This is an Accepted Manuscript, which has been through the Royal Society of Chemistry peer review process and has been accepted for publication.

Accepted Manuscripts are published online shortly after acceptance, before technical editing, formatting and proof reading. Using this free service, authors can make their results available to the community, in citable form, before we publish the edited article. We will replace this Accepted Manuscript with the edited and formatted Advance Article as soon as it is available.

You can find more information about Accepted Manuscripts in the Information for Authors.

Please note that technical editing may introduce minor changes to the text and/or graphics, which may alter content. The journal’s standard Terms & Conditions and the Ethical guidelines still apply. In no event shall the Royal Society of Chemistry be held responsible for any errors or omissions in this Accepted Manuscript or any consequences arising from the use of any information it contains.
Stable Analogues of Nojirimycin – Synthesis and Biological Evaluation of Nojiristegine and manno-Nojiristegine

Agnete H. Viuff, a Louise M. Besenbacher, a Akiko Kamoric, c Mikkel T. Jensen, a Mogens Kilian, b Atsushi Kato c and Henrik H. Jensen*, a

a Department of Chemistry, Aarhus University, Langelandsgade 140, 8000, Aarhus C, Denmark.

b Department of Biomedicine, Aarhus University, Wilhelm Meyers allé 4, 8000 Aarhus C, Denmark.

c Department of Hospital Pharmacy, University of Toyama, 2630 Sugitani, Toyama 930-01940, Japan.

hhj@chem.au.dk

Abstract

Two novel iminosugars called nojiristegines, being structural hybrids between nor-tropane alkaloid calystegine and nojirimycins, have been synthesised and found to be stable molecules despite the presence of a hemiaminal functionality. The synthesised iminosugars were evaluated against a panel of glycosidases and the best inhibition (IC50), found against α-glucosidases, was in the micromolar region. The compounds were also evaluated as potential antibiotics but no useful level of activity was observed.

Introduction

In 1965, the first naturally occurring iminosugar 5-amino-5-deoxy-glucospyranose was found1 and subsequently named nojirimycin, after its isolation from Streptomyces nojiriensis. 2 In 1984, the mannose derivative of nojirimycin (manno-nojirimycin or nojirimycin B), was discovered by Niwa et al.3 who isolated it from Streptomyces lavendulae SF-425. One of the main reasons for the
interest in these molecules was their potent glycosidase inhibition established in 1970 by Niwa et al. for nojirimycin⁴ and by Legler and Julich for manno-nojirimycin.⁵ These two prominent alkaloids of the iminosugar/azasugar family⁶ have spawned a whole area of research mainly due to their potent inhibition of glycosidases. Several analogous molecules have been found in Nature or synthesised by creative chemists."⁷,⁸,⁹

Figure 1. Structure of known iminosugars/azasugars. Box shows the structural hybrid molecules synthesised and evaluated in this study.

In general, iminosugars/azasugars have been reported to have important therapeutic potential in diseases such as diabetes, cancer, lysosomal storage diseases, viral and bacterial diseases.¹⁰,¹¹,¹² Recently Warren et al. published their work on the iminosugar BCX4430 which is a broad spectrum antiviral agent that inhibits infection with e.g. Ebola and other highly fatal viruses.¹³

Despite their potent glycosidase inhibition, both nojirimycin and manno-nojirimycin have little potential to be therapeutic candidates, due to their instability caused by their hemiaminal functionality, which eventually undergoes the Amadori rearrangement.¹⁴,¹⁵ This inherent drawback of the nojirimycins has been addressed by Inouye et al. through chemical reduction of nojirimycin to arrive at 1-deoxynojirimycin (Figure 1),¹⁶ which was later found by Yagi et al. to be naturally present in mulberry trees.¹⁷ The stability issue of nojirimycins has probably caused the scientific community to mainly focus on 1-deoxy-nojirimycin and its analogues at the expense of
nojirimycins. However, one finding often cited in the literature is the antibiotic activity of the nojirimycins, which was recognised very shortly after their discovery. Ishida et al. reported nojirimycin to have antibiotic activity against bacterial strains from the otherwise dissimilar genera, Shigella flexneri, Sarcina lutea and Xanthomonas oryzae. Surprisingly, the compound had weak or no activity towards closely related bacteria such as Escherichia coli and even other strains of Shigella. Manno-nojirimycin was found by their discoverers to have weak antibiotic activity against Xanthomonas oryzae but no activity against other bacteria tested.

The hemi-aminal functionality of alkaloids is not always as unstable as that of nojirimycins. Among the naturally occurring iminosugars, the calystegines possess a bicyclic structure that prevents imine formation and thereby Amadori rearrangement. Calystegines are glycosidase inhibitors, but have a hydroxyl group in the place of the hydroxymethyl group of the natural substrate. Inspired by the calystegine core structure and the unstable noeuromycins from the Bols laboratories we previously reported on the synthesis of both glucose, galactose and glucuronic acid configured synthetic hybrids of calystegines and noeuromycins (Figure 1). These compounds have been christened noeurostegines.

In this report we describe the synthesis, initial inhibitory data and potential antibiotic activity of stable hybrids between nojirimycin/manno-nojirimycin and calystegines. We here refer to these molecules as nojiristegine (1) and manno-nojiristegine (2).

**Synthesis**

The strategy for the synthesis of the nojiristegines was largely inspired by our previously reported synthesis of noeurostegine and calystegine A. The synthesis of both target molecules (1 and 2) was performed from methyl α-D-glucopyranoside (3) and methyl α-D-mannopyranoside (4), respectively. In a series of known steps these were separately converted into 2,3,4-tri-O-benzyl-6-iodo-6-deoxy pyranosides 5 and 6 (Scheme 1).
Scheme 1. Synthesis of known 6-deoxy-6-iodo glycosides.

For the synthesis of glucose-configured nojiristegine (1), the 6-iodo-glucoside 5 was subjected to zinc-mediated fragmentation under sonication in THF-H$_2$O giving an aldehyde, which was used directly in the following Barbier reaction, without further purification. This resulted in two diastereoisomeric allylic alcohols (8R and 8S) as an inseparable 2:3 mixture in 76 % yield over 2 steps (Scheme 2).

Scheme 2. Synthesis of nojiristegine (1).
The mixture of dienes 8 possessing a free secondary alcohol proved incapable of undergoing ring-closing metathesis with the Grubbs-Hoveyda 2nd generation catalyst. Subsequent to alcohol acetylation with acetic anhydride, triethyl amine and DMAP in 85% yield, the stereoisomers 9 could if needed be separated and ring closing metathesis promoted by Grubbs-Hoveyda 2nd generation catalyst went smoothly in 93% yield on the diastereoisomeric mixture from the previous reaction. When working with the single isomers, the C6 stereochemistry could be deduced by 1D and 2D 1H-NMR experiments and assigned based on $^{3}J_{HH}$ coupling constants for 10 and 11. This established the major product from the Barbier allylation to be 8S, but since both allylic alcohols (8R and 8S) eventually would be oxidised to the same ketone, the synthesis of nojiristegine (1) progressed with a mixture of isomers. The acetyl groups were then removed from a mixture of ring closed acetates 10 and 11 by Zemplén conditions and the resulting alcohol was next protected by the more robust $p$-methoxybenzyl (PMB) ether with PMB-Cl/NaH in 57% yield over 2 steps. The cycloheptene was next subjected to a hydroboration/oxidation sequence to give the two regioisomeric secondary alcohols 13 and 14, of which each was found to be a single diastereoisomer. The ratio of the two secondary alcohols was found to be different depending on whether the reaction was conducted on the R or the S-isomer. The S-isomer gave 1:2.7 in favour of the desired alcohol, while the R-isomer gave 1.7:1 in favour of the undesired alcohol. Performing the reaction on the mixture of stereoisomers originating from the Barbier reaction gave the desired alcohol in a yield of 46%. The alcohol could now be oxidised using Dess-Martin periodinane (DMP) to give the ketones 15, which could be used as a precursor for the introduction of both an azide and a hydroxymethyl substituent. Accordingly, ketone 15 was first converted into the chloroepoxides by reaction with lithiated dichloromethane followed by treatment with DBU. These compounds (17) were then reacted with tetrabutylammonium azide to give aldehydes 18, which were reduced with NaBH$_4$ and the resulting alcohols protected as benzyl ethers to give the desired azides 20 in 34% over 6 steps. This sequence of known reactions was easily monitored by TLC analysis but intermediate structural assignment by NMR spectroscopy was highly challenging. Intermediates 16-18 were found to be unstable and chromatographic purification was therefore omitted. Identity was established by mass spectrometry alone and the yield reported is for the full sequence (15-20).

Given the nature of these reactions it cannot be ruled out, that other isomers than the one shown in Scheme 2 was formed but none could be isolated. The stereochemistry of the product could not be
verified until the last step of the synthesis, since it was not possible to obtain crystals of high
enough quality for a crystal structure.

The PMB ether could now be deprotected using TFA and the resulting alcohol oxidised using DMP
in 65 % over two steps. Basic hydrogenation with triethylamine resulted in reduction of the azide
and spontaneous cyclisation into the benzyl protected nojiristegine (22), as established by a
hemiaminal \(^{13}\text{C}\)-NMR chemical shift of 92 ppm. Hydrogenolysis using Pearlman’s catalyst was
then used to remove the remaining benzyl groups in quantitative yield. The presence of exclusively
large coupling constants as a result of axial-axial \(^1\text{H}-^1\text{H}\) geometry verifies the stereochemistry of the
quaternary centre originating from the introduction of the azido group.

Nojiristegine (1) was hence successfully synthesised in 21 linear steps and was found to be fully
shelf stable and stable in solution over several weeks.

The synthesis of the epimeric manno-nojirimycin (2) followed a similar synthetic route. The iodide
6 (Scheme 1) was subjected to zinc-mediated fragmentation and subsequent Barbier reaction with
allyl bromide.\(^{27}\) This resulted in two inseparable diastereoisomers in a 2.2:1 mixture in 76% yield
(Scheme 3). The stereochemical outcome was established via derivatisation at a later stage (vide
infra). The diastereoisomeric mixture was then acetylated using acetic anhydride and DMAP in
pyridine and subsequently subjected to ring-closing metathesis. It was possible to separate the two
stereoisomers (26 and 27) as the ring-closed acetates in 19% and 50% over two steps, respectively
(Scheme 3).
Scheme 3. Synthesis towards manno-nojiristegine (2) and establishing the stereochemical outcome of the Barbier reaction.
For structural assignment, the two acetates (26 and 27) were then individually deacylated by Zemplén conditions, O-benzylated by NaH/BnBr and the double bond reduced. This gave two isomers (32 and 33) where one could be established by $^{13}$C-NMR to be symmetrical. Consequently, the major/minor isomer from the Barbier reaction was established as the 6R/6S isomer, respectively (Scheme 3). As for the synthesis of nojiristegine (1, Scheme 2) both diastereoisomers (26 and 27) could principally be used in the next steps but the synthesis was completed with 27 alone (Scheme 4).

The alcohol 29 was protected as a PMB ether in 84% over two steps to give cycloheptene 34, which was subjected to hydroboration and oxidative work up to afford two regioisomeric alcohols in a 2.1:1 ratio as sole diastereoisomers in a combined yield of 78%. The desired major alcohol isomer 36 was then oxidised using Dess-Martin periodinane (DMP). The resulting ketone underwent the same sequence of reactions as in the nojiristegine synthesis (Scheme 2) to install benzyloxymethyl and azide substituents. Hence, 37 was treated with lithiated dichloromethane and then DBU to give the chloro-epoxide 39, which could be opened to compound 40 by treatment with tetrabutylammonium azide in DMSO. The aldehyde function was then reduced using sodium borohydride and the resulting alcohol protected as a benzyl ether. This gave the desired product 42 in a 1.7:1 mixture with its epimer 43 in a total yield of 38% over 5 steps from ketone 37. The PMB ether of 42 could now be deprotected using 2,3-dichloro-5,6-dicyano-1,4-benzoquinone (DDQ) and the resulting alcohol oxidised to the ketone (44) in 39 % over two steps. Azide reduction first afforded the spontaneously cyclised 45 as shown by the presence of a hemiaminal C-atom ($\delta_c$ 91.2 ppm) and next hydrogenolysis to remove benzyl ether protection gave manno-nojiristegine (2) in quantitative yield. The stereochemistry originating from the azide introduction step could now be verified by analysis of coupling constants originating from the $^1$H nuclei of the bicyclic molecule 2. The $^1$H-NMR spectrum showed one small (4.0 Hz) and one large coupling constant (9.2 Hz) as expected from an equatorial/axial (H2/H3) and axial/axial (H3/H4) arrangement, respectively. If the quaternary carbon atom bearing the nitrogen had been opposite (46), two small coupling constants would be expected upon cyclisation (Scheme 5).
Scheme 4. Synthesis of *manno*-nojiristegine (2) from cycloheptenol 29.
Scheme 5. The different products formed from the two possible azide isomers 44 and 46.

The synthesis produced manno-nojiristegine in 21 steps from the unprotected methyl manno-pyranoside (4). The compound was stable and did not show any degradation after several weeks on the shelf.

Concerning the modest stereoselectivity in the Barbier reactions of aldehydes 7 and 23, we found the two major products (8S and 24R) to possess opposite stereochemistry of the newly formed stereogenic centre (Entry 1 and Entry 2). Interestingly, Skaanderup and Madsen found a more pronounced selectivity for both indium, magnesium and zinc in the analogous Barbier reaction with allyl bromide on the corresponding N-benzyl imines (Entry 6-8).\textsuperscript{31} Except for the metal zinc in reaction with the N-benzyl imine of 23, the stereochemical outcome was reversed compared to the aldehyde substrates (7 and 23).

For a complete comparison, we also prepared the aldehyde analogous to 7 and 23 from D-galactose\textsuperscript{26} (Entry 3) and again found the reaction to be selective for the C6-S isomer as compound 7 originating from D-glucose demonstrating the dependence on C-\(\alpha\) stereochemistry. This was also opposite to the corresponding N-benzyl imine (Entry 8).\textsuperscript{31}

Interestingly, the indium mediated Barbier allylation was previously found to be dependent on the C-\(\beta\) stereochemistry in the synthesis of noeurostegine and galacto-noeurostegine (Entry 4 and Entry 5), where the aldehyde is flanked by a benzyloxy group.\textsuperscript{22,24} For these examples, the aldehyde leading to noeurostegine (Entry 4) having the same stereochemistry as 7, the S-product dominated, while the opposite was true for the aldehyde leading to galacto-noeurostegine (Entry 5).
Table 1. The stereoselectivity of the Barbier allylation of aldehydes using different metals.

<table>
<thead>
<tr>
<th>Entry</th>
<th>Reference</th>
<th>Starting material</th>
<th>Product</th>
<th>Metal</th>
<th>R/S</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>This work</td>
<td>![Image of starting material 7]</td>
<td>![Image of product 8]</td>
<td>In</td>
<td>2:3</td>
</tr>
<tr>
<td>2</td>
<td>This work</td>
<td>![Image of starting material 23]</td>
<td>![Image of product 24]</td>
<td>In</td>
<td>2.2:1</td>
</tr>
<tr>
<td>3</td>
<td>This work</td>
<td>![Image of starting material]</td>
<td>![Image of product]</td>
<td>In</td>
<td>1:2.1</td>
</tr>
<tr>
<td>4</td>
<td>22</td>
<td>![Image of starting material]</td>
<td>![Image of product]</td>
<td>In</td>
<td>1:1.7</td>
</tr>
<tr>
<td>5</td>
<td>24</td>
<td>![Image of starting material]</td>
<td>![Image of product]</td>
<td>In</td>
<td>3:2</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>6</td>
<td>31</td>
<td>![Image of starting material]</td>
<td>![Image of product]</td>
<td>Mg</td>
<td>16:1</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td>Zn</td>
<td>5:1</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td>In</td>
<td>0:1</td>
</tr>
<tr>
<td>7</td>
<td>31</td>
<td>![Image of starting material]</td>
<td>![Image of product]</td>
<td>Mg</td>
<td>1:8</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td>Zn</td>
<td>8:1</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td>In</td>
<td>1:0</td>
</tr>
<tr>
<td>8</td>
<td>31</td>
<td>![Image of starting material]</td>
<td>![Image of product]</td>
<td>Mg</td>
<td>8:1</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td>Zn</td>
<td>2:1</td>
</tr>
</tbody>
</table>
Table 2. $K_i$ and IC$_{50}$ values of iminosugars against various glycosidases

<table>
<thead>
<tr>
<th>Enzyme</th>
<th>Values in µM</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td><img src="image1.png" alt="Chemical Structures" /> <img src="image2.png" alt="Chemical Structures" /> <img src="image3.png" alt="Chemical Structures" /> <img src="image4.png" alt="Chemical Structures" /></td>
</tr>
<tr>
<td>α-Glucosidase</td>
<td><img src="image1.png" alt="Structure 1" /> <img src="image2.png" alt="Structure 2" /> <img src="image3.png" alt="Structure 3" /> <img src="image4.png" alt="Structure 4" /></td>
</tr>
<tr>
<td>Yeast</td>
<td>$^a$NI (16%) NI (20%) $K_i$: 6.3$^5$ 85% at 100 µM$^{32}$</td>
</tr>
<tr>
<td>Rice</td>
<td>IC$_{50}$: 27 NI (12%)</td>
</tr>
<tr>
<td>Rat intestinal maltase</td>
<td>IC$_{50}$: 181 NI (11%)</td>
</tr>
<tr>
<td>Rabbit small intestine maltase</td>
<td>IC$_{50}$: 4.6$^{33}$</td>
</tr>
<tr>
<td>Rat intestinal sucrase</td>
<td>IC$_{50}$: 14 NI (23%)</td>
</tr>
<tr>
<td>Rabbit small intestine sucrase</td>
<td>$K_i$: 0.13$^{34}$</td>
</tr>
<tr>
<td>Amyloglucosidase</td>
<td><img src="image1.png" alt="Alga glucosidase" /> <img src="image2.png" alt="Alga glucosidase" /> <img src="image3.png" alt="Alga glucosidase" /> <img src="image4.png" alt="Alga glucosidase" /></td>
</tr>
<tr>
<td>Aspergillus niger</td>
<td>NI (0%) NI (0%)</td>
</tr>
<tr>
<td>Alglucosidase alfa</td>
<td><img src="image1.png" alt="Structure" /> <img src="image2.png" alt="Structure" /> <img src="image3.png" alt="Structure" /> <img src="image4.png" alt="Structure" /></td>
</tr>
<tr>
<td>Human lysosome</td>
<td>IC$_{50}$: 30 NI (2%)</td>
</tr>
<tr>
<td>α-Amylase</td>
<td><img src="image1.png" alt="Structure" /> <img src="image2.png" alt="Structure" /> <img src="image3.png" alt="Structure" /> <img src="image4.png" alt="Structure" /></td>
</tr>
<tr>
<td>Porcine pancreas</td>
<td>NI (26%) NI (9%)</td>
</tr>
<tr>
<td>β-Glucosidase</td>
<td><img src="image1.png" alt="Structure" /> <img src="image2.png" alt="Structure" /> <img src="image3.png" alt="Structure" /> <img src="image4.png" alt="Structure" /></td>
</tr>
<tr>
<td>Sweet almond</td>
<td>NI (28%) NI (21%) $K_i$: 0.9$^5$ $K_i$: 20$^5$</td>
</tr>
<tr>
<td>Bovine liver</td>
<td>NI (44%) NI (35%)</td>
</tr>
<tr>
<td>β-Glucocerebrosidase</td>
<td><img src="image1.png" alt="Structure" /> <img src="image2.png" alt="Structure" /> <img src="image3.png" alt="Structure" /> <img src="image4.png" alt="Structure" /></td>
</tr>
<tr>
<td>Human lysosome</td>
<td>IC$_{50}$: 440 NI (20%)</td>
</tr>
<tr>
<td>Enzyme Type</td>
<td>Source</td>
</tr>
<tr>
<td>----------------------</td>
<td>-----------------------</td>
</tr>
<tr>
<td>α-Galactosidase</td>
<td>Green coffee beans</td>
</tr>
<tr>
<td></td>
<td></td>
</tr>
<tr>
<td>α-Galactosidase A</td>
<td>Human lysosome</td>
</tr>
<tr>
<td></td>
<td></td>
</tr>
<tr>
<td>β-Galactosidase</td>
<td>Bovine liver</td>
</tr>
<tr>
<td></td>
<td></td>
</tr>
<tr>
<td>α-Mannosidase</td>
<td>Jack bean</td>
</tr>
<tr>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
</tr>
<tr>
<td>β-Mannosidase</td>
<td>Snail</td>
</tr>
<tr>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td><em>Aspergillus wentii</em></td>
</tr>
<tr>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
</tr>
<tr>
<td>α-L-Rhamnosidase</td>
<td><em>Penicillium decumbens</em></td>
</tr>
<tr>
<td></td>
<td></td>
</tr>
<tr>
<td>α-L-Fucosidase</td>
<td>Bovine kidney</td>
</tr>
<tr>
<td></td>
<td></td>
</tr>
<tr>
<td>β-Glucuronidase</td>
<td><em>E. coli</em></td>
</tr>
<tr>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>Bovine liver</td>
</tr>
<tr>
<td></td>
<td></td>
</tr>
<tr>
<td>α,α-Trehalase</td>
<td>Porcine kidney</td>
</tr>
<tr>
<td></td>
<td></td>
</tr>
<tr>
<td>β-N-Acetyl-</td>
<td></td>
</tr>
</tbody>
</table>
Inhibition studies

The compounds 1 and 2 were tested for inhibitory activity against a panel of glycosidases (Table 2), but in many cases no inhibition was found. Nojiristegine (1) was, however, found to be moderately active as an α-glucosidase inhibitor with an \( IC_{50} \) of 14 µM for rat intestinal sucrase, 27 µM for rice and 30 µM for recombinant α-glucosidase alglycosidase alpha of which the latter enzyme is used in enzyme replacement therapy for Pompe disease. In comparison, 1-deoxynojirimycin is a strong inhibitor of these enzymes (\( K_i \) 0.024 µM, \( K_i \) 0.01 µM, \( K_i \) 0.55 µM, respectively)\(^{35,36,37}\) which indicates that the steric bulk of the ethylene bridge in nojiristegine (1) might interfere with binding to the enzymes. Yeast α-glucosidase and sweet almond β-glucosidase are both potently inhibited by nojirimycin,\(^5\) whereas nojiristegine was largely inactive. Human glucocerebrosidase was modestly inhibited by nojiristegine (1, \( IC_{50} \) 440 µM) but much less than what has been found for noeurostegine (\( IC_{50} \) 0.4 µM).\(^{38}\) Interestingly, nojiristegine (1) was not found to be an inhibitor of coffee bean α-galactosidase despite the fact that both calystegine B$_2$ (\( K_i \) 0.86 µM),\(^{39}\) noeurostegine (\( K_i \) 2.5 µM)\(^{22}\) and 1-deoxynojirimycin (\( K_i \) 23 µM)\(^{40}\) are all potent inhibitors of this enzyme (Figure 2). Since \textit{galacto}-noeurostegine (4-epi-noeurostegine) is not an inhibitor of green coffee bean α-galactosidase, but calystegine and noeurostegine inhibit this enzyme potently, we have previously speculated that the two former azasugars would bind in a nojirimycin (NJ) binding mode\(^{24}\) placing the ethylene bridge in the same position as nojiristegine (1). The observation, that nojiristegine is not an inhibitor of this enzyme, brings our previous suggestion into question.

