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Chemoenzymatic syntheses of (−)-1-deoxymannojirimycin (DMJ) and its naturally occurring 6-*O*-α-L-rhamnopyranosyl glycoside

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The naturally occurring sugar mimetic alkaloids 1-deoxymannojirimycin (DMJ, 1) and 6-*O*-α-L-rhamnopyranosyl-DMJ (2) have each been prepared in a completely stereoselective manner from the *cis*-1,2-dihydrocatechol 3, itself obtained in enantiomerically pure form by microbial oxidation of chlorobenzene.

The alkaloid 1-deoxymannojirimycin (DMJ, **1**) has been isolated from the seeds of *Lonchocarpus sericeus*[1] and *L. costaricensis* (Leguminosae)[2] as well as from cultures of *Streptomycese lavendulae*.[3] Like a number of other naturally occurring polyhydroxylated piperidines,[4] this compound displays various rather important biological properties. In particular, it is a potent inhibitor of several α-mannosidases.[5] Interestingly, derivatisation of DMJ can result in significant enhancement of its enzyme inhibiting properties. For example, one of us has recently reported[6] the isolation of 6-*O*-α-L-rhamnopyranosyl-DMJ (Rha-DMJ, **2**), as well as DMJ itself,[6b] from the bark of *Anglyocalyx pynaertii* (Leguminosae) and shown that this first naturally occurring glycoside of DMJ is a significantly more potent inhibitor of α-L-fucosidase than parent **1**. Given the therapeutic potential arising from a capacity to control glycosylation,[7] DMJ and its derivatives have attracted attention as agents for the treatment of, *inter alia*, cancer, diabetes and viral infections.[7,8] As a result various ingenious synthetic routes to polyhydroxylated piperidines, including DMJ, have been developed.[4,9] One of the most elegant and effective methods has been described by Hudlicky and co-workers[10] who were able to convert the *cis*-1,2-dihydrocatechol **3** into various *endo*-nitrogenous aza sugars including (+)-kifunensine and mannojirimycin. A great attraction of this approach is that compound **3**[11] is available in large quantity and enantiomerically pure form *via* microbial oxidation of chlorobenzene and it can be readily elaborated to various differentially protected and, therefore, synthetically valuable forms of several aza sugars. Here we report on the application of this approach to the preparation of DMJ and a differentially protected derivative that has been exploited in the (first) total synthesis of 6-*O*-α-L-rhamnopyranosyl-DMJ (**2**). Since various disaccharide derivatives of DMJ have been identified as potential cancer remedies[12] the present work should provide useful methods for accessing members of this interesting class of pseudo-trisaccharide.

The synthesis of DMJ from *cis*-1,2-dihydrocatechol **3** is shown in Scheme 1 and follows the early steps of Hudlicky's mannojirimycin synthesis. Thus, the readily available acetonide derivative[10] of diol **3** was subject to epoxidation with *m*-CPBA and the resulting epoxide **4**[10] (81% from **3**) converted, in a completely regioselective fashion, into the chlorohydrin **5** (98%) upon reaction with lithium chloride. Treatment of the last compound with lithium azide resulted in an S_N2-type displacement and formation of the previously reported[10] *cis*-azido-alcohol **6** (91%, mp 93–94 °C) which was *O*-benzylated under standard conditions to give the expected ether **7**† {100%, $[\alpha]_D$ −111.0 (*c* 0.8)‡}. Ozonolytic cleavage of a methanolic solution of this last compound followed by reductive work-up with sodium borohydride and subsequent protection of the ensuing 1°-alcohol using TBS-Cl then afforded the azido-ether **8** {89%, $[\alpha]_D$ −10.2 (*c* 1.3)}. Two-fold hydrogenolysis of this last compound using 5% Pd on C as catalyst resulted in formation of lactam **9**[13] {86%, mp 110–111 °C, $[\alpha]_D$ +15.3 (*c* 1.0)} which was converted into the corresponding piperidine **10**[13] {73%, mp 86–87 °C, $[\alpha]_D$ −66 (*c* 0.6)} upon exposure to the borane–dimethyl sulfide complex followed by cleavage of the resulting borane–amine complex with methanol in the presence of 10% palladium on carbon.[14] Treatment of compound **10** with 80% v/v trifluoroacetic acid (TFA)–H_2O at 18 °C for 18 h then gave the TFA salt, **1**·TFA, of DMJ. The spectral and physical data derived from this material proved an excellent match with those obtained on an authentic sample.

