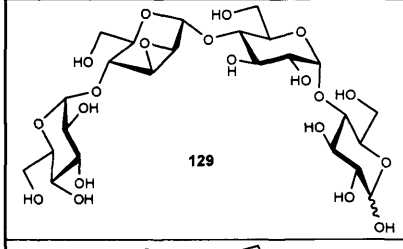
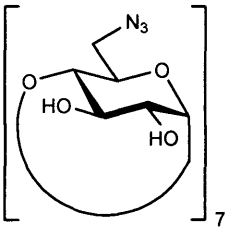
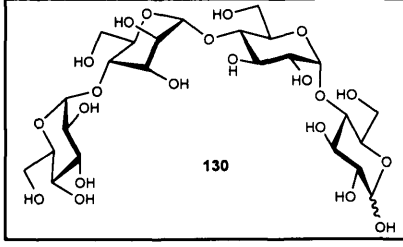
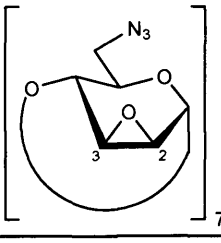
 **21**	**21**	 **31**	**31**
 **22**	**22**	 **32** **a** R = H **b** R = Ts	**32**
 **23**	**23**	 **33** 3-[$^2H_1$]-allose	**33**
 **24a** Gluco	**24** Methyl-α-D-glucopyranoside	 **34** **a** R = Bz **b** R = H	**34**
 **25**	**25**	 **35** 3-[$^2H_1$]-glucose	**35**
 **26**	**26**	 **36**	**36** (4,6-*O*-benzylidene)-methyl-α-D-glucopyranoside
 **27**	**27**	 **37**	**37**
 **28**	**28**	 **38**	**38**
 **29** D-Glucose	**29** D-Glucose	 **39**	**39** 4,6-*O*-benzylidene)-methyl-β-D-glucopyranoside
 **30**	**30** Diacetone glucose (DAG)	 **40**	**40**

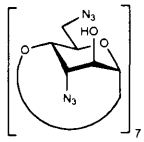
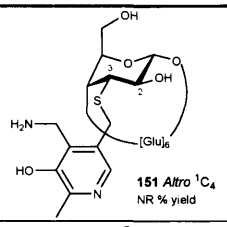
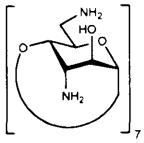
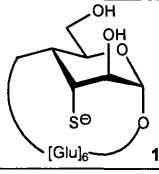
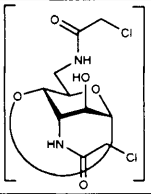
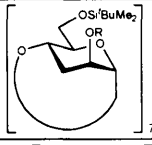
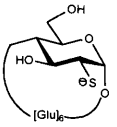
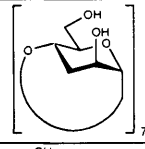
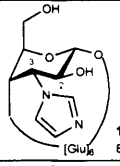
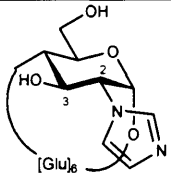
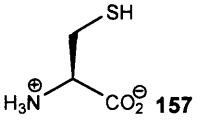
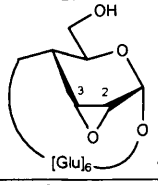
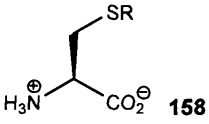
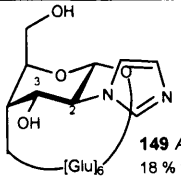
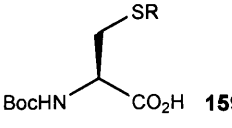
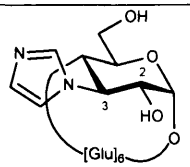
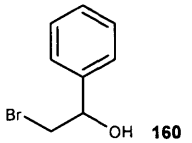
 **41**	**41**	 **51** *Galacto*	**51**
 **42**	**42**	 **52** *Gluco*	**52**
 **43**	**43**	 **53** bound to catalyst	**53**
 **44**	**44**	 **54a** *Manno*	**54**
 **45**	**45**	 **55** *myo*-inositol	**55** *myo*-Inositol
 **46**	**46**	 **56** *scyllo*-inositol	**56** *scyllo*-Inositol
 **47**	**47**	 **57** *chiro*-inositol	**57** *chiro*-Inositol
 **48** (*cf* **24a**)	**48**	 **58** *epi*-inositol	**58** *epi*-Inositol
 **49**	**49**	 **59** 5-Deoxy-5-fluoro-*myo*-inositol	**59**
 **50a** *Galacto*	**50** Methyl-α-D-galactopyranoside	 **60**	**60**

