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SYNTHESIS FROM D-MANNOSE OF 1,4-DIDEOXY-1,4-IMINO-L-RIBITOL AND OF THE α-MANNOSIDASE INHIBITOR 1,4-DIDEOXY-1,4-IMINO-D-TALITOL

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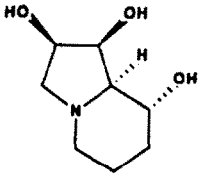
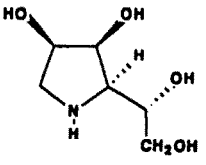
The syntheses of 1,4-dideoxy-1,4-imino-L-ribitol and of 1,4-dideoxy-1,4-imino-D-talitol from D-mannose are described. 1,4-Dideoxy-1,4-imino-D-talitol is a specific and competitive inhibitor of human liver α-mannosidase *in vitro* and also blocks the lysosomal catabolism of asparigine-linked glycans of glycoproteins *in vivo*.

Swainsonine (1) has potential as an agent for stimulation of the immune response[1] and for the prevention of the metastasis of cancer.[2] The biological effects of swainsonine on viruses and on animal and plant cells arise from inhibition of a mannosidase of glycoprotein processing.[3] The open chain analogue of swainsonine, 1,4-dideoxy-1,4-imino-D-mannitol (2), is the azafuranose analogue of mannose and is a powerful and specific inhibitor of several mannosidases,[4] including glycoprotein processing mannosidases;[5] also, (2) as well as other hydroxylated pyrrolidines has been shown to possess immunostimulating activity.[6] There has been considerable interest in the synthesis[7] and enzyme inhibitory properties[8] of stereoisomers of swainsonine; the 8a epimer of swainsonine (3) has been shown to be almost as potent an inhibitor of a human α-mannosidase as is swainsonine.[9] This paper describes the synthesis from diacetone mannose (6) of 1,4-dideoxy-1,4-imino-D-talitol (4), the open chain analogue of 8a-epi-swainsonine, and a preliminary evaluation of (4) as an inhibitor of α-mannosidase *in vitro* and *in vivo*; the synthesis of 1,4-dideoxy-1,4-imino-L-ribitol (5) is also reported. Recently, alternative syntheses of (4) and (5) from D-mannose, together with their evaluation as immunostimulatory agents, have been reported.[6]



(1)



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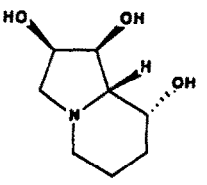

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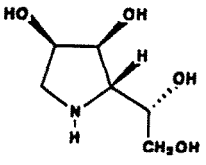
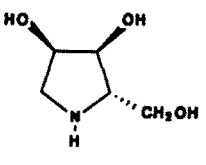


(4)



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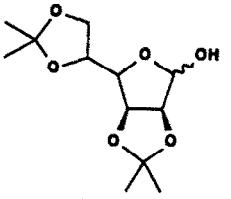
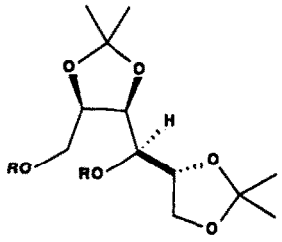
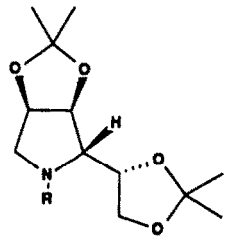


(6)

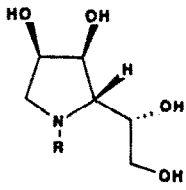
Diacetone mannose (6) was converted into 1,2:4,5-di-O-isopropylidene-D-mannitol (7) in 81% yield as previously described.[10,11] Esterification of the diol (7) with methanesulphonyl chloride gave the dimesylate (8) which with benzylamine gave the protected pyrrolidine (9) [71% yield - 57% overall from D-mannose]; the ease of this cyclisation is presumably due to the relative ease of initial nucleophilic attack at the primary mesylate since a similar cyclisation to a 2,5-dideoxy-2,5-iminohexitol reported by Shing[12] required the use of a bistrifluoromethane sulphonate. The N-benzyl group in (9) was removed by hydrogenolysis in the presence of 10% palladium on charcoal to give the diacetonide (10) [95% yield] which was treated with aqueous trifluoroacetic acid to afford 1,4-dideoxy-1,4-imino-D-talitol (4) most easily handled as the crystalline hydrochloride [95% yield - 52% overall yield from D-mannose]. This synthesis of (4) from D-mannose in which the pyrrolidine ring is formed directly from the bis-mesylate is shorter than the sequential introduction of leaving groups on C-1 and C-4 of the protected mannitol previously reported.[6]



