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Potent glycosidase inhibitors via hetero Diels–Alder reactions: asymmetric synthesis of 5-methyl-trihydroxypyrrolidines

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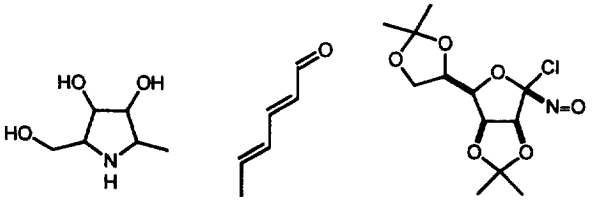
Abstract: Some straightforward chemical transformations of oxazine diol **4**, which was obtained from sorbaldehyde **2** by an asymmetric hetero Diels–Alder reaction followed by osmylation, led to the protected dihydroxypyrrolidine-aldehyde **8a** and, after basic epimerisation, to **8b**. Reduction of the aldehyde moiety and deprotection gave the potent glycosidase inhibitors 2,5,6-trideoxy-2,5-imino-D-altritol and D-allitol **9a** and **9b**. © 1997 Elsevier Science Ltd. All rights reserved.

Some type **1** 5-methyl-polyhydroxypyrrolidines, also called ω-deoxy-azasugars, are powerful L-fucosidase inhibitors; they were prepared via chemo-enzymatic syntheses by Wong *et al.*[1–3]. Recently the all-*trans* stereoisomer was isolated from a tree of the *leguminoseae* family (*Sophoreae* tribu)[4] and shown to possess β-mannosidase inhibitor properties[4] (Scheme 1).

We describe herein the chemical[5] synthesis of two type **1** pyrrolidinetriols starting from the enantiomerically pure oxazine-diol **4** which has been obtained previously with excellent enantioselectivity (>98%) from sorbaldehyde **2** via an asymmetric hetero Diels–Alder reaction using chiral chloronitroso derivative **3**[6] (obtained from D-mannose according to the Kresze and Vasella procedure[7,8]). The key steps are N-cyclisation after reductive cleavage of the N–O bond and base-induced epimerisation of the formyl group.

Synthesis

Protection of the *cis*-diol moiety as the acetonide **5** (dimethoxypropane, Amberlyst-15, 2 h, 45°C, quant.) followed by hydrogenolysis of the N–O bond (Pd/C, EtOH, 50°C) gave a linear amino acetal which was N-protected again to **6a** ($ClCO_2Bn$, NaOH, rt; 15 h, 84% from **5**). Mesylation to **6b** (MsCl, NEt_3 in CH_2Cl_2) and cyclisation (aq. N NaOH, 80°C, 1.5 d, 65% from **6a**) gave the protected



1 **2** **3**

Scheme 1.

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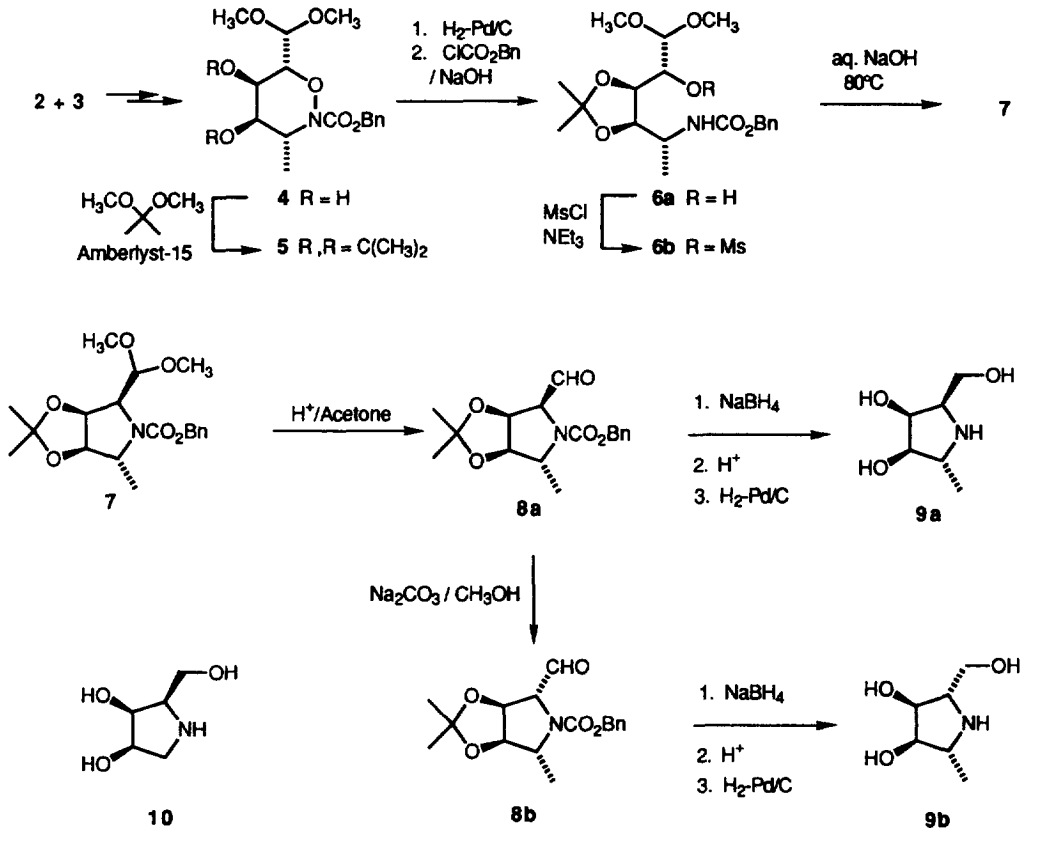