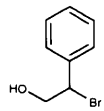
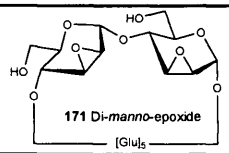
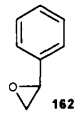
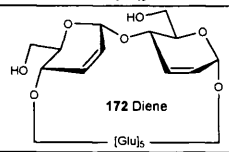
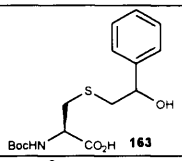
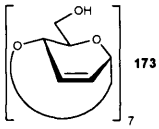
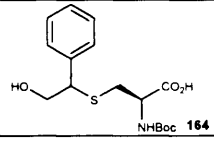
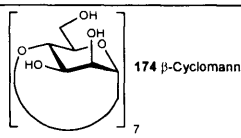
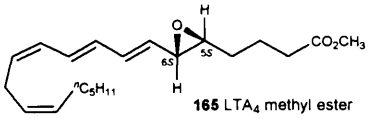
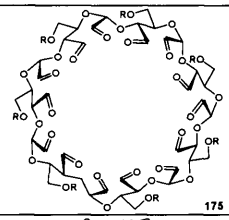
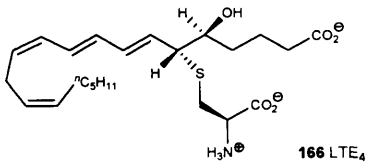
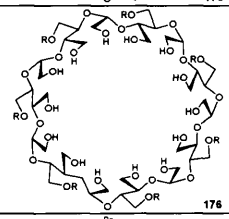
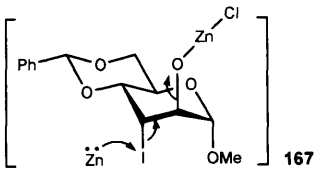
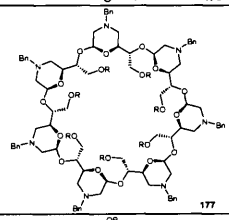
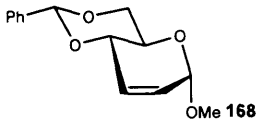
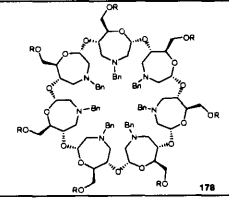
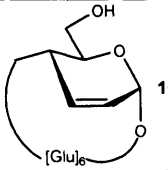
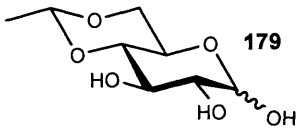
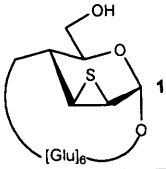
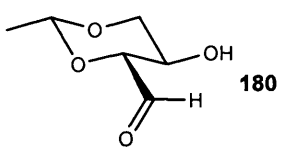
 **61**	**61**	 **71a** (2,4-$^2H_2$)-methyl-α-D-glucopyranoside	**71**
 **62** Trehalose	**62**	 **72a** (2,3,4-$^2H_3$)-methyl-α-D-glucopyranoside	**72**
 **63** Also partial exchange at C-6	**63**	 **73a** (2,3,4-$^2H_3$)-methyl-α-D-mannopyranoside	**73**
 **64** Sucrose	**64**	 **74b**, 4-D_1	**74**
 **65** Also partial exchange at C-6 & C-6'	**65**	 **75b**, 2,4-D_2	**75**
 **66** 1,6-Anhydro-β-cellobiose	**66**	 **76b**, 2-D_1	**76**
 **67** 22 % yield, >= 90 % deuteration	**67**	 **77b**, 2,3-D_2	**77**
 **68a**	**68**	 **78b**, 2-D_1 identical to **76b**	**78**
 **69**	**69** β-Methyl-D-galactopyranoside	 **79a**, 2,3,4,6-D_4	**79**
 **70**	**70**	 **80b**, 2,4,6-D_3	**80**