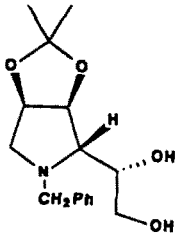
(7) R = H
(8) R = SO_2Me



(9) R = CH_2Ph
(10) R = H



(4) R = H
(11) R = CH_2Ph



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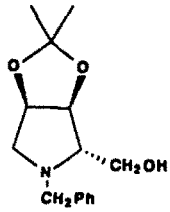
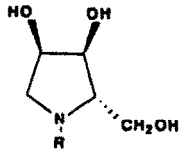


(13)



(5) R = H
(14) R = CH_2Ph

Whereas hydrolysis of the fully protected amine (9) by aqueous trifluoroacetic acid caused efficient removal of both isopropylidene protecting groups to give (11), treatment of (9) with aqueous acetic acid resulted in selective hydrolysis of the side chain acetonide to give the diol (12) in quantitative yield. Confirmation that the ring closure in the formation of the pyrrolidine (9) had proceeded with inversion of configuration at C-4 was obtained by the conversion of the diol (12) to 1,4-dideoxy-1,4-imino-L-ribitol (5). Oxidative cleavage of the diol (12) by treatment with sodium periodate, followed by reduction of the resulting aldehyde by sodium borohydride gave the protected imino-L-ribitol (13) [78% yield]. Hydrolysis of the acetonide (13) gave the N-benzyl derivative (14). Removal of the N-benzyl protecting group from (13) followed by trifluoroacetic acid hydrolysis gave 1,4-dideoxy-1,4-imino-L-ribitol (5). As in the case of other hydroxylated pyrrolidines,[13] the hydrochloride of (5) is easier to handle and to crystallise than is the free base. The spectroscopic and analytical data for the L-ribitol derivatives (5), (13) and (14) were identical, save for the specific rotation, to those properties recorded for the corresponding D-ribitol compounds prepared from D-gulonolactone.[14]

1,4-Dideoxy-1,4-imino-D-talitol (4) is a specific competitive inhibitor of human liver lysosomal α-mannosidase with a K_i of 1.2×10^{-4} M at pH 4 (the optimum pH of the enzyme).[15] None of ten other human liver lysosomal glycosidases, including β-mannosidase, was inhibited by more than 4% by iminotalitol (4) at a concentration of 1 mM; at this concentration, 80% inhibition of α-mannosidase activity was observed. The inhibition of lysosomal α-mannosidase increased with pH over the range pH 3 to pH 6 as has been observed for other α-mannosidase inhibitors.[16] Human liver neutral α-mannosidase, which had been separated from the lysosomal enzyme by affinity chromatography on concanavalin A-sepharose,[17] was also inhibited by (4) but only by 50% at pH 6.5 (the pH optimum of the enzyme) with 1 mM concentrations of the inhibitor. In order to investigate whether (4) inhibits lysosomal and other α-mannosidases *in vivo* as well as *in vitro*, normal human fibroblasts were grown in culture medium containing 1 mM (4) for seven days.[18] Neutral oligosaccharides were extracted from harvested cells and analysed by TLC. The concentration of oligosaccharides in the cells grown in the presence of the iminotalitol (4) was much higher than in control cells grown in the absence of the inhibitor. This indicates that (4) has blocked the lysosomal catabolism of asparagine-linked glycans of glycoproteins. The pattern of the storage products was compared with those in genetically and swainsonine-induced mannosidosis in fibroblasts to deduce which cellular α-mannosidases had been inhibited. The predominant oligosaccharides had the composition $Man_2GlcNAc$ and $Man_3GlcNAc$, the same as those found in genetic mannosidosis. Therefore, it is deduced that these oligosaccharides arise by incomplete catabolism of complex N-glycans, due to inhibition of lysosomal α-mannosidase. The absence of oligosaccharides of composition $Man_9GlcNAc$ and $Man_5GlcNAc$ indicates that (4) is not a strong inhibitor *in vivo* of Golgi α-mannosidase I, or of endoplasmic reticulum α-mannosidase, or of Golgi α-mannosidase II. These observations are consistent with those of the *in vitro* specificity of the iminotalitol (4).

