COMMUNICATION

ChemComm
www.rsc.org/chemcomm

Synthesis of the aminocyclopentitol moieties of the hopanoids of *Zymomonas mobilis* and '*Anacystis montana*'

Jose Luis Chiara,* Isabel Storch de Gracia and Ágatha Bastida
Instituto de Química Orgánica General, CSIC, Juan de la Cierva, 3. Madrid, Spain.
E-mail: jl.chiara@iqog.csic.es; Fax: 34 915644853

Received (in Cambridge, UK) 28th April 2003, Accepted 3rd June 2003
First published as an Advance Article on the web 20th June 2003

The first synthesis of the cyclopentitol units in bacterial hopanoids has been accomplished from D-glucosamine and the possible biological activity of the free cyclitols as glycosidase inhibitors has been studied.

The triterpenoids of the hopane family are widely distributed among prokaryotes and are ubiquitous in the organic matter of virtually all sediments and petroleum.[1] In spite of their abundance, their biological function is not well known except for their role as membrane stabilizers, similar to that of sterols in eukaryotes.[2] Biosynthetic studies on the hopane nucleus led to the discovery of the long overlooked methylerythritol phosphate pathway for isoprenoid biosynthesis.[3] Bacteriohopanepolyols are the most widespread and structurally diverse of all hopanoids. They are most frequently found substituted with polar moieties derived from amino acids or carbohydrates. Particularly interesting are those containing a cyclopentitol unit linked through an ether bond to the C-35 hydroxy group (Fig. 1), which in some bacterial strains constitute up to 40% of the total hopanoids.[4] Two structural types have been identified that differ in the stereochemistry of two of the five stereogenic centers of the cyclopentitol ring.[5] Bacteriohopanetetrol ether **1a** has been found in *Methylobacterium organophylum*,[6] *Rhodopseudomonas acidophila*[7] and *Zymomonas mobilis*[6,8] while the diastereoisomeric bacteriohopanetetrol ether **1b** has been isolated only from cyanobacterium '*Anacystis montana*'.[8] Recent biosynthetic studies in *Z. mobilis*[9] have shown that *N*-acetyl-D-glucosamine is the precursor of the cyclopentitol moiety in **1a**. To our knowledge, no synthesis of these abundant cyclopentitols has been described yet in the literature.

Intrigued by the structural similarity between the cyclopentitol units in bacterial hopanoids and that found in the α,α-trehalase inhibitor of microbial origin trehazolin (**2**), we decided to undertake the first synthesis of the cyclitols in **1a** and **1b** and study their possible biological activity as glycosidase inhibitors. Our synthetic approach follows a similar strategy to that developed previously by us for the preparation of **2** (Scheme 1).[10,11] The cyclitol moiety is constructed starting from D-glucosamine by forming a carbon–carbon bond between C-1 and C-5, in parallel to the biosynthetic outcome. This key synthetic transformation consists of a double Swern oxidation followed by a samarium diiodide-promoted intramolecular pinacol coupling reaction in a one-pot sequence[12] performed on

