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Higher Acyclic Nitrogen Containing Deoxy Sugar Derivatives: A New Lead in the Generation of Antimycobacterial Chemotherapeutics†

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Abstract—Syntheses of higher acyclic nitrogen containing deoxy sugar derivatives via nitroaldol reaction of different nitroalkanes with 2,3-dideoxy-α,β-unsaturated aldehydo sugars obtained from glycals namely acetylated glucal and galactal and their in vitro antimycobacterial activity are presented. © 2002 Elsevier Science Ltd. All rights reserved.

Introduction

Mycobacterium tuberculosis is directly responsible for more human deaths than any other single infectious agent.[1] There are 8 million new infections and 3 million deaths from TB every year; it is expected that in the next decade 90 million new cases will occur, resulting in 30 million deaths.[2] The mycobacterial cell wall is the primary permeability barrier responsible for the intrinsic resistance to many hydrophobic antibiotic substances.[3,4] A survey of literature[5] reveals that frontline antitubercular chemotherapeutic agents interfere with the biosynthesis of the cell wall of *M. tuberculosis* though their mechanism of action is not known completely. A close inspection of the mycobacterial cell wall indicates a dense network of crosslinked sugar residues esterified with mycolic acid at their ends.[6] This complex cell wall structure of *M. tuberculosis* contribute to its defence against foreign bodies. The complex network of sugar residues in the mycobacterial cell wall offers new targets for development of a drug against TB. Keeping in mind the importance of these sugar residues present in the cell wall of *M. tuberculosis*, some reports of interference by sugar derivatives, either in the form of iminoalditols[7] or as an appendage to known drugs,[8–10] have come up. With the resurgence of multidrug resistant *Mycobacterium*, the present therapy is proving ineffective and there is a need to explore completely new chemical entities which can work as antimycobacterial agents. With reference to the structure of ethambutol which being a long chain acyclic molecule is very flexible and has many energetically readily accessible conformations likely to be populated to sit well upon the arabinan site of action,[10] it could be presumed that nitrogen containing alkanols should work as antimycobacterial agents. Since syntheses of polyhydroxylated nitro or amino alkanes involve the generation of a number of chiral centers and a convenient strategy demands the possibility of preparing them from the compounds with well defined stereochemistry like sugars. There are reports of syntheses of acyclic amino alditols in the literature but, to date, no effort has been made towards assessing their property as antimycobacterial agents. To the best of our knowledge, the present communication is the first report that describes the syntheses of certain acyclic nitro and amino deoxy alditols from sugars and their bioevaluation as antimycobacterial agents.

Chemistry

Previously we reported the syntheses of higher acyclic nitrogen containing deoxy sugar derivatives by nitroaldol reaction (Henry reaction) of nitromethane with

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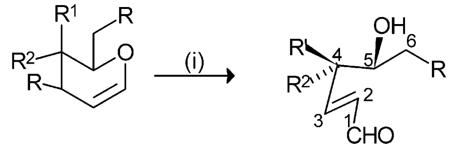
4,5,6-tri-*O*-acetyl-2,3-dideoxy-*aldehydo*-D-*erythro*-hex-2-enose.[11] This strategy was extended to another aldehydo sugar obtained from galactal and different nitroalkanes like nitromethane and nitroethane. The standardized protocol was followed for the syntheses of prototype compounds. The synthesis was initiated with (2*E*)-4,5,6-tri-*O*-acetyl-2,3-dideoxy-*aldehydo*-hex-2-enose (**5** and **6**), which was easily prepared from 3,4,6-tri-*O*-acetyl-D-glycal (**1** and **2**)[12] (Scheme 1).

Reaction of nitroalkanes with **5** and **6** in the presence of triethylamine yielded nitroaldols **7**–**10** as diastereomeric mixture (Scheme 2).

Subsequently, the acetylation of nitroaldols **7**–**10** furnished the conjugated derivatives **11**–**14** (Scheme 3). The *E* stereochemistry at the newly formed double bond was confirmed by nOe difference study.

