Piperidine analogues of D-galactose as potent inhibitors of α-galactosidase: Synthesis by stannane-mediated hydroxymethylation of 5-azido-1,4-lactones. Structural relationships between D-galactosidase and L-rhamnosidase inhibitors

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The syntheses of the polyhydroxylated piperidines deoxygalactonojirimycin **2**, homogalactonojirimycins **7** and **9**, and other 2,6-iminoheptitol derivatives, including an analogue of L-altropyranose, are reported. 5-Azidoaldono-1,4-lactones undergo chain extension to afford azido lactols by the addition of a hydroxymethyllithium species **18**, generated by transmetallation of a protected stannylmethanol derivative **17**. Hydrogenation results in azide reduction with subsequent intramolecular reductive amination to give piperidine ring systems. The deprotected iminogalactopyranose analogues are potent and selective α-galactosidase inhibitors. Observations on the structural features determining selectivity of inhibition of α-galactosidases over naringinase (L-rhamnosidase) are also reported.

Introduction

Deoxynojirimycin analogues[1] of D-galactose, galactostatin **1** and deoxygalactostatin (deoxygalactonojirimycin—DGJ) **2** are extremely potent inhibitors of both α- and β-D-galactosidases.[2] Similarly, the enantiomeric 6-deoxy compound, deoxyfuconojirimycin DFJ **3** is a potent inhibitor of L-fucosidases.[3] Piperidine analogues of D-mannopyranose such as deoxymannojirimycin DMJ **4** and its C-1 homologues are generally weak inhibitors of D-mannosidases but are often potent inhibitors of L-fucosidases.[4] L-Fucopyranose and D-mannopyranose are structurally related in that their ring hydroxy groups (C-2 to C-4) have the same relative and absolute configuration (Fig. 1). A requirement for L-fucosidase inhibition by piperidine analogues of pyranoses is that they should possess ring hydroxy groups in this configuration. In the mirror image series, L-rhamnopyranose and D-galactopyranose are similarly related by the configuration of their secondary hydroxy groups.

Investigations into the synthesis and evaluation of homonojirimycin analogues of L-rhamnopyranose have indicated that α-homorhamnojirimycin (HRJ) **5** is a selective and potent inhibitor of naringinase (L-rhamnosidase) whereas β-homorhamnojirimycin **6** is a selective and potent inhibitor of α-galactosidase.[5] The difference in selectivity has been rationalized on the basis that **6** possesses four stereogenic centres in common with D-galactose and may be viewed as the β-1-methyl derivative of DGJ **2**.

This paper reports the synthesis of a range of homonojirimycin analogues of D-galactopyranose in order to evaluate the effects of varying the configuration of C-2 and C-6 substituents on their ability to inhibit either galactosidases or naringinase and to determine structure–activity relationships for their inhibition. A unified synthetic methodology for accessing DGJ **2**, β-HGJ (homogalactonojirimycin) **7** (enantiomer of the known β-homomannojirimycin **8**[6]), α-HGJ **9** and α-1-methyl-DGJ **10**, and the other related 2,6-iminoheptitols **11** and **12** is also described.

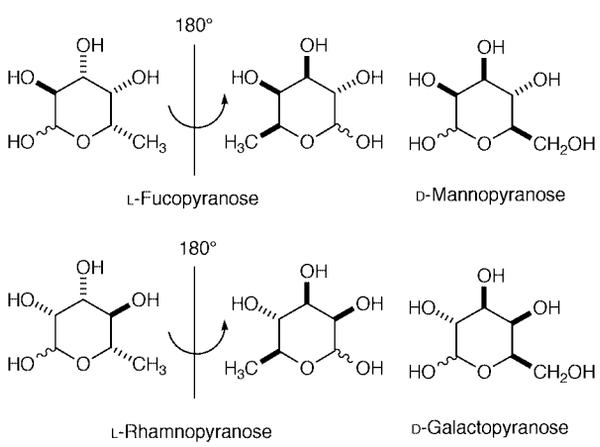
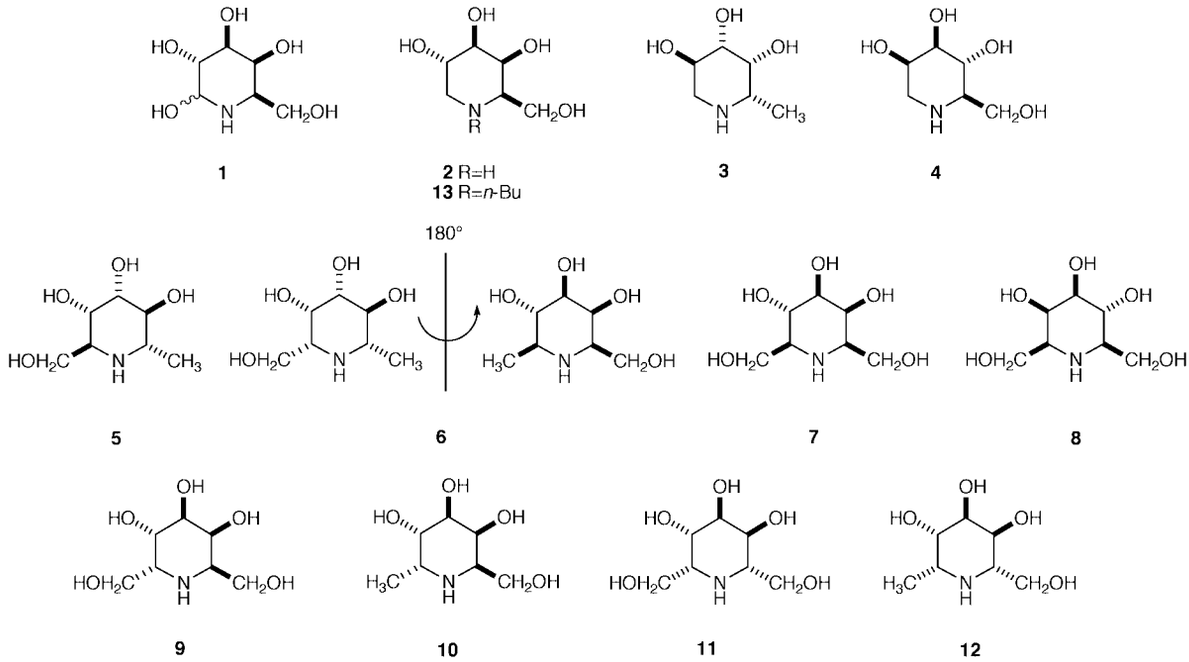


Fig. 1 Relative and absolute configurations of various pyranoses.

N-Butyl-DGJ **13** is known to be an inhibitor of the Golgi glucosyltransferanse responsible for the biosynthesis of glucocerebroside, a precursor to the biologically important glycosphingolipids, gangliosides.[7] A number of *N*-butylated iminosugars, including *N*-butyl-DGJ, NBDGJ **13**,[8] have been used in studies related to a range of disorders where inherited defects in catabolic lysosomal glycosidases—such as Tay-Sachs[9] and Gauchers diseases—result in the storage of complex glycosphingolipids. Storage may be prevented by inhibiting the rate of glucocerebroside biosynthesis with *N*-butyl-DGJ **13** thereby lowering the lysosomal concentration of gangliosides, and allowing their degradation by the defective enzyme to be carried out at acceptable rates.[9,10] Although there are a number of such analogues, including **13**, which may have potential for the treatment of Tay-Sachs and Gauchers diseases, all the currently described inhibitors are themselves potent inhibitors of various glycosidases. It is therefore highly desirable to find an analogue which more selectively and

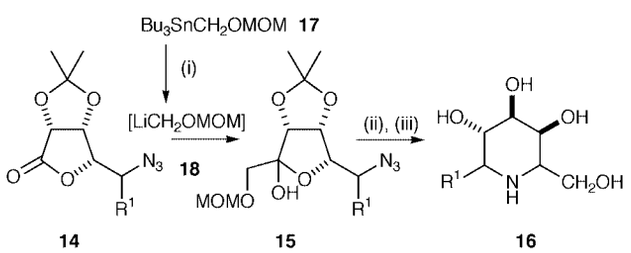


specifically inhibits the transferase rather than the hydrolases. The synthesis of a series of homonojirimycin analogues of D-galactopyranose therefore makes available a range of substances for future *N*-alkylation studies for investigating selective activity against Golgi glucosyltransferase.