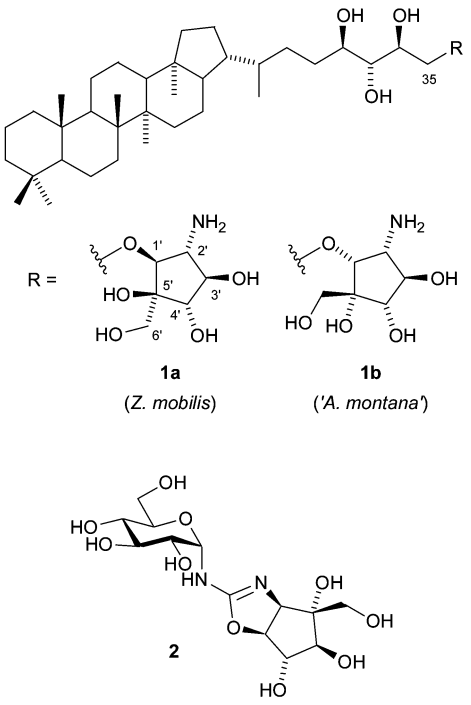
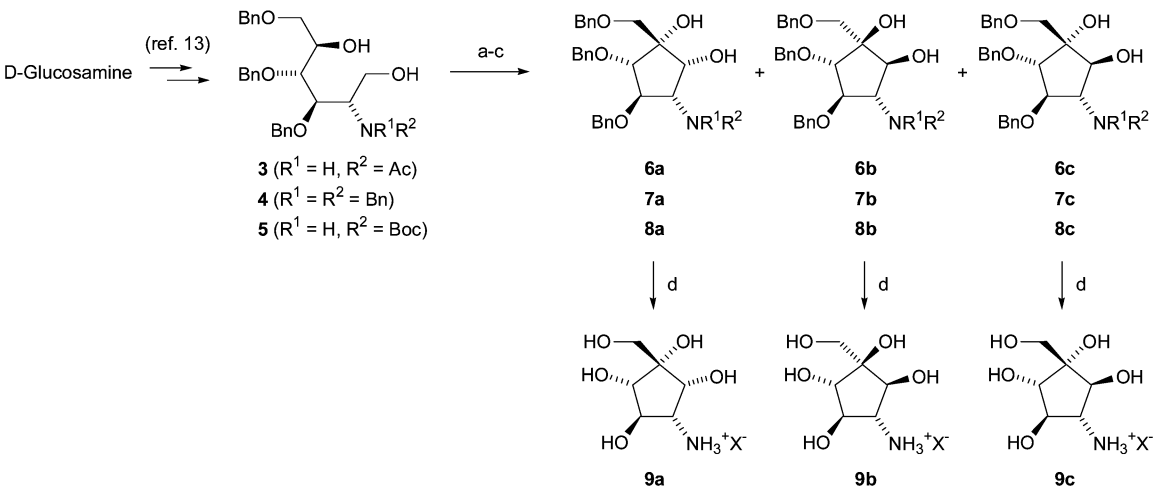


Fig. 1



Scheme 1 *Reagents and conditions*: (a) $(COCl)_2$, DMSO, THF, −60 °C. (b) Et_3N, −60 °C to rt. (c) SmI_2, THF/*t*BuOH, temp. (see Table 1). (d) **6a,b**: i. H_2, Pd/C, EtOH/EtOAc, rt. ii. 2 M aq. HCl, 80 °C. **7a–c**: H_2, Pd/C, EtOH/EtOAc/TFA, rt. **8a,b**: i. H_2, Pd/C, EtOH/EtOAc, rt. ii. 2 M aq HCl, rt.

DOI: 10.1039/b304576e

readily prepared[13] 1,5-diols **3**–**5**. Different amino-protecting groups (amide, carbamate and *N*,*N*-dialkylamino) were assayed to study their effect on the diastereoselectivity of the carbocyclization reaction. Table 1 shows the yields and diastereoselectivities obtained in each case. The carbocyclization process proceeded smoothly to provide the expected carbocycles as a mixture of diastereoisomers in good overall yield ranging from 70% to 82%. As anticipated,[10] a mixture of two *cis*-diols (**6a–8a** and **6b–8b**) was obtained as shown by their ready transformation into the corresponding isopropylidene acetals (acetone/$Me_2C(OMe)_2$, cat. PPTS (pyridinium *p*-toluenesulfonate), 92–96%). In the case of substrate **4**, with a *N*,*N*-dibenzylamino group, a minor *trans*-diastereoisomer (**7c**), which did not form an isopropylidene acetal, was also obtained. The diastereoselectivity of the cyclization was almost independent of the nature of the *N*-protecting groups (Table 1, entries 1, 2 and 5) and improved only slightly at lower temperatures (Table 1, entries 3–6). Complete deprotection by hydrogenolysis (compounds **7a–c**) followed by acid treatment (compounds **6a**, **6b**, **8a**, and **8b**) afforded the free aminocyclopentitols **9a–c**† in good yield as their corresponding ammonium salts. NOESY experiments performed on the isopropylidene acetal derivatives or on the free cyclitols allowed an unambiguous assignment of the stereochemistry of each carbocyclic diastereoisomer (Fig. 2).