Scheme 2.

pyrrolidine-triol **7**. Selective deprotection of the dimethylacetal function by Amberlyst-15 in acetone led in good yield (*ca.* 80%) to 2,5-*trans*-aldehyde **8a** as the key product (Scheme 2).

Compound **8a** presents a severe steric bulkiness, so that its ^{1}H-NMR spectrum showed distinct resonance for two rotamers. Epimerisation with Na_2CO_3 in MeOH (1 h, rt, *ca.* 65%) gave the sterically less crowded and thermodynamically more stable 2,5-*cis*-aldehyde **8b**. Crude compounds **8a** and **8b** were characterised by ^{1}H-NMR[9]. Reduction with $NaBH_4$ in EtOH of the aldehyde function to the corresponding alcohols, deprotection (Amberlyst-15 $H^{\oplus}$, EtOH, 80°C) followed by catalytic hydrogenolysis (on Pd/C) led to the expected pyrrolidinetriols **9a** and **9b** (50% yield from **7**)[9].

Glycosidases inhibition assays[10,13]

Pyrrolidines **9a,b** proved to be glycosidases inhibitors: **9a** is a strong competitive inhibitor of α-D-mannosidase and α-L-fucosidase (83% and 95% inhibition at 1 mM, Ki=53 μM and 9 μM respectively), but a weak inhibitor of β-D-glucosidase (40% inhibition at 1 mM) and has no effect on α-D-glucosidase. Isomer **9b** seems to be a specific α-L-fucosidase inhibitor (85% inhibition at 1 mM).

Pyrrolidine **9a** is an α-D-mannosidase inhibitor as potent as 1-deoxy-*manno*-nojirimycin[11]. Its activity is similar to that of unmethylated pyrrolidine compound **10**, except that the latter has no effect on α-L-fucosidase[12]. The 5-methyl substituent seems to be responsible for the α-L-fucosidase inhibitory activity, but has a marginal effect on α- and β-D-glucosidase and α-D-mannosidase.