<table>
<thead>
<tr>
<th>hexosaminidase</th>
<th>Human placenta</th>
<th>NI (8%)</th>
<th>NI (2%)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>NI: No inhibition (Less than 50% inhibition at 1000 µM)</td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td>( ): Inhibition % at 1000 µM</td>
<td></td>
</tr>
</tbody>
</table>
Figure 2. Inhibitory data for inhibition of α-galactosidase from green coffee beans.

Manno-nojiristegine (2) did not show any activity against tested mannosidases and most other glycosidases. Weak inhibition of β-glucoronidase from *E. coli* and α,α-trehalase from porcine kidney (*IC₅₀* of 810 µM and 400 µM, respectively) was obtained.

**Antibiotic studies**

Nojiristegines 1 and 2 and the close structural analogue 1-deoxyojirimycin were evaluated for potential antibiotic activity against a series of bacterial strains based on availability, clinical relevance and their close relation to original strains tested with nojirimycin (*Pseudomonas aeruginosa* strain PAO1, *Escherichia coli* strains VK98 and VK144, and *Staphylococcus aureus* strain VK75). Nojirimycin has been found largely inactive against non-resistant *Shigella flexneri* 2a, *Shigella flexneri* 6 Mita, *Staphylococcus aureus* Terashima and 209 P and *E. coli* Umezawa, but highly active against *Xanthomonas oryzae*, resistant *Shigella flexneri* 2a and *Sarcina lutea* PCI 1001.¹⁸

Neither 1-deoxyojirimycin nor nojiristegine (1) showed any antibiotic activity against the tested strains of *Pseudomonas aeruginosa*, *Staphylococcus aureus* or *E. coli* up to concentrations of 500 µg/mL. Lack of activity against *E. coli* and *Staphylococcus aureus* was also seen in the original results for nojirimycin,¹⁸ but activity against *E. coli* could have been expected due its close genetic relation to *Shigella flexneri*.

Niwa *et al.* reported manno-nojirimycin to be weakly active (50-fold less than nojirimycin) against *Xanthomonas oryzae* and also tested other unspecified bacterial strains but found no activity.¹⁹ Assuming that Niwa *et al.* tested the activity against the same bacteria as Ishida *et al.*,¹⁸ there seems to be a noteworthy difference between the antibiotic activity of nojirimycin and its manno-epimer, but this cannot, to the best of our knowledge, be confirmed.
In our test, \textit{manno}-nojiristegine (2) did not show any activity against the strains as investigated for 1.

While glycosidase inhibition has been carefully investigated, our literature study has revealed that only little is known about the antibiotic activity of nojirimycin and \textit{manno}-nojirimycin. In order to take advantages of this antibiotic activity, it is evident that further studies must be conducted in order to establish the mechanism for this activity.

\textbf{Conclusion}

Two new iminosugars of the hemiaminal nortropane type have been synthesised through 21 steps from methyl $\alpha$-$D$-glucopyranoside and methyl $\alpha$-$D$-mannopyranoside. The key transformation was the installation of a nitrogen atom in form of an azide at a quaternary carbon atom, which was performed over several steps from a ketone functionality.

The synthesised highly functionalised iminosugars were named nojiristegine and \textit{manno}-nojiristegine due to their resemblance with both calystegines and nojirimycin. As a result of the ethylene bridge inherited from calystegines, the synthesised molecules were found to be stable in aqueous medium for several days.

Nojiristegine (1) and its \textit{manno}-configured isomer (2) were both evaluated as potential glycosidase inhibitors, but only weak inhibition was found. This shows that only limited space is available in the glycosidase binding pocket, which only in certain cases can accommodate the ethylene bridge. Glycosidase inhibition by calystegines and noeuromycins has in general been found to be more potent that what was found for the two nojiristegines investigated in this study.

The synthesised compounds (1 and 2) were also evaluated as a potential antibiotics against a few bacterial strains, resulting in no activity against any of the strains of \textit{Pseudomonas aeruginosa}, \textit{E. coli}, and \textit{Staphylococcus aureus} for neither 1 nor 2. A careful literature survey revealed that literature only holds very little information about the antibiotic activity of nojirimycin despite the fact that this compound is often referred to as an antibiotic. Given the current threat of resistant bacteria a modern study of the nojirimycins would be in order to confirm its activity and activity profile.
Experimental

Inhibition studies

The ability of the compounds to inhibit glycosidase activity was carried out as previously described. 23

Antibiotic studies

The ability of the compounds to inhibit bacterial growth was tested on 5 % blood agar plates (Statens Serum Institut, Copenhagen). The inoculum was a suspension of the respective bacterial strains in sterile 0.9% saline at a density of approximately $10^7$ bacteria per ml. The suspension was added to the entire surface of the agar plate whereupon excess fluid was removed with a pipette. After drying of the plate filter paper discs with a diameter of 5 mm were placed on the agar surface and 200 ul of the respective dilutions of the compounds were added. Potential inhibition zones were read after overnight incubation at 37°C.

The bacterial strains used in the inhibition experiments were *Pseudomonas aeruginosa* strain PAO1, *Escherichia coli* strains VK98 and VK144, and *Staphylococcus aureus* strain VK75.

Organic synthesis

General methods

All reactions with air- and moisture sensitive compounds were conducted in an atmosphere of nitrogen or argon. Dichloromethane, toluene, acetonitrile and THF were dried over aluminum oxide via an MBraun SPS-800 solvent purification system, other dry solvents were dried over molecular sieves. Evaporation of solvents was done under reduced pressure at 40 °C. Flash column chromatography was carried out with a Merck silica gel (230-400 Mesh). TLC analysis was carried out on silica gel on aluminum foil (Merck Kieselgel 60 F254). TLC plates were visualized by using ceric sulfate/ammonium molybdate in 10 % $\text{H}_2\text{SO}_4$ or ninhydrin in n-butanol/acetic acid and successive heating to dryness.

Apparatus

NMR spectra were recorded on a Varian Mercury 400 spectrometer. $^1\text{H}$-, gCOSY-, and gHMOC-NMR were recorded at 400 MHz and $^{13}\text{C}$ NMR and DEPT-135 at 100 MHz. Chemical shifts ($\delta$) are given in ppm relative to the residual solvent signals (CDCl$_3$ ($\delta$ 7.26 ppm for proton and 77.16 ppm
for carbon resonances) and D$_2$O (δ 4.79 ppm for proton). Assignment of NMR spectra is based on gCOSY, gHMQC and DEPT-135 experiments. Mass spectra were recorded on a Micromass LC-TOF spectrometer with positive electrospray ionization. Melting points were measured on a Büchi B-450, and are not corrected. Optical rotations were measured on an ADP440+ polarimeter and reported in units of deg•cm$^2$•g$^{-1}$. Concentrations are given in g/100mL. IR spectra were obtained as neat on a PerkinElmer FT-IR spectrometer (Spectrum TWO/UATR TWO) and the wavelengths ($
u_{max}$) are reported in cm$^{-1}$.

Synthesis:

**Methyl 4,6-0-Benzylidene-α-D-glucopyranoside (48):**

Methyl α-D-glucopyranoside (3) (5.11 g, 26.3 mmol) was dissolved in freshly distilled acetonitrile (100 mL) and the mixture was stirred vigorously under a nitrogen atmosphere. Benzaldehyde dimethyl acetal (7 mL, 46.6 mmol, 1.8 eq) and 10-camphorsulfonic acid (298 mg, 1.28 mmol, 5 %) were added, and the mixture was heated to reflux. After 45 min the reaction mixture was allowed to cool to r.t., neutralized by adding a few drops of triethylamine and concentrated under reduced pressure. The resulting white powder was dissolved in ethyl acetate (250 mL), washed with brine, dried over MgSO$_4$, filtered and concentrated under reduced pressure. The resulting crystals were mixed with diethyl ether and filtered to remove excess benzaldehyde dimethyl acetal. Methyl 4,6-O-benzylidene-α-D-glucopyranoside (48) was isolated as white crystals (7.43 g, 74 %)

$R_f$ (methanol/EtOAc 1:9) 0.55; $^1$H-NMR (400 MHz, CDCl$_3$): δ$_H$ 7.51-7.34 (5H, m, Ar H), 5.53 (1H, s, CHPh), 4.79 (1H, d, $J_{1,2}$ 4.0 Hz, H1), 4.29 (1H, dd, $J_{6eq,5}$ 4.8 Hz, $J_{6eq,6ax}$ 9.8 Hz, H6eq), 3.92 (1H, t, $J_{3,2}$ = $J_{3,4}$ 9.2 Hz, H3), 3.81 (1H, dt, $J_{5,6ax}$ = $J_{5,4}$ 10.4 Hz, $J_{5,6eq}$ 4.5, H5), 3.74 (1H, t, $J_{6ax,6eq}$ = $J_{6ax,5}$ 9.7 Hz, H6ax), 3.62 (1H, bred s, H2), 3.49 (1H, t, $J_{4,5}$ = $J_{4,3}$ 9.2 Hz, H4), 3.44 (3H, s, CH$_3$), 2.82 (1H, bs, OH), 2.34 (1H, bs, OH). $^{13}$C-NMR (100 MHz, CDCl$_3$): δ$_C$ 137.2 (Ar), 129.4, 128.5, 126.6 (Ar), 102.2 (CHPh), 100.0 (C1), 81.2 (C4), 73.0 (C2), 71.9 (C3), 69.2 (C6), 62.6 (C5), 55.8 (CH$_3$) LRMS(ESI): calcd. for C$_{14}$H$_{18}$O$_6$Na 305.0996; found 304.9;

NMR data were in accordance with previously reported values.$^{41}$

**Methyl 2,3-di-O-benzyl-4,6-O-benzylidene-α-D-glucopyranoside (49):**

NaH (60 %, 3.00 g 75.0 mmol, 3.9 eq) was slowly added to a stirred solution of methyl 4,6-O-benzylidene-α-D-glucopyranoside 48 (5.35 g 19.0 mmol) in DMF (40 mL) under a nitrogen atmosphere. The reaction mixture was cooled to 0 °C and BnBr (7 mL, 58.9 mmol, 3.1 eq) was
added drop wise under formation of hydrogen gas. The mixture was allowed to slowly heat to r.t. and after 45 min the mixture was diluted with water and diethyl ether and extracted with diethyl ether. The combined organic phases were washed with water, dried over MgSO$_4$, filtered and concentrated under reduced pressure. The crude product was purified by flash column chromatography (pentane/ethyl acetate 8:1 → 6:1) to give the benzyl protected compound 49 as white crystals (7.63g, 87 %)

$R_f$ (pentane/ethyl acetate 6:1) 0.39; $M_p$ (uncorr.) 96.8-97.8°C (lit. 94-95°C crystalised from MeOH); $^1$H-NMR (400 MHz, CDCl$_3$): $\delta$H 7.51 -7.26 (15H, ArH), 5.55 (1H, s, CHPh), 4.92 (1H, d, J 11.3 Hz, CHHPh), 4.86 (1H, d, J 12.1 Hz, CHHPh), 4.84 (1H, d, CHHPh) 4.70 (1H, d, CHHPh), 4.60 (1H, d, J$_{1,2}$ 3.6 Hz, H1), 4.27 (1H, dd, J$_{6eq,5}$ 4.8 Hz J$_{6eq,6ax}$ 9.6 Hz, H6eq), 4.05 (1H, t, J$_{3,4}$ = J$_{3,4}$ 9.4 Hz, H3), 3.82 (1H, dt, J$_{5,6eq}$ 4.6 Hz J$_{5,6ax}$ = J$_{5,6}$ 9.7 Hz, H5), 3.71 (1H, t, J$_{6ax}$ = J$_{6ax,6eq}$ 10.2 Hz, H6ax), 3.60 (1H, d, J$_{4,5}$ = J$_{4,3}$ 10.2 Hz, H4), 3.56 (1H, dd J$_{2,3}$ 10.0 Hz, J$_{2,3}$ 10.0 Hz, H2), 3.41 (3H, s, $CH_3$). $^{13}$C-NMR (100 MHz, CDCl$_3$): $\delta$C 138.9 (Ar), 138.4 (Ar), 137.6 (Ar), 129.1 (Ar), 128.6 (Ar), 128.5 (Ar), 128.4 (Ar), 128.3 (Ar), 128.2 (Ar), 128.0 (Ar), 127.8 (Ar), 126.2 (Ar), 104.1 (CHPh), 99.4 (C1), 82.3 (C4), 79.4 (C2), 78.8 (C3), 75.5 (CH$_2$Ph), 73.9 (CH$_2$Ph), 69.3 (C6), 62.5 (C5), 55.5 (CH$_3$). LRMS(ESI): calcd. for C$_{28}$H$_{30}$O$_6$Na 485.1935; found 484.9

NMR data was in accordance with previously reported values.$^{41}$

**Methyl 2,3,4-tri-O-benzyl-$\alpha$-D-glucopyranoside (50):**

BH$_3$·THF complex (1M solution in THF, 80 mL, 80 mmol, 4.9 eq) was mixed with methyl 2,3-di-O-benzyl-4,6-O-benzylidene-$\alpha$-D-glucopyranoside (49) (7.54 g, 16.3 mmol) under a nitrogen atmosphere. The mixture was stirred for 10 min before Cu(OTf)$_2$ (294 mg 0.810 mmol, 5 %) was added and the reaction was then allowed to stir overnight. The reaction was quenched by adding triethylamine (2.3 mL) and methanol (29 mL), concentrated under reduced pressure, added additional methanol (25 mL) and concentrated again. Toluene was added and the mixture was concentrated and purified by flash column chromatography (pentane/ethyl acetate 2:1). Methyl 2,3,4-tri-O-benzyl-$\alpha$-D-glucopyranoside (50) was isolated as white crystals (6.48 g, 86 %)

$R_f$ (pentane/ethyl acetate 2:1) 0.29; $^1$H-NMR (400 MHz, CDCl$_3$): $\delta$H 7.38-7.27 (15H, m, ArH), 4.99 (1H, d, J 10.8 Hz, CHHPh), 4.89 (1H, d, CHHPh), 4.84 (1H, d, J 11.2 Hz, CHHPh), 4.81 (1H, d, J 12.0 Hz, CHHPh), 4.67 (1H, d, CHHPh), 4.65 (1H, d, CHHPh), 4.58 (1H, d, J$_{1,2}$ 3.6 Hz, H1), 4.01 (1H, t, J$_{3,4}$ = J$_{3,4}$ 9.4 Hz, H3), 3.77 (1H, dd, J$_{6a,5}$ 2.4 Hz, J$_{6a,6b}$ 11.7 Hz, H6a) 3.73-3.63 (2H, m, H6b,
H5), 3.53 (1H, t, J4,5 = J4,3 9.2 Hz, H4), 3.51 (1H, dd, J2,1 3.6 Hz, J2,3 9.2 Hz, H2), 3.37 (3H, s, CH3).

$^{13}$C-NMR (100 MHz, CDCl3): δC 138.8 (Ar), 138.2 (Ar), 138.2 (Ar), 128.5 (Ar), 128.4 (Ar), 128.2 (Ar), 128.1 (Ar), 128.0 (Ar), 127.9 (Ar), 127.7 (Ar), 98.2 (C1), 82.0 (C3), 80.0 (C2), 76.8 (C4), 75.8 (CH2Ph), 75.1 (CH2Ph), 73.4 (CH2Ph), 70.7 (C5), 61.9 (C6), 55.2 (CH3).

LRMS(ESI): calcd. for C28H32O6Na 487.2091; found 487.0.

NMR data were in accordance with previously reported values.41

Methyl 2,3,4-tri-O-benzyl-6-deoxy-6-iodo-α-D-glucopyranoside (5):

Triphenylphosphine (8.88 g, 33.8 mmol, 2.5 eq), imidazole (1.91 g, 28.0 mmol, 2.1 eq) and iodine (3.78 g, 14.9 mmol, 1.1 eq) were added to a solution of methyl 2,3,4-tri-O-benzyl-α-D-glucopyranoside (50) (6.30 g, 13.6 mmol) in anhydrous toluene (150 mL) under a nitrogen atmosphere. The mixture was heated to reflux for 2 hours, then cooled to r.t. and added 5% aqueous sodium thiosulfate (130 ml). The organic phase was washed with brine, dried over MgSO4 and concentrated. The crude product was purified by flash column chromatography (pentane/ethyl acetate 15:1) to yield the iodide 5 as a white solid (5.35 g, 69%).

$R_f$ (pentane/ethyl acetate 15:1) 0.16; $^1$H-NMR (400 MHz, CDCl3): δH 7.37-7.23 (15H, m, ArH), 4.98 (1H, d, J 10.8 Hz, CH2Ph) 4.93 (1H, d, J 10.9 Hz, CH2Ph), 4.80 (1H, d, J 10.8 Hz, CH2Ph) 4.79 (1H, d, J 12.2 Hz, CH2Ph), 4.68 (1H, d, J 9.1 Hz, CH2Ph), 4.65 (1H, d, J 10.2 Hz, CH2Ph), 4.62 (1H, d, J1,2 2.6 Hz, H1), 4.02 (1H, t, J3,2 = J3,4 9.6 Hz, H3), 3.58 (1H, dd, J2,1 3.6 Hz J2,3 9.6 Hz, H2), 3.49-3.42 (2H, m, H5, H6a), 3.42 (3H, s, CH3), 3.36-3.26 (2H, m, H4, H6b). $^{13}$C-NMR (100 MHz, CDCl3): δC 138.7 (Ar), 138.2 (Ar), 138.1 (Ar), 128.7 (Ar), 128.7 (Ar), 128.6 (Ar), 128.2 (Ar), 128.1 (Ar), 127.9 (Ar), 98.3 (C1), 81.7 (C4), 81.6 (C3), 80.3 (C2), 75.9 (CH2Ph), 75.5 (CH2Ph), 73.6 (CH2Ph), 69.5 (C5), 55.6 (CH3), 7.9 (C6). LRMS(ESI): calcd. for C28H31O5Na 597.1108; found 596.6

NMR data was in accordance with previously reported values.42

(2R,3S,4R)-2,3,4-Tri-O-benzylhex-5-enal (7):
Pre-activated Zn dust\(^a\) (6.42 g, 98.2 mmol, 11 eq) and water (25 mL) were added to a solution of the iodide 5 (5.14 g, 8.95 mmol) in THF (230 mL). The mixture was sonicated at 40 °C for 1h 35 min, then diluted with water and diethyl ether and filtered through a pad of Celite \(\textregistered\). The organic phase was washed with water and brine, dried over Na\(_2\)SO\(_4\), filtered and concentrated under reduced pressure to yield the aldehyde 7 (3.67 g) without further purification.