Results and discussion

Synthesis

Imino-analogues of L-fucopyranose have previously been accessed by incorporation of the methyl group *via* methyllithium addition to 5-azido-1,4-lactones.[11] The corresponding D-galactopyranose mimics were correspondingly available from nucleophilic addition of a hydroxymethyl anion equivalent to azido lactones **14** bearing the required absolute configuration (Scheme 1). Hydrogenation of the resulting 6-azido-2-keto

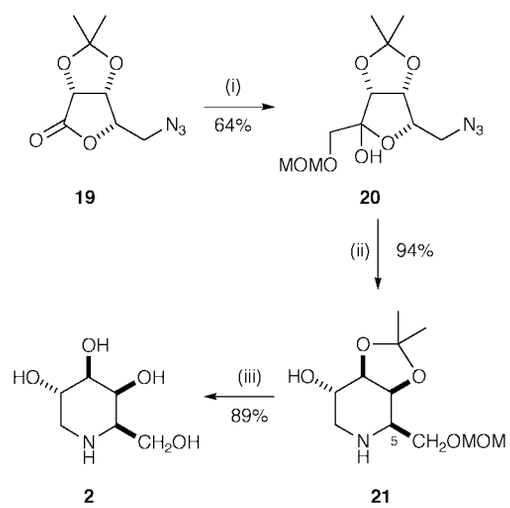


Scheme 1 *Reagents and conditions*: (i) BuLi, THF, −78 °C; (ii) H_2, Pd, EtOH; (iii) HCl, MeOH.

furanoses **15** induced intramolecular reductive amination to give the desired piperidines **16**.

The transmetallation of protected stannylmethanol derivatives using butyllithium is a reliable source of hydroxymethyl anion equivalents,[12] and their use in one-carbon-chain extensions of carbohydrate lactones has been reported.[13] Tributyl-[(methoxymethoxy)methyl]stannane **17** was used as the reagent for this purpose.[14] Part of this work has been previously reported in a communication.[15]

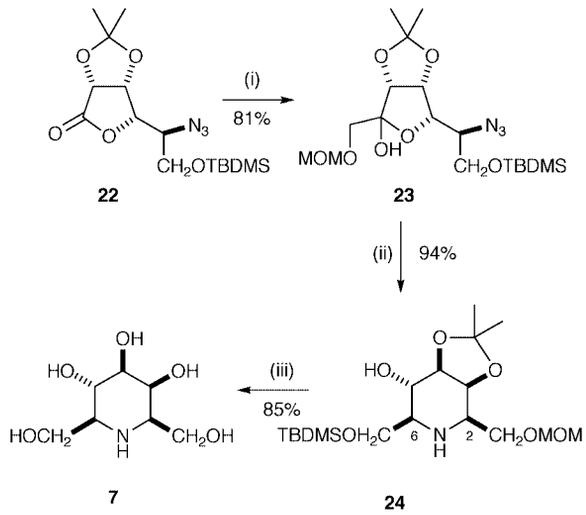
The potent galactosidase inhibitor deoxygalactonojirimycin **2** (DGJ or deoxygalactostatin) was synthesized using this strategy. Addition of the hydroxymethyllithium species **18** to the protected 5-azido-L-lyxono-1,4-lactone **19**[16] in THF at −78 °C gave the anomeric azido lactols **20** (7:1 mixture) in 64% yield (Scheme 2). Catalytic hydrogenation of the lactols **20** gave



Scheme 2 *Reagents and conditions*: (i) [$LiCH_2OMOM$] **18**, THF, −78 °C; (ii) H_2, Pd, EtOH; (iii) HCl, MeOH.

the protected D-*galacto* piperidine **21**, as a single diastereoisomer in 94% yield *via* reduction of the azido group and subsequent intramolecular reductive amination. Deprotection of the *O*-isopropylidene and MOM-group protection in methanolic hydrogen chloride, followed by purification on acidic ion-exchange resin, gave DGJ **2** in 89% yield, identical to the known material,[17] $[\alpha]_D^{23}$ +51.6 (*c* 1.24 in H_2O) lit.,[17d] {$[\alpha]_D^{23}$ +52.0 (*c* 0.4 in H_2O)}.

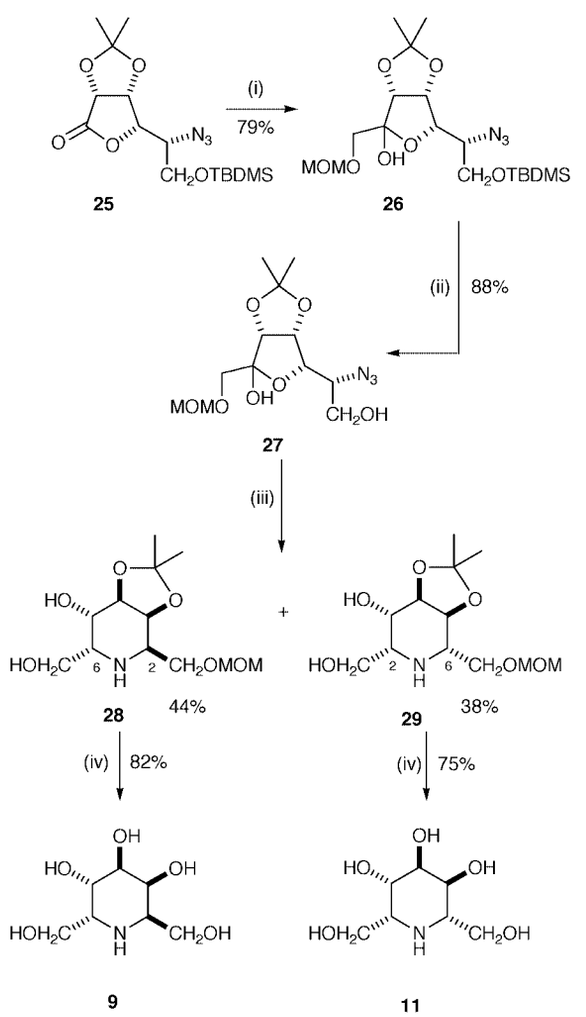
The homogalactonojirimycins were available *via* hydroxymethyl anion equivalent addition to the homologous 5-azidohexono-1,4-lactones. β-Homogalactonojirimycin (β-HGJ) **7** is the enantiomer of β-homomannojirimycin (β-HMJ) **8**,[6] and was synthesized *via* hydroxymethylation of the 5-azido-L-mannono-1,4-lactone **22**[18] to give the anomeric lactones **23** (8:1 mixture) in 81% yield (Scheme 3). Hydrogenation of the lactols **23** by using palladium black in ethanol gave the piperidine **24** as a single diastereoisomer in 94% yield. Deprotection of **24** with methanolic hydrogen chloride, followed by ion-exchange chromatography, produced β-HGJ **7** in 85% yield. Comparison of **7** with its enantiomer β-HMJ **8** showed them to possess identical 500 MHz ^1H NMR spectra, confirming the 2,6-*cis* configuration in **24**. The optical rotations were also equal in magnitude and opposite in sign: {$[\alpha]_D^{24}$ +4.0 (*c* 1.0



Scheme 3 *Reagents and conditions*: (i) [$LiCH_2OMOM$] **18**, THF, −78 °C; (ii) H_2, Pd, EtOH; (iii) HCl, MeOH.

in MeOH)} for β-HGJ **7** and {$[\alpha]_D^{20}$ −4.3 (*c* 1.3 in MeOH)} for β-HMJ **8**.[6b] The formation of the 2,6-*cis*-substituted piperidine **24** is consistent with delivery of hydrogen to the intermediate cyclic imine from the face opposite to the *O*-isopropylidene protecting group.

The epimeric material α-homogalactonojirimycin (α-HGJ) **9**[19] was obtained from hydroxymethylation of 5-azido-D-gulono-1,4-lactone **25**[20] to give the anomeric lactols **26** (12.5 : 1 mixture) in 79% yield (Scheme 4). Cleavage of the TBDMS



Scheme 4 *Reagents and conditions*: (i) [$LiCH_2OMOM$] **18**, THF, −78 °C; (ii) TBAF, THF; (iii) H_2, 10% Pd-C, EtOAc; (iv) HCl, MeOH.