Amino alkanols **15**–**22** were obtained by hydrogenation of nitroaldols **7**–**10** (Scheme 4) and conjugated derivatives **11**–**14** (Scheme 5).

The yields of the aminoalkanols obtained from **11**–**14** were found to be poor owing to stronger adsorption of diene to the catalyst surface which led to a number of side products, therefore, an indirect procedure to obtain **19**–**22** was adopted by first synthesizing **23**–**26** which were then reduced by hydrogenation over 10% Pd/C to yield **19**–**22** (Scheme 6).



1. R = R^2 = OAc, R^1 = H
2. R = R^1 = OAc, R^2 = H

3. R = R^2 = OAc, R^1 = H
4. R = R^1 = OAc, R^2 = H

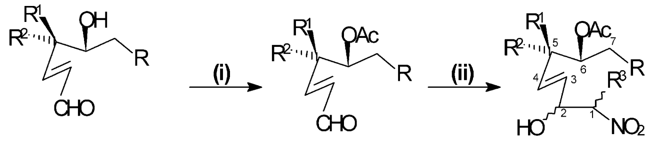
Scheme 1. (i) 0.01 N H_2SO_4, $HgSO_4$ (cat.), dioxane, 4 h, rt.

Results and Discussion

All the new compounds were evaluated for their antimycobacterial activity in an agar dilution assay system[13] with comparison to oxofloxacin as the standard drug. The results of the antimycobacterial assay carried out clearly indicated that the designed prototype molecules exhibited the desired activity. The structure of compounds along with the nature of substituents influenced the activity profile of these compounds. Here, it may be noted that the fluidity and permeability of the mycobacterial cell wall depends on the structure of the mycolic acid present in it and the difference in its structure leads to different sensitivity levels of mycobacterial species to lipophilic inhibitors.[4] The better antimycobacterial activity of protected disaccharides as compared to deprotected sugars was recently reported by Reynolds et al.[14] Thus, all acetyl protected compounds were evaluated for their antimycobacterial activity with the presumption that these compounds, after their passage through the cell wall, would be hydrolyzed by non-specific cell esterases resulting in the hydrophilic compounds.

The growth of mycobacterial cells was seen for complete inhibition at different concentration levels (6.25–100 µg/mL) of the synthesized compounds. Among the β-hydroxy nitro compounds **7**–**9**, only compound **8** showed activity up to 12.5 µg/mL and the rest did not show appreciable activity. These nitro compounds were converted to their corresponding amino alditols **15**–**18** to evaluate their antimycobacterial activity but were found to be inactive at all concentrations.

Similarly, when the compounds **11**–**14** were made deoxy at the β position with respect to the nitro group by

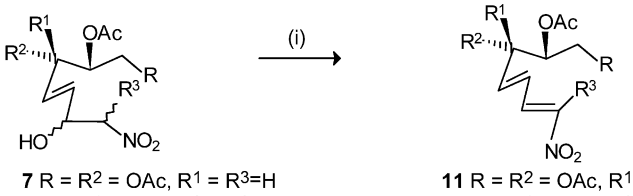


3 R = R^2 = OAc, R^1 = H
4 R = R^1 = OAc, R^2 = H

5 R = R^2 = OAc, R^1 = H
6 R = R^1 = OAc, R^2 = H

7 R = R^2 = OAc, R^1 = R^3=H
8 R = R^1 = OAc, R^2 = R^3=H
9 R = R^2 = OAc, R^1 = H, R^3=CH$_3$
10 R = R^1 = OAc, R^2 = H, R^3=CH$_3$

Scheme 2. (i) Ac_2O, Pyr, 10 h, 0–4 °C; (ii) Et_3N, $R^3CH_2NO_2$, N_2, 10 h, 0–4 °C, 48 h, rt.