\(R_f\) (pentane/ethyl acetate 1:5) 0.74; \(^1\)H-NMR (400 MHz, CDCl\(_3\)): \(\delta \ H\) 9.61 (1H, s, CHO), 7.33-7.18 (15H, m, ArH), 5.79 (1H, ddd, \(J_{5.4}\) 7.61 Hz, \(J_{5.6E}\) 11.0 Hz, \(J_{5.6Z}\) 16.7 Hz, H5), 5.24 (1H, d, \(J_{6Z,5}\) 11.0 Hz, H6Z), 5.23 (1H, d, \(J_{6E,5}\) 16.6 Hz, H6E), 4.68 (1H, d, \(J_{11.7}\) Hz, CHHPh), 4.67 (d, \(J_{11.8}\) Hz, 1H, CHHPh), 4.54 (1H, d, CHHPh), 4.50 (1H, d, \(J_{11.5}\) Hz, CHHPh), 4.45 (1H, d, CHHPh), 4.33 (1H, d, CHHPh), 4.11 (1H, dd, \(J_{4.5}\) 7.6 Hz, \(J_{4.3}\) 5.0 Hz, H4), 3.84 (1H, dd, \(J_{2.3}\) 4.2 Hz, \(J_{2.1}\) 0.6 Hz, H2), 3.76 (1H, t, \(J_{3.2}\) = \(J_{3.4}\) 4.6 Hz, H3). \(^{13}\)C-NMR (100 MHz, CDCl\(_3\)): \(\delta \ C\) 201.7 (CHO), 137.9 (Ar), 137.8 (Ar), 137.3 (Ar), 134.9 (C5), 128.7 (Ar), 128.6 (Ar), 128.5 (Ar), 128.4 (Ar), 128.3 (Ar), 128.2 (Ar), 128.0 (Ar), 127.7 (Ar), 119.5 (C6), 82.6 (C4), 82.0 (C3), 80.0 (C2), 74.7 (CH\(_2\)Ph), 73.4 (CH\(_3\)Ph), 71.1 (CH\(_2\)Ph). HRMS(ESI): calcd. for C\(_{27}\)H\(_{38}\)O\(_4\)Na 439.1880; found 439.1885.

NMR data was in accordance with previously reported values\(^43\)

\((3R,4S,5S,6R/S)-3,4,5-Tri-O-benzyl-6-hydroxy-1,8-nonadiene (8):

Indium dust (1.96 g, 17.1 mmol, 1.9 eq) and allyl bromide (2.3 mL, 27.1 mmol, 3 eq) were added to a stirred solution of the crude aldehyde 7 (3.67 g, 8.81 mmol) in 50% aqueous THF (64 mL). The reaction was stirred overnight at r.t. after which sat. aq. NH\(_4\)Cl was added to the reaction mixture and the aqueous phase was neutralized with sat. aq. NaHCO\(_3\). The resulting mixture was extracted with dichloromethane and the combined organic phases dried over MgSO\(_4\), filtered and concentrated under reduced pressure. The crude product was purified by flash column chromatography (pentane/ethyl acetate 11:1) to give the diene 8 as an oil (3.13 g, 76 % over 2 steps). According to NMR the product was a 2:3 mixture of R- and S-diastereoisomers.

\(R_f\) (pentane/ethyl acetate 11:1) 0.19, \(^1\)H-NMR (400 MHz, CDCl\(_3\)): \(\delta \ H\) 7.43-7.21 (48.7H, m, ArH), 6.03-5.74 (3.9H, m), 5.67 (1.9H, ddt, \(J_{17.4}\) Hz, \(J_{10.6}\) Hz, \(J_{7.0}\) Hz), 5.38 - 5.24 (6.1H, m), 5.10-4.99 (6.1H, m), 4.83 (3.2H, dd, \(J_{11.3}\) Hz, \(J_{15.3}\) Hz), 4.72 (3.2H, d, \(J_{11.1}\) Hz), 4.68 - 4.51 (9.8H, m),

\(^a\) Zinc dust (1.01 g) was mixed with 1M HCl (10 mL) and stirred at r.t for 30 min. The mixture was then filtered and the zinc dust was washed with water and diethyl ether. At last the zinc dust was dried under high vacuum with a heat gun.
The alcohol 8 (1.09 g, 2.39 mmol) was dissolved in anhydrous dichloromethane under a nitrogen atmosphere before Ac₂O (0.41 mL, 4.34 mmol, 1.8 eq), Et₃N (0.66 mL, 4.74 mmol, 2 eq) and DMAP (27.7 mg, 0.23 mmol, 0.09 eq) were added. The mixture was allowed to stir overnight after which a small amount of DMAP was added. After stirring for additional 2 h the reaction was quenched by adding methanol and diluted with dichloromethane. The organic phase was washed with 1M aq. HCl and sat. aq. NaHCO₃, dried over MgSO₄, filtered and concentrated under reduced pressure. The crude product was purified by flash column chromatography (pentane/ethyl acetate 25:1) yielding the R (9R) and S (9S) isomers of the product (1.02 g, 85 %) which could be partly separated.

9S: colourless oil (0.550 g, 46 %), Rf (pentane/ethyl acetate 11:1) 0.45; [α]D293K -14.4 (c 1, CHCl₃); 1H NMR (400 MHz, CDCl₃): δH 7.34-7.26 (15H, m, ArH), 5.91 (1H, ddd, J₂,3 7.7 Hz, J₂,1Z 10.3 Hz, J₁,2E 16.7 Hz, H2), 5.65 (1H, m, H8) 5.33 (1H, dd, J₁Z,1E 1.1 Hz, H1Z), 5.30 (1H, dd, H1E), 5.21 (1H, dt, J₆,7a = J₆,5 3.5 Hz, J₆,7b 9.1 Hz, H6), 4.99 (1H, dt, J₆E,₈ 16.1 Hz J₆E,₇a = J₆E,₇b 1.6 Hz, H9E) 4.98 (1H, dt, J₉Z,₈ 10.3 Hz J₉Z,₇a = J₉Z,₇b 0.94 Hz, H9Z), 4.75 (1H, d, J 11.2 Hz, CHHPh), 4.71 (1H, d, J 11.4 Hz, CHHPh), 4.66 (1H, d, CHHPh), 4.61 (2H, d, J 12.2 Hz, CHHPh, CHHPh) 4.36 (1H, d, CHHPh), 4.06 (1H, d, J₄,₃ 5.6 Hz, H3), 3.76 (1H, dd, J₅,₄ 5.6 Hz, H5), 3.62 (1H, t, H4), 2.48 (2H, m, H7), 1.98 (1H, s, CH₃). 13C-NMR (100 MHz, CDCl₃): δC 170.1 (C=O), 138.1 (Ar), 138.3 (Ar), 138.6 (Ar), 134.8 (C8/C2), 134.4 (C2/C8), 128.2 (Ar), 128.2 (Ar), 127.9 (Ar), 127.7 (Ar), 127.5 (Ar), 127.4 (Ar), 119.4 (C1), 117.1 (C9), 81.8 (C3), 81.1 (C5), 80.5 (C4), 74.7 (CH₂Ph), 74.1 (CH₂Ph), 73.9 (C6), 70.7 (CH₂Ph), 34.0 (C7), 21.0 (CH₃). HRMS(ESI): calcd. for C₃₂H₆₀O₅Na 523.2455; found 523.2466.
9R: colourless oil (0.250 g, 21%), $R_f$ (pentane/ethyl acetate 11:1) 0.31; $[\alpha]_D^{293K}$ -9.2 (c 1, CHCl$_3$); $^1$H-NMR (400 MHz, CDCl$_3$): $\delta_H$ 7.36-7.25 (15H, m, ArH), 5.97 – 5.83 (1H, m, H2), 5.58 – 5.42 (1H, m, H8), 5.28 (1H, bs, H1a), 5.27 (1H, ddd, $J$ 6.8 Hz, J 1.8 Hz, J 0.9 Hz, H1b), 4.99 - 4.96 (1H, m, H9Z), 4.96 – 4.92 (1H, m, H9E), 4.90 (1H, ddd, $J$ 6.7b $J$ 5.8 Hz, $J$ 6,5 7.4 Hz, H6), 4.82 (1H, d, J 11.5 Hz, CHPh), 4.76 (1H, d, CHPh), 4.63 (1H, d, CHPh), 4.31 (1H, d, CHPh), 4.28 (1H, d, CHPh), 4.19 (1H, d, CHPh), 3.97 (1H, d, C HHPh), 3.94 (1H, d, C HHPh), 3.89 (1H, d, C HHPh), 3.86 (1H, d, C HHPh). $^{13}$C-NMR (100 MHz, CDCl$_3$): $\delta_C$ 170.4 (C=O), 138.5 (Ar), 138.2 (Ar), 135.5 (C2/C8), 133.6 (C8/C2), 128.3 (Ar), 128.3 (Ar), 128.2 (Ar), 128.0 (Ar), 127.7 (Ar), 127.6 (Ar), 118.7 (C1), 117.7 (C9), 81.6 (C4), 79.8 (C3), 78.8 (C5), 74.9 (CH$_2$Ph), 74.8 (CH$_2$Ph), 72.6 (C6), 70.5 (CH$_2$Ph), 35.5 (C7), 21.0 (CH$_3$). HRMS(ESI): calcd. for C$_{32}$H$_{36}$O$_5$Na 523.2455; found 523.2463.

(3R,4S,5S,6S)- 6-O-Acetyl-3,4,5-tri-O-benzyl-cycloheptene (10) and (3R,4S,5S,6R)- 6-O-Acetyl-3,4,5-tri-O-benzyl-cycloheptene (11):

The diene 9 (2.98 g, 5.94 mmol) was dissolved in anhydrous toluene (21 mL) under a nitrogen atmosphere. Grubbs-Hoveyda 2nd generation catalyst (75 mg, 0.12 mmol, 2 %) was added to the reaction flask, which was immediately put in a preheated 80°C oil bath. The mixture was stirred for 1h 30 min, cooled down to r.t. and concentrated under reduced pressure. The crude product was purified by flash column chromatography (pentane/ethyl acetate 15:1) to give the cycloheptenes 10 and 11 (2.04 g, 73 %) as a slightly yellow oil.

(3R,4S,5S,6S)- 6-O-Acetyl-3,4,5-tri-O-benzyl-cycloheptene (10)

$R_f$ (pentane/ethyl acetate 5:1) 0.56; $[\alpha]_D^{293K}$ -6.4 (c 1, CHCl$_3$); $^1$H-NMR (400 MHz, CDCl$_3$): $\delta_H$ 7.37-7.26 (15H, m, ArH), 5.70 (1H, dd, $J$ 2,1 11.8 Hz, $J$ 2,3 1.0 Hz, H2), 5.62 (1H, dt, $J$ 1.7a = $J$ 1.7b 5.5 Hz, $J$ 1.2 11.8 Hz, H1), 5.33 (1H, ddd, $J$ 6,7b/5 1.6 Hz, $J$ 6,7b/5 3.0 Hz, $J$ 6,7a 9.8 Hz, H6), 4.77 (1H, d, J 11.1 Hz, CHPh), 4.69 (2H, s, CH$_2$Ph), 4.66 (2H, s, CH$_2$Ph), 4.63 (1H, d, CH/FPh), 4.45 (1H, d, J 9.2 Hz, H5), 3.79 (1H, dd, $J$ 3,4 4.7 Hz, H3), 3.74 (1H, dd, H4), 2.62 (1H, ddd, $J$ 7a,7b 16.7 Hz, H7a), 2.35 (1H, d, $J$ 7b,7a 16.7 Hz, H7b), 2.04 (3H, s, CH$_3$). $^{13}$C-NMR (100 MHz, CDCl$_3$): $\delta_C$ 170.1 (C=O), 138.6 (Ar), 138.5 (Ar), 138.3 (Ar), 131.2 (C2), 128.3 (Ar), 128.3 (Ar), 128.0 (Ar), 127.9 (Ar), 127.6 (Ar), 127.5 (Ar), 126.0 (C1), 83.3 (C4), 82.6 (C3), 79.0 (C5), 74.2 (CH$_2$Ph), 72.8 (CH$_2$Ph), 73.1 (CH$_2$Ph), 71.2 (C6), 29.6 (C7), 21.2 (CH$_3$). HRMS(ESI): calcd. for C$_{30}$H$_{32}$O$_3$Na 495.2142; found 495.2154.
(3R,4S,5S,6R)-6-O-Acetyl-3,4,5-tri-O-benzyl-cycloheptene (11):  

$R_f$ (pentane/ethyl acetate 11:1) 0.33; [α]$_D^{203K}$ -2.9 (c 1, CHCl$_3$); $^1$H-NMR (400 MHz, CDCl$_3$): δ$_H$ 7.36-7.21 (m, 15H, ArH), 5.84 (dd, $J_{2,1}$ 11.3 Hz, $J_{2,2}$ 3.6 Hz, J 1.6 Hz, 1H, H2), 5.76-5.66 (m, 1H, H1), 4.95 (dd, $J_{6,7b}$ 10.0 Hz, $J_{6,5}$ 7.2 Hz, $J_{6,7a}$ 2.8 Hz, 1H, H6), 4.87 (d, J 10.6 Hz, 1H, CH/Ph), 4.82 (d, J 11.3 Hz, 1H, CH/Ph), 4.69 (d, J 10.7 Hz, 2H, 2xCH/Ph), 4.68 (d, J 11.5 Hz, 2H, 2xCH/Ph), 4.20 (m, 1H, H4), 3.70-3.58 (m, 2H, H5, H3), 2.37 (ddd, $J_{7a,7b}$ 15.6 Hz, $J_{7a,1}$ 6.6 Hz, 1H, H7a), 2.32-2.22 (m, 1H, H7b), 1.93 (s, 3H, CH$_3$). $^{13}$C-NMR (100 MHz, CDCl$_3$): δ$_C$ 170.1 (C=O), 138.8 (Ar), 138.7 (Ar), 138.6 (Ar), 134.0 (C2), 128.4 (Ar), 128.2 (Ar), 127.8 (Ar), 127.6 (Ar), 127.6 (Ar), 125.3 (C1), 85.8 (C5/C3), 84.3 (C5/C3), 77.9 (C4), 75.8 (CH$_2$Ph), 75.3 (CH$_2$Ph), 73.0 (CH$_2$Ph), 72.7 (C6), 29.4 (C7). HRMS(ESI): calcd. for C$_{30}$H$_{32}$O$_5$Na 495.2142; found 495.2146.

(3R,4S,5S,6S/R)-3,4,5-Tri-O-benzyl-6-hydroxy-cycloheptene (51S):

The acetyl protected compounds (10 and 11) (2.62 g, 5.54 mmol) was dissolved in anhydrous methanol (86 mL) under a nitrogen atmosphere. Sodium (52 mg) was added and the mixture was stirred for 6h 20 min. The mixture was then concentrated under reduced pressure, added toluene and concentrated again. The crude product (51S) was used directly in the next step without further purification.

(3R,4S,5S,6S)-3,4,5-Tri-O-benzyl-6-hydroxy-cycloheptene (51S)

$R_f$ (pentane/ethyl acetate 5:1) 0.25; [α]$_D^{203K}$ +2.1 (c 1, CHCl$_3$); $^1$H-NMR (400 MHz, CDCl$_3$): δ$_H$ 7.37-7.25 (15H, m, ArH), 5.81 (1H, ddt, $J_{2,2}$ 1.5 Hz, $J_{2,7}$ 3.2 Hz, J 11.5 Hz, H2), 5.72 (1H, ddt, $J_{1,7a}$ 1.9 Hz, $J_{1,7b} = J_{1,3}$ 5.7 Hz, H1), 4.92 (1H, d, J 10.8 Hz, CH/Ph), 4.85 (1H, d, J 11.5 Hz, CH/Ph), 4.70 (1H, d, CH/Ph), 4.70 (2H, s, CH$_2$Ph), 4.66 (1H, d, CH/Ph), 4.16 (1H, ddd, $J_{3,4}$ 9.1 Hz, H3), 4.06 (1H, d, $J_{6,7a}$ 8.4 Hz, H6), 3.78 (1H, ddt, $J_{4,5}$ 7.2 Hz, H4), 3.67 (1H, ddd, $J_{5,6}$ 3.2 Hz, H5), 2.63 (1H, bs, OH), 2.52 (1H, ddt, $J_{7a,7b}$ 16.5 Hz, H7a), 2.19 (1H, ddd, H7b). $^{13}$C-NMR (100 MHz, CDCl$_3$): δ$_C$ 138.8 (Ar), 138.5 (Ar), 138.5 (Ar), 132.3 (C2), 128.6 (Ar), 128.5 (Ar), 128.2 (Ar), 128.0 (Ar), 127.9 (Ar), 127.8 (Ar), 127.7 (C1), 85.2 (C5/C3), 82.6 (C4), 75.3 (CH$_2$Ph), 78.6 (C3), 74.1 (CH$_2$Ph), 72.9 (CH$_2$Ph), 69.0 (C6), 31.4 (C7). HRMS(ESI): calcd. for C$_{28}$H$_{30}$O$_4$Na 453.2036; found 453.2044.

(3R,4S,5S,6R)-3,4,5-Tri-O-benzyl-6-hydroxy-cycloheptene (51R)
$R_f$ (pentane/ethyl acetate 5:1) 0.36; $^1$H-NMR (400 MHz, CDCl$_3$): $\delta$H 7.45-7.29 (m, 15H, ArH), 5.91-5.82 (m, 2H, H1, H2), 5.08 (d, J 11.3 Hz, 1H, CHHPh), 4.99 (d, J 10.7 Hz, 1H, CHHPh), 4.78 (d, 1H, CHHPh), 4.74 (s, 2H, CH$_2$Ph), 4.69 (d, 1H, CHHPh), 4.23 (d, J 8.8 Hz, 1H, H3), 3.67-3.56 (m, 2H, H4+H6), 3.48 (dd, J 18.1 Hz, J 9.8 Hz, 1H, H5), 2.50-2.37 (m, 1H, H7a), 2.29-2.11 (m, 1H, H7b).

$^1^3$C-NMR (100 MHz, CDCl$_3$): $\delta$C 138.6 (Ar), 138.4 (Ar), 138.4 (Ar), 133.7 (C2), 128.6 (Ar), 128.3 (Ar), 128.0 (Ar), 127.8 (Ar), 127.7 (Ar), 127.5 (Ar), 127.5 (Ar), 126.8 (C1), 89.3 (C5), 83.4 (C4/C6), 78.4 (C3), 75.9 (CH$_2$Ph), 75.4 (CH$_2$Ph), 72.7 (CH$_2$Ph), 70.6 (C4/C6), 31.1 (C7).

HRMS(ESI): calcd. for C$_{28}$H$_{30}$O$_4$Na 453.2036; found 453.2044.

(3R,4S,5S,6S/R)-3,4,5-Tri-O-benzyl-6-O-p-methoxybenzyl-cycloheptene (12):

NaH (60%, 0.454 g, 18.92 mmol, 2 eq) and PMBCl (1.5 mL, 11.05 mmol, 2 eq) were added to a cooled (0 °C) and stirred solution of the crude alcohol (51) (5.54 mmol) in anhydrous DMF (27 mL) under a nitrogen atmosphere. The reaction mixture was heated to r.t. and stirred overnight. The next day more NaH was added. After 20 min the reaction mixture was quenched by addition of n-butylamine, diluted with EtOAc, washed with water and brine, dried over MgSO$_4$, filtered and concentrated under reduced pressure. The crude product was purified by flash column chromatography (pentane/ethyl acetate 14:1) yielding the PMB ether (12) (1.75 g, 57 % over 2 steps).