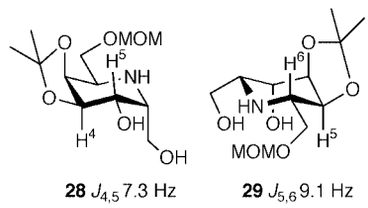
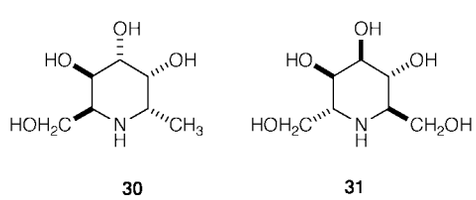


Fig. 2 ^{1}H NMR assignment of configuration for diastereoisomeric piperidines **28** and **29**.

ether was effected using TBAF in THF to give the azido lactols **27** in 88% yield. This partial deprotection was carried out based on the reported synthesis of α-homofuconojirimycin (α-HFJ) **30**[11b] from a similar 1-deoxy system bearing the opposite absolute configuration, in which case removal of the terminal silyl ether protection was required in order to ensure the formation of the 2,6-*trans*-substituted piperidine during intramolecular reductive amination. Hydrogenation of lactols **27** in the presence of 10% palladium on charcoal in ethyl acetate produced the separable epimeric piperidines **29** and **28** in 38% and 44% yield respectively. Deprotection of **29** in methanolic hydrogen chloride followed by purification on acidic ion-exchange resin gave β-homo-L-altronojirimycin **11** in 75% yield. Similar deprotection of **28** provided α-HGJ **9** in 82% yield.

The 500 MHz ^{1}H NMR spectra of **29** and **28** were used to assign the configurations of the newly formed stereogenic centres, assuming that both materials adopt chair-like conformations in solution. The less polar material **29** was assigned as the 2,6-*cis*-substituted piperidine, owing to *trans*-diaxial coupling between H-5 and H-6, $J_{5,6}$ 9.1 Hz (Fig. 2). The more polar material **28** displayed a *trans*-diaxial coupling between H-4 and H-5, $J_{4,5}$ 7.3 Hz, consistent with a 2,6-*trans*-substituted piperidine.

The formation of the epimeric piperidines in this case is in contrast to high selectivity for the formation of the 2,6-*trans*-substituted piperidine observed in the corresponding transformation in the enantiomeric deoxy system for the synthesis of α-HFJ **30**.[11b] However, a lack of selectivity has been reported in an attempt to form a 2,6-*trans*-substituted piperidine in the synthesis of α-homomannojirimycin (α-HMJ) **31**.[6b]



A recently reported synthesis of homonojirimycin analogues of L-rhamnose provided three of the possible four stereoisomers with respect to C-2 and C-6, **5**, **6**, **10** and **12**.[5] One isomer, β-homorhamnojirimycin (β-HRJ) **6**, was found to be a weak rhamnosidase inhibitor but a potent α-galactosidase inhibitor, since it may also be viewed as the β-1-methyl-substituted derivative of DGJ **2**. In order to extend structure–activity relationships for these materials, the α-1-methyl epimer **10**, not available using the previously reported methodology, was prepared using the hydroxymethylation–intramolecular reductive amination strategy from the 6-deoxy-5-azido-D-gulono-1,4-lactone **32**.[21] Hydroxymethylation of **32** gave the azido lactols **33** (14 : 1 mixture) in 76% yield and subsequent hydrogenation the separable epimeric piperidines **34** and **35** in 44% and 46% yield respectively (Scheme 5). Deprotection of **34** in methanolic hydrogen chloride provided the homoiminosugar **12** in 79% yield, identical to the 5-*epi*-L-rhamnopyranose analogue previously reported,[5] confirming its 2,6-*cis* configuration. Similarly, deprotection of **35** gave α-1-methyl-DGJ **10** in 73% yield.

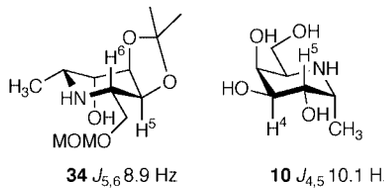
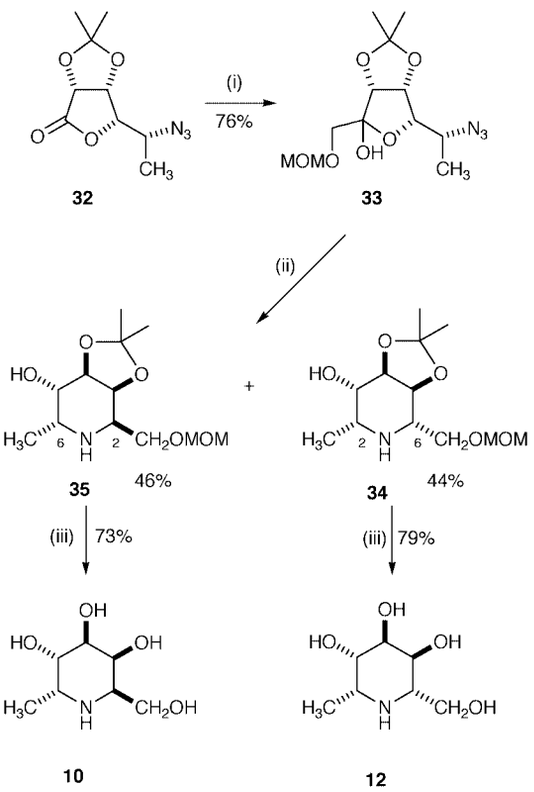


Fig. 3 ^{1}H NMR assignment of configuration for compounds **10** and **34**.



Scheme 5 *Reagents and conditions*: (i) [$LiCH_2OMOM$] **18**, THF, −78 °C; (ii) H_2, 10% Pd-C, EtOAc; (iii) HCl, MeOH.

The less polar epimer **34** was assigned as the 2,6-*cis*-substituted piperidine based on the *trans*-diaxial coupling between H-5 and H-6, $J_{5,6}$ 8.9 Hz, in the 500 MHz ^{1}H NMR spectrum (Fig. 3), consistent with the piperidine ring adopting a chair-like conformation in solution. The more polar epimer **35** was accordingly assigned as the 2,6-*trans*-substituted piperidine. The 500 MHz ^{1}H NMR spectrum of **10** in D_2O at pH 8–9 was consistent with a 2,6-*trans* assignment, based on the observed *trans*-diaxial coupling between H-4 and H-5, $J_{4,5}$ 10.1 Hz.

In summary, the addition of a hydroxymethyl anion equivalent to 5-azido carbohydrate lactones provides a method for their chain extension to the corresponding azido-lactols, which may be converted to piperidines *via* azide reduction and *in situ* intramolecular reductive amination. This work demonstrates that application of this methodology to suitably substituted 5-azido carbohydrate lactones allows the efficient synthesis of piperidine analogues of D-galactopyranose.

Biological assays

The homoiminosugars **7**, **9**, **11**, and **10** were assayed for inhibition of α-galactosidase (green coffee bean) and a range of other glycosidases including naringinase [L-rhamnosidase (*Penicillium decumbens*)] [others were α-glucosidase (Brewers' yeast, rice), β-glucosidase (almond), β-galactosidase (*Escherichia coli*, *Aspergillus niger* and bovine liver) and α-mannosidase (Jack bean)] (see Table 1). All compounds were found to be potent and selective competitive inhibitors of

Table 1

Compound	Inhibition of naringinase (*Penicillium decumbens*)	Inhibition of α-galactosidase (green coffee bean)
5[5]	IC_{50} 15 μM, K_i 5.3 μM	46% (0.1 mM)[a]
6[5]	IC_{50} 750 μM	IC_{50} 0.34 μM, K_i 0.31 μM
7	NT	IC_{50} 1.8 μM, K_i 0.43 μM
9	IC_{50} 150 μM	K_i 4.8 nM
10	IC_{50} 120 μM	K_i 0.20 μM
11	IC_{50} > 500 μM	IC_{50} 11 μM

[a] Inhibitor concentration. NT - not tested.

α-galactosidase: β-HGJ **7** (IC_{50} 1.8 μM, K_i 0.43 μM); α-HGJ **9** (K_i 4.8 nM); 1α-methyl-DGJ **10** (K_i 0.20 μM); and β-homo-L-altronojirimycin **11** (IC_{50} 11 μM). This compares with DGJ **2** (K_i 2.7 nM) and 1β-methyl-DGJ **6** (IC_{50} 0.34 μM, K_i 0.31 μM). DGJ **2** is also known to inhibit β-galactosidases; however, the imino-*C*-glycosides caused no inhibition. Compounds **9**, **10** and **11** were found to be weak inhibitors of naringinase at high concentrations (IC_{50} 120–>500 μM).