In summary this paper describes the synthesis of 1,4-dideoxy-1,4-imino-D-talitol (4) and of 1,4-dideoxy-1,4-imino-L-ribitol (5) from D-mannose. It is becoming clear that several diastereoisomers of swainsonine and the corresponding open chain analogues may prove useful in dissecting the pathways for the post-translational modification and catabolism of glycoproteins. Potential antiviral properties of the hydroxylated pyrrolidines are currently being evaluated and will be published elsewhere.

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Experimental

M.p.s were recorded on a Kofler block. Infra red spectra were recorded on a Perkin-Elmer 297 spectrophotometer. ^1H NMR spectra were run at 300 MHz on a Bruker WH 300 spectrometer (500 MHz on a Bruker AM 500 spectrometer); ^{13}C NMR spectra were recorded on a Bruker AM 250 (at 62.9 MHz) or on a Bruker AM 500 (at 125 MHz) spectrometer. For ^{13}C NMR spectra in D_2O, 1,4-dioxane (δ 67.6) was used as the internal standard. Mass spectra were recorded on VG Micromass ZAB 1F or MM 30F spectrometers; in order to obtain satisfactory mass spectra for the majority of compounds reported in this paper it was necessary to use ACE, DCI or FAB techniques. Microanalyses were performed by the microanalytical services of the Dyson Perrins Laboratory. TLC was performed on glass plates coated with silia gel Blend 41, and compounds were visualised with a spray of 5% v/v sulphuric acid in methanol or a solution of 5% dodecamolybdophosphoric acid in methanol. Flash chromatography was carried out using Merck Kieselgel 60, 230-400 mesh. Unless otherwise stated, all

organic solvents were dried before use; tetrahydrofuran was distilled from a solution dried with sodium in the presence of benzophenone under dry nitrogen. D-Mannose was obtained from the Sigma Chemical Company and was used without purification.

1,2:4,5-Di-O-isopropylidene-D-mannitol (7). D-Mannose was converted to the diacetonide (6) as previously described.[10] Concentrated sulphuric acid (2.8 ml) was added to a vigorously stirred suspension of D-mannose (12 g, 66.6 mmol) in acetone (200 ml) at room temperature. After 3 h, the reaction mixture was neutralised with anhydrous sodium carbonate (12 g), filtered and evaporated to give, after recrystallisation from ethyl acetate:hexane (1:5), 2,3:5,6-di-O-isopropylidene-D-mannofuranose (6), (14 g, 81%), m.p. 124°-125°C [lit.[10] m.p. 122°-123°C]. The acetonide (6) (3.9 g, 15.0 mmol) was added to a stirred solution of sodium borohydride (567 mg, 15.0 mmol) in ethanol (30 ml) at room temperature. After 30 min, ammonium chloride was added to the reaction mixture in order to decompose the excess hydride; the solvent was then removed and the residue purified by flash chromatography (hexane:ethyl acetate, 2:3) to afford 1,2:4,5-di-O-isopropylidene-D-mannitol (7), [3.9 g, 81% overall yield from D-mannose], m.p. 47°-49°C, $[\alpha]_D^{20}$ -9.2° (c, 1.05 in $CHCl_3$) [lit.[11] m.p. 47°-48°C]. ν_{max} (KBr) 3600-3100 cm^{-1}. ^1H NMR ($CDCl_3$) δ 1.37 (3H, s, Me), 1.41 (3H, s, Me), 1.42 (3H, s, Me), 1.53 (3H, s, Me), 3.60 (1H, d, J 6.3 Hz), 3.84 (1H, dd, J 4.4 and 12.1 Hz), 3.91 (1H, dd, J 4.4 and 12.1 Hz), 4.09 (3H, m), 4.32 (1H, m), 4.40 (1H, dd, J 1.5 and 7.2 Hz). m/z (CI, NH_3): 263 ($M+H^+$, 100%), 247 ($M-Me^+$, 60%). (Found C, 54.67; H, 8.21. $C_{12}H_{22}O_6$ requires C, 54.96; H, 8.40%).