(3R,4S,5S,6S)-3,4,5-Tri-O-benzyl-6-O-p-methoxybenzyl-cycloheptene (12S):

$R_f$ (pentane/ethyl acetate 14:1) 0.23; $[\alpha]_{D}^{293K}$ - 6.8 (c 1, CHCl$_3$); $^1$H-NMR (400 MHz, CDCl$_3$): $\delta$H 7.38-7.25 (15H, m, ArH), 7.24-7.20 (2H, m, ArH from PMB), 6.87-6.82 (2H, m, ArH from PMB), 5.69-5.56 (2H, m, H1, H2), 4.76-4.59 (5H, m, CHHAr), 4.54 (1H, d, J 11.2, CHHAr), 4.45 (2H, s, CH$_2$Ar), 4.21 (1H, d, J$_{5,4}$ 9.2 Hz, H3), 3.94 (1H, ddd, J$_{6,5}$ 1.0 Hz, J$_{6,7}$ 3.8 Hz, J$_{6,7}$ 10.0 Hz, H6), 3.88 (1H, dd, J$_{5,4}$ 3.8 Hz, H5), 3.8 (3H, s, CH$_3$), 3.74 (1H, dd, H4), 2.61-2.51 (1H, m, H7a), 2.37 (1H, d, J$_{7b,7a}$ 18.0, H7b). $^1^3$C-NMR (100 MHz, CDCl$_3$): $\delta$C 159.1 (Ar), 158.4 (Ar from PMB), 128.9 (Ar), 128.7 (Ar), 132.0 (Ar), 130.6 (C1/C2), 129.9 (Ar), 129.8 (Ar), 129.1 (Ar), 128.3 (Ar), 128.2 (Ar), 128.0 (Ar), 127.8 (Ar), 127.6 (Ar), 127.5 (Ar), 127.4 (C1/C2), 127.0 (Ar), 113.5 (Ar from PMB), 84.1 (C4), 82.0 (C5), 79.3 (C3), 76.0 (C6), 73.7 (CH$_2$Ar), 73.2 (CH$_2$Ar), 72.5 (CH$_2$Ar), 70.6 (CH$_2$Ar), 57.4 (C7), 55.2 (CH$_3$). HRMS(ESI): calcd. for C$_{36}$H$_{38}$O$_5$Na 573.2611; found 573.2619.

(3R,4S,5S,6R)-3,4,5-Tri-O-benzyl-6-O-p-methoxybenzyl-cycloheptene (12R):
$R_f$ (pentane/ethyl acetate 13:1) 0.33; $[\alpha]_D^{293K}$ = 4.1 (c 1, CHCl$_3$); $M_p$ (uncorr.) 73.8-78.4 °C; $^1$H-NMR (400 MHz, CDCl$_3$): $\delta$H 7.31-7.19 (15H, m, ArH), 7.19-7.14 (2H, m, ArH from PMB), 6.85-6.76 (2H, m, ArH from PMB), 5.79 (1H, ddd, $J_{2,1}$ 11.4 Hz, $J_{2,7}$ 3.5 Hz, $J_{2,3}$ 1.0 Hz, H2), 5.65 (1H, dtd, $J_{1,7}$ 6.1 Hz, J 2.0 Hz, H1), 4.85 (1H, d, J 10.8 Hz, CHHAr), 4.76 (1H, d, J 11.0 Hz, CHHAr), 4.73 (1H, d, CHHAr), 4.70 (1H, d, CHHAr), 4.70 (1H, d, J 12.1 Hz, CHHAr), 4.63 (1H, d, CHHAr), 4.48 (2H, s, CH$_2$Ar), 4.26 (1H, dd, $J_{3,4}$ 8.9 Hz, H3), 3.76 (3H, s, CH$_3$), 3.72-3.60 (2H, m, H4, H5), 3.55 (1H, ddd, $J_{2,1}$ 6.2 Hz, H7a), 2.32-2.21 (1H, m, H7b). $^{13}$C-NMR (100 MHz, CDCl$_3$): $\delta$C 159.2 (Ar from PMB), 139.1 (Ar), 138.8 (Ar), 133.4 (C2), 130.7 (Ar), 129.4 (Ar), 128.4 (Ar), 128.4 (Ar), 128.4 (Ar), 127.9 (Ar), 127.9 (Ar), 127.6 (Ar), 125.9 (C1), 113.9 (Ar from PMB), 87.0 (C4/C5), 84.6 (C4/C5), 78.7 (C6), 78.2 (C3), 75.6 (CH$_2$Ar), 75.2 (CH$_2$Ar), 73.1 (CH$_2$Ar), 71.7 (CH$_2$Ar), 55.4 (CH$_3$), 29.3 (C7). HRMS(ESI): calcd. for C$_{36}$H$_{38}$O$_5$Na 573.2611; found 573.2617.

**Oil; $R_f$ (pentane/ethyl acetate 4:1) 0.17; $[\alpha]_D^{293K}$ = +12.8 (c 1, CHCl$_3$)**; $^1$H-NMR (400 MHz, CDCl$_3$): $\delta$H 7.38-7.22 (15H, m, ArH), 7.22-7.18 (2H, m, ArH from PMB), 6.87-6.82 (2H, m, ArH from PMB), 4.77 (1H, d, J 12.2 Hz, CHHAr), 4.75 (1H, d, J 11.2 Hz, CHHAr), 4.63 (1H, d, CHHAr), 4.54 (1H, d, CHHAr), 4.47 (2H, s, CH$_2$Ar), 4.44 (1H, d, J 11.8 Hz, CHHAr), 4.38 (1H, d, CHHAr), 3.98 (1H, dd, J 2.3 Hz, J 9.6 Hz, H1/H5), 3.96 (1H, dd, J 8.3 Hz, J 4.2 Hz, H4/H2), 3.82 (1H, dd, J 9.5 Hz, J 4.1 Hz, H3), 3.79 (3H, s, CH$_3$) 3.62 (1H, dd, J 10.8 Hz, J 3.3 Hz, H5/H1), 3.32 (1H, dd, J 8.8 Hz, J 4.1 Hz, H2/H4), 2.82 (1H, bs, OH), 2.13-1.99 (2H, m, H6a, H7a), 1.86-1.77 (1H, m, H10a).
H6b/H7b), 1.50-1.37 (1H, m, H6b/H7b). $^{13}$C-NMR (100 MHz, CDCl$_3$): δC 159.14 (Ar from PMB), 138.7 (Ar), 138.2 (Ar), 138.0 (Ar), 130.8 (Ar), 129.1 (Ar), 128.6 (Ar), 128.6 (Ar), 128.4 (Ar), 127.9 (Ar), 127.8 (Ar), 127.6 (Ar), 113.8 (Ar from PMB), 89.3 (C2/C4), 82.8 (C3), 78.7 (C1/C5), 78.03 (C2/C4), 74.22 (CH$_2$Ar), 72.84 (CH$_2$Ar), 72.32 (CH$_2$Ar), 70.72 (CH$_2$Ar), 69.44 (C1/C5), 53.3 (CH$_3$), 31.0 (C6/C7), 24.8 (C6/C7). HRMS(ESI): calcd. for C$_{36}$H$_{40}$O$_6$Na 591.2717; found 591.2715.

(2R,3S,4S,5R)-2,3,4-tri-O-benzyl-5-O-p-methoxybenzyl-cycloheptane-1-ol (13R):

Colourless oil; $R_f$ (pentane/ethyl acetate 4:1) 0.42; $[\alpha]_D^{293K}$ +20.4 (c 1, CHCl$_3$); $^1$H-NMR (400 MHz, CDCl$_3$): δH 7.30 – 7.18 (m, 15H, ArH), 7.14 (d, J 8.3, 2H, ArH from PMB), 6.79 – 6.73 (m, 2H, ArH from PMB), 4.69 – 4.52 (m, 5H, CH$_2$Ar), 4.51 – 4.42 (m, 2H, CH$_2$Ar), 4.37 (d, J 10.9 Hz, 1H, CH$_2$Ar), 4.11 – 4.01 (m, 1H, H1/H5), 3.99 (dt, J 6.7 Hz, J 3.3 Hz, 1H, H1/H5), 3.77 (dd, J 7.4 Hz, J 2.5 Hz, 1H, H2/H4), 3.74 (s, 3H, CH$_3$), 2.12 – 1.94 (m, 4H, H6/H7). $^{13}$C-NMR (100 MHz, CDCl$_3$): δC 159.3 (Ar from PMB), 138.8 (Ar), 138.7 (Ar), 138.5 (Ar), 130.3 (Ar), 129.6 (Ar), 128.5 (Ar), 128.4 (Ar), 128.0 (Ar), 127.9 (Ar), 127.9 (Ar), 127.7 (Ar), 127.5 (Ar), 113.9 (Ar from PMB), 84.2 (C2/C4), 81.6 (C3), 78.8 (C2/C4), 77.0 (C1/C5), 73.3 (CH$_2$Ar), 72.7 (CH$_2$Ar), 72.0 (CH$_2$Ar), 7.17 (CH$_2$Ar), 67.0 (C1/C5), 55.4 (CH$_3$), 36.6 (C6/C7), 35.8 (C6/C7). HRMS(ESI): calcd. for C$_{36}$H$_{40}$O$_6$Na 591.2717; found 591.2710.

(3R,4S,5S,6S)-3,4,5-tri-O-benzyl-6-O-p-methoxybenzyl-cycloheptane-1-ol (14S):

Oil; $R_f$ (pentane/ethyl acetate 4:1) 0.06; $[\alpha]_D^{293K}$ -0.9 (c 1, CHCl$_3$); $^1$H-NMR (400 MHz, CDCl$_3$): δH 7.375 – 7.25 (15H, m, ArH), 7.235 – 7.19 (2H, m, ArH from PMB), 6.875 – 6.82 (2H, m, ArH from PMB), 4.68 (1H, d, J 12.2 Hz, CH$_2$Ar), 4.61 (1H, d, J 11.6 Hz, CH$_2$Ar), 4.61 (1H, d, CH$_2$Ar) 4.61 (2H, s, CH$_2$Ar), 4.55 (1H, d, CH$_2$Ar), 4.42 (2H, s, CH$_2$Ar), 4.18 (1H, bd, J 4.1 Hz, H1), 3.94 (1H, dd, J 7.3 Hz, J 9.5 Hz, H3/H6) 3.90 (1H, d, J 2.5 Hz, H5/H4), 3.85 (1H, dd, J 2.7 Hz, J 11.5 Hz, H6/H3), 3.80 (3H, s, CH$_3$), 3.73 (1H, dd, J 7.1 Hz, J 2.7 Hz, H4/H5), 2.38 (1H, d, J 14.8 Hz, J 11.6 Hz, J 4.0 Hz, H7a/H2a), 2.17 (1H, d, J 1.3 Hz, J 11.0 Hz, J 14.3 Hz, H2a/H7a), 1.94 (1H, dd, J 14.3 Hz, J 6.0 Hz, H2b/H7b), 1.84 (1H, d, J 14.2 Hz, H7b/H2b). $^{13}$C-NMR (100 MHz, CDCl$_3$): δC 159.1 (Ar from PMB), 138.9 (Ar), 138.7 (Ar), 138.6 (Ar), 130.8 (Ar), 129.2 (Ar), 128.4 (Ar), 128.4 (Ar), 128.3 (Ar), 127.9 (Ar), 127.9 (Ar), 127.7 (Ar), 127.5 (Ar), 113.8 (Ar from PMB), 83.5 (C4/C5), 79.8 (C5/C4), 77.4 (C6/C3), 74.5 (C3/C6), 72.7 (CH$_2$Ar), 72.4 (CH$_2$Ar), 71.9 (CH$_2$Ar), 70.9 (CH$_2$Ar), 65.1(C1), 55.3 (CH$_3$), 37.2 (C2/C7), 35.6 (C7/C2). HRMS(ESI): calcd. for C$_{36}$H$_{40}$O$_6$Na 591.2717; found 591.2726.
(3R,4S,5S,6R)-3,4,5-tri-O-benzyl-6-O-p-methoxybenzyl-cycloheptane-1-ol (14R)

Colourless oil; Rf (pentane/ethyl acetate 4:1) 0.14; [α]D293K +1.0 (c 1, CHCl3); 1H-NMR (400 MHz, CDCl3): δH 7.35 – 7.15 (m, 18H, ArH), 6.82 (2H, m, ArH from PMB), 4.92 (1H, d, J 11.1 Hz, CHAr), 4.72 (1H, d, J 11.2 Hz, CHAr), 4.65 (1H, d, CHAr), 4.59 (1H, d, J 11.5 Hz, CHAr), 4.54 (1H, d, CHAr), 4.53 (1H, d, CHAr) 4.48 (1H, d, J 11.5 Hz, CHAr), 4.44 (1H, d, CHAr), 3.76 (3H, s, CH3), 3.76 – 3.72 (2H, m), 3.70 (1H, d, J 8.3 Hz) 3.65 (1H , dd, J 8.4 Hz, J 4.4 Hz), 3.63 – 3.58 (1H, m), 1.92 – 1.81 (2H, m, H2/H7), 1.81 – 1.71 (2H, m, H2/H7).

13C-NMR (100 MHz, CDCl3): δC 159.2 (Ar from PMB), 138.6 (Ar), 138.5 (Ar), 130.7 (Ar), 129.8 (Ar), 128.6 (Ar), 128.5 (Ar), 128.1 (Ar), 127.9 (Ar), 127.7 (Ar), 127.7 (Ar), 113.8 (Ar from PMB), 84.8, 83.9, 83.0, 77.4, 75.5 (C2Ar), 74.5 (C3/C4/C5), 73.1 (C2Ar), 71.7, 70.8 (C2Ar), 55.4 (CH2Ar), 27.3 (C2/C7), 22.8 (C2/C7). HRMS(ESI): calcd. for C36H40O6Na 591.2717; found 591.2718

(2S,3R,4S,5S/R)-2,3,4-tri-O-benzyl-5-p-methoxybenzyl-cycloheptanone (15):

The alcohol 13 (830 mg, 1.46 mmol) was dissolved in dichloromethane (45 mL) and Dess-Martin periodinane (1.24 g, 2.93 mmol, 2 eq) was added. The mixture was stirred for 40 min after which it was quenched by adding Na2S2O3 (s) and sat. aq. NaHCO3. The mixture was allowed to stir for 35 min and was then extracted with dichloromethane, dried over MgSO4, filtered and concentrated under reduced pressure giving the ketone 15 (919 mg). The crude product was used directly in the next step without further purification.

(2S,3R,4S,5S)-2,3,4-tri-O-benzyl-5-p-methoxybenzyl-cycloheptanone (15S):

Rf (pentane/ethyl acetate 4:1) 0.26; [α]D293K –15.3 (c 1, CHCl3); 1H-NMR (400 MHz, CDCl3): δH 7.36–7.20 (13H, m, ArH), 7.18–7.12 (4H, m, ArH from PMB), 6.83–6.78 (2H, m, ArH from PMB), 4.77 (1H, d, J 11.9 Hz, CHAr), 4.65 (1H, d, J 12.3 Hz, CHAr), 4.57 (1H, d, CHAr), 4.51 (1H, d, CHAr), 4.44–4.37 (4H, m, CH2Ar), 4.08 (1H, d, J2,3 3.7 Hz, H2) 3.92–3.84 (3H, m, H3, H4, H5), 3.76 (3H, s, CH3), 2.54 (1H, ddd, J7a,6a 4.5 Hz, J7a,7b 7.3 Hz, J7b,7a 13.2 Hz, H7a), 2.31 (1H, ddd, J7b,7a 10.4 Hz, H7b), 2.08 (1H, ddt, J6a,5 10.4 Hz, J6a,6b 14.0 Hz, H6a), 1.84 (1H, ddt, J6b,5 7.3 Hz, H6b). 13C-NMR (100 MHz, CDCl3): δC 206.7 (C=O), 159.3 (Ar from PMB), 138.4 (Ar), 137.9 (Ar), 129.3 (Ar), 128.5 (Ar), 128.3 (Ar), 127.9 (Ar), 127.7 (Ar), 127.7 (Ar), 127.5 (Ar), 113.9 (Ar), 86.5 (C2), 80.4 (2xC, C3/C4/C5), 76.2 (C3/C4/C5), 73.2 (CH2Ar), 72.7 (CH2Ar), 72.5
(CH$_2$Ar), 70.8 (CH$_2$Ar), 55.4 (CH$_3$), 37.0 (C7), 24.2 (C6). HRMS(ESI): calcd. for C$_{36}$H$_{38}$O$_6$Na 589.2561; found 589.2569.

(2S,3R,4S,5R)-2,3,4-tri-O-benzyl-5-p-methoxybenzylcycloheptanon (15R):

$R_T$ (pentane/ethyl acetate 3:1) 0.63; $[\alpha]_D^{293\text{K}}$ -1.8 (c 1, CHCl$_3$); $^1$H-NMR (400 MHz, CDCl$_3$): $\delta_H$ 7.35-7.18 (m, 15H, ArH), 7.18-7.13 (m, 2H, ArH from PMB), 6.83-6.77 (m, 2H ArH from PMB), 4.66 (1H, d, $J$ 11.8 Hz, CHAr), 4.66 (1H, d, CHAr), 4.58 (1H,d, $J$ 10.5 Hz, CHAr), 4.56 (1H, d, CHAr), 4.49 (1H, d, $J$ 11.3, CHAr), 4.47-4.40 (4H, m), 3.86 (1H, dd, $J$ 7.2 Hz, $J$ 3.4 Hz), 3.81 (1H, t, $J$ 4.8 Hz), 3.80- 3.76 (1H, m) 3.77 (3H, s, CH$_3$), 2.58-2.39 (2H, m, H7), 2.12-1.95 (2H, m, H6). $^{13}$C-NMR (100 MHz, CDCl$_3$): $\delta_C$ 207.3 (C=O), 159.3 (Ar from PMB), 138.2 (Ar), 138.1 (Ar), 137.7 (Ar), 130.5 (Ar), 129.4 (Ar), 128.5 (Ar), 128.4 (Ar), 128.4 (Ar), 128.1 (Ar), 128.0 (Ar), 127.9 (Ar), 127.8 (Ar), 127.8 (Ar), 113.8 (Ar from PMB), 86.1, 81.3, 81.0, 78.8, 73.6 (CH$_2$Ar), 73.1 (CH$_2$Ar), 72.5 (CH$_2$Ar), 71.4 (CH$_2$Ar), 55.4 (CH$_3$), 37.4 (C7), 24.5 (C6). HRMS(ESI): calcd. for C$_{36}$H$_{38}$O$_6$Na 589.2561; found 589.2569.

(1R,2R,3S,4S,5R/S)-1-azido-2,3,4-tri-O-benzyl-1-(benzyloxy)methyl-5-O-p-methoxybenzylcycloheptane (20)

Anhydrous dichloromethane (0.23 mL, 3.60 mmol, 9.8 eq) was added to a solution of freshly prepared LDA in THF (~0.37 M, 2.0 mL, 0.740 mmol, 2 eq) at -78 °C. The mixture was stirred for 10 min and then a solution of the ketone 15 (208 mg, 0.367 mmol) in THF (1.8 mL) was added. After stirring for 8 h more LDA was added (2 mL) at -78 °C and the reaction was left to stir overnight. The next morning more LDA (2 mL) and dichloromethane (0.2 mL) was added and the reaction heated to 0 °C. After 2h the reaction was quenched with sat. aq. NH$_4$Cl, diluted with ethyl acetate, washed with water and brine, dried over MgSO$_4$, filtered and concentrated under reduced pressure to give the crude alcohol (16).

Crude alcohol 16 was dissolved in anhydrous DMSO (2 mL) and added DBU (90 µL, 0.602 mmol, 1.7 eq) and stirred for 1h 40 min. The mixture was then diluted with ethyl acetate and washed with water and brine. The combined aqueous phases were extracted with ethyl acetate and the combined organic phases were now dried over MgSO$_4$, filtered and concentrated under reduced pressure to give the crude chloro-epoxide (17).

Crude 17 was dissolved in anhydrous DMF (2.4 mL) and added TBAN$_3$ (322 mg, 1.13 mmol, 3.1 eq) under a nitrogen atmosphere. The mixture was heated to 50 °C and after 10 min cooled to r.t,
diluted with ethyl acetate, washed with water, dried over MgSO₄, filtered and concentrated under reduced pressure.

The resulting aldehyde 18 was then dissolved in methanol (3.5 mL) and NaBH₄ (98.6 mg, 2.60 mmol, 7 eq) was added and the mixture stirred at 0 °C for 2h. More NaBH₄ was then added and after another 10 min at 0 °C and 15 min at room temperature the mixture was quenched with water, concentrated under reduced pressure, re-dissolved in ethyl acetate, washed with water, dried over MgSO₄, filtered and concentrated under reduced pressure to give the crude alcohol (19).