7 R = R^2 = OAc, R^1 = R^3=H
8 R = R^1 = OAc, R^2 = R^3=H
9 R = R^2 = OAc, R^1 = H, R^3=CH$_3$
10 R = R^1 = OAc, R^2 = H, R^3=CH$_3$

11 R = R^2 = OAc, R^1 = R^3=H
12 R = R^1 = OAc, R^2 = R^3=H
13 R = R^2 = OAc, R^1 = H, R^3=CH$_3$
14 R = R^1 = OAc, R^2 = H, R^3=CH$_3$

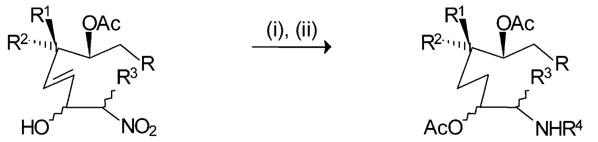
Scheme 3. (i) Ac_2O, Pyr, 10 h, 0–4 °C.

generating a double bond at α position (i.e., 1,3-diene system), no appreciable activity was seen in them except that **12** and **14** were active at 100 μg/mL. However, when one of the double bonds was removed to generate compounds **23**–**26**, activity up to 6.25 μg/mL was observed for compounds **23** and **25** having *S* configuration at C-5 and C-6, respectively.

Compounds **23**–**26** were converted to amino alditols **19**–**22**, which were screened for antimycobacterial activity. Among them chiraly pure compounds **19** and **20** showed activity up to 6.25 μg/mL. The details of the activity are presented in Table 1.

From the above observation, it could be argued that the compounds which had deoxy sites in succession having less restricted rotation and which were chiraly pure, namely **19**, **20** and **23**, were most active. It is intriguing that compounds **24** and **26** obtained from aldehyde **6**, which in turn was obtained from galactose, did not show appreciable activity.

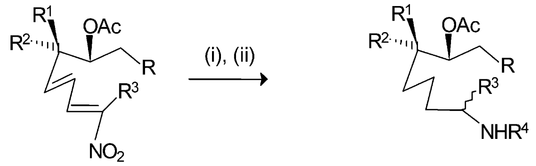
These results clearly indicated the potential of chain extended nitrogen containing deoxy sugar derivatives as antimycobacterial agents. The compounds **19**, **20** and **23** were also tested in microwell plate Alamar blue assay[15] and were found to be active below 12.5 μg/mL. Thus it



7 R = R^2 = OAc, R^1 = R^3=H
8 R = R^1 = OAc, R^2 = R^3=H
9 R = R^2 = OAc, R^1 = H, R^3=CH$_3$
10 R = R^1 = OAc, R^2 = H, R^3=CH$_3$

15 R = R^2 = OAc, R^1 = R^3=H, R^4=Ac
16 R = R^1 = OAc, R^2 = R^3=H, R^4=Ac
17 R = R^2 = OAc, R^1 = H, R^3=CH$_3$, R^4=Ac
18 R = R^1 = OAc, R^2 = H, R^3=CH$_3$, R^4=Ac

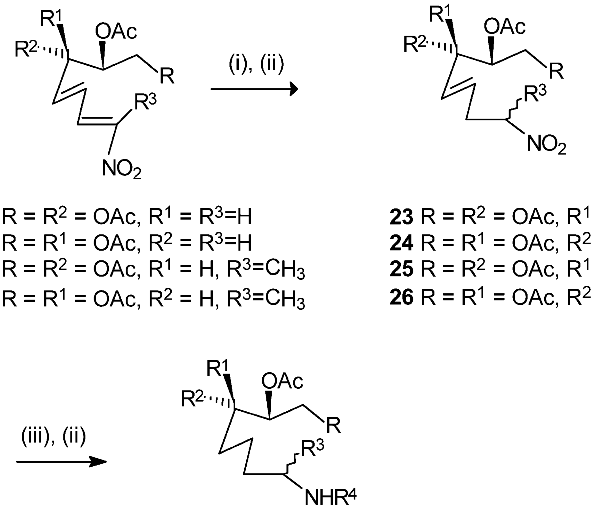
Scheme 4. (i) H_2, 10% Pd/C, CH_3OH, 8 h, 379 KPa, rt; (ii) Ac_2O, Pyr, 10 h, 0–4 °C.