The differentially protected DMJ derivative **10** served as an effective precursor to 6-*O*-α-L-rhamnopyranosyl-DMJ (**2**) (Scheme 2). Thus, the former compound was *N*- and *O*-benzylated under standard conditions and the TBS-ether moiety then cleaved with tetra-*n*-butylammonium fluoride (TBAF) to give the 1°-alcohol **11**[15] {89% from **10**, $[\alpha]_D$ +10.5 (*c* 1.0)} which could be coupled with the L-rhamnopyranosyl bromide **12**[16] in the presence of silver triflate.[17] Coupling product **13** {87%, $[\alpha]_D$ −42.5 (*c* 1.0)} was deprotected in a stepwise fashion by sequential exposure to TFA (resulting in removal of the acetonide group), methanolic sodium methoxide (to remove the acetate groups) and dihydrogen in the presence of 5% Pd on C. The sample of Rha-DMJ (**2**) {80% from **13**, $[\alpha]_D$ −51.7 (*c* 0.6, H_2O)} thus obtained after this final deprotection step gave spectral data in full accord with the assigned structure and in excellent agreement with those derived from the natural material.[6a]

Synthetic **2** was evaluated for its glycosidase inhibitory activity (Table 1) and shown to be 5–10 times less active than

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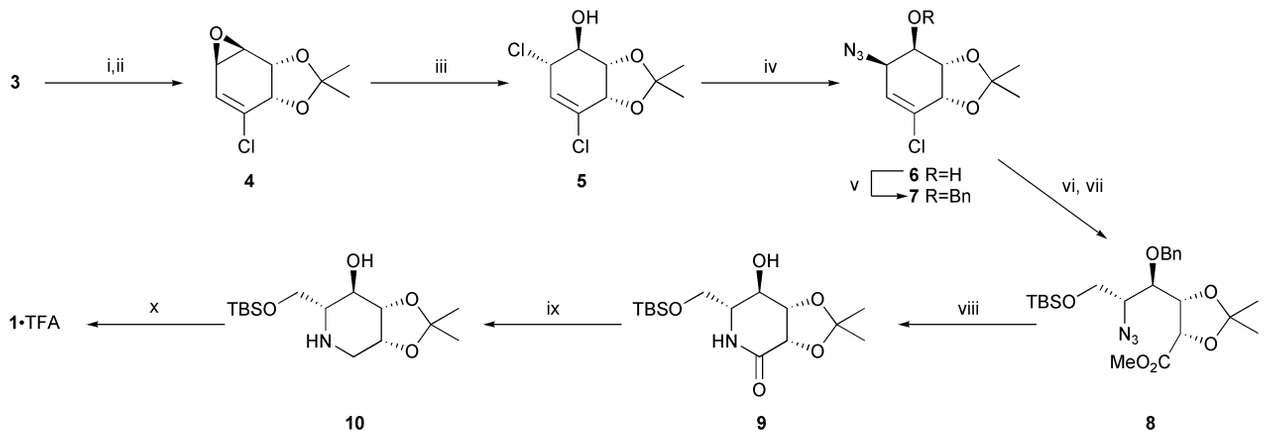
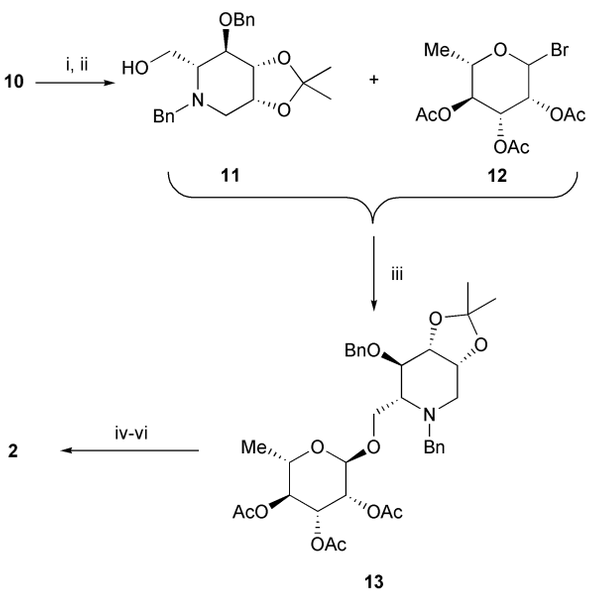


Scheme 1 *Reagents and conditions*: (i) 2,2-DMP, *p*-TsOH·H_2O (cat.), 18 °C, 1 h; (ii) *m*-CPBA (1.0 mole equiv.), CH_2Cl_2, 0–18 °C, 11 h; (iii) LiCl (5 mole equiv.), HOAc (3 mole equiv.), THF, 18 °C, 17 h; (iv) LiN_3 (3 mole equiv.), DMF, 18 °C, 72 h; (v) BnBr (3.0 mole equiv.), KI (0.04 mole equiv.), NaH (1.6 mole equiv.), THF, 0–18 °C, 24 h; (vi) O_3 (excess), pyridine (4.9 mole equiv.), MeOH, −78 °C, 1 h then $NaBH_4$ (11 mole equiv.), −10 °C, 3 h; (vii) TBS-Cl (1.5 mole equiv.), imidazole (5 mole equiv.), CH_2Cl_2, 18 °C, 2 h; (viii) H_2 (1 atm), 5% Pd on C (cat.), EtOAc, 18 °C, 36 h; (ix) BH_3·DMS (10 mole equiv.), THF, 18 °C, 4.5 h then 10% Pd on C, MeOH, 18 °C, 38 h; (x) 80% v/v TFA–H_2O, 18 °C, 20 h.