1,4-Bis(O-methanesulphonyl)-2,3:5,6-di-O-isopropylidene-D-mannitol (8). To a stirred solution of the diol (7) (3.0 g, 11.5 mmol) in pyridine (20 ml) at 0°C, methanesulphonyl chloride (3.5 ml, 45.8 mmol) together with a catalytic amount of 4-dimethylaminopyridine (0.28 g, 2.3 mmol) were added. After 2 h, pyridine was removed in vacuo; the residue was dissolved in chloroform (100 ml) and the organic solution was then washed with water (2 x 100 ml) and dried (magnesium sulphate). Evaporation of the solvent gave a residue which was purified by flash chromatography (hexane:ethyl acetate, 1:2) to give 1,4-bis(O-methanesulphonyl)-2,3:5,6-di-O-isopropylidene-D-mannitol (8), (4.8 g, quantitative yield), colourless oil, $[\alpha]_D^{20}$ +30.7° (c, 2.12 in $CHCl_3$), ν_{max} (film) 1350 cm^{-1}. ^1H NMR ($CDCl_3$) δ 1.37 (3H, s, Me), 1.41 (3H, s, Me), 1.46 (3H, s, Me), 1.54 (3H, s, Me), 3.10 (3H, s, $MeSO_2$), 3.15 (3H, s, $MeSO_2$), 4.03 (1H, dd, J 5.9 and 7.3 Hz), 4.16 (2H, m), 4.43 (4H, m), 4.78 (1H, dd, J 7.2 and 7.8 Hz). m/z (DCI, NH_3): 436 ($M+NH_4^+$, 100%), 419 ($M+H^+$, 65%).

N-Benzyl-1,4-dideoxy-2,3:5,6-di-O-isopropylidene-1,4-imino-D-talitol (9). The dimesyl compound (8) in benzylamine (10 ml) was stirred at 60°-70°C for 60 h. The reaction mixture was then partitioned between brine (50 ml) and chloroform (120 ml). The organic layer was then separated, washed with water (2 x 100 ml), dried (magnesium sulphate) and the solvent removed to give, after purification by flash chromatography (ether:hexane, 2:3), N-benzyl-1,4-dideoxy-2,3:5,6-di-O-isopropylidene-1,4-imino-D-talitol (9), (2.7 g, 71%), pale yellow oil, $[\alpha]_D^{20}$ +60.1° (c, 1.65 in $CHCl_3$), ^1H NMR ($CDCl_3$) δ 1.32 (3H, s, Me), 1.36 (3H, s, Me), 1.45 (3H, s, Me), 1.53 (3H, s, Me), 2.53 (1H, dd, H-1, $J_{1,1'}$ 10.1 Hz, $J_{1,2}$ 4.5 Hz), 2.94 (1H, dd, H-4, $J_{3,4}$ 3.8 Hz, $J_{4,5}$ 6.2 Hz), 3.20 (1H, dd, H-1', $J_{1',2}$ 6.0 Hz), 3.62 (1H of AB, $PhCH_2$, J_{AB} 13.3 Hz), 3.85 (1H, dd, H-6, $J_{6,6'}$ 8.3 Hz, $J_{5,6}$ 7.5 Hz), 4.02 (1H, dd, H-6', $J_{5,6'}$ 6.5 Hz), 4.21 (1H, q, H-5), 4.23 (1H of AB, $PhCH_2$), 4.41 (1H, dd, H-3, $J_{2,3}$ 6.8 Hz), 4.61 (1H, dt, H-2), 7.28 (5H, m, ArH). ^{13}C NMR ($CDCl_3$) δ 24.95 (q), 25.31 (q), 26.51 (q), 27.27 (q), 59.12 (t), 66.39 (t), 70.61 (d), 77.21 (d), 78.86

(d), 82.26 (d), 109.44 (s), 112.85 (s), 126.93 (d), 128.19 (d), 128.76 (d), 139.07 (s). m/z (ACE, NH_3): 334 ($M+H^+$, 100%), 232 (97%). (Found C, 68.80; H, 8.55; N, 4.51. $C_{19}H_{27}NO_4$ requires C, 68.47; H, 8.11; N, 4.20%).