11 R = R^2 = OAc, R^1 = R^3=H
12 R = R^1 = OAc, R^2 = R^3=H
13 R = R^2 = OAc, R^1 = H, R^3=CH$_3$
14 R = R^1 = OAc, R^2 = H, R^3=CH$_3$

19 R = R^2 = OAc, R^1 = R^3=H, R^4=Ac
20 R = R^1 = OAc, R^2 = R^3=H, R^4=Ac
21 R = R^2 = OAc, R^1 = H, R^3=CH$_3$, R^4=Ac
22 R = R^1 = OAc, R^2 = H, R^3=CH$_3$, R^4=Ac

Scheme 5. (i) H_2, 10% Pd/C, CH_3OH, 8 h, 379 KPa, rt; (ii) Ac_2O, Pyr, 10 h, 0–4 °C.



11 R = R^2 = OAc, R^1 = R^3=H
12 R = R^1 = OAc, R^2 = R^3=H
13 R = R^2 = OAc, R^1 = H, R^3=CH$_3$
14 R = R^1 = OAc, R^2 = H, R^3=CH$_3$

23 R = R^2 = OAc, R^1 = R^3=H,
24 R = R^1 = OAc, R^2 = R^3=H,
25 R = R^2 = OAc, R^1 = H, R^3=CH$_3$
26 R = R^1 = OAc, R^2 = H, R^3=CH$_3$

19 R = R^2 = OAc, R^1 = R^3=H, R^4=Ac
20 R = R^1 = OAc, R^2 = R^3=H, R^4=Ac
21 R = R^2 = OAc, R^1 = H, R^3=CH$_3$, R^4=Ac
22 R = R^1 = OAc, R^2 = H, R^3=CH$_3$, R^4=Ac

Scheme 6. (i) $NaBH_4$, CH_3CN/H_2O (1:1), 30 min, rt; (ii) Ac_2O, Pyr, 10 h, 0–4 °C; (iii) H_2, 10% Pd/C, CH_3OH, 7 h, 379 KPa, rt.

Table 1. Antimycobacterial activity against *M. tuberculosis* $H_{37}Rv$ by agar dilution method

Compd	Concentration in μg/mL				
	100	50	25	12.5	6.25
7	NA	NA	NA	—	—
8	CI	CI	CI	NA	—
9	CI	CI	NA	—	—
11	NA	NA	NA	NA	—
12	CI	NA	—	—	—
13	NA	NA	NA	—	—
14	CI	NA	—	—	—
15	NA	NA	NA	NA	—
16	NA	NA	—	—	—
17	—	NA	NA	—	—
18	—	NA	NA	—	—
19	—	CI	CI	CI	CI
20	—	CI	CI	CI	CI
21	—	CI	CI	CI	NA
22	—	CI	CI	—	—
23	—	CI	CI	CI	CI
24	NA	NA	NA	—	—
25	—	CI	CI	CI	CI
26	—	CI	NA	—	—

CI, complete inhibition; NA, not active; — not tested at particular concentration.

may be presumed that the chiraly pure compounds **19**, **20** and **23**, showing activity in two different test systems, bear a little resemblance with clinically used antitubercular agent ethambutol and thus may have a similar site of action as that of ethambutol in the biosynthesis of the cell wall of *M. tuberculosis.*

Conclusion

In conclusion, we have shown herein that higher acyclic nitrogen containing deoxy sugar derivatives can conveniently be synthesized from easily available glycals as new leads for development of antimycobacterial agents. Our findings embodied in the present paper certainly give impetus for search of carbohydrate derived new antitubercular agents. In vivo screening of the compounds (**19**, **20**, **23** and **25**) is under way.