Cyclopentitols **9a–c** were assayed as inhibitors against a panel of commercially available *exo*-glycosidases of different origin and specificity. Table 2 shows the IC_{50} values measured using a spectrophotometric method to follow the enzymatic hydrolysis of the corresponding *p*-nitrophenyl glycoside as substrates for the different enzymes. Cyclitol **9a** showed weak inhibitory activity for both glucosidases and galactosidases without any anomeric specificity, while cyclitols **9b** and **9c** showed no activity ($IC_{50} > 10^3$ M) for any of the enzymes assayed.

In summary, we have accomplished the first synthesis of the cyclopentitol units in bacterial hopanoids and investigated the possible biological activity of the free cyclitols as glycosidase inhibitors.

Financial support by the Ministry of Science and Technology of Spain (project BQU2000-1501-C02-01) is gratefully acknowledged.

Table 1 One-pot two-step carbocyclization of diols **3–5**.

Entry	Substrate	*T*/°C	Yield (%)[a]	Products (ratio)[b]
1	**3**	−50	74	**6a,b,c** (2 : 1 : 0)
2	**4**	−50	75	**7a,b,c** (6 : 3 : 2)
3	**5**	23	82	**8a,b,c** (2 : 1 : 0)
4	**5**	0	76	**8a,b,c** (2 : 1 : 0)
5	**5**	−50	82	**8a,b,c** (2.5 : 1 : 0)
6	**5**	−78	70	**8a,b,c** (3.5 : 1 : 0)

[a] Overall isolated yields. [b] Diastereoisomeric ratios were determined from the ^{1}H NMR of the crude reaction mixtures.

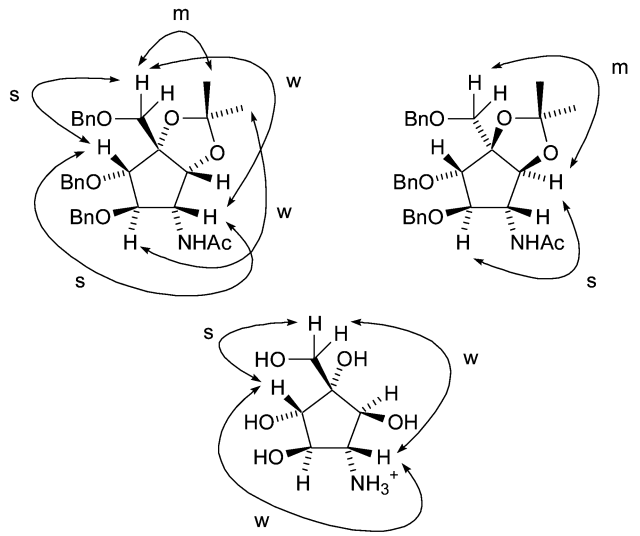


Fig. 2 Relative intensities of selected cross-peaks in NOESY spectra (s: strong; m: medium; w: weak).

Table 2 Inhibitory activity (IC_{50} values) of cyclitols **9a–c** against a panel of commercial exo-glycosidases.

	IC_{50}/M[a]		
Enzyme	**9a**	**9b**	**9c**
α-Glucosidase (baker's yeast)	6×10^{-4}	—[b]	—[b]
β-Glucosidase (almonds)	3×10^{-4}	—[b]	—[b]
α-Mannosidase (Jack beans)	—[b]	—[b]	—[b]
α-Galactosidase (coffee beans)	2×10^{-5}	—[b]	—[b]
β-Galactosidase (*E. coli*)	—[b]	—[b]	—[b]
β-Glucosidase (bovine liver)	2×10^{-4}	—[b]	—[b]

[a] Measured at 37 °C at the optimal pH described for each enzyme. [b] $IC_{50} > 10^{-3}$ M.