1,4-Dideoxy-2,3:5,6-di-O-isopropylidene-1,4-imino-D-talitol (10). The cyclised tertiary amine (9) (336 mg, 1.01 mmol) in ethanol (20 ml) was stirred under an atmosphere of hydrogen in the presence of 10% palladium on charcoal (150 mg) at room temperature for 2 h. The reaction mixture was filtered through celite to remove the catalyst and the solvent then removed to give, after purification by flash chromatography (ethyl acetate), 1,4-dideoxy-2,3:5,6-di-O-isopropylidene-1,4-imino-D-talitol (10), (233 mg, 95%), m.p. 60°C, $[\alpha]_D^{20}$ -44.1° (c, 0.37 in $CHCl_3$), ν_{max} (KBr) 3320 (m, NH) cm^{-1}. ^1H NMR ($CDCl_3$) δ 1.32 (3H, s, Me), 1.34 (3H, s, Me), 1.42 (3H, s, Me), 1.48 (3H, s, Me), 3.08 (2H, m, H-1, H-1'), 3.16 (1H, dd, H-4, J 1.2 and 5.6Hz), 3.84 (1H, t, H-5), 4.05 (2H, m, H-6, H-6'), 4.49 (1H, dd, H-3, J 1.5 and 5.8 Hz), 4.73 (1H, m, H-2). m/z (DCI, NH_3): 244 ($M+H^+$, 100%), 142 (33%). This compound is rather unstable in air.

1,4-Dideoxy-1,4-imino-D-talitol (4). The acetonide (10) (233 mg, 0.96 mmol) in 50% aqueous trifluoroacetic acid (10 ml) was stirred at room temperature for 15 h. The solvent was removed; the resulting trifluoroacetate salt was neutralised with dilute aqueous sodium hydroxide and purified by ion exchange chromatography (Dowex 50 x, 8-100, H^+ form, eluted with 0.5 molar aqueous ammonium hydroxide) to give 1,4-dideoxy-1,4-imino-D-talitol (4), oil [^1H NMR (D_2O) δ 2.62 (1H, dd, H-1, $J_{1,1'}$ 12.5 Hz, $J_{1,2}$ 3.4 Hz), 2.78 (1H, dd, H-4, $J_{3,4}$ 7.9 Hz, $J_{4,5}$ 4.2 Hz), 3.02 (1H, dd, H-1', $J_{1',2}$ 5.1 Hz), 3.40 (1H, dd, H-6, $J_{6,6'}$ 11.8 Hz, $J_{5,6}$ 7.7 Hz), 3.51 (1H, dd, H-6', $J_{5,6'}$ 4.1 Hz), 3.62 (1H, m, H-5), 3.78 (1H, dd, H-3, $J_{2,3}$ 5.2 Hz), 3.95 (1H, dt, H-2)]; the free base (4) was dissolved in water (5 ml) and the solution was adjusted to pH 4 with dilute aqueous hydrochloric acid to afford, after freeze drying, 1,4-dideoxy-1,4-imino-D-talitol hydrochloride, (181 mg, 95%), m.p. 144°-145°C, $[\alpha]_D^{20}$ -56.3° (c, 0.41 in H_2O), ν_{max} (KBr) 3600-2500 (br, NH and OH) cm^{-1}. ^1H NMR (D_2O) δ 3.19 (1H, dd, H-1, $J_{1,1'}$ 13.0 Hz, $J_{1,2}$ 1.6 Hz), 3.30 (1H, dd, H-1', $J_{1',2}$ 3.8 Hz), 3.41 (1H, dd, H-4, $J_{3,4}$ 8.9 Hz, $J_{4,5}$ 4.3 Hz), 3.50 (1H, dd, H-6, $J_{6,6'}$ 12.1 Hz, $J_{5,6}$ 4.9 Hz), 3.61 (1H, dd, H-6', $J_{5,6'}$ 3.7 Hz), 3.84 (1H, q, H-5), 4.10 (1H, dd, H-3, $J_{2,3}$ 4.1 Hz), 4.20 (1H, dt, H-2). ^{13}C NMR (D_2O) δ 50.90 (t), 62.80 (d), 64.41 (t), 68.79 (d), 70.27 (d), 73.15 (d). m/z (FAB, Gly/MeOH): 164 ($M+H^+$, 100%). (Found C, 36.04; H, 7.04; N, 6.96. $C_6H_{13}NO_4$.HCl requires C, 36.09; H, 7.02; N, 7.02%).