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References

1. Wang, Y.-F.; Dumas, D. P.; Wong, Ch.-H., *Tetrahedron Lett.* **1993**, *34*, 403.
2. Dumas, D. P.; Kajimoto, T.; Liu, K. K.-C.; Wong, Ch.-H.; Berkowitz, D. B.; Danishefsky, S. J., *Bioorg. Med. Chem. Lett.* **1992**, *2*, 33.
3. Liu, K. K.-C.; Kajimoto, T.; Chen, L.; Zhong, Z.; Ichikawa, Y.; Wong, Ch.-H., *J. Org. Chem.* **1991**, *56*, 6280.
4. Molyneux, R. J.; Pan, Y. T.; Tropea, J. E.; Elbein, A. D.; Lawyer, C. H.; Hughes, D. J.; Fleet, G. W. J., *J. Nat. Products* **1993**, *56*, 1356.
5. It is noteworthy that enantiospecific chemical syntheses are more accurate than enantioselective chemio-enzymatic syntheses for assays of glycosidases inhibition: Provencher, L.; Steensma, D. H.; Wong, Ch.-H., *Bioorg. Med. Chem.* **1994**, *2*, 1179; Shilvock, J. P.; Wheatley, J. R.; Davis, B.; Nash, R. J.; Griffiths, Rh. C.; Jones, M. G.; Müller, M.; Crook, S.; Watkin, D. J.; Smith, C.; Besra, G. S.; Brennan, P. J.; Fleet, G. W. J., *Tetrahedron Lett.* **1996**, *37*, 8569.
6. Defoin, A.; Sarazin, H.; Streith, J., *Synlett* **1995**, 1187.
7. Felber, H.; Kresze, G.; Prewo, R.; Vasella, A., *Helv. Chim. Acta* **1986**, *69*, 1137; *corrigendum*: Braun, H.; Charles, R.; Kresze, G.; Sabuni, M.; Winkler, J., *Liebigs Ann. Chem.* **1987**, 11298.
8. Braun, H.; Felber, H.; Kresze, G.; Schmidtchen, Fr. P.; Prewo, R.; Vasella, A., *Liebigs Ann. Chem.* **1993**, 261.
9. **8a**: ^{1}H-NMR (250 MHz, $CDCl_3$, 300 K), major rotamer: 9.43, d, CHO; 7.36, m, 5 H arom.; 5.16, 5.06, 2d, J=12.2, CH_2 benzyl; 4.99, t, H-3; 4.47, d, H-4; 4.28, q, H-5; 4.15, dd, H-2; 1.46, 1.28, 2s, 2 Me; 1.14, d, Me-5; J(CHO,2)=3.4, J(2,3)=6.4, J(3,4)=5.8, J(4,5)=0, J(5,Me-5)=6.9; minor rotamer: 9.32, d, CHO; 7.36, m, 5 H arom.; 5.12, 5.08, 2d, J=12.2, CH_2 benzyl; 5.03, t, H-3; 4.47, d, H-4; 4.36, q, H-5; 4.15, dd, H-2; 1.43, 1.28, 2s, 2 Me; 1.23, d, Me-5; J(CHO,2)=3.3, J(2,3)=7.0, J(3,4)=5.8, J(4,5)=0, J(5,Me-5)=6.9. **8b**: ^{1}H-NMR (250 MHz, $CDCl_3$, 336 K): 9.66, s, CHO; 7.33, s, 5 H arom.; 5.19, s, CH_2 benzyl; 4.92, dd, H-3; 4.57, s broad, H-2; 4.34, d, H-4; 4.33, q, H-5; 1.45, 1.32, 2s, 2Me; 1.15, d, Me-5; J(2,3)=1.9, J(3,4)=5.6, J(4,5)=0, J(5,Me-5)=7.0. **9a**: colourless crystals; m.p.=118–120°C; $[\alpha]_d^{20}$=+36 (c=0.83, MeOH); ^{1}H-NMR (250 MHz, D_2O): 4.20, t, H-3; 3.79, dd, Ha-1′; 3.68, dd, H-4; 3.60, dd, Hb-1′; 3.39, dt, H-2; 3.06, dq, H-5; 1.20, d, Me-5; J(1a′,1b′)=11.0, J(1a′,2)=J(1b′,2)=6.8, J(2,3)=4.4, J(3,4)=4.4, J(4,5)=8.8, J(5,Me)=6.4. Anal. calc. for $C_6H_{13}NO_3$: C 48.96, H 8.90, N 9.52; found: C 48.6, H 8.9, N 9.3. **9b**: colourless resin; $[\alpha]_d^{20}$=−2 (c=1.0, MeOH); ^{1}H-NMR (250 MHz, D_2O): 3.92, t, H-3; 3.71, dd, Ha-1′; 3.67, dd, Hb-1′; 3.60, dd, H-4; 3.09, q, H-2; 3.05, dq, H-5; 1.22, d, Me-5; J(1a′,1b′)=11.6, J(1a′,2)=5.1, J(1b′,2)=5.6, J(2,3)=5.2, J(3,4)=6.1, J(4,5)=7.3, J(5,Me)=6.5. Anal. calc. for $C_6H_{13}NO_3$: C 48.96, H 8.90, N 9.52; found: C 48.7, H 8.9, N 9.4.
10. Glycosidase activity of **9a** and **9b** were determined according to the literature[13] at 37°C in 0.05 M Na citrate–phosphate buffer against α-D-glucosidase (EC 3.2.1.20) from *Bacillus stearothermophilus* at pH 6.8, β-D-glucosidase (EC 3.2.1.21) from almond at pH 5.0, α-D-mannosidase (EC 3.2.1.24) from Jack beans at pH 4.5 and α-L-fucosidase (EC 3.2.1.51) from bovine kidney at pH 5.5. Glycosidases and corresponding *p*-nitrophenyl glycopyranosides were obtained from Sigma Chemical Co. The amount of enzyme added in each essay was adjusted so that less than 10% of the substrate would be consumed. Inhibitors were incorporated variously to give a final concentration in the range of 10^{-3} to 10^{-5} M. The release of *p*-nitrophenol was measured at 400 nm in a spectrophotometer Gilford 'respons' *versus p*-nitrophenol calibration solutions. Dissociation constants for inhibitors were calculated in absence and presence of inhibitors according to the Lineweaver–Burck method.

11. Legler, G.; Jülich, E., *Carbohydr. Res.* **1984**, *128*, 61.
12. Fleet, G. W. J.; Nicholas, S. J.; Smith, P. W.; Evans, S. V.; Fellows, L. E.; Nash, R. J., *Tetrahedron Lett.* **1985**, *26*, 3127.
13. Lundblad, G.; Bjare, U.; Lybing, S.; Måhlén, A., *Int. J. Biochem.* **1983**, *15*, 835.

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