19 was then dissolved in anhydrous DMF (1.9 mL) and cooled to 0 °C. NaH (60%, 53.7 mg, 1.34 mmol, 3.7 eq) and then benzyl bromide (65 µL, 0.547 mmol, 1.5 eq) was added under a nitrogen atmosphere. The reaction was stirred overnight and then quenched with water, diluted with ethyl acetate, washed with water, dried over MgSO₄, filtered and concentrated under reduced pressure. The crude product was purified by flash column chromatography (pentane/ethyl acetate 20:1) to give the product (20) (81 mg, 34% over 6 steps).

\((1R,2R,3S,4S,5R)-1\text{-azido-2,3,4-tri-O-benzyl-1-(benzyloxy)methyl-5-O-p-methoxybenzyl-cycloheptane (20R)}\)

\(R_f\) (pentane/ethyl acetate 20:1) 0.23; \([\alpha]_D^{293K} +6.0 \text{ (c 1, CHCl}_3)\); \(^1\)H-NMR (400 MHz, CDCl₃): \(\delta_H\)

7.39-7.19 (m, 20H, ArH), 7.19-7.12 (m, 2H, ArH from PMB), 6.86-6.78 (m, 2H, ArH from PMB), 4.84 (d, J 10.7 Hz, 1H, CHAr), 4.73 (d, J 11.5 Hz, 1H, CHAr), 4.72 (d, 1H, CHAr), 4.69 (d, J 12.0 Hz, 1H, CHAr), 4.61 (d, 1H, CHAr), 4.67 (d, J 11.1 Hz, 1H, CHAr), 4.57 (d, J 11.9 Hz, 2H, CHAr), 4.47 (d, 1H, CHAr), 4.28 (d, 1H, CHAr), 3.94 (d, J 4.4 Hz, 1H, H2/H3), 3.87 (dd, J 8.4 Hz, J 6.9 Hz, 1H, H2/H3), 3.81 - 3.76 (m, 4H, CH₂, H4), 3.63 (d, J 8.6 Hz, 1H, H5), 3.57 (td, J 8.6 Hz, J 2.7 Hz, 1H, H5), 1.93-1.71 (m, 3H, H6/H7), 1.55 (dd, J 11.3 Hz, J 6.7 Hz, 1H, H6/H7). \(^{13}\)C-NMR (100 MHz, CDCl₃): \(\delta_C\)

159.2 (Ar from PMB), 139.1 (Ar), 138.6 (Ar), 138.1 (Ar), 138.0 (Ar), 131.1 (Ar), 129.4 (Ar), 128.5 (Ar), 128.4 (Ar), 128.4 (Ar), 128.2 (Ar), 127.8 (Ar), 127.8 (Ar), 127.8 (Ar), 127.6 (Ar), 127.6 (Ar), 113.8 (Ar from PMB), 87.7 (C2/C3), 82.9 (C4), 80.4 (C2/C3), 78.8 (C5), 75.5 (CH₂Ar), 74.5 (CH₂Ar), 73.8 (CH₂Ar), 73.8 (C8), 73.4 (CH₂Ar), 72.5 (CH₂Ar), 67.1 (C1), 55.4 (CH₃), 26.9 (C6/C7), 26.7 (C6/C7). HRMS(ESI): calcd. for C₄₄H₄₇N₃O₆Na 736.3357; found 736.3363.
(1R,2R,3S,4S,5S)-1-azido-2,3,4- tri-O- benzyl-1-(benzyloxy)methyl-5-O-p-methoxybenzyl-cycloheptane (20S)

Rf (pentane/ethyl acetate 20:1) 0.10; 1H-NMR (400 MHz, CDCl3): δH 7.46-7.20 (m, 23H, ArH), 6.82 - 6.71 (m, 2H, ArH from PMB), 4.73-4.52 (m, 9H), 4.48 (d, J 11.1 Hz, 1H), 4.07-3.95 (m, 4H), 3.81 (s, 3H, CH3), 3.83-3.78 (m, 1H) 3.73 (d, J 9.8 Hz, 1H), 2.31 (ddd, J 14.7 Hz, J 11.0 Hz, J 4.1 Hz, 1H, H6a/H7), 2.04-1.90 (m, 1H, H6a/H7), 1.79 (ddt, J 14.7 Hz, J 11.2 Hz, J 3.9 Hz, 1H, H6b), 1.40-1.27 (m, 1H, H6a/H7). HRMS(ESI): calcd. for C44H47N3O6Na 736.3357; found 736.3385.

(2R,3S,4R,5R)-5-Azido-2,3,4- tri-O-benzyl-5-(benzyloxy)methyl-cycloheptanone (21)

The PMB ether (20) (69.6 mg, 97.6 µmol) was mixed with trifluoroacetic acid/dichloromethane 1:1 (7.6 mL) at 0 °C. After 1h 10 min the mixture had turned dark purple and was concentrated under reduced pressure. The crude alcohol was redisolved in dichloromethane (4.4 mL) and added Dess-Martin periodinate (109 mg, 0.255 mmol, 2.6 eq). After 30 min the reaction was quenched with sat. aq. NaHCO3 and Na2S2O3. The mixture was left to stir for 1 h and then extracted with dichloromethane, dried over MgSO4, filtered and concentrated under reduced pressure. The crude product was combined with another batch (18 µmol) and purified by flash column chromatography (pentane/ethyl acetate 8:1) to give the ketone (21) (43.2 mg, 65% over 2 steps).

Rf (pentane/ethyl acetate 6:1) 0.36; [α]D293K +33.0 (c 1, CHCl3); 1H-NMR (400 MHz, CDCl3): δH 7.45-7.21 (m, 18H, ArH), 7.08 (dd, J 6.6 Hz, J 2.9 Hz, 2H, ArH), 4.78 (d, J 11.8 Hz, 1H, CHPh), 4.71 (d, J 11.6 Hz, 2H, 2xCCHPh), 4.69 (d, J2,3 5.6 Hz, 1H, H2), 4.48 (d, J 12.0 Hz, 1H, CHPh), 4.45 (d, J 11.3 Hz, 2H, 2xCCHPh), 4.42 (d, J 10.3 Hz, 1H, CHPh), 4.11 (d, J 11.4 Hz, 1H, CHPh), 4.01 (d, J3,4 3.7 Hz, 1H, H4), 3.89 (dd, 1H, H3), 3.61 (d, J8a,8b 9.6 Hz, 1H, H8a), 3.47 (d, H8b), 2.60-2.38 (m, 2H, H6/H7), 1.99 (ddd, J 14.1 Hz, J 9.1 Hz, J 4.7 Hz, 1H, H6/H7), 1.75 (ddd, J 14.5 Hz, J 7.9 Hz, J 5.3 Hz, 1H, H6/H7). 13C-NMR (100 MHz, CDCl3): δC 205.9 (C=O), 137.9 (Ar), 137.7 (Ar), 137.3 (Ar), 128.5 (Ar), 128.4 (Ar), 128.2 (Ar), 128.2 (Ar), 128.0 (Ar), 127.9 (Ar), 127.6 (Ar), 87.3 (C2), 80.0 (C3), 78.4 (C4), 74.4 (C8), 74.0 (CH2Ar), 73.4 (CH2Ar), 73.2 (CH2Ar), 72.5 (CH2Ar), 66.1 (C5), 37.6 (C6/C7), 26.0 (C6/C7). HRMS(ESI): calcd. for C36H37N3O5Na 614.2625; found 614.2623.

(1S,2R,3S,4R,5R)-2,3,4-tris(benzyloxy)-5-((benzyloxy)methyl)-8-azabicyclo[3.2.1]octan-1-ol (22)
The ketone (21) (42.2 mg, 71.3 µmol) was dissolved in methanol/ethyl acetate 1:1 (2.6 mL). Three drops of triethylamine and then 20% Pearlmans catalyst (12.5 mg, 0.0178 mmol, 20%) were added. The mixture was bubbled through with hydrogen and stirred under a hydrogen atmosphere for 15 min. The catalyst was then filtered through a syringe filter and the filtrate concentrated under reduced pressure to give the crude hemiaminal (22) (38.8 mg).  

Rf (pentane/ethyl acetate 1:1) 0.36; [α]D<sup>293K</sup> +15.4 (c 1, CHCl₃); <sup>1</sup>H-NMR (400 MHz, CDCl₃): δH 7.43 – 7.16 (m, 20H, ArH), 5.04 (d, J 11.4 Hz, 1H, CHHPh), 4.92 (d, J 10.8 Hz, 1H, CHHPh), 4.81 (d, J 11.1 Hz, 1H, CHHPh), 4.79 (d, J 10.8 Hz, 2H, CH2Ph, CHHPh), 4.60 (d, 1H, CHHPh), 4.53 (d, J 11.7 Hz, 1H, CHHPh), 4.43 (d, 1H, CHHPh), 3.75 (d, J<sub>8a,6b</sub> 9.0 Hz, 1H, H6a), 3.74 (d, J<sub>2,3</sub> 8.2 Hz, 1H, H2), 3.65 (t, J<sub>3,4</sub> 8.3 Hz, 1H, H3), 3.53 (d, 1H, H4), 3.36 (d, 1H, H6b), 2.34 (ddd, J<sub>8a,8b</sub> 12.0 Hz, J<sub>8a,7a</sub> 9.9 Hz, J<sub>8a,7b</sub> 4.0 Hz, 1H, H8a), 1.90 (ddd, J<sub>7a,7b</sub> 12.6 Hz, J<sub>7a,8a</sub> 4.5 Hz, 1H, H7a), 1.66 (td, J<sub>7b</sub>, 12.2 Hz, 1H, H8b), 1.56 (td, 1H, H7b). <sup>13</sup>C-NMR (100 MHz, CDCl₃): δC 139.1 (Ar), 138.9 (Ar), 138.9 (Ar), 138.1 (Ar), 128.5 (Ar), 128.5 (Ar), 128.4 (Ar), 128.1 (Ar), 127.9 (Ar), 127.8 (Ar), 127.7 (Ar), 127.6 (Ar), 127.6 (Ar), 91.7 (C1), 87.2(C2), 84.9 (C3), 82.8(C4), 77.4 (CH2Ph), 75.7 (CH2Ph), 74.7 (CH2Ph), 73.5 (CH2Ph), 73.2 (CH2Ph), 62.9 (C5), 31.7 (C8), 25.5 (C7). HRMS(ESI): calcd. for C₃₆H₃₉NO₅H 566.2901; found 566.2910.

(1S,2R,3S,4R,5R)-5-(hydroxymethyl)-8-azabicyclo[3.2.1]octane-1,2,3,4-tetraol - Nojiristegine (1)

The crude benzyl protected compound (21) (66.8 µmol) was dissolved in methanol/ethyl acetate 1:1 (2.6 mL) and added 20% Pearlmans catalyst (10.2 mg, 14.5 µmol, 22%). The mixture was bubbled through hydrogen and stirred under a hydrogen atmosphere for 3h 50 min. 2 drops of concentrated hydrochloric acid was then added and the reaction left overnight. The next morning the catalyst was filtered of through a syringe filter and the filtrate concentrated under reduced pressure. The crude compound was purified by flash column chromatography (5% NH₄OH in ethanol) to give nojiristegine (1) (16.5 mg, quant.)
$R_f$ (5% NH$_4$OH in ethanol) 0.31; $[\alpha]_D^{293K}$ -2.4 (c 1, H$_2$O); $^1$H-NMR (400 MHz, D$_2$O): $\delta$H 3.78 (d, $J_{6a,6b}$ 12.2 Hz, 1H, H6a), 3.68 (d, $J$ 8.7 Hz, 1H, H2), 3.58 (d, $J$ 8.7 Hz, 1H, H4), 3.56 (d, 1H, H6b), 3.46 (t, $J_{2,3}$ 8.7 Hz, 1H, H3), 2.28 – 2.14 (m, 1H, H8a), 1.88 – 1.73 (m, 2H, H7a+H8b), 1.73 – 1.62 (m, 1H, H7b). $^{13}$C-NMR (100 MHz, D$_2$O): $\delta$C 91.5 (C1), 75.9 (C2), 74.9 (C3), 72.1 (C4), 65.3 (C5), 62.3 (C6), 28.3 (C8), 23.2 (C7). HRMS(ESI): calcd. for C$_8$H$_{15}$NO$_5$H 206.1023; found 206.1023.

**Methyl 6-O-trityl-$\alpha$-D-mannopyranoside (52)**

Commercially available methyl $\alpha$-D-mannopyranoside (4) (4.96 g, 25.5 mmol) was dissolved in anhydrous pyridine (40 mL). Trityl chloride (10.8 g, 38.7 mol, 1.5 eq) and DMAP (634 mg, 5.15 mmol, 0.2 %) were added and the reaction mixture was stirred at room temperature overnight under a nitrogen atmosphere. The pyridine was then removed under reduced pressure and the residue was diluted with dichloromethane. The organic phase was washed with 1M aq. HCl, sat. aq. NaHCO$_3$ and brine and the organic layer was dried over MgSO$_4$, filtered and concentrated under reduced pressure giving the tritylated crude compound (52) (18.1 g) as a white solid.

$R_f$ (ethyl acetate/pentane 1:1) 0.39. HRMS(ESI): calcd. for C$_{26}$H$_{28}$O$_6$Na 459.1778; found 459.1782.

**Methyl 2,3,4-Tri-O-benzyl-6-O-trityl-$\alpha$-D-mannopyranoside (53)**

The crude compound 52 was dissolved in anhydrous DMF (110 mL) under a nitrogen atmosphere. The mixture was cooled to 0 °C and 60 % sodium hydride in mineral oil (6.20 g, 156 mmol, 6 eq) was slowly added, followed by dropwise addition of benzyl bromide (13.9 mL, 117 mmol, 4.5 eq) under formation of hydrogen gas. The reaction mixture was then allowed slowly to warm to room temperature and react overnight. The mixture was quenched by addition of methanol (30 mL) and then evaporated under reduced pressure. The residue was diluted with diethyl ether and washed with water. The aqueous phase was extracted with diethyl ether and the combined organic phases were washed with brine, dried over MgSO$_4$, filtered and concentrated under reduced pressure yielding the crude compound (53) (32.6 g) as a brown oil.

$R_f$ (ethyl acetate/pentane 1:1) 0.39. HRMS(ESI): calcd. for C$_{47}$H$_{50}$O$_6$NH$_4$ 724.3633; found 724.3640.

**Methyl 2,3,4-tri-O-benzyl-6-hydroxy-$\alpha$-D-mannopyranoside (54)**

The crude compound (53) was dissolved in methanol/dichloromethane 2:1 and then $p$-toluenesulfonic acid was added until pH < 4. The reaction was stirred overnight. The mixture was
then neutralised with triethylamine and evaporated under reduced pressure. The residue was diluted with ethyl acetate and washed with water. The aqueous phase was extracted with ethyl acetate and the combined organic phases were washed with brine, dried with MgSO₄, filtered and concentrated. The crude product was purified by flash column chromatography (ethyl acetate/pentane 1:10 → 1:1) to give 54 as a yellow oil (7.86 g, 65% over 3 steps).

Rf (ethyl acetate/pentane 1:3) 0.27; ¹H-NMR (400 MHz, CDCl₃): δH 7.40–7.27 (15H, m, ArH), 4.97 (1H, d, J 10.9 Hz, CH2Ph), 4.81 (1H, d, J 12.3 Hz, CH2Ph), 4.72 (1H, s, H1), 4.71 (1H, d, CH2Ph), 4.68 (1H, d, CH2Ph), 4.66 (2H, s, CH2Ph), 4.00 (1H, t, J4,3 = J4,5 9.5 Hz, H4), 3.93 (1H, dd, J3,2 2.9 Hz, H3), 3.87 (1H, dd, J2,1 9.2 Hz, H2), 3.83–3.78 (1H, m, H6), 3.65 (1H, ddd, J5,6a 4.4 Hz, J5,6b 3.1 Hz, H5), 3.22 (3H, s, CH3), 2.16 (1H, bs, OH). ¹³C-NMR (100 MHz, CDCl₃): δC 138.6 (Ar), 138.5 (Ar), 138.3 (Ar), 128.5 (Ar), 128.3 (Ar), 128.1 (Ar), 127.8 (Ar), 127.7 (Ar), 99.4 (C1), 80.3 (C3), 75.3 (CH2Ph), 75.0 (C4), 74.8 (C2), 73.0 (CH2Ph), 72.3 (CH2Ph), 72.2 (C6), 62.5 (C6), 54.9 (CH3). HRMS(ESI): calcd. for C28H32O6N4 482.2537; found 482.2546.

NMR data was in accordance with previously reported values.⁴⁴

**Methyl 2,3,4-tri-O-benzyl-6-deoxy-6-iodo-α-D-mannopyranoside (6)**

Methyl 2,3,4-tri-O-benzyl-α-D-mannopyranoside (54) (24.3 g, 52.5 mmol) was dissolved in anhydrous toluene (300 mL) under a nitrogen atmosphere. Triphenylphosphine (34.5 g, 131 mmol, 2.5 eq), imidazole (8.59 g, 126 mmol, 2.4 eq) and iodine (14.7 g, 57.8 mmol, 1.1 eq) were added and the reaction mixture was heated to reflux. The reaction was stirred for 1 h 30 min., and then cooled to room temperature. Na₂S₂O₃ (aq) (300 mL) was added and the mixture was washed with water and brine, dried with MgSO₄, filtered and concentrated. The crude product was purified by flash column chromatography (ethyl acetate/pentane 1:20 → 1:2) to give the iodide (6) as a yellow oil (28.1 g, 93%).

Rf (ethyl acetate/pentane 1:19) 0.32; ¹H-NMR (400 MHz, CDCl₃): δH 7.44–7.28 (15H, m, ArH), 5.01 (1H, d, J 11.0 Hz, CH2Ph), 4.78 (d, J 12.5 Hz, 1H, CH2Ph), 4.77 (1H, d, J 1.8 Hz, H1). 4.73 (1H, d, CH2Ph), 4.70 (1H, d, CH2Ph), 4.62 (2H, s, CH2Ph) 3.91 (1H, dd, J3,4 9.3 Hz, J3,2 3.0 Hz, H3), 3.84–3.77 (2H, m, H4, H2), 3.63–3.51 (2H, m, H6a, H5) 3.39 (3H, s, CH3) 3.35 (1H, ddd, J6a,6b 10.2 Hz, J5,6b 2.4 Hz, H6b). ¹³C-NMR (100 MHz, CDCl₃): δC 138.4 (Ar), 138.3 (Ar), 138.3 (Ar), 128.6 (Ar), 128.5 (Ar), 128.1 (Ar), 127.9 (Ar), 127.8 (Ar), 127.8 (Ar), 127.7 (Ar), 99.1 (C1),
80.0 (C3), 78.7 (C4), 75.5 (CH₂Ph), 74.6 (C2), 72.8 (CH₂Ph), 72.2 (CH₂Ph), 71.5 (C5), 55.2 (CH₃), 7.2 (C6). HRMS(ESI): calcd. for C₂₈H₃₁O₅IN₄ 592.1554; found 592.1554.

NMR data was in accordance with previously reported values.⁴²

(2S,3S,4R)-2,3,4-Tri-O-benzylhex-5-enal (23)

A solution of iodide 6 (541 mg, 0.934 mmol) in anhydrous THF (20 mL) and water (5 mL) was degassed in a sonication bath. Then preactivated zinc dust (625 mg, 10.3 mmol, 11 eq)⁵ was added and the mixture sonicated at 40 °C for 1 h 30 min. The reaction mixture was diluted with water and diethyl ether and filtered through a pad of Celite®. The organic phase was washed with water and brine, dried over Na₂SO₄, filtered and concentrated under reduced pressure to yield the aldehyde (23) (394 mg) as a clear oil without further purification.