N-Benzyl-1,4-dideoxy-1,4-imino-D-talitol Hydrochloride (11). The fully protected amine (9) (210 mg, 0.63 mmol) was stirred at room temperature in 50% aqueous trifluoroacetic acid (10 ml) for 24 h. The solvent was removed and the residue purified by ion exchange chromatography (Dowex 50 x, 8-100, H^+ form, eluted with 0.5 molar aqueous ammonium hydroxide) to give N-benzyl-1,4-dideoxy-1,4-imino-D-talitol (11) as a syrup; the free base (11) was dissolved in water (5 ml) and the solution was adjusted to pH 4 with dilute aqueous hydrochloric acid to afford, after freeze drying, N-benzyl-1,4-dideoxy-1,4-imino-D-talitol hydrochloride, (164 mg, 90%), a very hygroscopic solid, $[\alpha]_D^{20}$ -10.1° (c, 0.94 in H_2O), ^1H NMR (D_2O) δ 3.20 (1H, dd, H-1, $J_{1,1'}$ 12.9 Hz, $J_{1,2}$ 4.2 Hz), 3.28 (1H, dd, H-1', $J_{1',2}$ 3.9 Hz), 3.44 (1H, dd, H-6, $J_{6,6'}$ 12.3 Hz, $J_{5,6}$ 4.9 Hz), 3.53 (2H, m, H-4, H-6'), 3.80 (1H, m, H-5), 4.11 (1H, dd, H-3, $J_{2,3}$ 4.2 Hz, $J_{3,4}$ 6.3 Hz), 4.21 (1H, q, H-2), 4.26 (1H of AB, $PhCH_2$, J_{AB} 13.3 Hz), 7.3 (5H, m, ArH); the other benzylic signal was suppressed with the HOD signal. ^{13}C NMR (D_2O) δ 56.03 (t), 63.63 (t), 64.48 (t), 70.64 (d), 70.65 (d),

73.35 (d), 73.89 (d), 130.37 (d), 130.88 (s), 131.32 (d), 132.08 (d). m/z (DCI, NH_3): 254 ($M+H^+$, 100%), 192 (60%).

N-Benzyl-1,4-dideoxy-1,4-imino-2,3-O-isopropylidene-D-talitol (12). The fully protected amine (9) (1.22 g, 3.66 mmol) was stirred at 50°C in 80% aqueous acetic acid (20 ml) for 36 h. The solvent was removed and the residue purified by flash chromatography (ethyl acetate) to give N-benzyl-1,4-dideoxy-1,4-imino-2,3-O-isopropylidene-D-talitol (12), (1.10 g, quantitative yield), pale yellow oil, $[\alpha]_D^{20}$ -15.2° (c, 1.22 in $CHCl_3$), ν_{max} (film) 3600-3240 (br, OH) cm^{-1}. ^1H NMR ($CDCl_3$) δ 1.34 (3H, s, Me), 1.63 (3H, s, Me), 3.11 (2H, m), 3.38 (1H, d, J 6.6 Hz), 3.45 (1H, m), 3.53 (1H, dd, J 3.4 and 11.7 Hz), 3.73 (1H, dd, J 3.4 and 11.7 Hz), 4.13 (1H of AB, $PhCH_2$, J_{AB} 12.5 Hz), 4.19 (1H of AB, $PhCH_2$), 4.69 (1H, d, J 6.1 Hz), 4.81 (1H, m), 7.33 (5H, m, ArH). ^{13}C NMR ($CDCl_3$) δ 23.47 (q), 26.63 (q), 57.69 (t), 61.73 (t), 64.31 (t), 69.39 (d), 72.01 (d), 82.24 (d), 84.27 (d), 111.76 (s), 127.54 (d), 128.55 (d), 129.31 (d), 138.45 (s). m/z (DCI, NH_3): 294 ($M+H^+$, 100%), 232 (33%).