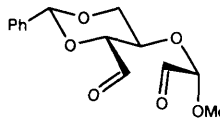
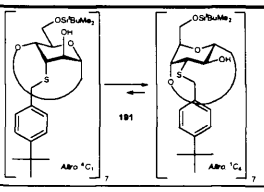
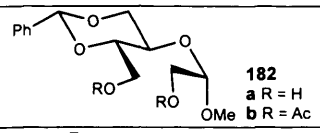
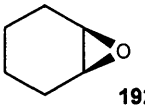
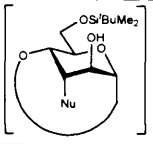
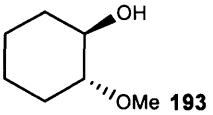
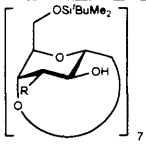
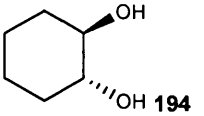
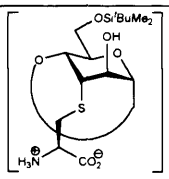
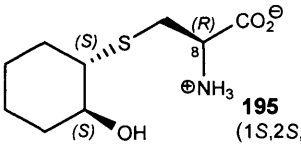
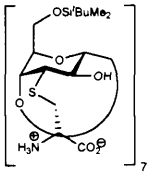
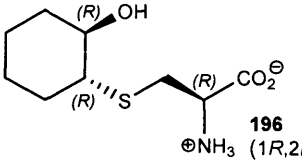
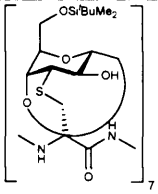
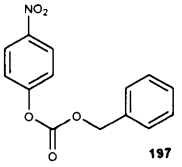
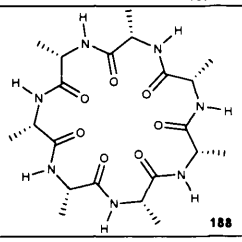
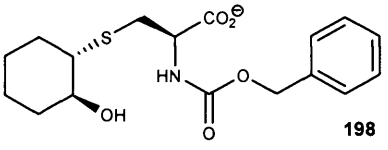
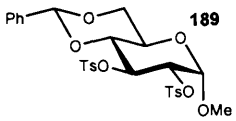
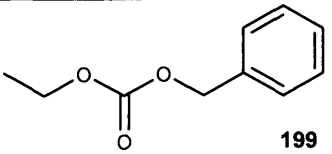
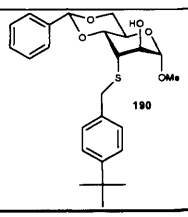
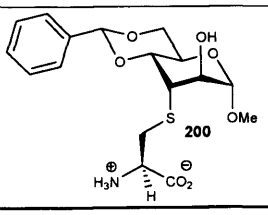
 **81b**, 4,6-D_2	**81**	 **91a**	**91**
 **82** (*cf* **46**)	**82**	 **92** α-Cyclodextrin	**92** α-Cyclodextrin
 **83**	**83**	 **93** Hexakis-(2,3,6,6'-[$^2H_4$]-) α-cyclodextrin	**93**
 **84**	**84**	 **94** β-Cyclodextrin	**94** β-Cyclodextrin
 **85**	**85**	 **95** Heptakis-(2,3,6,6'-[$^2H_4$])-β-cyclodextrin	**95**
 **86**	**86**	 **96** 6-*O*-^{t}butyl-dimethylsilyl-β-cyclodextrin	**96**
 **87**	**87**	 **96** Heptakis-(6-*O*-^{t}butyl-dimethylsilyl)-β-cyclodextrin	**97**
 **88**	**88**	 **98**	**98**
 **89**	**89**	 **99a** Methyl-β-D-glucopyranoside (*cf* **48**)	**99**
 **90a**	**90**	 **100**	**100**