Conclusions

Whilst α-HGJ **9** retains the potency of deoxygalactostatin **2** as an α-galactosidase inhibitor it appears that incorporation of either an α- or β-methyl group, or a β-hydroxymethyl group (compounds **6**, **7** and **10**) into the structure of deoxygalactostatin (DGJ) **2** results in a decrease in potency for the inhibition of α-galactosidase. A complete loss of ability to inhibit β-galactosidase is observed for all compounds. The actual configurations at C-2 and C-6 in this series of compounds bearing the same ring hydroxy group configurations seem to be important in determining selectivity for the inhibition of α-galactosidase over naringinase. This is illustrated by comparing inhibition by α-HRJ **5** (a potent and selective naringinase inhibitor) with β-HRJ **6**[5] (β-1-methyl-DGJ) and the other D-galactopyranose analogues β-HGJ **7**, α-HGJ **9** and α-1-methyl-DGJ **10** (potent and selective α-galactosidase inhibitors). All compounds possess the same ring hydroxy-group configuration but differing configurations for C-2 and C-6 substituents. Compounds with four stereogenic centres corresponding to those of C-2 to C-5 in D-galactose are selective α-galactosidase inhibitors. Additionally, β-HRJ **6** (β-1-methyl-DGJ) which also possesses four stereogenic centres in common with an L-rhamnose configuration, causes only weak inhibition of naringinase. The L-altropyranose analogue **11** possesses four stereogenic centres in common with α-HRJ **5**, but is a selective inhibitor of α-galactosidase, further suggesting that the configuration of the three ring hydroxy groups corresponding to those in D-galactose is still an important factor in determining the ability to inhibit α-galactosidase but that it is the C-2, C-6 substitution and relative configuration which governs α-galactosidase–naringinase selectivity. These phenomena are currently under investigation using molecular modelling studies.[22]

Experimental

THF was distilled under an atmosphere of dry nitrogen from sodium benzophenone ketyl; hexane refers to the fraction of petroleum ether which boils in the range 60–80 °C and was redistilled before use. All other solvents were used as supplied (Analytical or HPLC grade), without prior purification. Reactions were performed under an atmosphere of nitrogen or argon maintained by an inflated balloon. Butyllithium was used as a solution in hexanes at the molarity stated; tributyl-[(methoxymethoxy)methyl]stannane **17** was prepared according to a procedure by Johnson;[14] hydrogenations were performed using an atmosphere of hydrogen gas maintained by an inflated

balloon. All other reagents were used as supplied, without prior purification. Flash chromatography was performed on Sorbsil C60, and ion-exchange chromatography was performed on Amberlite IR-120 (H^+). Melting points were recorded on a Kofler hot block and are uncorrected. Nuclear magnetic resonance (NMR) spectra were recorded on a Bruker AC 200 (^{1}H: 200 MHz and ^{13}C: 50.3 MHz) or Bruker AM 500 or AMX 500 (^{1}H: 500 MHz and ^{13}C: 125.8 MHz) spectrometer for samples in the deuterated solvent stated. All chemical shifts (δ) are quoted in ppm and coupling constants (J) in Hz. Residual signals from the solvents were used as an internal reference and ^{13}C NMR spectra in D_2O were referenced to 1,4-dioxane (δ_C 67.4). ^{13}C Multiplicities were assigned using a DEPT sequence. IR spectra were recorded on a Perkin-Elmer 1750 IR Fourier Transform, or Perkin-Elmer Paragon 1000 spectrophotometer using either thin films on NaCl plates (film) or KBr discs (KBr) as stated. For clarity, only the salient, characteristic peaks are quoted. Low-resolution mass spectra (*m*/*z*) were recorded on VG MASS LAB 20-250, BIO Q, VG Platform or VG Autospec spectrometers, and high-resolution mass spectra (HRMS *m*/*z*) were measured on a VG Autospec spectrometer. Techniques used were chemical ionization (CI, NH_3), desorption chemical ionization (DCI, NH_3), electrospray, electron impact (EI) or atmospheric pressure chemical ionization (APCI) using partial purification by HPLC with methanol–acetonitrile–water (40 : 40 : 20) as eluent, as stated. Specific optical rotations $[\alpha]_D$ are quoted in 10^{-1} deg cm^2 g^{-1} and were recorded on a Perkin-Elmer 241 polarimeter with a path length of 1 dm. Concentrations are quoted in 10^{-2} g cm^{-3}. Elemental analyses were performed by the microanalysis service of the Dyson Perrins Laboratory.

General procedure 1

Butyllithium (2.5 M solution in hexanes; 1.2 equiv.) was added to a stirred solution of tributyl[(methoxymethoxy)methyl]-stannane **17** (1.5 equiv.) in THF (approx. 1 cm^3 mmol^{-1}) at −78 °C. After 5 min, a solution of the requisite azido-1,4-lactone (1 equiv.) in THF (approx. 1 cm^3 mmol^{-1}) was added. The solution was stirred at −78 °C for 30 min and the reaction mixture was quenched by the addition of saturated ammonium chloride solution. The mixture was partitioned between water and ethyl acetate and the combined organic phase was washed with brine, dried (magnesium sulfate), filtered and concentrated *in vacuo*. The residue was purified by flash chromatography on silica (ethyl acetate–hexane).

General procedure 2

A solution of the requisite protected piperidine in 3% methanolic hydrogen chloride was stirred at room temperature for 24 h. The solution was concentrated *in vacuo* (with further co-evaporation with methanol, three times) and the residue subsequently purified by ion-exchange chromatography on Amberlite (IR 120, H^+-form) (1.0 M ammonium hydroxide).

6-Azido-6-deoxy-3,4-*O*-isopropylidene-1-*O*-methoxymethyl-α,β-L-*lyxo*-hex-2-ulofuranose 20

Azido lactols **20** were prepared according to general procedure 1 using butyllithium (2.5 M solution in hexanes; 1.22 cm^3, 3.05 mmol), tributyl[(methoxymethoxy)methyl]stannane **17** (1.27 g, 3.52 mmol) and azido-1,4-lactone **19**[16] (500 mg, 2.35 mmol). Purification of the crude material using column chromatography (ethyl acetate–hexane, 1 : 2) afforded **20** as a colourless oil (435 mg, 64%), $[\alpha]_D^{23}$ −25.1 (*c* 0.97 in $CHCl_3$) (Found: C, 45.7; H, 6.6; N, 15.3. $C_{11}H_{19}N_3O_6$ requires C, 45.7; H, 6.4; N, 14.5%); ν_{max}(film)/cm^{-1} 3414 (br, OH), 2102 (s, N_3); δ_H(500 MHz; $CDCl_3$) Major anomer: 1.31, 1.46 (6H, 2s, C(CH_3)$_2$), 3.42 (3H, s, CH_2OCH_3), 3.52 (1H, dd, $J_{5,6'}$ 6.1, $J_{6,6'}$ 12.6, H-6′), 3.56 (1H, dd, $J_{5,6}$ 7.0, $J_{6,6'}$ 12.6, H-6), 3.79 (2H, AB system, $J_{1,1'}$ 11.1, H_2-1), 3.81 (1H, s, OH), 4.26 (1H, ddd, $J_{5,6}$ 7.0, $J_{5,6'}$ 6.1, $J_{4,5}$ 3.8, H-5), 4.59 (1H, d, $J_{3,4}$ 5.8, H-3), 4.69 (2H, AB system, *J* 6.5, CH_2OCH_3), 4.8 (1H, dd, $J_{3,4}$ 5.8, $J_{4,5}$ 3.8, H-4); δ_C(50 MHz; $CDCl_3$) Major anomer: 24.6, 25.9 (2q, C(CH_3)$_2$), 49.6 (t, C-6), 55.5 (q, CH_2OCH_3), 69.4 (t, C-1), 77.9, 80.1 (2d, C-4, C-5), 85.3 (d, C-3), 97.1 (t, CH_2OCH_3), 104.0 (s, C-2), 113.1 (s, C(CH_3)$_2$); *m*/*z* (CI, NH_3) 244 ($MH^+ - N_2 - H_2O$, 100%).

1,5-Dideoxy-1,5-imino-3,4-*O*-isopropylidene-6-*O*-methoxymethyl-D-galactitol 21

A solution of azido lactols **20** (300 mg, 1.03 mmol) in ethanol (5 cm^3) was stirred under an atmosphere of hydrogen in the presence of palladium black (100 mg) for 72 h. The reaction mixture was filtered through Celite and concentrated *in vacuo*. The residue was subjected to flash chromatography on silica (methanol–ethyl acetate, 1 : 9) to afford the piperidine **21** as a white solid (255 mg, 94%), which was recrystallized from ethyl acetate, mp 114–115 °C; $[\alpha]_D^{26}$ +37.6 (*c* 1.23 in $CHCl_3$) (Found: C, 53.7; H, 8.9; N, 5.7. $C_{11}H_{21}NO_5$ requires C, 53.4; H, 8.6; N, 5.7%); ν_{max}(KBr)/cm^{-1} 3309 (br, NH, OH); δ_H(500 MHz; $CDCl_3$) 1.35, 1.53 (6H, 2s, C(CH_3)$_2$), 2.44 (1H, dd, $J_{1^a,2}$ 10.9, $J_{1^a,1^e}$ 12.3, H-1^a), 3.17 (2H, m, H-1^e, H-5), 3.38 (3H, s, CH_2OCH_3), 3.63 (1H, m, H-6′), 3.73 (1H, m, H-2), 3.76 (1H, dd, $J_{5,6}$ 4.5, $J_{6,6'}$ 10.2, H-6), 3.90 (1H, dd, $J_{3,4}$ 5.4, $J_{2,3}$ 7.2, H-3), 4.15 (1H, dd, $J_{3,4}$ 5.4, $J_{4,5}$ 2.7, H-4), 4.67 (2H, s, CH_2OCH_3); δ_C(50 MHz; $CDCl_3$) 26.3, 28.2 (2q, C(CH_3)$_2$), 48.8 (t, C-1), 55.2 (q, CH_2OCH_3), 56.1 (d, C-5), 68.4 (t, C-6), 71.3, 74.0, 80.8 (3d, C-2, C-3, C-4), 96.6 (t, CH_2OCH_3), 109.5 (s, C(CH_3)$_2$); *m*/*z* (APCI$^+$) 248 (MH^+, 100%).