Rₛ(ethyl acetate/pentane 1:11) 0.34; ¹H-NMR (400 MHz, CDCl₃): δ₉ 9.66 (1H, s, CHO), 7.35-7.26 (15H, m, ArH), 5.87 (1H, ddd, J₅,₄ 8.0 Hz, J₅,₆ 10.4 Hz, J₅,₆ 17.6 Hz, H₅), 5.37 (1H, d, J₆E,₅ 17.3 Hz, H₆E), 5.35 (1H, d, J₆Z,₅ 10.3 Hz, H₆Z) 4.72-4.61 (4H, m, 2xC₇H₇Ph), 4.49 (1H, d, J 11.7 Hz, CH₇Ph), 4.37 (1H, d, J 11.8 Hz, CH₈Ph), 4.11 (1H, m, H₄) 4.07 (1H, dd, J 3,₂ 3.7 Hz, J₂,₁ 1.2 Hz, H₂), 3.89 (1H, dd, J₃,₄ 5.6 Hz, H₃). ¹³C-NMR (100 MHz, CDCl₃): δ₁ 201.8 (CHO), 138.2 (Ar), 138.0 (Ar), 137.5 (Ar), 135.3 (C₅), 128.6 (Ar), 128.5 (Ar), 128.4 (Ar), 128.2 (Ar), 128.1 (Ar), 128.0 (Ar), 127.9 (Ar), 127.7 (Ar), 120.0 (C₆), 83.8 (C₄), 83.0 (C₃), 80.7 (C₂), 74.3 (CH₂Ph), 73.0 (CH₃), 70.9 (CH₂Ph). HRMS(ESI): calcd. for C₂₇H₂₈O₅Na 439.1880; found 439.1880.

NMR data was in accordance with previously reported values.⁴³

(4R/S,5R,6S,7R)-5,6,7-Tri-O-benzyl-1,8-nonadiene-4-ol (24)

To a stirred solution of the crude aldehyde (23) (0.934 mmol) in 50% aqueous THF (14 mL) was added indium dust (227 mg, 1.98 mmol, 2 eq) and allyl bromide (0.30 mL, 3.47 mmol, 4 eq). The mixture was stirred overnight at room temperature. Sat. aq. NaHCO₃ (5 mL) was added to neutralise the reaction mixture, which was then extracted with chloroform and the combined organic phases dried over MgSO₄, filtered and concentrated. The crude product was purified by flash column chromatography (ethyl acetate/pentane 1:20) to give the product (24) as a colourless oil (310 mg, b Zinc dust (2.06 g) was mixed with 1M HCl (20 mL) and stirred at room temperature for 1 hour. Then the mixture was filtered and the zinc dust was washed with water and diethyl ether. The zinc dust was dried under high vacuum at about 30 min and in the first 5 min heated with a heat gun.)
76% over 2 steps). According to NMR the product was a 1:2.2 mixture of S- and R-diastereoisomers.

\( R_f (\text{ethyl acetate/pentane 1:9}) 0.40; ^1\text{H-NMR} (400 \text{ MHz, CDCl}_3): \delta_H 7.27-7.09 (30\text{H, m, ArH}), 5.87-5.58 (4\text{H, m}), 5.28-5.19 (4\text{H, m}), 4.66 (4\text{H, dt, } J \text{ 18.6 Hz, } J \text{ 11.5 Hz}), 4.55 (2\text{H, d, } J \text{ 11.8 Hz}), 4.39-4.34 (4\text{H, m}), 4.28 (q, } J \text{ 11.0 Hz), 4.04 (1\text{H, dd, } J \text{ 7.4 Hz, } J \text{ 5.8 Hz}), 3.97 (1\text{H, dd, } J \text{ 7.9 Hz, } J \text{ 5.0 Hz}), 3.85 (2\text{H, dd, } J \text{ 10.2 Hz, } J \text{ 7.4 Hz}), 3.74-3.69 (2\text{H, m}), 3.53-3.47 (2\text{H, m}), 2.96 (1\text{H, s}), 2.57 (1\text{H, s}), 2.43-2.36 (1\text{H, m}), 2.27-2.11 (3\text{H, m}). \) Major isomer: \(^{13}\text{C-NMR} (100 \text{ MHz, CDCl}_3): \delta_C 138.3 (\text{Ar}), 138.3 (\text{Ar}), 138.3 (\text{Ar}), 136.0 (\text{CH=CH}_2), 119.1 (\text{CH=CH}_2), 117.4 (\text{CH=CH}_2), 82.9, 81.5, 81.5, 74.7, 72.9, 71.1, 70.6, 37.7 (\text{C3}). \) Minor isomer: \(^{13}\text{C-NMR} (100 \text{ MHz, CDCl}_3): \delta_C 138.2 (\text{Ar}), 138.1 (\text{Ar}), 138.0 (\text{Ar}), 135.7 (\text{CH=CH}_2), 135.2 (\text{CH=CH}_2), 119.3 (\text{CH=CH}_2), 117.2 (\text{CH=CH}_2), 82.1, 81.0, 78.1, 75.3, 73.0, 70.7, 70.3, 38.7 (\text{C3}). \) Major isomer + minor isomer: \(^{13}\text{C-NMR} (100 \text{ MHz, CDCl}_3): \delta_C 128.4 (\text{Ar}), 128.4 (\text{Ar}), 128.3 (\text{Ar}), 128.1 (\text{Ar}), 128.0 (\text{Ar}), 128.0 (\text{Ar}), 127.9 (\text{Ar}), 127.8 (\text{Ar}), 127.8 (\text{Ar}), 127.6 (\text{Ar}), 127.6(\text{Ar}). \) HRMS(ESI): calcd. for C\(_{30}\)H\(_{34}\)O\(_4\)Na 481.2349; found 481.2350.

(4\text{R,5R,6S,7R})-5,6,7-Tri-O-benzyl-1,8-nonadiene-4-acetate (25)

The alcohol (24) (2.67 g, 5.81 mmol) was dissolved in anhydrous pyridine (20 mL) under an atmosphere of nitrogen. Acetic anhydride (20 mL) and DMAP (35.2 mg, 0.293 mmol, 5 mol%) were added and stirred at room temperature for 3 h 30 min. The reaction was cooled on a water bath and quenched by slow addition of water (20 mL) and then diluted with ethyl acetate. The organic phase was washed with 1M aq. HCl and sat. aq. NaHCO\(_3\), dried over MgSO\(_4\), filtered and concentrated under reduce pressure to yield the acetylated compound (25) (2.93 g) without further purification.

\( R_f (\text{ethyl acetate/pentane 1:15}) 0.32; ^1\text{H-NMR} (400 \text{ MHz, CDCl}_3): \delta_H 7.33 – 7.13 (m, 20.4\text{H}), 5.86 (\text{tdd, } J \text{ 17.7 Hz, } J \text{ 10.3 Hz, } J \text{ 7.7 Hz, 1.3H}), 5.61 (\text{ddt, } J \text{ 17.3 Hz, } J \text{ 10.6 Hz, } J \text{ 7.1 Hz, 1.3H}), 5.36 – 5.17 (m, 3.7\text{H}), 4.96 – 4.89 (m, 2.4\text{H}), 4.71 (d, } J \text{ 11.4 Hz, 0.9H)), 4.64 (d, } J \text{ 11.4 Hz, 1H}), 4.63 – 4.47 (m, 3.1\text{H}), 4.48 (s, 0.5\text{H}), 4.31 (d, } J \text{ 11.3 Hz, 1H}), 4.27 (d, } J \text{ 11.9 Hz, 1.1H}), 4.09 (dd, } J \text{ 7.9 Hz, } J \text{ 4.0 Hz, 0.3H}), 4.04 (dd, } J \text{ 7.7 Hz, } J \text{ 4.9 Hz, 0.9H}), 3.77 (dd, } J \text{ 6.0 Hz, } J \text{ 3.3 Hz, 1H}), 3.69 (dd, } J \text{ 6.8 Hz, } J \text{ 2.8 Hz, 0.3H}), 3.62 – 3.58 (m, 0.2H), 3.56 (dd, } J \text{ 6.0 Hz, } J \text{ 4.9 Hz, 1H}), 2.39 (t, } J \text{ 6.7 Hz, 0.9H}), 2.34 (t, } J \text{ 7.0 Hz, 0.4H}), 1.89 (s, 1.1H), 1.89 (s, 2.7H). \) Major isomer: \(^{13}\text{C-NMR} (100 \text{ MHz, CDCl}_3): \delta_C 170.1 (\text{C=O}), 138.6 (\text{Ar}), 138.5 (\text{Ar}), 138.4 (\text{Ar}), 136.0 (\text{CH=CH}_2), 134.6 (\text{CH=CH}_2), 119.0 (\text{CH=CH}_2), 117.4 (\text{CH=CH}_2), 81.9, 81.1, 79.6, 74.5 (\text{CH=CH}_2), 73.5 (\text{CH=CH}_2), 73.0, 70.6
(CH₂Ph), 34.6 (C3), 21.2 (CH₃). Minor isomer: ¹³C-NMR (100 MHz, CDCl₃): δC 170.7 (C=O), 138.5 (Ar), 138.4 (Ar), 138.3 (Ar), 136.3 (CH₂=CH), 134.0 (CH₂=CH), 119.1 (CH₂=CH), 117.9 (CH₂=CH), 81.7, 80.8, 78.1, 74.9 (CH₂Ph), 73.8, 72.4 (CH₂Ph), 70.5 (CH₂Ph), 36.3 (C3), 21.4 (CH₃). Major isomer + minor isomer: ¹³C-NMR (100 MHz, CDCl₃): δC 128.5 (Ar), 128.5 (Ar), 128.4 (Ar), 128.4 (Ar), 128.4 (Ar), 128.2 (Ar), 128.1 (Ar), 128.1 (Ar), 127.9 (Ar), 127.9 (Ar), 127.8 (Ar), 127.7 (Ar), 127.7 (Ar), 127.6 (Ar). HRMS(ESI): calcd. for C₃₂H₃₆O₅NH₄ 518.2901; found 518.2903.

(3R,4S,5R,6S)- 6-O-Acetyl-3,4,5-tri-O-benzyl-cycloheptene (26) and (3R,4S,5R,6R)- 6-O-Acetyl-3,4,5-tri-O-benzyl-cycloheptene (27)

The crude acetate (25) (5.81 mmol) was dissolved in anhydrous toluene (30 mL) under an atmosphere of nitrogen. Grubbs-Hoveyda 2nd generation catalyst (35.7 mg, 54.8 µmol, 1 mol%) was added to the mixture which was immediately put on a preheated 80 °C oil bath. The mixture stirred for 2 h before more catalyst was added. After additional 1 h 30 min the reaction was concentrated under reduced pressure and the crude product was purified by flash column chromatography (diethyl ether/chloroform 0:1 → 1:40) yielding 27 and 26 (1.96 g, 71% over 2 steps) which were partly separated.

(3R,4S,5R,6S)- 6-O-Acetyl-3,4,5-tri-O-benzyl-cycloheptene (26):

Colourless oil (53.7 mg, 19% over 2 steps); Rf (ethyl acetate/pentane 1:13) 0.38; [α]D²⁹⁸K -1.2 (c 1.0, CHCl₃). [H-NMR (400 MHz, CDCl₃): δH 7.38 – 7.23 (m, 15H, ArH), 5.72 (1H, ddd, J₁,7b 6.9 Hz, J₁,7a 1.2 Hz, H1), 5.15 (1H, td, J₆,₅ = J₆,₇b 6.9 Hz, J₆,₇a 3.2 Hz, H6), 4.78 (1H, d, J 9.7 Hz, CHHPh), 4.75 (1H, d, CHHPh), 4.73 (1H, d, J 12.2 Hz, CHHPh), 4.66 (1H, d, CHHPh), 4.62 (1H, d, J 11.8 Hz, CHHPh), 4.55 (1H, d, CHHPh), 4.35 (1H, dd, J₃,₄ 7.2 Hz, H3), 3.96 (1H, dd, J₃,₅,₄ 1.8 Hz, H5), 3.91 (1H, dd, H4), 2.56 (1H, ddd, J₇₈,₇₉ 15.6 Hz, H7a), 2.33 (1H, dt, H7b), 1.96 (3H, s, CH₃). ¹³C-NMR (100 MHz, CDCl₃): δC 169.9 (C=O), 138.6 (Ar), 138.6 (Ar), 138.5 (Ar), 132.2 (CH=CH), 128.2 (Ar), 128.2 (Ar), 128.7 (Ar), 127.9 (Ar), 127.5 (Ar), 127.4 (Ar), 126.2 (CH=CH), 80.5 (C5), 78.3 (C4), 75.9 (C3), 73.4 (CH₂Ph), 73.3 (CH₂Ph), 71.8 (CH₂Ph / C6), 71.8 (CH₂Ph/C6), 28.2 (C7), 21.1 (CH₃). HRMS(ESI): calcd. for C₃₀H₃₂O₃Na 495.2142; found 495.2147.

(3R,4S,5R,6R)- 6-O-Acetyl-3,4,5-tri-O-benzyl-cycloheptene (27): colourless oil (1.37 g, 50% over 2 steps); Rf (ethyl acetate/pentane 1:13) 0.38; [α]D²⁹⁸K -4.0 (c 1.0, CHCl₃). [H-NMR (400
MHz, CDCl$_3$): δ$_H$ 7.42-7.23 (15H, m, ArH), 5.76 (1H, dt, $J_{2,1}$ 12.4 Hz, $J_{2,3}$ 2.7 Hz, H2), 5.62 (1H, m, H1), 4.92 (1H, d, $J_{11.9}$ Hz, C$H$HPh), 4.84 (1H, d, $J_{11.7}$ Hz, C$H$HPh), 4.79 (1H, d, C$H$HPh), 4.76 (1H, d, $J_{11.4}$ Hz, C$H$HPh), 4.75 (1H, dt, $J_{6.7a}$ 11.4 Hz, $J_{6.7b}$ = $J_{6.5}$ 1.3 Hz, H6), 4.68 (1H, d, C$H$HPh), 4.60 (1H, dq, $J_{3,4}$ 9.2 Hz, H3), 4.14 (1H, s, H5), 3.56 (1H, dd, $J_{4,5}$ 1.4 Hz H4), 2.78 (1H, ddq, $J_{7a,7b}$ 14.6 Hz, $J_{7a,1}$ 2.9 Hz, H7a), 2.05 (1H, ddd, $J_{7b,1}$ 8.8 Hz, $J_{7b,1}$ 3.5 Hz, H7b), 1.99 (3H, s, C$H$3).

$^{13}$C-NMR (100 MHz, CDCl$_3$): δ$_C$ 170.4 (C=O), 139.0 (Ar), 138.9 (Ar), 134.5 (C$H$=CH), 128.4 (Ar), 128.3 (Ar), 128.0 (Ar), 127.9 (Ar), 127.7 (Ar), 127.6 (Ar), 124.1 (C$H$=CH), 83.1 (C4), 80.1 (C5), 77.1 (C3), 74.7 (C$H$2Ph), 73.7 (C$H$2Ph), 73.4 (C$H$2Ph), 73.2 (C6), 26.9 (C7), 21.4 (C$H$3). HRMS(ESI): calcd. for C$_{30}$H$_{32}$O$_5$Na 495.2142; found 495.2143.

(3R,4S,5R,6R)-3,4,5-Tri-O-benzyl-6-hydroxy-cycloheptene (29)

Sodium (70.2 mg, 2.96 mmol, 1 eq.) was dissolved in anhydrous methanol (10 mL). The sodium methoxide solution (10 mL) was added to the acetylated compound (27) (1.36 g, 2.88 mmol) under an atmosphere of nitrogen. The reaction mixture was stirred overnight at room temperature. The mixture was concentrated under reduced pressure, diluted with dichloromethane and washed with water, dried with MgSO$_4$, filtered and concentrated. The deacetylated crude product (29) (1.22 g) was used without further purification.

$R_f$ (diethyl ether/chloroform 1:40) 0.23; $^1$H-NMR (400 MHz, CDCl$_3$): δ$_H$ 7.39-7.27 (15H, m, ArH), 5.87-5.75 (2H, m, C$H$=CH), 4.92 (1H, d, $J_{11.9}$ Hz, C$H$HPh), 4.84 (1H, d, $J_{11.9}$ Hz, C$H$HPh), 4.78 (1H, d, C$H$HPh), 4.73 (1H, d, C$H$HPh), 4.65 (1H, d, $J_{11.8}$ Hz, C$H$HPh), 4.58 (1H, d, C$H$HPh), 4.42 (1H, dd, $J_{3,4}$ 8.0 Hz, $J_{3,2}$ 3.7 Hz, H3), 4.04 (1H, t, $J_{5,4}$ = $J_{5,6}$ 1.8 Hz, H5), 3.86 (1H, ddt, $J_{6,OH}$ 9.6 Hz, $J_{6,7b}$ 7.2 Hz, $J_{6,5}$ 2.4 Hz, H6), 3.80 (1H, dd, H4), 2.89 (1H, d, $J_{OH,6}$ 9.7 Hz, OH), 2.64-2.55 (1H, m, H7a), 2.19 (1H, ddd, $J_{7b,7a}$ 14.3 Hz, $J_{7b,1}$ 2.6 Hz, H7b). $^{13}$C-NMR (100 MHz, CDCl$_3$): δ$_C$ 139.0 (Ar), 138.7 (Ar), 138.7 (Ar), 132.8 (C$H$=CH), 128.5 (Ar), 128.5 (C$H$=CH), 128.4 (Ar), 128.2 (Ar), 128.0 (Ar), 127.8 (Ar), 127.8 (Ar), 127.8 (Ar), 127.7 (Ar), 127.7 (Ar), 81.9 (C4, C5), 75.1 (C3), 74.2 (C$H$2Ph), 73.6 (C$H$2Ph), 72.2 (C$H$2Ph), 70.8 (C6), 31.3 (C7). HRMS(ESI): calcd. for C$_{28}$H$_{30}$O$_4$Na 453.2036; found 453.2040.

(3R,4S,5R,6R)-3,4,5-Tri-O-benzyl-6-O-p-methoxybenzyl-cycloheptene (34)

The crude alcohol (29) (2.88 mmol) was dissolved in anhydrous DMF (20 mL) under an atmosphere of nitrogen and cooled to 0 °C. Sodium hydride (234 mg, 5.74 mmol, 2 eq) and p-methoxybenzyl chloride (0.77 mL, 5.71 mmol, 2 eq) were added to the reaction mixture which was
heated to room temperature. The mixture was stirred for 4 h 30 min and was quenched by addition of n-butylamine (5 mL), diluted with ethyl acetate, washed with 1M aq. HCl and sat. aq. NaHCO₃, dried over MgSO₄, filtered and concentrated under reduced pressure. The crude product was purified by flash column chromatography (ethyl acetate/pentane 1:25 → 1:15) yielding the PMB-ether (34) (1.33 g, 84% over 2 steps) as a clear oil.

Rf(Ethyl acetate/pentane 1:12) 0.28; [α]D²⁹⁸K +7.6 (c 1.0, CHCl₃); ¹H-NMR (400 MHz, CDCl₃): δH 7.35-7.15 (15H, m, ArH), 7.09 (2H, d, J 8.6 Hz, ArH from PMB), 6.77 (2H, d, J 8.6 Hz, ArH from PMB), 5.60 (1H, dt, J 12.5 Hz, J 2.5 Hz, H2), 5.50 (1H, m, H1), 4.81 (2H, s, CH₂Ph), 4.72 (1H, d, J 11.9 Hz, CHPH), 4.68 (1H, d, J 11.6 Hz, CHPH), 4.59 (1H, d, CHPH), 4.53-4.46 (2H, m, H3, CH₂Ph), 4.30 (1H, d, J 11.7 Hz, CHPH), 4.26 (1H, d, CHPH), 4.08 (1H, s, H5), 3.71 (3H, s, CH₃), 3.29 (1H, dd, J 9.3 Hz, J 1.0 Hz, H4), 3.21 (1H, dd, J 10.7 Hz, J 3.2, H6), 2.67-2.57 (1H, m, H7a), 2.07 (1H, ddd, J 15.1 Hz, J 8.6 Hz, J 2.9 Hz, H7b). ¹³C-NMR (100 MHz, CDCl₃): δC 159.3 (Ar from PMB), 139.5 (Ar), 139.1 (Ar), 139.0 (Ar), 133.4 (C2), 130.6 (Ar), 129.2 (Ar), 128.5 (Ar), 128.4 (Ar), 128.4 (Ar), 128.2 (Ar), 127.9 (Ar), 127.9 (Ar), 127.9 (Ar), 127.6 (Ar), 127.5 (Ar), 127.3 (Ar), 125.0 (C1), 113.9 (Ar from PMB), 83.3 (C4), 80.1 (C5), 78.5 (C6), 74.2 (C3), 73.6 (CH₂Ph), 73.4 (CH₂Ph), 70.5 (CH₂Ph), 55.4 (CH₃), 27.7 (C7). HRMS(ESI): calcd. for C₃₆H₃₈O₅Na 573.2611; found 573.2613.