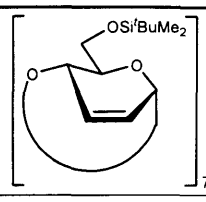
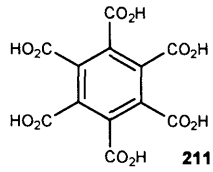
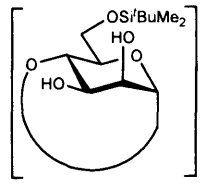
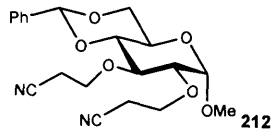
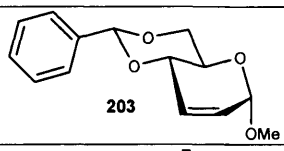
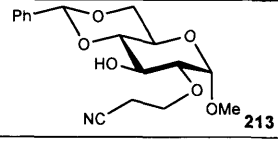
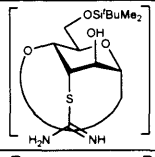
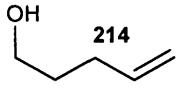
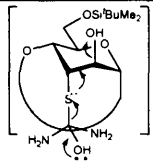
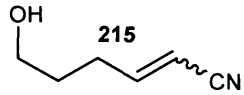
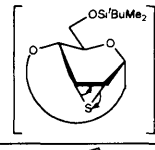
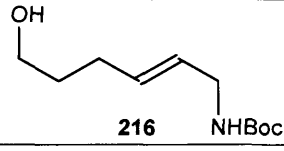
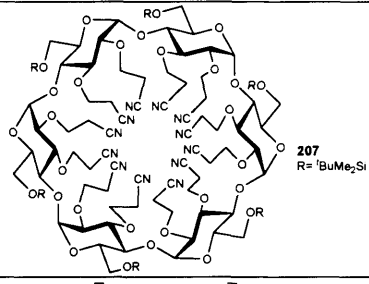
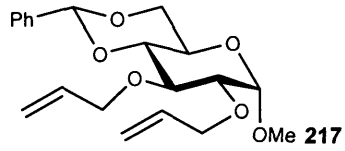
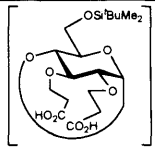
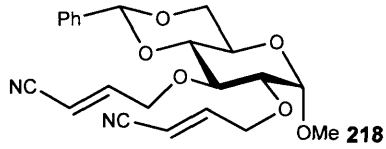
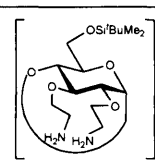
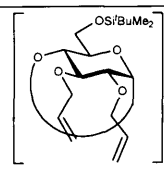
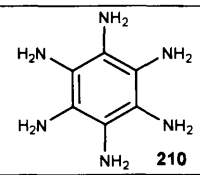
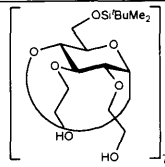
 **101a-g**	**101**	 **111** Hexakis (6-*O*-^{t}butyl-dimethylsilyl-2-*O*-*p*-toluene-sulfonyl)-α-cyclodextrin	**111**
 **102a-d**	**102**	 **112** Heptakis (6-*O*-^{t}butyl-dimethylsilyl-2-*O*-*p*-toluene-sulfonyl)-β-cyclodextrin	**112**
 **103**	**103**	 **113** γ-Cyclodextrin	**113** γ-Cyclodextrin
 **104**	**104**	 **114** Octakis (6-*O*-^{t}butyl-dimethyl-silyl)-γ-cyclodextrin	**114**
 **105**	**105**	 **115** Octakis (6-*O*-^{t}butyl-dimethylsilyl-2-*O*-*p*-toluene-sulfonyl)-γ-cyclodextrin	**115**
 **106**	**106**	 **116** Hexakis (2,3-anhydro-6-*O*-^{t}butyl-dimethylsilyl)-α-cyclodextrin	**116**
 **107**	**107**	 **117** Heptakis (2,3-anhydro-6-*O*-^{t}butyl-dimethylsilyl)-β-cyclodextrin	**117**
 **108**	**108**	 **118** Octakis (2,3-anhydro-6-*O*-^{t}butyl-dimethylsilyl)-γ-cyclodextrin	**118**
 **109**	**109**	 **119** **a** R^1 = Ts, R^2 = H **b** R^1 = Ts, R^2 = Bz **c** R^1 = I, R^2 = H	**119**
 **110** Hexakis (6-*O*-^{t}butyl-dimethylsilyl)-α-cyclodextrin	**110**	 **120**	**120** 3,6-anhydro-α-cyclodextrin