1,5-Dideoxy-1,5-imino-D-galactitol (1-deoxygalactonojirimycin) 2

21 (134 mg, 0.54 mmol) was deprotected according to general procedure 2 to afford **2** as a hygroscopic foam (79 mg, 89%) identical to the known material;[17] $[\alpha]_D^{23}$ +51.6 (*c* 1.24 in H_2O) {lit.,[17d] $[\alpha]_D^{23}$ +52.0 (*c* 0.4 in H_2O)}; δ_H(500 MHz; D_2O, pH 9) 2.35 (1H, dd, $J_{1^a,2}$ 10.9, $J_{1^a,1^e}$ 12.7, H-1^a), 2.73 (1H, m, H-5), 3.08 (1H, dd, $J_{1^e,2}$ 5.3, $J_{1^a,1^e}$ 12.7, H-1^e), 3.42 (1H, dd, $J_{3,4}$ 3.2, $J_{2,3}$ 9.7, H-3), 3.55 (1H, dd, $J_{5,6'}$ 6.8, $J_{6,6'}$ 11.3, H-6′), 3.59 (1H, dd, $J_{5,6}$ 6.6, $J_{6,6'}$ 11.3, H-6), 3.71 (1H, ddd, $J_{1^e,2}$ 5.3, $J_{1^a,2}$ 10.9, $J_{2,3}$ 9.7, H-2), 3.95 (1H, dd, $J_{3,4}$ 3.2, $J_{4,5}$ 1.0, H-4).

6-Azido-7-*O*-*tert*-butyldimethylsilyl-6-deoxy-3,4-*O*-isopropylidene-1-*O*-methoxymethyl-α,β-L-*manno*-hept-2-ulofuranose 23

Azido lactols **23** were prepared according to general procedure 1 using butyllithium (2.5 M solution in hexanes; 0.64 cm^3, 1.60 mmol), tributyl[(methoxymethoxy)methyl]stannane **17** (500 mg, 1.81 mmol) and azido-1,4-lactone **22**[18] (500 mg, 1.39 mmol). Purification of the crude material using column chromatography (ethyl acetate–hexane, 1 : 6) afforded **23** as a colourless oil (490 mg, 81%), $[\alpha]_D^{25}$ +10.4 (*c* 1.34 in $CHCl_3$) (Found: C, 50.2; H, 8.3. $C_{18}H_{35}N_3O_7Si$ requires C, 49.9; H, 8.1%); ν_{max}(film)/cm^{-1} 3421 (br, OH), 2099 (s, N_3); δ_H(500 MHz; $CDCl_3$) Major anomer: 0.10 (6H, s, Si(CH_3)$_2$), 0.92 (9H, s, SiC(CH_3)$_3$), 1.34, 1.50 (6H, 2s, C(CH_3)$_2$), 3.41 (3H, s, CH_2OCH_3), 3.66–3.69 (1H, m, H-6), 3.67 (1H, s, OH), 3.74–3.79 (3H, m, H_2-1, H-7), 4.02 (1H, dd, $J_{6,7'}$ 2.5, $J_{7,7'}$ 8.0, H-7′), 4.04 (1H, dd, $J_{5,6}$ 8.2, $J_{4,5}$ 3.6, H-5), 4.57 (1H, d, $J_{3,4}$ 5.9, H-3), 4.69 (1H, d, *J* 6.5, CH_2OCH_3), 4.73 (1H, d, *J* 6.5, CH_2OCH_3), 4.86 (1H, dd, $J_{3,4}$ 5.9, $J_{4,5}$ 3.6, H-4); δ_C(50 MHz; $CDCl_3$) Major anomer: −5.5 (q, Si(CH_3)$_2$), 18.2 (s, SiC(CH_3)$_3$), 24.7, 26.0 (2q, C(CH_3)$_2$), 25.8 (q, SiC(CH_3)$_3$), 55.5 (q, CH_2OCH_3), 61.1 (d, C-6), 64.3 (t, C-7), 69.5 (t, C-1), 77.0, 80.0 (2d, C-4, C-5), 85.0 (d, C-3), 97.1 (t, CH_2OCH_3), 104.1 (s, C-2), 112.9 (s, C(CH_3)$_2$); *m*/*z* (CI, NH_3) 388 ($MH^+ - N_2 - H_2O$, 100%); (+ve Electrospray) [Found: 451.2578 (MNH_4^+). $C_{18}H_{39}N_4O_7Si$ requires *m*/*z*, 451.2588].

7-*O*-*tert*-Butyldimethylsilyl-2,6-dideoxy-2,6-imino-3,4-*O*-isopropylidene-1-*O*-methoxymethyl-L-*glycero*-L-*galacto*-heptitol 24

A solution of azido lactols **23** (300 mg, 0.69 mmol) in ethanol (5 cm^3) was stirred under an atmosphere of hydrogen in the presence of palladium black (60 mg) for 48 h. The reaction mixture was filtered through Celite and concentrated *in vacuo*. The residue was subjected to flash chromatography on silica (gradient elution: ethyl acetate–hexane, 1:1 to ethyl acetate) to afford the piperidine **24** as a colourless oil (255 mg, 94%), $[\alpha]_D^{25}$ +23.0 (*c* 0.47 in $CHCl_3$) (Found: C, 55.3; H, 9.7. $C_{18}H_{37}NO_6Si$ requires C, 55.2; H, 9.6%); ν_{max}(film)/cm^{-1} 3452 (br, NH, OH); δ_H(500 MHz; $CDCl_3$), 0.07 (6H, 2s, Si(CH_3)$_2$), 0.89 (9H, s, SiC(CH_3)$_3$), 1.35, 1.52 (6H, 2s, C(CH_3)$_2$), 2.55 (1H, ddd, $J_{5,6}$ 10.0, $J_{6,7}$ 5.0, $J_{6,7'}$ 6.0, H-6), 3.22 (1H, ddd, $J_{2,3}$ 2.6, $J_{1,2}$ 5.0, $J_{1',2}$ 8.3, H-2), 3.38 (3H, s, CH_2OCH_3), 3.58 (1H, dd, $J_{4,5}$ 7.3, $J_{5,6}$ 10.0, H-5), 3.64 (1H, dd, $J_{1',2}$ 8.3, $J_{1,1'}$ 9.8, H-1′), 3.76 (1H, dd, $J_{1,2}$ 5.0, $J_{1,1'}$ 9.8, H-1), 3.79 (1H, dd, $J_{7,7'}$ 9.7, $J_{6,7'}$ 6.0, H-7′), 3.85 (1H, dd, $J_{7,7'}$ 9.7, $J_{6,7}$ 5.0, H-7), 3.95 (1H, dd, $J_{4,5}$ 7.3, $J_{3,4}$ 5.2, H-4), 4.14 (1H, dd, $J_{2,3}$ 2.6, $J_{3,4}$ 5.2, H-3), 4.67 (2H, s, CH_2OCH_3); δ_C(50 MHz; $CDCl_3$) −5.5 (q, Si(CH_3)$_2$), 18.1 (s, SiC(CH_3)$_3$), 25.8 (q, SiC(CH_3)$_3$), 26.5, 28.3 (2q, C(CH_3)$_2$), 55.3 (d and q, C-6, CH_2OCH_3), 58.6 (d, C-2), 64.4, 68.5 (2t, C-1, C-7), 78.4, 79.9, 81.2 (3d, C-3, C-4, C-5), 96.7 (t, CH_2OCH_3), 109.7 (s, C(CH_3)$_2$); *m*/*z* (APCI$^+$) 392 (MH^+, 100%); (CI^+) [Found: 392.2478 (MH^+). $C_{18}H_{38}NO_6Si$ requires *m*/*z*, 392.2468].