(3R,4S,5R,6R)-3,4,5-tri-O-benzyl-6-O-p-methoxybenzyl-cycloheptane-1-ol (35) and (2R,3S,4R,5R)-2,3,4-Tri-O-benzyl-5-O-p-methoxybenzyl-cycloheptane-1-ol (36)

The alkene (34) (1.33 g, 2.42 mmol, 1 eq) was dissolved in anhydrous THF (50 mL) under an atmosphere of nitrogen. The stirred reaction mixture was cooled to 0 °C and then BH₃·THF (1M in THF, 7.3 mL, 7.3 mmol, 3 eq) was added drop wise. The reaction was left at 0 °C for 2 hours 30 min, then 35% H₂O₂ (11 mL) and 2M NaOH (6.6 mL) were added drop wise at 0 °C and the mixture was stirred at room temperature overnight. The mixture was then diluted with diethyl ether (50 mL) and washed with water and brine, dried over MgSO₄, filtered and concentrated. The crude product was purified by flash column chromatography (ethyl acetate/pentane 1:3 → 1:1) to yield the two regioisomers 35 and 36 (1.07 g, 78%) as a 1:2.1 mixture which were partly separated.

(3R,4S,5R,6R)-3,4,5-Tri-O-benzyl-6-O-p-methoxybenzyl-cycloheptane-1-ol (35)

Colourless oil (61.4 mg, 4%); Rf(ethyl acetate/pentane 1:3) 0.24; ¹H-NMR (400 MHz, CDCl₃): δH 7.48–7.26 (15H, m, ArH), 7.26–7.20 (2H, m, ArH from PMB), 6.92–6.86 (2H, m, ArH from PMB),
4.87 (1H, d, J 12.2 Hz, CHHPh), 4.82 (d, CHHPh), 4.69 (d, J 11.4 Hz, CHHPh), 4.64 (d, CHHPh), 4.60 (d, J 11.8 Hz, CHHPh), 4.52 (d, CHHPh), 4.42 (d, J 11.6 Hz, CHHPh), 4.36 (d, CHHPh), 4.29 (1H, m, H1), 4.14 (1H, bs, H/H5), 3.99–3.91 (1H, m, H3/H6), 3.81 (3H, s, CH3), 3.74 (1H, dd, J 10.8 Hz, J 5.5 Hz, H/H6), 3.70 (dd, J 6.9 Hz, J 1.1 Hz, H/H5), 2.54 (1H, bs, OH), 2.29 (1H, dd, J 14.7 Hz, J 4.3 Hz, H2/H7), 2.21–2.07 (2H, m, H2/H7), 2.03–1.95 (1H, m, H2/H7).

13C-NMR (100 MHz, CDCl3): δc 159.2 (Ar from PMB), 139.2 (Ar), 138.7 (Ar), 138.6 (Ar), 130.7 (Ar), 129.3 (Ar), 128.5 (Ar), 128.4 (Ar), 128.2 (Ar), 127.9 (Ar), 127.8 (Ar), 127.7 (Ar), 127.7 (Ar), 127.6 (Ar), 127.3 (Ar), 113.9 (Ar PMB), 85.5 (C4/C5), 81.5 (C4/C5), 79.5 (C1), 77.0 (CH2Ph), 73.4 (CH2Ph), 72.7 (CH2Ph), 72.6 (CH2Ph), 71.0 (C3/C6), 66.7 (C3/C6), 55.4 (OCH3), 37.3 (C2/C7), 36.1 (C2/C7).

HRMS(ESI): calcd. for C36H40O6Na 591.2717; found 591.2724.

(2R,3S,4R,5R)-2,3,4-Tri-O-benzyl-5-O-p-methoxybenzyl-cycloheptane-1-ol (36)

Colourless oil (0.181 g, 13%); Rf (Ethyl acetate:pentane 1:5) 0.28; 1H-NMR (400 MHz, CDCl3): δH 7.45–7.24 (15H, m, ArH), 7.16 (2H, d, J 8.6 Hz, ArH from PMB), 6.88–6.82 (2H, m, ArH from PMB), 4.90 (1H, d, J 11.8 Hz, CHHPh), 4.86 (1H, d, CHHPh), 4.69 (2H, s, CH2Ph), 4.57 (1H, d, J 11.8 Hz, CHHPh), 4.46 (1H, d, CHHPh), 4.34 (1H, d, JOH, J 10.1 Hz, OH), 4.32 (1H, d, J 11.7 Hz, CHHPh), 4.26 (1H, d, CHHPh), 4.11 (1H, s, H2), 3.89 (1H, ddd, J 1.7a, J 7.2 Hz, J1.7b, 2.8 Hz, H1), 3.83 (1H, dd, J4.3, 7.3 Hz, J4.5, 2.4 Hz, H4), 3.75 (3H, s, CH3), 3.42 (1H, d, H3), 3.27 (1H, dd, J5.6a, 11.2 Hz, J5.6b, 5.0 Hz, H5), 2.04 (1H, dtd, J6a.6b, 14.3 Hz, J6a.5, 11.8 Hz, J6a.7a, 3.0 Hz, H6a), 1.93 (1H, dtd, J7a.7b, 14.5 Hz, J7a.1 = J7a.6b, 7.1 Hz, H7a), 1.86–1.73 (1H, m, H6b), 1.61 (1H, ddd, H7b).

13C-NMR (100 MHz, CDCl3): δc 159.2 (Ar from PMB), 138.5 (Ar), 138.4 (Ar), 137.7 (Ar), 130.4 (Ar), 129.1 (Ar), 129.0 (Ar), 128.4 (Ar), 128.4 (Ar), 128.2 (Ar), 127.5 (Ar), 127.5 (Ar), 127.7 (Ar), 127.7 (Ar), 113.8 (Ar from PMB), 87.5 (C4), 84.4 (C3), 80.1 (C2), 79.8 (C5), 73.8 (CH2Ph), 73.3 (CH2Ph), 73.0 (CH2Ph), 70.5 (CH2Ph), 68.8 (C1), 55.2 (CH3), 25.8 (C7), 22.3 (C6).

HRMS(ESI): calcd. for C36H40O6Na 591.2717; found 591.2720.

(2S,3R,4R,5R)-2,3,4-Tri-O-benzyl-5-p-methoxybenzyl-cycloheptanone (37)

The alcohol (36) (177 mg, 0.311 mmol) was dissolved in dichloromethane (9.4 mL) and stirred at room temperature. Freshly prepared Dess-Martin periodinane (269 mg 0.634 mmol, 2 eq) was added and after 20 min the reaction was quenched by addition of Na2S2O3(aq) and sat. aq. NaHCO3, stirred for 10 min and then extracted with dichloromethane, dried over MgSO4, filtered and concentrated under reduced pressure. The crude product was purified by flash column
chromatography (ethyl acetate/pentane 1:10) yielding the product (37) (0.152 g, 87%) as a colourless oil.

\( R_f \) (Ethyl acetate/pentane 1:6) 0.48; \([\alpha]_D^{208K} +0.6\) (c 1.0, CHCl\(_3\)); \(^{1}\)H-NMR (400 MHz, CDCl\(_3\)): \(\delta_H\) 7.40–7.29 (15H, m, ArH), 7.18 (2H, d, \(J\) 8.6 Hz, ArH from PMB), 6.88 (2H, d, \(J\) 8.6 Hz, ArH from PMB), 4.84 (2H, s, CH\(_2\)Ph), 4.68 (1H, d, \(J\) 12.0 Hz, CH\(_2\)Ph), 4.63 (1H, d, \(J\) 11.4 Hz, CH\(_2\)Ph), 4.58 (1H, d, CH\(_2\)Ph), 4.54 (1H, d, CH\(_2\)Ph), 4.34 (2H, s, CH\(_2\)Ph), 4.12 (1H, d, \(J\) 8.6 Hz, J\(_{5,6}\) 5.0 Hz, H5), 3.82 (3H, s, CH\(_3\)), 3.69 (1H, s, H4), 3.47 (1H, d, \(J\) 7.2 Hz, H2), 3.82 (3H, s, CH\(_3\)), 3.69 (1H, d, J\(_{3,2}\) 7.3 Hz, H3), 3.47 (1H, d, J\(_{5,6}\) 10.6 Hz, J\(_{5,6}\) 5.0 Hz, H5), 2.55 (td, J\(_{7a,7b}\) = J\(_{7a,6a}\) 11.8, J\(_{7a,6b}\) 4.2 Hz, H7a), 2.47 (1H, ddd, J\(_{6a,6b}\) 14.4 Hz, H6a), 2.09 – 1.98 (1H, d, H6b). \(^{13}\)C-NMR (100 MHz, CDCl\(_3\)): \(\delta_C\) 206.0 (C1), 159.3 (Ar from PMB), 138.6 (Ar), 138.3 (Ar), 137.7 (Ar), 130.2 (Ar), 129.2 (Ar), 128.4 (Ar), 128.2 (Ar), 128.1 (Ar), 127.9 (Ar), 127.8 (Ar), 127.7 (Ar), 127.4 (Ar), 113.9 (Ar from PMB), 86.3 (C2), 82.6 (C3), 79.8 (C5), 79.5 (C4), 73.1 (CH\(_2\)Ph), 73.1 (CH\(_2\)Ph), 72.7 (CH\(_2\)Ph), 70.8 (CH\(_2\)Ph), 55.3 (CH\(_3\)), 35.5 (C7), 26.8 (C6). HRMS(ESI): calcd. for C\(_{28}\)H\(_{32}\)O\(_6\)N\(_4\) 589.2561; found 589.2565.

(1R,2R,3S,4R,5R)-1-Azido-2,3,4-tri-O-benzyl-1-(benzyloxy)methyl-5-p-methoxybenzylcycloheptane (42) and (1S,2R,3S,4R,5R)-1-azido-2,3,4-tri-O-benzyl-1-(benzyloxy)methyl-5-p-methoxybenzyl-cycloheptane (43)

Anhydrous dichloromethane (1.02 mL, 15.9 mmol, 9.6 eq) was slowly added to a solution of freshly prepared LDA (~0.37 M in THF, 8.6 mL, 3.18 mmol, 1.9 eq) at -78 °C under nitrogen atmosphere. The mixture was stirred for 10 min and then a solution of the ketone (37) (936 mg, 1.65 mmol) in THF (7.8 mL) was added. After 50 min at -78 °C the reaction was removed from cooling, quenched with sat. aq. NH\(_4\)Cl and diluted with ethyl acetate. The mixture was washed with water and brine, dried over MgSO\(_4\), filtered and concentrated under reduced pressure to give the crude alcohol (38).

\( R_f \) (Ethyl acetate/pentane 1:10) 0.23; HRMS(ESI): calcd. for C\(_{37}\)H\(_{40}\)O\(_6\)Cl\(_2\)Na 673.2094; found 673.2099.

The crude alcohol (38) (1.65 mmol) was dissolved in anhydrous DMSO (11 mL) and stirred at room temperature under nitrogen atmosphere. DBU (0.40 mL, 2.69 mmol, 1.6 eq) was added and the reaction stirred until TLC analysis showed full consumption of starting material. The reaction was then quenched by addition of water and diluted with ethyl acetate. The organic phase was
washed with water, dried over MgSO₄, filtered and concentrated under reduced pressure to give the crude epoxide (39).

Rₛ(Ethyl acetate/pentane 1:4) 0.25.

The crude epoxide (39) (1.65 mmol) was dissolved in anhydrous DMSO (33 mL) and tetrabutylammonium azide (2.23 g, 7.9 mmol, 4.8 eq) was added. The reaction was stirred at 50°C under an atmosphere of nitrogen. After 3½ h the reaction was quenched by addition of water and diluted with ethyl acetate. The organic phase was washed with water, dried over MgSO₄, filtered and concentrated to give the crude aldehyde (40).

Rₛ(Ethyl acetate/pentane 1:7) 0.34; HRMS(ESI): calcd. for C₃₇H₃₉N₃O₆Na 644.2731; found 644.2727.

The crude aldehyde (40) (1.65 mmol) was dissolved in anhydrous methanol (40 mL) and stirred under an atmosphere of nitrogen at 0 °C. The mixture was added NaBH₄ (555 mg, 15.0 mmol, 9.1 eq) and stirred for 1 h 15 min before it was quenched by addition of water and diluted with dichloromethane. The mixture was extracted with dichloromethane, dried over MgSO₄, filtered and concentrated under reduced pressure to give the crude alcohol (41) (0.873 g).

Rₛ(Ethyl acetate : pentane 1:6) 0.23; ¹H-NMR (400 MHz, CDCl₃): δH 7.38 (1H, d, J 6.8 Hz, ArH), 7.28–7.18 (14H, m, ArH), 7.12 (2H, d, J 8.6 Hz, ArH from PMB), 6.79 (2H, d, J 8.6 Hz, ArH from PMB), 4.91–4.79 (3H, m, CH₂Ar), 4.65 (1H, d, J 12.0 Hz, CHAr), 4.58 (1H, d, J 10.2 Hz, CHAr), 4.47 (1H, d, CHAr), 4.37–4.24 (3H, m), 4.05 (1H, s, J 14.0 Hz), 3.74 (3H, s, CH₃), 3.71 (1H, d, J 5.2 Hz), 3.57 (1H, d, J 11.6 Hz), 3.40 (1H, d, J 9.2 Hz), 3.15 (1H, ddd, J 10.5 Hz, J 4.2 Hz, J 2.7 Hz), 1.95–1.65 (4H, m), 1.26 (1H, ddd, J 15.3 Hz, J 9.3 Hz, J 2.2 Hz), 0 0.84 – 0.72 (1H, m). HRMS(ESI): calcd. for C₃₇H₄₁N₃O₆NH₄ 641.3334; found 641.3334

The crude alcohol (41) (1.65 mmol) was dissolved in anhydrous DMF and cooled to 0 °C. Sodium hydride (2.8 mmol, 1.7 eq) was dissolved under a nitrogen atmosphere. Benzyl bromide (0.25 mL, 2.1 mmol, 1.3 eq) was then slowly added and the mixture stirred overnight. The reaction was quenched with methanol and concentrated under reduced pressure. The residue was redisolved in diethyl ether and washed with water. The combined aqueous phases were extracted with ether and the combined organic phases washed with brine, dried over MgSO₄, filtered and concentrated under reduced pressure. The crude product was purified by flash column chromatography (20:1 -> 10:1 pentane:ethyl acetate) to give an approximately 1:1.7 mixture of epimers (42 and 43), which was partly separated (444 mg, 38% over 5 steps).
(1R,2R,3S,4R,5R)-1-Azido-2,3,4-tri-O-benzyl-1-(benzyloxy)methyl-5-p-methoxybenzyl-cycloheptane (42): (154 mg, 13%), R_f (pentane/ethyl acetate 4:1) 0.5; 1H-NMR (400 MHz, CDCl_3): δ_H 7.41 (d, J 7.1 Hz, 2H, ArH), 7.37 – 7.09 (m, 20H, ArH), 6.83 (d, J 8.5 Hz, 2H, ArH), 4.87 (d, J 8.6 Hz, 1H, CHHAr), 4.86 (s, 2H, CH_2Ar), 4.69 (d, J 10.8 Hz, 1H, CHHAr), 4.66 (d, J 12.2 Hz, 1H, CHHAr), 4.58 (d, J 12.0 Hz, 1H, CHHAr), 4.42 (d, J 12.1 Hz, 1H, CHHAr), 4.41 (d, J 11.9 Hz, 1H, CHHAr), 4.32 (d, J 11.8 Hz, 1H, CHHAr), 4.26 (d, J 11.7 Hz, 1H, CHHAr), 4.15 (d, J 2,3 9.1 Hz, 1H, H_2), 3.74 (s, 3H, CH_3), 3.70 (d, J 3,4 8a,8b 10.1 Hz, 1H, H_8a), 3.66 (d, J 1H, H_8b), 3.51 (d, J 1H, H_7a), 2.13 – 1.97 (m, 1H, H_7a), 1.97 – 1.69 (m, 3H, H_6, H_7b).

13C-NMR (100 MHz, CDCl_3): δ_C 159.2 (Ar from PMB), 139.2 (Ar), 138.8 (Ar), 137.9 (Ar), 130.6 (Ar), 129.1 (Ar), 128.5 (Ar), 128.4 (Ar), 128.3 (Ar), 128.2 (Ar), 128.2 (Ar), 128.1 (Ar), 128.0 (Ar), 127.9 (Ar), 127.8 (Ar), 127.8 (Ar), 127.6 (Ar), 127.5 (Ar), 127.4 (Ar), 113.8 (Ar form PMB), 82.1 (C2), 80.7 (C3), 79.4 (C4), 79.3 (C5), 75.9 (CH_2Ar), 74.2 (CH_2Ar), 73.8 (CH_2Ar), 73.7 (CH_2Ar), 71.6 (C8), 70.4 (CH_2Ar), 68.8 (C1), 55.3 (CH_3), 29.0 (C7), 24.1 (C6).

HRMS(ESI): calcd. for C_{44}H_{47}N_3O_6 NH_4 731.3803; found 731.3612.

(1S,2R,3S,4R,5R)-1-Azido-2,3,4-tri-O-benzyl-1-(benzyloxy)methyl-5-p-methoxybenzyl-cycloheptane (43): (54.9 mg, 5%), R_f (pentane/ethyl acetate 4:1) 0.8; 1H-NMR (400 MHz, CDCl_3): δ_H 7.39 (d, J 6.5 Hz, 1H, ArH), 7.36 – 7.12 (m, 20H, ArH), 6.83 (d, J 8.5 Hz, 1H, ArH from PMB), 4.91 (d, J 11.4 Hz, 1H, CHHAr), 4.84 (s, 2H, CH_2Ar), 4.63 (d, 1H, CHHAr), 4.51 (d, J 11.2 Hz, 1H, CHHAr), 4.48 (d, 1H, CHHAr), 4.45 (s, 2H, CH_2Ar), 4.34 (d, J 11.6 Hz, 1H, CHHAr), 4.28 (d, 1H, CHHAr), 4.06 (s, 1H, H_2), 3.82 (d, J 3,4 8.9 Hz, 1H, H_4), 3.77 (s, 3H, CH_3), 3.66 – 3.56 (m, 4H, H_5+H_3+H_8), 2.24 – 2.14 (m, 1H, H_7a), 2.06 – 1.86 (m, 2H, H_6a, H_7b), 1.82 – 1.70 (m, 1H, H_6b).

13C-NMR (100 MHz, CDCl_3): δ_C 159.2 (Ar from PMB), 139.3 (Ar), 138.7 (Ar), 138.6 (Ar), 137.9 (Ar), 130.8 (Ar), 129.3 (Ar), 129.1 (Ar), 128.5 (Ar), 128.4 (Ar), 128.4 (Ar), 128.4 (Ar), 128.2 (Ar), 128.2 (Ar), 128.0 (Ar), 127.8 (Ar), 127.8 (Ar), 127.6 (Ar), 127.6 (Ar), 127.3 (Ar), 113.9 (Ar from PMB), 83.5 (C5/C3), 82.2 (C4), 79.6 (C2), 78.2 (C5/C3), 75.8 (CH_2Ar), 73.9 (2C, CH_2Ar), 73.7 (CH_2Ar), 73.5 (CH_2Ar), 70.8 (CH_2Ar), 67.1 (C1), 55.4 (CH_3), 28.1 (C7), 25.5 (C6).

HRMS(ESI): calcd. for C_{44}H_{47}N_3O_6 NH_4 731.3803; found 731.3611.

(1R,2R,3S,4R,5R)-1-Azido-2,3,4-tri-O-benzyl-1-(benzyloxy)methyl-cycloheptane (55)

The PMB-ether (42) (154 mg, 0.216 mmol) was dissolved in dichloromethane (2.8 mL) and water (0.12 mL) and DDQ (69 mg, 0.302 mmol, 1.4 eq) was added. The reaction was stirred vigorously for 4 h and then quenched with sat. aq. NaHCO_3, diluted with dichloromethane and washed with...
brine. The organic phase was dried over MgSO\(_4\), filtered and concentrated to give the crude alcohol (55) which was used directly in the next step without further purification.

\[ R_f (\text{pentane/ethyl acetate 4:1}) 0.32, \quad ^1H-NMR (400 MHz, CDCl\(_3\)): \delta_H 7.89 – 7.78 (m, 2H, ArH), 7.40 – 7.17 (m, 19H, ArH), 7.08 – 6.94 (m, 2H, ArH), 4.93 (d, J 11.8 Hz, 1H), 4.82 (t, J 10.8 Hz, 2H), 4.71 (d, J 11.7 Hz, 1H), 4.68 – 4.52 (m, 7H), 4.03 – 3.93 (m, 2H), 3.85 (s, 3H, CH\(_3\)), 3.78 (d, J 9.8 Hz, 1H), 3.72 – 3.63 (m, 1H), 1.97 – 1.80 (m, 2H), 1.73 (dddd, J 31.1 Hz, J 15.2 Hz, J 10.0 Hz, J 4.6 Hz, 2H). \]

\[ ^{13}C-NMR (100 MHz, CDCl\(_3\)): \delta_C 164.6, 138.4, 138.3, 138.2, 137.8, 134.4, 132.0, 129.9, 129.7, 129.0, 128.6, 128.5, 128.4, 128.4, 128.3, 128.3, 128.2, 128.2, 128.1, 127.9, 127.9, 127.9, 127.9, 127.8, 127.8, 127.7, 127.7, 127.7, 127.6, 127.6, 127.5, 114.3, 80.3, 79.6, 78.8, 75.2, 75.1, 74.2, 73.7, 73.6, 73.2, 71.5, 68.5, 55.5, 27.4, 26.4. \]

HRMS(ESI): calcd. for C\(_{36}\)H\(_{39}\)N\(_3\)O\(_5\)Na 616.2782; found 616.2785.