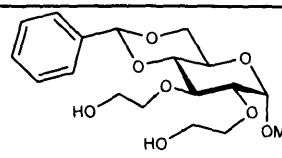
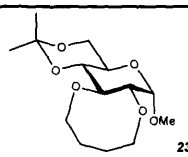
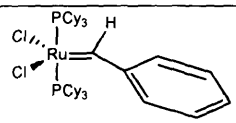
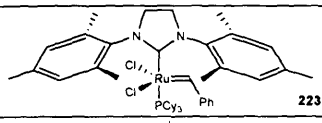
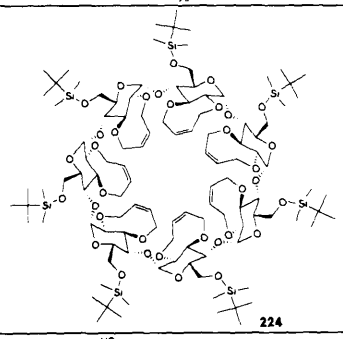
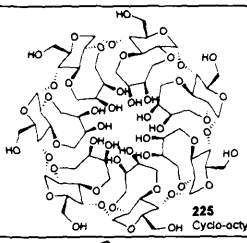
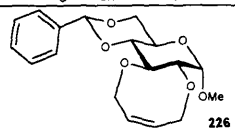
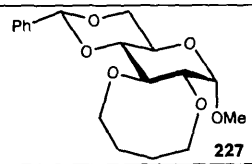
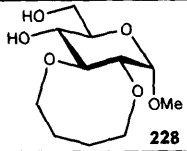
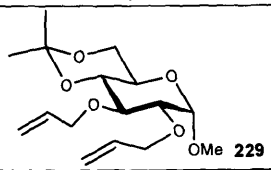
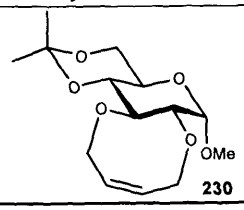
 **121** **a** R^1 = Ts, R^2 = H **b** R^1 = I, R^2 = H	**121**	 **131** D-Altrose	**131** D-Altrose
 **122**	**122**	 **132** Altrosan	**132** Altrosan
 **123**	**123**	 **133** Levoglucosan	**133** Levoglucosan
 **124**	**124**	 **134**	**134**
 **125**	**125**	 **135** **a** R = OH **b** R = NH_2	**135**
 **126**	**126**	 **136**	**136**
 **127**	**127**	 **137** $^4C_1$ **a** R = OH **b** R = NH_2	**137**
 **128** Methyl-α-D-altropyranoside	**128**	 **138** $^1C_4$ **a** R = OH **b** R = NH_2	**138**
 **129**	**129**	 **139**	**139**
 **130**	**130**	 **140**	**140**

 7 **141**	**141**	 **151** *Altro* $^1C_4$ NR % yield	**151**
 7 **142**	**142**	 **152**	**152**
 7 **143**	**143**	**153** 3,3'-Sulfide dimer 30 % yield	**153**
 **144** **a** R = H **b** R = Me **c** R = Bn 7	**144**	 **154**	**154**
 7 **145**	**145**	**155** 2,2'-Sulfide dimer 13 % yield	**155**
 **146** *Altro* $^1C_4$ 83 % yield	**146**	**156** 2,3'-Sulfide dimer 0 % yield	**156**
 **147** *Gluco* $^4C_1$ 3.8 % yield	**147**	 **157**	**157**
 **148** *Allo*	**148**	 **158**	**158**
 **149** *Altro* $^1C_4$ 18 % yield	**149**	 **159**	**159**
 **150** *Gluco* $^4C_1$ 55 % yield	**150**	 **160**	**160**

 161	**161**	 171 Di-*manno*-epoxide	**171**
 162	**162**	 172 Diene	**172**
 163	**163**	 173	**173**
 164	**164**	 174 β-Cyclomannin	**174**
 165 LTA_4 methyl ester	**165** Leukotriene A_4	 175	**175**
 166 LTE_4	**166** Leukotriene E_4	 176	**176**
 167	**167**	 177	**177**
 168	**168**	 178	**178**
 169 Alkene	**169**	 179	**179**
 170 Thiirane	**170**	 180	**180**

 **181**	**181**		**191**
 **182** **a** R = H **b** R = Ac	**182**	 **192**	**192**
 **183** $^4C_1$	**183**	 **193**	**193**
 **184** $^1C_4$	**184**	 **194**	**194**
 **185** *Altro* $^4C_1$	**185**	 **195** (1*S*,2*S*,8*R*)	**195**
 **186** *Altro* $^1C_4$	**186**	 **196** (1*R*,2*R*,8*R*)	**196**
 **187**	**187**	 **197**	**197**
 **188**	**188**	 **198**	**198**
 **189**	**189**	 **199**	**199**
 **190**	**190**	 **200**	**200**

	**201**		**211**
	**202**		**212**
	**203**		**213**
	**204**		**214**
	**205**		**215**
	**206**		**216**
	**207**		**217**
	**208**		**218**
	**209**		**219**
	**210**		**220**

 221	221	 231	231
 222	222		
 223	223		
 224	224		
 225 Cyclo-octyl-"homo"-β-cyclodextrin	225		
 226	226		
 227	227		
 228	228		
 229	229		
 230	230		

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