2,6-Dideoxy-2,6-imino-L-*glycero*-L-*galacto*-heptitol (β-homogalactonojirimycin) 7

24 (190 mg, 0.47 mmol) was deprotected according to general procedure 2 to afford **7** as a hygroscopic white foam (80 mg, 85%); the material was consistent with the known enantiomer,[6] $[\alpha]_D^{23}$ +4.0 (*c* 1.0 in MeOH) {enantiomer, lit.,[6b] $[\alpha]_D^{23}$ +4.3 (*c* 1.3 in MeOH)}; δ_H(500 MHz; CD_3OD) 2.50 (1H, ddd, $J_{5,6}$ 9.8, $J_{6,7}$ 3.0, $J_{6,7'}$ 6.0, H-6), 2.76 (1H, m, $J_{2,3}$ 1.3, H-2), 3.36 (1H, dd, $J_{4,5}$ 9.4, $J_{3,4}$ 3.1, H-4), 3.53 (1H, dd, $J_{4,5}$ 9.4, $J_{5,6}$ 9.8, H-5), 3.65 (2H, m, H_2-1), 3.67 (1H, $J_{7,7}$ 11.0, $J_{6,7'}$ 6.0, H-7′), 3.84 (1H, $J_{7,7}$ 11.0, $J_{6,7}$ 3.0, H-7), 3.87 (1H, dd, $J_{2,3}$ 1.3, $J_{3,4}$ 3.1, H-3); *m*/*z* (CI^+) [Found: 194.1024 (MH^+). $C_7H_{16}NO_5$ requires *m*/*z*, 194.1028].

6-Azido-7-*O*-*tert*-butyldimethylsilyl-6-deoxy-3,4-*O*-isopropylidene-1-*O*-methoxymethyl-α,β-D-*gulo*-hept-2-ulofuranose 26

Azido lactols **26** were prepared according to general procedure 1 using butyllithium (2.5 M solution in hexanes; 1.45 cm^3, 3.63 mmol), tributyl[(methoxymethoxy)methyl]stannane **17** (1.52 g, 4.19 mmol) and azido-1,4-lactone **25**[20] (1.0 g, 2.79 mmol). Purification of the crude material using column chromatography (ethyl acetate–hexane, 1:6) afforded the title compound as a colourless oil (960 mg, 79%), $[\alpha]_D^{25}$ −34.9 (*c* 0.96 in $CHCl_3$) (Found: C, 49.7; H, 8.1. $C_{18}H_{34}N_3O_7Si$ requires C, 49.9; H, 8.1%); ν_{max}(film)/cm^{-1} 3401 (br, OH), 2098 (s, N_3); δ_H(500 MHz; $CDCl_3$) Major anomer: 0.10, 0.11 (6H, s, Si(CH_3)$_2$), 0.92 (9H, s, SiC(CH_3)$_3$), 1.30 (3H, s, C(CH_3)$_2$), 1.46, 3.43 (6H, 2s, C(CH_3)$_2$), 3.65 (1H, ddd, $J_{5,6}$ 9.2, $J_{6,7}$ 2.9, $J_{6,7'}$ 5.2, H-6), 3.78 (1H, s, OH), 3.81 (2H, m, H_2-1), 3.83 (1H, dd, $J_{6,7'}$ 5.2, $J_{7,7'}$ 10.8, H-7′), 3.90 (1H, dd, $J_{6,7}$ 2.9, $J_{7,7'}$ 10.8, H-7), 4.23 (1H, dd, $J_{5,6}$ 9.2, $J_{4,5}$ 3.6, H-5), 4.60 (1H, d, $J_{3,4}$ 5.9, H-3), 4.71 (1H, d, *J* 6.5, CH_2OCH_3), 4.74 (1H, dd, $J_{3,4}$ 5.9, $J_{4,5}$ 3.6, H-4), 4.76 (1H, d, *J* 6.5, CH_2OCH_3); δ_C(50 MHz; $CDCl_3$) Major anomer: −5.6, −5.5 (2q, Si(CH_3)$_2$), 18.2 (s, SiC(CH_3)$_3$), 24.6, 26.0 (2q, C(CH_3)$_2$), 25.8 (q, SiC(CH_3)$_3$), 55.5 (q, CH_2OCH_3), 62.5 (d, C-6), 63.3 (t, C-7), 69.4 (t, C-1), 78.4, 79.9 (2d, C-4, C-5), 85.6 (d, C-3), 97.1 (t, CH_2OCH_3), 103.6 (s, C-2), 113.6 (s, C(CH_3)$_2$); *m*/*z* (CI, NH_3) 388 ($MH^+ - N_2 - H_2O$, 100%); (+ve Electrospray) [Found: 451.2595 (MNH_4^+). $C_{18}H_{39}N_4O_7Si$ requires *m*/*z*, 451.2588].

6-Azido-6-deoxy-3,4-*O*-isopropylidene-1-*O*-methoxymethyl-α,β-D-*gulo*-hept-2-ulofuranose 27

TBAF (1.0 M solution in THF; 1.8 cm^3, 1.8 mmol) was added to a solution of azido lactols **26** (600 mg, 1.38 mmol) in THF (5 cm^3), the mixture stirred at room temperature for 2 h and the reaction mixture subsequently concentrated *in vacuo*. The residue was purified by flash chromatography on silica (ethyl acetate–hexane, 3:2) to afford azido lactols **27** as a colourless oil (390 mg, 88%), $[\alpha]_D^{23}$ −32.1 (*c* 0.57 in $CHCl_3$) (Found: C, 45.4; H, 6.8; N, 13.5. $C_{12}H_{21}N_3O_7$ requires C, 45.1; H, 6.6; N, 13.2%); ν_{max}(film)/cm^{-1} 3393 (br, OH), 2101 (s, N_3); δ_H(500 MHz; $CDCl_3$) Major anomer: 1.30, 1.47 (6H, 2s, C(CH_3)$_2$), 3.43 (3H, s, CH_2OCH_3), 3.78, 3.84 (2H, AB system, *J* 11.3, H_2-1), 3.65–3.88 (4H, m, H_2-7, H-6, OH), 4.26 (1H, dd, $J_{5,6}$ 8.9, $J_{4,5}$ 3.6, H-5), 4.60 (1H, d, $J_{3,4}$ 5.9, H-3), 4.72 (1H, d, *J* 6.5, CH_2OCH_3), 4.77 (1H, d, *J* 6.5, CH_2OCH_3), 4.79 (1H, dd, $J_{3,4}$ 5.9, $J_{4,5}$ 3.6, H-4); δ_C(50 MHz; $CDCl_3$) Major anomer: 24.5, 25.9 (2q, C(CH_3)$_2$), 55.5 (q, CH_2OCH_3), 62.0 (t, C-7), 63.3 (d, C-6), 69.2 (t, C-1), 79.4, 79.9 (2d, C-4, C-5), 85.5 (d, C-3), 97.0 (t, CH_2OCH_3), 103.8 (s, C-2), 113.0 (s, C(CH_3)$_2$); *m*/*z* (CI, NH_3) 274 ($MH^+ - N_2 - H_2O$, 100%).

2,6-Dideoxy-2,6-imino-4,5-*O*-isopropylidene-7-*O*-methoxymethyl-L-*glycero*-L-*gluco*-heptitol 29 and 2,6-dideoxy-2,6-imino-3,4-*O*-isopropylidene-1-*O*-methoxymethyl-D-*glycero*-L-*galacto*-heptitol 28