Experimental

All the reactions were monitored by thin layer chromatography over silica gel coated TLC plates. The spots on TLC plates were visualized by warming the $CeSO_4$ (1% in 2 N H_2SO_4) sprayed plates in an oven at 100 °C. Other developers used were iodine vapors and potassium permanganate spray. For column chromatography, silica gel (60–120 mesh) was used. The melting point was recorded on electrically heated melting point apparatus and is uncorrected. IR spectra were recorded on Perkin-Elmer 881 and FTIR-8210 PC Shimadzu Spectrophotometers either as KBr discs or neat and the values are expressed in cm^{-1}. ^1H NMR, ^{13}C NMR, nOe spectra were recorded on Bruker Avance DRX-300 MHz and Avance DPX 200 FT Bruker Robotics Spectrometer using TMS as an internal reference and $CDCl_3$ (δ 77.0). FAB mass spectra were recorded on JEOL SX 102/DA 6000 mass spectrometer using Argon/Xenon (6 KV, 10 mA) as the FAB gas. EI mass spectra were recorded on JEOL JMS-D-300 spectrometer with the ionization potential of 70 eV. Elemental analyses were carried out on a Carlo-Erba-1108 instrument. Optical rotations were determined on Autopol III polarimeter using 1 dm cell at 28 °C in methanol as the solvent; concentrations mentioned are in g/100 mL.

For the spectral data of **7**, **11**, **15**, **19**, **23**, see ref 11.

General procedure for nitroaldol condensation to form compounds 7–10

To a stirred solution of nitroalkane (5 mL) and triethylamine (0.25 mL, 1.8 mmol) was added the solution of the required aldehyde **5** or **6** (500 mg, 1.8 mmol) dissolved in nitroalkane (1.8 mL) during half an hour at 0 °C under N_2 atmosphere. The reaction was allowed to continue at this temperature for 10 h and then at room temperature for another period of 48 h. The reaction mixture was concentrated in vacuo and the residue was chromatographed over silica gel using the specified solvent as the eluent to furnish **7**–**10**.

Mixture of (3*E*)-5,6,7-tri-*O*-acetyl-1,3,4-trideoxy-1-nitro-D-(*lyxo* and *xylo*)-hept-3-enitol (8). Pale yellow oil (62.3%). Eluent for column chromatography: hexane/ethylacetate = 4:1, v/v. R_f 0.46 (hexane/ethylacetate = 1:1, v/v). IR (neat, cm^{-1}) 3468 (–OH), 1744 (>C=O), 1556, 1426 (–NO_2), 974 (> = <). ^1H NMR[16] ($CDCl_3$, 200 MHz) δ 2.07, 2.10, 2.17 (3×s, 9H, $COCH_3$), 3.65 (brs, O*H*, exchangeable with deuterium), 4.04 (dd, $J_{7a,6} = 2.9$ Hz and $J_{7a,7b} = 11.9$ Hz, 1H, H_A-7a), 4.06 (dd_e, $J_{7a,6} = 2.5$ Hz, 1H, H_B-7a), 4.29 (dd, $J_{7b,6} = 3.2$ Hz and $J_{7b,7a} = 11.8$ Hz, 1H, H-7b), 4.39–4.55 (m, 2H, H-1), 4.89 (m, 1H, H-2), 5.22 (m, 1H, H_A-6), 5.36 (m, 1H, H_B-6), 5.49 (t, $J_{5,4} = J_{5,6} = 5.1$ Hz, 1H, H-5), 5.82–5.87 (m_e, 2H, H-3 and H-4). ^{13}C NMR[17] ($CDCl_3$, 50 MHz) δ 20.24, 20.39, 20.47, 20.54, 20.61, 20.71 ($COCH_3$), 61.94, 62.15 (C-7), 68.24, 70.97, 71.21 (C-2, C-5 and C-6), 79.54, 80.41 (C-1), 127.09, 127.17 (C-4), 131.67, 131.77 (C-3), 169.75, 170.14, 170.60 ($COCH_3$). FAB MS *m*/*z* 335 $[M+2]^+$, 289 $[M-NO_2+2]^+$, 274 $[M-OAc]^+$. Elemental analysis calculated for $C_{13}H_{19}NO_9$ (333.29) C: 46.85%, H: 5.75%, N: 4.20%. Found C: 46.68%, H: 5.75%, N: 4.93%.