N-Benzyl-1,4-dideoxy-1,4-imino-2,3-O-isopropylidene-L-ribitol (13). Sodium periodate (1.75 g, 8.19 mmol) was added to a stirred solution of N-benzyl-1,4-dideoxy-1,4-imino-2,3-O-isopropylidene-D-talitol (12) (800 mg, 2.73 mmol) in 80% aqueous ethanol (20 ml) at room temperature; when tlc showed all the starting material had been consumed after 30 min, the reaction mixture was treated with sodium borohydride (207 mg, 5.46 mmol) and stirred for a further 60 min. The excess hydride was then decomposed by addition of excess solid ammonium chloride and the solvent removed to give, after purification by flash chromatography (ethyl acetate:hexane, 8:3), N-benzyl-1,4-dideoxy-1,4-imino-2,3-isopropylidene-L-ribitol (13), (557 mg, 78%), yellow oil, $[\alpha]_D^{20}$ +45.7° (c, 1.0 in $CHCl_3$), ν_{max} (film) 3600-3200 (br, OH) cm^{-1}. ^1H NMR ($CDCl_3$) δ 1.33 (3H, s, Me), 1.55 (3H, s, Me), 2.65 (1H, dd, H-1, $J_{1,1'}$ 10.7 Hz, $J_{1,2}$ 3.8 Hz), 2.98 (1H, br dd, H-4), 3.22 (1H, dd, H-1', $J_{1',2}$ 5.7 Hz), 3.58 (1H, dd, H-5, $J_{5,5'}$ 11.2 Hz, $J_{4,5}$ 3.6 Hz), 3.61 (1H of AB, $PhCH_2$, J_{AB} 13.0 Hz), 3.71 (1H, dd, H-5', $J_{4,5'}$ 3.8 Hz), 3.99 (1H of AB, $PhCH_2$), 4.61 (2H, m, H-2, H-3), 7.31 (5H, m, ArH). ^{13}C NMR ($CDCl_3$) δ 24.80 (q), 27.16 (q), 58.11 (t), 58.43 (t), 59.54 (t), 69.88 (d), 78.42 (d), 82.49 (d), 112.61 (s), 127.26 (d), 128.39 (d), 128.64 (d), 138.27 (s). m/z (ACE, NH_3): 264 ($M+H^+$, 100%), 232 (41%). (Found C, 68.14; H, 8.27; N, 5.12. $C_{15}H_{21}NO_3$ requires C, 68.44; H, 7.98; N, 5.32%). The spectroscopic properties of (13) (with the exception of specific rotation) are identical to those of the D-enantiomer, N-benzyl-1,4-dideoxy-1,4-imino-2,3-O-isopropylidene-D-ribitol reported in the accompanying paper.[14]

1,4-Dideoxy-1,4-imino-L-ribitol (5). The protected ribitol (13) (257 mg, 0.98 mmol) in ethanol (10 ml) was stirred under an atmosphere of hydrogen in the presence of catalyst of 10% palladium on charcoal (120 mg) at room temperature for 2h. The reaction mixture was filtered through celite and the solvent removed; the crude product [^1H NMR showed that the benzyl group had been completely removed] was dissolved in 50% aqueous trifluoroacetic acid (6 ml) and the solution was allowed to stand at room temperature for 24 h. The solvent was removed; the resulting trifluoroacetate salt was neutralised with dilute aqueous sodium hydroxide and purified by ion exchange chromatography (Dowex 50 x, 8-100, H^+ form, eluted with 0.5 molar aqueous ammonium hydroxide) to give 1,4-dideoxy-1,4-imino-L-ribitol (5), as a hygroscopic yellow solid, [^1H NMR (D_2O) δ 2.58 (1H, dd, H-1, $J_{1,1'}$ 12.4 Hz, $J_{1,2}$ 3.9 Hz), 2.84 (1H, m, H-4), 2.94 (1H, dd, H-1', $J_{1',2}$ 5.3 Hz), 3.40 (1H, dd, H-5 $J_{5,5'}$ 11.6 Hz, $J_{4,5}$ 6.0 Hz), 3.51 (1H, dd, H-5', $J_{4,5'}$ 4.3 Hz), 3.64 (1H, dd, H-3 $J_{2,3}$ 5.2 Hz, $J_{3,4}$ 7.3 Hz), 3.92 (1H, dt, H-2).]; the free base (5) was dissolved in water (5 ml) and the solution was adjusted to pH 4 with dilute aqueous hydrochlori