A solution of azido lactols **27** (350 mg, 1.10 mmol) in ethyl acetate (5 cm^3) was stirred under an atmosphere of hydrogen in the presence of 10% palladium on charcoal (150 mg) for 72 h. The reaction mixture was filtered through Celite and concentrated *in vacuo*. The residue was subjected to flash chromatography on silica (methanol–ethyl acetate, 1:9) to give the piperidine **29**, first eluted, as a white amorphous solid (115 mg, 38%), $[\alpha]_D^{25}$ −72.8 (*c* 1.66 in $CHCl_3$); ν_{max}(KBr)/cm^{-1} 3414 (br, NH, OH); δ_H(500 MHz; $CDCl_3$) 1.41, 1.53 [6H, 2s, C(CH_3)$_2$], 2.92 (1H, ddd, $J_{5,6}$ 9.1, $J_{6,7}$ 2.7, $J_{6,7'}$ 7.2, H-6), 3.10 (1H, m, H-2), 3.42 (3H, s, CH_2OCH_3), 3.56 (1H, dd, $J_{6,7'}$ 7.2, $J_{7,7'}$ 9.9, H-7′), 3.83 (1H, dd, $J_{6,7}$ 2.7, $J_{7,7'}$ 9.9, H-7), 3.85 (1H, dd, $J_{1',2}$ 6.4, $J_{1,1'}$ 11.2, H-1′), 3.93 (1H, dd, $J_{1,2}$ 4.3, $J_{1,1'}$ 11.2, H-1), 3.99 (1H, dd, $J_{5,6}$ 9.1, $J_{4,5}$ 5.1, H-5), 4.11 (1H, m, H-3), 4.28 (1H, dd, $J_{3,4}$ 2.6, $J_{4,5}$ 5.1, H-4), 4.70 (2H, AB system, *J* 6.5, CH_2OCH_3); δ_C(50 MHz; $CDCl_3$) 26.3, 28.1 (2q, C(CH_3)$_2$), 55.4 (q, CH_2OCH_3), 56.1, 58.8 (2d, C-2, C-6), 63.5 (t, C-1), 68.7 (t, C-7), 67.5, 71.7, 77.1 (3d, C-3, C-4, C-5), 96.8 (t, CH_2OCH_3), 109.3 (s, C(CH_3)$_2$); *m*/*z* (APCI$^+$) 278 (MH^+, 100%); (+ve Electrospray) [Found: 278.1598 (MH^+). $C_{12}H_{24}NO_6$ requires *m*/*z*, 278.1604], and the piperidine **28**, second eluted, as a white amorphous solid (134 mg, 44%), $[\alpha]_D^{25}$ +17.7 (*c* 1.57 in $CHCl_3$); ν_{max}(KBr)/cm^{-1} 3432 (br, NH, OH); δ_H(500 MHz; $CDCl_3$) 1.37, 1.50 (6H, 2s, C(CH_3)$_2$), 3.26 (1H, m, H-6), 3.41 (3H, s, CH_2OCH_3), 3.63 (2H, m, H-2, H-1), 3.73 (1H, dd, $J_{1',2}$ 9.1, $J_{1,1'}$ 12.7, H-1′), 3.83 (2H, m, H_2-7), 4.02 (1H, m, H-5), 4.23 (1H, dd, $J_{4,5}$ 7.3, $J_{3,4}$ 3.1, H-4), 4.35 (1H, dd, $J_{3,4}$ 3.1, $J_{2,3}$ 1.4, H-3), 4.70 (2H, s, CH_2OCH_3); δ_C(50 MHz; $CDCl_3$) 24.2, 26.8 (2q, C(CH_3)$_2$), 49.7, 50.7 (2d, C-6, C-2), 55.3 (q, CH_2OCH_3), 63.9, 68.9 (2t, C-1, C-7), 70.5, 72.3, 75.2 (3d, C-3, C-4, C-5), 96.6 (t, CH_2OCH_3), 108.9 (s, C(CH_3)$_2$); *m*/*z* (APCI$^+$) 278 (MH^+, 100%); (CI^+) [Found: 278.1604 (MH^+). $C_{12}H_{24}NO_6$ requires *m*/*z*, 278.1604].

2,6-Dideoxy-2,6-imino-L-*glycero*-L-*gluco*-heptitol 11

29 (80 mg, 0.29 mmol) was deprotected according to general procedure 2 to afford **11** as a hygroscopic foam (42 mg, 75%), $[\alpha]_D^{23}$ −41.0 (*c* 0.69 in H_2O); δ_H(500 MHz; D_2O) 2.85 (1H, ddd, $J_{5,6}$ 10.6, $J_{6,7}$ 3.1, $J_{6,7'}$ 5.4, H-6), 3.04 (1H, ddd, $J_{2,3}$ 1.6, $J_{1,2} = J_{1',2}$ = 6.6, H-2), 3.61, 3.63 (2H, 2dd, $J_{1,2} = J_{1',2}$ = 6.6, $J_{1,1'}$ 11.1, H_2-1), 3.68 (1H, dd, $J_{6,7'}$ 5.4, $J_{7,7'}$ 11.7, H-7′), 3.73 (1H, dd, $J_{5,6}$ 10.6, $J_{4,5}$ 3.2, H-5), 3.77 (1H, dd, $J_{6,7}$ 3.1, $J_{7,7'}$ 11.7, H-7), 3.89 (1H, dd, $J_{2,3}$ 1.6, $J_{3,4}$ 3.9, H-3), 3.96 (1H, dd, $J_{3,4}$ 3.9, $J_{4,5}$

3.2, H-4); δ_C(50 MHz; D_2O) 54.6, 56.0 (2d, C-2, C-6), 62.1 (t, C-1, C-7), 66.7, 70.1, 71.7 (3d, C-3, C-4, C-5); *m/z* (CI^+) [Found: 194.1037 (MH^+). $C_7H_{16}NO_5$ requires *m/z*, 194.1028].

2,6-Dideoxy-2,6-imino-D-*glycero*-L-*galacto*-heptitol (α-homogalactonojirimycin) 9

28 (90 mg, 0.32 mmol) was deprotected according to general procedure 2 to afford **9** as a hygroscopic foam (51 mg, 82%), $[\alpha]_D^{23}$ +72.0 (*c* 0.54 in H_2O); δ_H(500 MHz; D_2O) 3.02 (1H, m, $J_{2,3}$ 2.0, H-2), 3.27 (1H, m, H-6), 3.61 (3H, m), 3.77 (2H, m, CH_2), 3.97 (1H, dd, $J_{2,3}$ 2.0, H-3), 4.00 (1H, dd, $J_{5,6}$ 6.0, $J_{4,5}$ 9.8, H-5); δ_C(50 MHz; D_2O) 53.9, 57.2 (2d, C-6, C-2), 57.5, 62.6 (2t, C-1, C-7), 69.6, 71.6 (3d, C-3, C-4, C-5); *m/z* (CI^+) [Found: 194.1038 (MH^+). $C_7H_{16}NO_5$ requires *m/z*, 194.1028].

6-Azido-6,7-dideoxy-3,4-*O*-isopropylidene-1-*O*-methoxymethyl-α,β-D-*gulo*-hept-2-ulofuranose 33

Azido lactols **33** were prepared according to general procedure 1 using butyllithium (2.5 M solution in hexanes; 1.14 cm^3, 2.86 mmol), tributyl[(methoxymethoxy)methyl]stannane **17** (1.19 g, 3.30 mmol) and azido-1,4-lactone **32**[21] (500 mg, 2.20 mmol). Purification of the crude material using column chromatography (ethyl acetate–hexane, 1:2) afforded the title compound as a colourless oil (505 mg, 76%), $[\alpha]_D^{25}$ −56.7 (*c* 1.54 in $CHCl_3$) (Found: C, 47.3; H, 6.9; N, 13.6. $C_{12}H_{21}N_3O_6$ requires C, 47.5; H, 6.9; N, 13.9%); ν_{max}(film)/cm^{-1} 3401 (br, OH), 2089 (s, N_3); δ_H(500 MHz; $CDCl_3$) Major anomer: 1.28 (3H, d, $J_{6,7}$ 6.6, H_3-7), 1.30, 1.46 (6H, 2s, $C(CH_3)_2$), 3.43 (3H, s, CH_2OCH_3), 3.76 (1H, s, OH), 3.81 (3H, m, $J_{1,1'}$ 11.1, H_2-1, obscured H-6), 4.00 (1H, dd, $J_{5,6}$ 9.3, $J_{4,5}$ 3.6, H-5), 4.58 (1H, d, $J_{3,4}$ 5.8, H-3), 4.71 (1H, d, *J* 6.5, CH_2OCH_3), 4.73 (1H, dd, $J_{3,4}$ 5.8, $J_{4,5}$ 3.6, H-4), 4.76 (1H, d, *J* 6.5, CH_2OCH_3); δ_C(50 MHz; $CDCl_3$) Major anomer: 16.0 (q, C-7), 24.6, 25.9 (2q, $C(CH_3)_2$), 55.4 (q, CH_2OCH_3), 57.2 (d, C-6), 69.2 (t, C-1), 80.1, 83.0, 85.4 (3d, C-3, C-4, C-5), 97.0 (t, CH_2OCH_3), 103.9 (s, C-2), 112.8 (s, $C(CH_3)_2$); *m/z* (CI, NH_3) 258 ($MH^+ - N_2 - H_2O$, 100%).