Mixture of (4*E*)-6,7,8-tri-*O*-acetyl-1,2,4,5-tetradeoxy-2-nitro-D-(*allo, altro, gluco* and *manno*)-oct-4-enitol (9). Pale yellow oil (58.8%). Eluent for column chromatography: hexane/ethylacetate = 4:1, v/v. R_f 0.53 (hexane/ethylacetate = 1:1, v/v). IR (neat, cm^{-1}) 3473 (–OH), 1743 (>C=O), 1552, 1371 (–NO_2), 977 (> = <). ^1H NMR ($CDCl_3$, 200 MHz) δ 1.50, 1.54 (2×d, $J_{1,2} = 4.2$ Hz, 6H, H_A-1 and H_B-1), 2.06, 2.08 (s, 9H, $COCH_3$), 3.22 (brs, O*H*, exchangeable with deuterium), 4.14 (dd, $J_{8a,7} = 6.7$ Hz and $J_{8a,8b} = 12.1$ Hz, 1H, H-8a), 4.27 (dd, $J_{8b,7} = 2.4$ Hz and $J_{8b,8a} = 11.9$ Hz, 1H, H-8b), 4.53–4.59 (m_e, 1.5H, H-2 and H-3), 4.70 (brs, 0.5H, H-3), 5.20 (m, 1H, H-7), 5.48 (m, 1H, H-6), 5.81–5.85

(m$_e$, 2H, H-4 and H-5). ^{13}C NMR (CDCl$_3$, 50 MHz) δ 12.50, 12.59, 15.76 (C-1), 20.61, 20.75, 20.81 (CO*C*H$_3$), 61.63 (C-8), 71.31, 71.37, 71.42, 71.50, 71.56, 71.62, 71.87, 73.07, 73.15 (C-3, C-6 and C-7), 85.67, 86.78 (C-2), 125.81, 127.37, 128.25, 128.43 (C-5), 131.83, 131.91, 132.0, 132.11 (C-4), 169.66, 170.14, 170.68 (*C*OCH$_3$). EI MS *m*/*z* 348 [M+1]$^+$, 288 [M−OAc]$^+$. Elemental analysis calculated for $C_{14}H_{21}NO_9$ (347.306) C: 48.41%, H: 6.09%, N: 4.03%. Found C: 48.29%, H: 6.32%, N: 4.11%.

Mixture of (4*E*)-6,7,8-tri-*O*-acetyl-1,2,4,5-tetradeoxy-2-nitro-D-(*gulo, ido, galacto* and *talo*)-oct-4-enitol (10). Pale yellow oil (59%). Eluent for column chromatography: hexane/ethylacetate = 4:1, v/v. R_f 0.50 (hexane/ethylacetate = 1:1, v/v). IR (neat, cm^{-1}) 3759 (–OH), 1743 (>C=O), 1552, 1517 (–NO$_2$), 974 (> = <). ^{1}H NMR[16] (CDCl$_3$, 200 MHz) δ 1.49 (d, J = 3.0 Hz, 3H, H$_A$-1), 1.52 (d, $J_{1,2}$ = 2.8 Hz, 3H, H$_B$-1), 2.07, 2.10 (2×s, 9H, COC*H*$_3$), 2.88 (brs, O*H*, exchangeable with deuterium), 4.07 (m, 1H, H-8a), 4.31 (m, 1H, H-8b), 4.5 (m$_e$, 1.5H, H-3 and H-2), 4.72 (m, 0.5H, H-2), 5.22 (m, 1H, H-7), 5.48 (t, $J_{6,5}$ = $J_{6,7}$ = 4.8 Hz, 1H, H-6), 5.78–5.79 (m$_e$, 2H, H-4 and H-5). ^{13}C NMR (CDCl$_3$, 50 MHz)[17] δ 12.44, 12.52, 15.53, 15.62 (C-1), 20.41, 20.57, 20.67 (CO*C*H$_3$), 61.90, 62.25 (C-8), 70.19, 70.92, 71.20, 71.30, 71.44, 71.78, 72.99 (C-3, C-6 and C-7), 85.47, 86.63, 86.71 (C-2), 127.24, 127.31, 128.27 (C-4), 131.55, 131.72, 131.84, 131.9 (C-5), 169.77, 170.13, 170.59 (*C*OCH$_3$). FAB MS *m*/*z* 348 (M+1)$^+$. Elemental analysis calculated for $C_{14}H_{21}NO_9$ (347.306) C: 48.41%, H: 6.09%, N: 4.03%. Found C: 48.24%, H: 5.99%, N: 4.32%.