Notes and references

† *Selected characterization data:* **9a** (X = CF_3CO_2): $[\alpha]_D^{22}$ +3.3 (*c* 1.0, CH_3OH); ^{1}H NMR (300 MHz, CD_3OD) δ 4.21 (d, 1H, *J* = 7.4 Hz, H-1′), 4.05 (dd, 1H, *J* = 7.7, 6.4 Hz, H-3′), 3.73 (d, 1H, *J* = 7.7 Hz, H-4′), 3.49 (s, 2H, CH_2-6′), 3.26 (dd, 1H, *J* = 7.4, 6.4 Hz, H-2′); ^{13}C NMR (75 MHz, CD_3OD) δ 79.9, 79.1, 76.6, 66.2, 63.6, 58.1. **9b** (X = CF_3CO_2): $[\alpha]_D^{22}$ +6.2 (*c* 1.2, CH_3OH); ^{1}H NMR (300 MHz, CD_3OD) δ 3.97 (d, 1H, *J* = 9.5 Hz, H-1′), 3.82(d, 1H, *J* = 6.0 Hz, H-4′), 3.75 (dd, 1H, *J* = 9.0, 6.0 Hz, H-3′), 3.67 (d, 1H, *J* = 11.2 Hz, H-6′), 3.58 (d, 1H, *J* = 11.2 Hz, H-6′), 3.25 (t, 1H, *J* = 9.2 Hz, H-2′); ^{13}C NMR (75 MHz, CD_3OD) δ 84.1, 78.3, 77.5, 72.8, 64.7, 61.1. **9c** (X = Cl): $[\alpha]_D^{22}$ + 4.4 (*c* 1.0, CH_3OH); ^{1}H NMR (500 MHz, CD_3OD) δ 3.72 (d, 1H, *J* = 8.1 Hz, H-4′), 3.64 (d, 1H, *J* = 7.7 Hz, H-1′), 3.59 (s, 2H, CH_2-6), 3.56 (dd, 1H, *J* = 8.6, 8.1Hz, H-3′), 2.70 (dd, 1H, *J* = 8.6, 7.7 Hz, H-2′); ^{13}C NMR (75 MHz, CD_3OD) δ 82.8, 79.8, 78.2, 76.8, 65.1, 60.5.

1 G. Ourisson, P. Albrecht and M. Rohmer, *Pure Appl. Chem.*, 1979, **51**, 709.
2 G. Ourisson, M. Rohmer and K. Poralla, *Ann. Rev. Microbiol.*, 1987, **41**, 301.
3 M. Rohmer, *Nat. Prod. Rep.*, 1999, **16**, 565.
4 M. A. F. Herrmans, B. Neus and H. Sahm, *J. Bacteriol.*, 1991, **173**, 5592.
5 D. Herrmann, P. Bisseret, J. Connan and M. Rohmer, *Tetrahedron Lett.*, 1996, **37**, 1791.
6 J.-M. Renoux and M. Rohmer, *Eur. J. Biochem.*, 1985, **151**, 405.
7 S. Neunlist, P. Bisseret and M. Rohmer, *Eur. J. Biochem.*, 1988, **171**, 245.
8 D. Hermann, P. Bisseret, J. Connan and M. Rohmer, *Tetrahedron Lett.*, 1996, **37**, 1791.
9 S. P. Vicent, P. Sinaÿ and M. Rohmer, *Chem. Commun.*, 2003, 782; M. Rohmer, B. Sutter and H. Sahm, *J. Chem. Soc., Chem. Commun.*, 1989, 1471.
10 I. Storch de Gracia, H. Dietrich, S. Bono and J. L. Chiara, *J. Org. Chem.*, 1998, **63**, 5883.
11 I. Storch de Gracia, S. Bobo, M. D. Martín-Ortega and J. L. Chiara, *Org. Lett.*, 1999, **1**, 1705.
12 J. L. Chiara and M. Martín-Lomas, *Tetrahedron Lett.*, 1994, **35**, 2969.
13 The preparation of 1,5-diols **3–5** will be described elsewhere.