1,2,6-Trideoxy-2,6-imino-4,5-*O*-isopropylidene-7-*O*-methoxymethyl-L-*glycero*-L-*gluco*-heptitol 34 and 2,6,7-trideoxy-2,6-imino-3,4-*O*-isopropylidene-1-*O*-methoxymethyl-D-*glycero*-L-*galacto*-heptitol 35

A solution of azido lactols **33** (300 mg, 0.99 mmol) in ethyl acetate (5 cm^3) was stirred under an atmosphere of hydrogen in the presence of 10% palladium on charcoal (200 mg) for 72 h. The reaction mixture was filtered through Celite and concentrated *in vacuo*. The residue was subjected to flash chromatography on silica (methanol–ethyl acetate, 1:9) to afford the piperidine **34**, first eluted, as a white solid (115 mg, 44%), $[\alpha]_D^{25}$ −107.1 (*c* 0.85 in $CHCl_3$) (Found: C, 55.5; H, 9.05. $C_{12}H_{23}NO_5$ requires C, 55.2; H, 8.9%); ν_{max}(KBr)/cm^{-1} 3298 (br, NH, OH); δ_H(500 MHz; $CDCl_3$) 1.17 (3H, d, $J_{1,2}$ 6.6, H_3-1), 1.36, 1.50 (6H, 2s, $C(CH_3)_2$), 2.91 (1H, ddd, $J_{5,6}$ 8.9, $J_{6,7}$ 2.8, $J_{6,7'}$ 9.3, H-6), 3.12 (1H, dq, $J_{2,3}$ 1.4, $J_{1,2}$ 6.6, H-2), 3.35 (1H, dd, $J_{7,7'}$ 9.7, $J_{6,7'}$ 9.3, H-7′), 3.38 (3H, s, CH_2OCH_3), 3.76 (1H, m, H-3), 3.77 (1H, dd, $J_{4,5}$ 5.2, $J_{5,6}$ 8.9, H-5), 3.79 (1H, dd, $J_{7,7'}$ 9.7, $J_{6,7}$ 2.8, H-7), 3.27 (1H, dd, $J_{4,5}$ 5.2, $J_{3,4}$ 2.6, H-4), 4.65 (2H, AB system, *J* 6.4, CH_2OCH_3); δ_C(50 MHz; $CDCl_3$) 16.8 (q, C-1), 26.3, 28.1 [2q, $C(CH_3)_2$], 50.8 (d, C-2), 55.3 (q, CH_2OCH_3), 58.6 (d, C-6), 69.4 (t, C-7), 68.1, 72.1, 77.1 (3d, C-3, C-4, C-5), 96.7 (t, CH_2OCH_3), 109.4 (s, $C(CH_3)_2$); *m/z* ($APCI^+$) 262 (MH^+, 100%); (+ve Electrospray) [Found: 262.1654 (MH^+). $C_{12}H_{24}NO_5$ requires *m/z*, 262.1654], and the piperidine **35**, second eluted as a white solid (120 mg, 46%), $[\alpha]_D^{25}$ +39.2 (*c* 0.83 in $CHCl_3$); ν_{max}(KBr)/cm^{-1} 3435 (br, NH, OH); δ_H(500 MHz; $CDCl_3$) 1.13 (3H, d, $J_{6,7}$ 6.9, H_3-7), 1.33, 1.50 (6H, 2s, $C(CH_3)_2$), 3.32 (1H, dq, $J_{5,6}$ 3.7, $J_{6,7}$ 6.9, H-6), 3.37 (3H, s, CH_2OCH_3), 3.47 (1H, ddd, $J_{2,3}$ 2.9, $J_{1,2}$ 5.5, $J_{1',2}$ 8.0, H-2), 3.59 (1H, dd, $J_{1,1'}$ 9.6, $J_{1',2}$ 8.0, H-1′), 3.70 (1H, dd, $J_{5,6}$ 3.7, $J_{4,5}$ 5.2, H-5), 3.73 (1H, dd, $J_{1,1'}$ 9.6, $J_{1,2}$ 5.5, H-1), 4.12 (1H, dd, $J_{3,4}$ 6.2, $J_{4,5}$ 5.2, H-4), 4.25 (1H, dd, $J_{3,4}$ 6.2, $J_{2,3}$ 2.9, H-3), 4.66 (2H, AB system, *J* 6.4, CH_2OCH_3); δ_C(50 MHz; $CDCl_3$) 14.8 (q, C-7), 25.3, 27.6 (2q, $C(CH_3)_2$), 47.9, 49.2 (2d, C-6, C-2), 55.2 (q, CH_2OCH_3), 68.9 (t, C-1), 72.0, 72.7, 77.7 (3d, C-3, C-4, C-5), 96.6 (t, CH_2OCH_3), 108.8 (s, $C(CH_3)_2$); *m/z* ($APCI^+$) 262 (MH^+, 100%); (+ve Electrospray) [Found: 262.1655 (MH^+). $C_{12}H_{24}NO_5$ requires *m/z*, 262.1654].

1,2,6-Trideoxy-2,6-imino-L-*glycero*-L-*gluco*-heptitol 12

34 (90 mg, 0.34 mmol) was deprotected according to general procedure 2 to afford **12** as a hygroscopic foam (48 mg, 79%), material identical to the known compound,[5] $[\alpha]_D^{23}$ −40.7 (*c* 0.71 in H_2O) {lit.,[5] $[\alpha]_D^{25}$ −38.1 (*c* 0.9 in H_2O)}; δ_H(200 MHz; D_2O, pH 8) 1.11 (3H, d, $J_{1,2}$ 6.8, H_3-1), 2.93 (1H, ddd, *J* 4.0, *J* 10.7, H-6), 3.20 (1H, dq, $J_{1,2}$ 6.8, H-2), 3.71–3.77 (4H, m), 3.98 (1H, dd).

2,6,7-Trideoxy-2,6-imino-D-*glycero*-L-*galacto*-heptitol 10

35 (85 mg, 0.33 mmol) was deprotected according to general procedure 2 to afford **10** as a foam (42 mg, 73%), $[\alpha]_D^{23}$ +67.3 (*c* 0.63 in H_2O); δ_H(500 MHz; D_2O, pH 9) 1.06 (3H, d, $J_{6,7}$ 7.1, H_3-7), 2.96 (1H, ddd, $J_{2,3}$ 1.9, $J_{1,2}$ 6.5, $J_{1',2}$ 6.7, H-2), 3.26 (1H, dq, $J_{5,6}$ 6.0, $J_{6,7}$ 7.1, H-6), 3.54 (1H, dd, $J_{1,1'}$ 11.1, $J_{1,2}$ 6.5, H-1), 3.59 (1H, dd, $J_{1,1'}$ 11.1, $J_{1',2}$ 6.7, H-1′), 3.66 (1H, dd, $J_{3,4}$ 3.3, $J_{4,5}$ 10.1, H-4), 3.82 (1H, dd, $J_{5,6}$ 6.0, $J_{4,5}$ 10.1, H-5), 3.94 (1H, dd, $J_{3,4}$ 3.3, $J_{2,3}$ 1.9, H-3); δ_C(50 MHz; D_2O, pH 9) 11.5 (q, C-7), 51.3, 53.0 (2d, C-6, C-2), 62.1 (t, C-1), 70.2, 70.6, 70.8 (3d, C-3, C-4, C-5); *m/z* (CI^+) [Found: 178.1080 (MH^+). $C_7H_{16}NO_4$ requires *m/z*, 178.1079].

Biological assays

β-HGJ 7. Enzymes were purified as described[23] and assayed using the corresponding *p*-nitrophenyl glycoside. Coffee bean α-galactosidase was assayed using 0.8 mM (for IC_{50}-values) or 0.06–0.4 mM (for K_i-values) *p*-nitrophenyl α-galactopyranoside in 0.1 M citrate phosphate buffer,[24] pH 6.0 for 30 min at 37 °C in the presence of test compound (0–0.75 μM). The reaction was stopped with 0.5 M sodium carbonate and the absorbance measured at 400 nm. Inhibition constants were calculated from the slope of inhibitor concentration *versus* activity (IC_{50}) or from Lineweaver–Burk plots (K_i). Jack bean β-galactosidase was assayed using 0.5 mM *p*-nitrophenyl β-galactopyranoside in 0.1 M citrate phosphate buffer, pH 3.8 for 30 min at 37 °C in the presence of 0–0.9 mM compound. The reaction was stopped, absorbance measured and the data plotted as described.[23]

9, 11 and 10. Tested at 113 μM (123 μM for **10**) against a range of commercially available (Sigma) hydrolases including α-glucosidase (Brewers' yeast and rice), β-glucosidase (almond), α-mannosidase (Jack bean), α-galactosidase (green coffee bean), β-galactosidase (*E. coli*, *A. niger* and bovine liver) and naringinase (*Penicillium decumbens*). Enzymes were approx. 0.02 unit per assay volume, substrates were 3.5 mM *p*-nitrophenyl glycosides, and assays were conducted as previously described.[25] K_i-Values were determined for α-galactosidase using Lineweaver–Burk analysis and substrate concentrations ranging from 0.18 to 1.8 mM.

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