(2R,3S,4R,5R)-5-Azido-2,3,4-tri-O-benzyl-1-(benzyloxy)methyl-cycloheptanone (44)

The crude alcohol (55) (0.216 mmol) was dissolved in dichloromethane (9 mL) and added Dess-Martin periodinane (146 mg, 0.345 mmol, 1.6 eq). Stirred for 50 min and then quenched with Na\(_2\)S\(_2\)O\(_3\) and sat. aq. NaHCO\(_3\). Stirred for 10 min at r.t and left at 4 °C for 2 weeks. The mixture was then extracted with dichloromethane, dried over MgSO\(_4\), filtered and concentrated. The crude compound was purified by flash column chromatography to give the ketone (44) (50.3 mg, 39% over 2 steps)

\[ R_f (\text{pentane/ethyl acetate 8:1}) 0.32; \quad [\alpha]_D^{298K} + 19.2 (c 1, CHCl\(_3\)); \quad ^1H-NMR (400 MHz, CDCl\(_3\)): \delta_H 7.37 – 7.13 (m, 18H, ArH), 7.05 – 6.96 (m, 2H, ArH), 4.76 (d, J 11.9 Hz, 1H, CH\(_2\)Ar), 4.67 (d, J 12.1 Hz, 1H, CH\(_2\)Ar), 4.67 (s, 1H, H2), 4.53 (d, 1H, CH\(_2\)Ar), 4.50 (d, 1H, CH\(_2\)Ar), 4.45 (d, J 11.9 Hz, 1H, CH\(_2\)Ar), 4.43 (d, J 11.0 Hz, 1H, CH\(_2\)Ar), 4.38 (d, 1H, CH\(_2\)Ar), 4.27 (d, 1H, CH\(_2\)Ar), 4.05 (d, J 5.0 Hz, 1H, H3/H4), 4.00 (d, J 5.1 Hz, 1H, H3/H4), 3.78 (d, J 9.1 Hz, 1H, H8a), 3.29 (d, J 9.1 Hz, 1H, H8b), 2.74 (ddd, J\(_{6a,6b}\) 19.3 Hz, J\(_{6a,7a}\) 12.6 Hz, J\(_{6a,7b}\) 3.2 Hz, 1H, H7a), 2.21 (ddd, J\(_{6b,7b}\) 5.5 Hz, J\(_{6b,7a}\) 2.7 Hz, 1H, H7b), 1.88 (ddd, J\(_{7a,7b}\) 15.2 Hz, 1H, H6a), 1.27 (bd, 1H, H6b). \]

\[ ^{13}C-NMR (100 MHz, CDCl\(_3\)): \delta_C 207.2 (C=O), 137.9 (Ar), 137.9 (Ar), 137.7 (Ar), 137.6 (Ar), 128.6 (Ar), 128.5 (Ar), 128.5 (Ar), 128.3 (Ar), 128.3 (Ar), 128.1 (Ar), 127.9 (Ar), 127.9 (Ar), 127.8 (Ar), 127.8 (Ar), 127.7 (Ar), 82.9 (C2), 79.2 (C3/C4), 76.4 (C8), 75.3 (C3/C4), 74.0 (CH\(_2\)Ar), 73.6 (CH\(_2\)Ar), 73.5 (CH\(_2\)Ar), 72.3 (CH\(_2\)Ar), 66.0 (C5), 35.6 (C7), 23.9 (C6). \]

HRMS(ESI): calcd. for C\(_{36}\)H\(_{37}\)N\(_3\)O\(_5\)Na 609.3071; found 609.3078.
(1S,2S,3S,4R,5R)-2,3,4-tris(benzyloxy)-5-((benzyloxy)methyl)-8-azabicyclo[3.2.1]octan-1-ol (45)

The ketone (44) (50.3 mg, 85 µmol) was dissolved in methanol/ethyl acetate 1:1 (3 mL) and added 3 drops of triethyl amine and 20 % Pearlman’s catalyst (11.8 mg, 16.8 µmol, 20 %). The atmosphere was exchanged for hydrogen and the reaction stirred for 5 h. The mixture was then filtered through Celite® and concentrated under reduced pressure, to give the crude product (45) (48.3 mg)

$R_f$ (pentane/ethylacetate 1:1 + 1 % triethyl amine) 0.27; $[\alpha]_{D}^{298K}$ -12 (c 1, CHCl$_3$); $^1$H-NMR (400 MHz, CDCl$_3$): $\delta$H 7.38 – 7.09 (m, 2H, ArH), 4.96 (d, $J$ 11.9, 1H, CHHAr), 4.83 (d, $J$ 10.8 Hz, 1H, CHHAr), 4.81 (d, 1H, 11.7 CHHAr), 4.60 (d, $J$ 11.6 Hz, 1H, CHHAr), 4.56 (d, $J$ 11.7 Hz, 2H, 2xCHHAr), 4.51 (d, $J$ 11.9 Hz, 1H, CHHAr), 4.41 (d, 1H, CHHAr), 3.91 (d, $J_2,3$ 9.2 Hz, 1H, H2), 3.79 (d, $J_3,4$ 3.7 Hz, 1H, H4), 3.74 (d, $J_5a,5b$ 8.9 Hz, 1H, H6a), 3.62 (dd, 1H, H1, H3), 3.27 (d, 1H, H6b), 1.83 – 1.72 (m, 2H, H7a, H8a), 1.70 – 1.59 (m, 1H, H7b/H8b), 1.48 – 1.30 (m, 1H, H7b/H8b). $^1$C-NMR (100 MHz, CDCl$_3$): $\delta$C 139.2 (Ar), 138.9 (Ar), 138.5 (Ar), 138.3 (Ar), 128.6 (Ar), 128.5 (Ar), 128.4 (Ar), 128.4 (Ar), 128.3 (Ar), 127.9 (Ar), 127.8 (Ar), 127.8 (Ar), 127.7 (Ar), 127.7 (Ar), 127.5 (Ar), 91.2 (C1), 80.9 (C3), 79.9 (C2), 79.8 (C4), 74.9 (CH$_2$Ar), 74.7 (CH$_2$Ar), 73.6 (CH$_2$Ar), 73.4 (C6), 72.5 (CH$_2$Ar), 63.3 (C5), 33.6 (C7/C8), 24.5 (C7/C8). HRMS(ESI): calcd. for C$_{36}$H$_{39}$NO$_5$H 566.2901; found 566.2907.

(1S,2S,3S,4R,5R)-5-(hydroxymethyl)-8-azabicyclo[3.2.1]octane-1,2,3,4-tetraol - manno-nojiristegine (2)

The crude benzyl protected compound (45) (85 µmol) was dissolved in 1:1 methanol:ethyl acetate (3 mL) and added 20 % Pearlman’s catalyst (12.1 mg, 17 µmol, 20 %) and 1 drop of concentrated hydrochloric acid. The mixture was bubbled through with hydrogen and stirred under a hydrogen atmosphere. After 5 h the reaction was concentrated under reduced pressure, redissolved in methanol and filtered through Celite®. The filtrate was concentrated under reduced pressure, dissolved in water and filtered through a syringe filter (VWR 0.2 µM cellulose acetate) to give the product hydrochloric salt with no further purification (2) (20.6 mg, quant.) as a yellow solid.

$R_f$ (5 % NH$_4$OH in ethanol) 0.22; $[\alpha]_{D}^{298K}$ -16.2 (c 1, H$_2$O); $^1$H-NMR (400 MHz, D$_2$O): $\delta$H 4.00 (d, $J_2,3$ 4.0 Hz, 1H, H2), 3.86 (d, $J_4,3$ 9.2 Hz, 1H, H4), 3.84 (d, $J_{6a,6b}$ 12.5 Hz, 1H, H6a), 3.78 (dd, 1H, H3), 3.69 (d, 1H, H6b), 2.25 – 2.08 (m, 2H, H7/H8), 2.03 – 1.93 (m, 1H, H8/H7), 1.93 – 1.81 (m,
1H, H8/H7). $^{13}$C-NMR (100 MHz, D$_2$O): $\delta$C 96.5 (C1), 75.4 (C2), 72.3 (C3), 71.8 (C4), 69.3 (C5), 63.2 (C6), 31.8 (C7/C8), 24.1 (C8/C7). HRMS(ESI): calcd. for C$_8$H$_{15}$NO$_5$H 206.1023; found 206.1024.

(3R,4S,5R,6S)-3,4,5,6-Tetra-O-benzyl-cycloheptene (30)

The alcohol (28) (50.2 mg, 0.116 mmol) was dissolved in anhydrous DMF (4 mL) under a nitrogen atmosphere and stirred at 0 °C. 60% NaH in mineral oil (9.76 mg, 0.232 mmol, 2 eq) was slowly added and then BnBr (29.1 mg, 0.170 mmol, 1.5 eq) was under the formation of hydrogen gas. The mixture was stirred overnight. The reaction was quenched by addition of methanol (2 mL) and the mixture was then diluted with diethyl ether and washed with water. The aqueous phase was extracted with diethyl ether and the combined organic phases were washed with brine, dried with MgSO$_4$, filtered and concentrated under reduced pressure. The crude product was purified by flash column chromatography (ethyl acetate/pentane 1:15) yielding the product (30) (28.1 mg, 47% over 2 steps) as a clear oil.

$R_f$ (ethyl acetate/pentane 1:10) 0.41; $[\alpha]_D^{298K} +7.8$ (c 1.0, CHCl$_3$); $^1$H-NMR (400 MHz, CDCl$_3$): $\delta$H 7.28–7.17 (20H, m, ArH), 5.73 (1H, ddd, $J_{1,2}$ 11.7 Hz, $J_{2,3}$ 3.8 Hz, $J_{1}$ 1.0 Hz, CH=CH), 5.65 – 5.58 (1H, m, CH=CH), 4.73 (2H, d, $J$ 11.9 Hz, 2xCH/Ph), 4.63 (1H, d, CH/Ph), 4.61 (1H, d, CH/Ph), 4.55 (2H, s, CH$_2$/Ph), 4.43 (1H, d, $J$ 11.9 Hz, CH/Ph), 4.38 (1H, d, CH/Ph), 4.42 – 4.32 (m, 1H, H3), 3.90 (2H, ddd, $J_{4,3}$ 9.8 Hz, $J_{5,6}$ 7.1 Hz, $J_{4,5}$ 1.8 Hz, H4, H5), 3.62 (1H, td, $J_{6,7}$ 2.8 Hz, 1H), 2.50–2.37 (1H, m, H7a), 2.34–2.18 (1H, m, H7b). $^{13}$C-NMR (100 MHz, CDCl$_3$): $\delta$C 139.3 (Ar), 139.2 (Ar), 139.0 (Ar), 138.8 (Ar), 132.8 (Ar), 128.4 (Ar), 128.4 (Ar), 128.4 (Ar), 128.4 (Ar), 127.9 (Ar), 127.9 (Ar), 127.8 (Ar), 127.7 (Ar), 127.6 (Ar), 127.5 (Ar), 127.5 (Ar), 126.3 (Ar), 81.7 (C4/C5), 79.7 (C4/C5), 77.8 (C6), 76.9 (CH$_3$/Ph), 73.9 (C3), 73.8 (CH$_2$/Ph), 72.3 (CH$_2$/Ph), 71.6 (CH$_2$/Ph), 28.1 (C7). HRMS(ESI): calcd. for C$_{28}$H$_{32}$O$_6$NH$_4$ 543.2506; found 543.2506.

(3R,4S,5R,6R)-3,4,5,6-Tetra-O-benzyl-cycloheptene (31)

The alcohol (29) (39.4 mg, 91.1 mmol) was dissolved in anhydrous DMF (4 mL) under a nitrogen atmosphere and stirred at 0 °C. 60% NaH in mineral oil (7.53 mg, 0.181 mmol, 2 eq) was slowly added and then BnBr (23.8 mg, 0.136 mmol, 1.5 eq) was added under formation of hydrogen gas. The mixture was stirred overnight. The reaction was quenched by addition of methanol (2 mL) and the mixture was then diluted with diethyl ether and washed with water. The aqueous phase was extracted with diethyl ether and the combined organic phases were washed with brine, dried with
MgSO₄, filtered and concentrated under reduced pressure. The crude product was purified by flash column chromatography (ethyl acetate/pentane 1:25 → 1:15) yielding the product (31) (20.9 mg, 45% over 2 steps) as a clear oil.

\( R_f \) (Ethyl acetate/pentane 1:10) 0.41; \( [\alpha]_D^{298K} \) +4.0 (c 1.0, CHCl₃); \(^1\)H-NMR (400 MHz, CDCl₃): \( \delta \) H 7.35–7.15 (20 H, m, ArH), 5.60 (1H, dt, \( J \) 12.5 Hz, \( J \) 2.5 Hz, H2), 5.54–5.47 (1H, m, H1), 4.82 (2H, s, \( CH_2 \)Ph), 4.73 (1H, d, \( J \) 12.5 Hz, \( CHH \)Ph), 4.68 (1H, d, \( J \) 11.6 Hz, \( CHH \)Ph), 4.60 (1H, d, \( CHH \)Ph), 4.52 (1H, CHPh), 4.54–4.46 (1H, m, H3), 4.37 (1H, d, \( J \) 12.1 Hz, \( CHH \)Ph), 4.32 (1H, d, \( CHH \)Ph), 4.11 (1H, s, H5), 3.31 (1H, dd, \( J \) 9.3 Hz, \( J \) 1.0 Hz, H4), 3.23 (1H, m, H4), 2.73–2.53 (1H, m, H7a), 2.10 (1H, ddd, \( J \) 15.1 Hz, \( J \) 8.6 Hz, \( J \) 2.8 Hz, H7b). \(^13\)C-NMR (100 MHz, CDCl₃): \( \delta \) C 139.5 (Ar), 139.1 (Ar), 139.1 (Ar), 138.7 (Ar), 133.6 (CH=CH), 128.5 (Ar), 128.4 (Ar), 128.2 (Ar), 127.9 (Ar), 127.9 (Ar), 127.7 (Ar), 127.6 (Ar), 127.5 (Ar), 127.3 (Ar), 124.9 (CH=CH), 83.4 (C4), 80.3 (C5), 79.0 (C6), 77.4 (CH₂Ph), 74.3 (CH₂Ph), 73.7 (C3), 73.4 (CH₂Ph), 70.9 (CH₂Ph), 27.6 (C7). HRMS(ESI): calcd. for C₃₅H₃₆O₄Na 543.2506; found 543.2506.

(1R,2S,3R,4S)-1,2,3,4-Tetra-O-benzyl-cycloheptane (32)

The alkene (30) (25 mg, 48 µmol) was dissolved in anhydrous methanol (0.5 mL) and stirred under a nitrogen atmosphere at room temperature. Triethylamine (5 µL, 37 µmol, 0.77 eq) and a catalytic amount of Pearlman’s catalyst were added to the mixture. The atmosphere was exchanged for hydrogen gas and the reaction was stirred for 1h 30 min hours before exchanging the atmosphere to nitrogen and diluting with methanol. The mixture was then filtrated through cotton wool and concentrated under reduced pressure. The crude product was purified by flash column chromatography (ethyl acetate/pentane 1:30) yielding the product (32) (15.2 mg, 61%) as a colourless oil.

\( R_f \) (Ethyl acetate/pentane 1:20) 0.35; \(^1\)H-NMR (400 MHz, CDCl₃): \( \delta \) H 7.40–7.15 (10H, m, ArH), 4.72 (1H, d, \( J \) 12.0 Hz, \( CHH \)Ph), 4.67 (1H, d, \( CHH \)Ph), 4.58 (d, \( J \) 11.8 Hz, \( CHH \)Ph), 4.50 (1H, d, \( CHH \)Ph), 4.09 (1H, d, \( J \) 6.4 Hz, H1/H2), 3.81 (1H, dd, \( J \) 6.3 Hz, \( J \) 3.8 Hz, H1/H2), 1.99-1.90 /1H, m, H5/H6/H7) 1.87–1.68 (2H, m, H5/H6/H7). \(^13\)C-NMR (100 MHz, CDCl₃): \( \delta \) C 139.2 (Ar), 139.2 (Ar), 128.4 (Ar), 128.4 (Ar), 127.7 (Ar), 127.7 (Ar), 127.5 (Ar), 127.4 (Ar), 81.8 (C1/C2), 73.1 (CH₂Ph), 71.5 (CH₂Ph), 31.0 (C5/C6/C7), 17.4 (C5/C6/C7). HRMS(ESI): calcd. for C₃₅H₃₈O₄Na 545.2662; found 545.2660.

(1R,2S,3R,4R)-1,2,3,4-Tetra-O-benzyl-cycloheptane (33)
The alkene (31) (20.9 mg, 40 µmol) was dissolved in anhydrous methanol (0.5 mL) and stirred under a nitrogen atmosphere at room temperature. Triethylamine (4 µL, 30 µmol, 0.77 eq) and a catalytic amount of Pearlman’s catalyst were added to the mixture. The atmosphere was exchanged for hydrogen gas and the reaction was stirred for 3 hours before exchanging the atmosphere to nitrogen and diluting with methanol. The mixture was then filtrated through cotton wool and concentrated under reduced pressure. The crude product was purified by flash column chromatography (ethyl acetate/pentane 1:30) yielding the (33) (9.52 mg, 45%) as a colourless oil.

_Rf_ (Ethyl acetate/pentane 1:20) 0.35. ¹H-NMR (400 MHz, CDCl₃): δ_H 7.36–7.15 (20H, m, ArH), 4.79 (1H, d, J 12.3 Hz, CH₂Ph), 4.75 (1H, d, J 12.3 Hz, CH₂Ph), 4.55 (1H, d, J 11.8 Hz, CH₂Ph), 4.54 (1H, d, J 11.4 Hz, CH₂Ph) 4.48 (1H, d, J 11.4 Hz, CH₂Ph), 4.46 (1H, d, J 11.8 Hz, CH₂Ph), 4.31 (2H, s, CH₂Ph), 4.05 (1H, s, C₂/C₃), 3.78-3.71 (1H, m, C₂/C₃), 3.39 (1H, d, J 11.8 Hz, C₂/C₃), 4.46 (1H, d, J 11.4 Hz, C₂/C₃), 4.45 (1H, d, J 11.4 Hz, C₂/C₃), 4.31 (2H, s, CH₂Ph), 4.30–3.24 (1H, m, C₁/C₄), 2.00–1.50 (6H, m, C₅, C₆, C₇). ¹³C-NMR (100 MHz, CDCl₃): δ_C 139.5 (Ar), 139.2 (Ar), 139.0 (Ar), 138.8 (Ar), 128.5 (Ar), 128.4 (Ar), 128.2 (Ar), 128.0 (Ar), 127.9 (Ar), 127.7 (Ar), 127.6 (Ar), 127.3 (Ar), 127.5 (Ar), 85.3 (C₁/C₄), 82.5 (C₂/C₃), 81.1 (C₁/C₄), 79.9 (C₂/C₃), 73.4 (CH₂Ph), 73.0 (CH₂Ph), 72.1 (CH₂Ph), 71.0 (CH₂Ph), 30.2 (C₅/C₆/C₇), 29.8 (C₅/C₆/C₇), 18.6 (C₅/C₆/C₇). HRMS(ESI): calcd. for C₃₅H₃₈O₄H 523.2843; found 523.2843.

Acknowledgements

We are grateful for support from the graduate school of science and technology, Aarhus University

References

6. By IUPAC rules _iminosugars_ are defined as cyclic sugar derivatives in which the ring oxygen has been replaced by N, whereas _azasugars_ are cyclic sugar derivatives where C has been replaced by N. Combined these families of molecules are often potent inhibitors of glycosidases, see ref. 9.


26 See supporting information for details.