Scheme 2 *Reagents and conditions*: (i) BnBr (4.3 mole equiv.), KI (0.5 mole equiv.), NaH (6.8 mole equiv.), THF, 18 °C, 24 h; (ii) TBAF (1.2 mole equiv.), THF, 18 °C, 2 h; (iii) AgOTf (1.2 mole equiv,), 4 Å molecular sieves, CH_2Cl_2, −10 °C, 0.5 h; (iv) 1 : 5 v/v TFA–CH_2Cl_2, 18 °C, 24 h; (v) NaOMe (0.2 M solution in MeOH), 18 °C, 2 h; (vi) H_2 (1 atm), 5% Pd on C (cat.), EtOH, H_2O (trace), 18 °C, 48 h.

the natural product. These results call into question the previously reported[6] glycosidase inhibitory activity of the naturally occurring **2** which may have been compromised by contamination of the assay sample with a more powerful inhibitor like β-L-homofuconojirimycin. Despite this, compound **2** must still be regarded as a rather potent inhibitor of bovine epididymis-derived α-fucosidase.

Table 1 Comparison of the glycosidase inhibitory properties of synthetic and natural samples of compound **2**[a]

Enzyme	IC_{50}/μM	
	Synthetic	Natural
α-Fucosidase		
Bovine epididymis	2.8	0.5
Human placenta	23	4.6
Rat epididymis	28	3.2
α-Mannosidase		
Rat epididymis	>1000	460
α-L-Rhamnosidase		
Penicillium decumbens	>1000	740

[a] The enzymes α-L-fucosidases from bovine epididymis and human placenta, and α-L-rhamnosidase from *Penicillium decumbens* were purchased from Sigma Chemical Co. The rat epididymal fluid prepared from rat epididymis according to the method of Skudlarek *et al.*[18] was used as the enzyme source of rat epididymis glycosidases. The glycosidase activities were determined using an appropriate *p*-nitrophenyl glycoside as substrate at the optimum pH of each enzyme. The reaction was stopped by adding 400 mM Na_2CO_3. The released *p*-nitrophenol was measured spectrometrically at 400 nm.

Experimental

Compound 13

A solution of silver triflate (203 mg, 0.79 mmol) in toluene (5 mL) was added dropwise to a magnetically stirred solution of alcohol **11** (253 mg, 0.66 mmol) and bromide **12** (297 mg, 0.84 mmol) in CH_2Cl_2 (20 mL) containing activated 4 Å molecular sieves (2 g) and maintained under a nitrogen atmosphere at −10 °C. After 0.5 h the reaction mixture was treated with Et_3N (1.5 mL) then CH_2Cl_2 (50 mL). The ensuing mixture was filtered through a 2 cm deep pad of Celite™ and the filtrate concentrated under reduced pressure. The light-yellow oil thus obtained was subjected to flash chromatography (silica, 25–35% v/v ethyl acetate–hexane gradient elution) and concentration of the appropriate fractions (R_f 0.5 in 4 : 2.5 : 5.5 v/v/v ethyl acetate–CH_2Cl_2–hexane) afforded glycoside **13** (377 mg, 87%) as a white foam (Found: $M^{+\bullet}$, 655.2971. $C_{35}H_{45}NO_{11}$ requires $M^{+\bullet}$, 655.2993). ν_{max} (NaCl)/cm^{-1} 2985, 2936, 1750, 1371, 1223, 1053, 735, 699; δ_H (300 MHz, $CDCl_3$) 7.40–7.10 (10 H, complex m), 5.32–5.18 (3 H, complex m), 5.04 (1 H, t, *J* 9.8 Hz), 4.72 (1 H, d, *J* 11.7 Hz), 4.64 (1 H, broad s), 4.63 (1 H, d, *J* 11.7 Hz, partially obscured), 4.30 (2 H, broad s), 4.04–3.90 (2 H, complex m), 3.81 (2 H, m), 3.62 (1 H, d, *J* 14.0 Hz), 3.54 (1 H, dd, *J* 10.0 and 4.8 Hz), 2.88 (1 H, m), 2.74 (1 H, broad d, *J* 14.0 Hz), 2.13 (3 H, s), 2.04 (3 H, s), 1.99 (3 H, s), 1.55 (3 H, s), 1.34 (3 H, s), 1.14 (3 H, d, *J* 6.3 Hz); δ_C (75 MHz, $CDCl_3$) 169.9, 169.8, 169.7, 139.0, 138.2, 128.4, 128.3, 128.2, 127.8, 127.6, 126.8, 109.0, 97.5, 76.5, 75.2, 72.8, 72.1, 71.0, 69.9, 69.2, 66.7, 66.5, 60.8, 58.7, 49.9, 27.4, 25.5, 21.1, 21.0, 20.9, 17.5; *m/z* (EI) 655 (<1%, $M^{+\bullet}$), 640 (5), 352 (100), 91 (60).

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Notes and references

† All new and stable compounds had spectroscopic data (IR, NMR, mass spectrum) consistent with the assigned structure. Satisfactory combustion and/or high-resolution mass spectral analytical data were obtained for new compounds and/or suitable derivatives.
‡ Unless specified otherwise, all optical rotations were determined in chloroform solution at 18–26 °C.

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