acid to afford, after freeze drying, 1,4-dideoxy-1,4-imino-L-ribitol hydrochloride, (165 mg, 76%), m.p. 126°-131°C, $[\alpha]_D^{20}$ -59.0° (c, 0.59 in H_2O), ν_{max} (film) 3600-2800 (br, NH andOH) cm^{-1}. ^1H NMR (D_2O) δ 3.20 (1H, dd, H-1, $J_{1,1'}$ 13.0 Hz, $J_{1,2}$ 2.0 Hz), 3.33 (1H, dd, H-1', $J_{1',2}$ 4.0 Hz), 3.46 (1H, m, H-4), 3.66 (1H, dd, H-5, $J_{5,5'}$ 12.6 Hz, $J_{4,5}$ 6.0 Hz), 3.81 (1H, dd, H-5', $J_{4,5'}$ 3.5 Hz), 4.04 (1H, dd, H-3, $J_{2,3}$ 4.1 Hz, $J_{3,4}$ 8.5 Hz), 4.22 (1H, dt, H-2). ^{13}C NMR (D_2O) δ 50.63 (t), 59.01 (t), 62.80 (d), 70.42 (d), 72.17 (d). m/z (DCI, NH_3): 134 ($M+H^+$, 100%), 102 (18%). The spectroscopic properties of the imino-L-ribitol (5) (with the exception of specific rotation) are identical to those of the D-enantiomer, 1,4-dideoxy-1,4-imino-D-ribitol hydrochloride reported in the accompanying paper.[14]

N-Benzyl-1,4-dideoxy-1,4-imino-L-ribitol (14). The acetonide (13) (100 mg, 0.38 mmol) in 50% aqueous trifluoroacetic acid (5 ml) was stirred at room temperature for 20 h. The solvent was removed; the resulting trifluoroacetate salt was neutralised with dilute aqueous sodium hydroxide and purified by ion exchange chromatography (Dowex 50 x, 8-100, H^+ form, eluted with 0.5 molar aqueous ammonium hydroxide) to give N-benzyl-1,4-dideoxy-1,4-imino-L-ribitol (14), a very hygroscopic solid, $[\alpha]_D^{20}$ +33.0° (c, 0.32 in H_2O), ^1H NMR (D_2O) δ 2.37 (1H, dd, H-1, $J_{1,1'}$ 10.0 Hz, $J_{1,2}$ 8.0 Hz), 2.59 (1H, q, H-4), 2.87 (1H, dd, H-1', $J_{1',2}$ 6.0 Hz), 3.35 (2H, m, H-5), 3.44 (1H of AB, $PhCH_2$, J_{AB} 12.4 Hz), 3.78 (2H, m, H-3 and 1H of $PhCH_2$), 3.88 (1H, dt, H-2), 7.2 (5H, m, ArH). ^{13}C NMR (D_2O) δ 57.66 (t), 60.40 (t), 62.27 (t), 70.03 (d), 71.51 (d), 73.86 (d), 128.82 (d), 129.61 (d), 131.04 (d), 138.00 (s). m/z (ACE, NH_3): 224 ($M+H^+$, 100%), 192 (83%). The spectroscopic properties of the N-benzylimino-L-ribitol (14) (with the exception of specific rotation) are identical to those of the D-enantiomer, N-benzyl-1,4-dideoxy-1,4-imino-D-ribitol reported in the accompanying paper.[14]

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