General procedure for acetylation

To a solution of the compound to be acetylated (3.3 mmol) in pyridine (5 mL) was added acetic anhydride (15 mL) at 0 °C and the resulting reaction mixture was left for 10 h at 0–4 °C. Crude reaction mixture was poured in acidurated (1 N HCl, 50 mL) ice, extracted with chloroform (3×20 mL), washed with NaHCO$_3$ and brine, dried over anhydrous sodium sulphate (Na$_2$SO$_4$) and concentrated in vacuo to give crude product which was purified by column chromatography over silica gel.

(1*E*, 3*E*)-5,6,7-Tri-*O*-acetyl-1,2,3,4-tetradeoxy-1-nitro-D-*threo*-hept-1,3-dienitol (12). Yellow solid (33.3%). Mp 101 °C. Eluent for column chromatography: benzene/diethylether = 93:7, v/v. R_f 0.40 (benzene/diethylether = 4:1, v/v; KMnO$_4$ used as spraying agent). [α]$_D$ +32.18° (*c* 0.63, methanol). IR (KBr, cm^{-1}) 1747 (>C=O), 1523, 1433 (–NO$_2$), 968 (> = <). ^{1}H NMR (CDCl$_3$, 200 MHz) δ 2.07, 2.10, 2.15 (3×s, 9H, COC*H*$_3$), 4.04 (dd, $J_{7a,6}$ = 6.4 Hz and $J_{7a,7b}$ = 11.9 Hz, 1H, H-7a), 4.34 (dd, $J_{7b,6}$ = 4.4 Hz and $J_{7b,7a}$ = 11.9 Hz, 1H, H-7$_b$), 5.29 (td, $J_{6,5}$ = $J_{6,7b}$ = 4.7 Hz and $J_{6,7a}$ = 6.3 Hz, 1H, H-6), 5.63 (t, $J_{5,4}$ = $J_{5,6}$ = 5.1 Hz, 1H, H-5), 6.30 (dd, $J_{4,5}$ = 5.3 Hz and $J_{4,3}$ = 15.3 Hz, 1H, H-4), 6.40 (dd, $J_{3,2}$ = 10.4 Hz and $J_{3,4}$ = 15.6 Hz, 1H, H-3), 7.14 (d, $J_{1,2}$ = 13.2 Hz, 1H, H-1), 7.54 (dd, $J_{2,3}$ = 10.6 Hz and $J_{2,1}$ = 13.2 Hz, 1H, H-2). ^{13}C NMR (CDCl$_3$, 50 MHz) δ 20.55, 20.61 (CO*C*H$_3$), 61.66 (C-7), 70.55 (C-6), 70.98 (C-5), 125.82 (C-4), 136.46 (C-3), 140.2 (C-2), 140.32 (C-1), 169.39, 169.74, 170.38 (*C*OCH$_3$). FAB MS *m*/*z* 315 [M]$^+$, 256 [M−OAc]$^+$, 214 [M−(OAc+COCH$_2$)]$^+$. Elemental analysis calculated for $C_{13}H_{17}NO_8$ (315.28) C: 49.52%, H: 5.43%, N: 4.44%. Found C: 49.97%, H: 5.53%, N: 3.98%.

(2*E*,4*E*)-6,7,8-Tri-*O*-acetyl-1,2,3,4,5-pentadeoxy-2-nitro-D-*erythro*-oct-2,4-dienitol (13). Pale yellow oil (45.2%). Eluent for column chromatography: benzene/diethyl ether = 23:2, v/v. R_f 0.44 (benzene/diethylether = 4:1, v/v; KMnO$_4$ used as spraying agent). [α]$_D$ +36.2° (*c* 0.09, methanol). IR (neat, cm^{-1}) 1743 (>C=O), 1519, 1427 (–NO$_2$), 974 (> = <). ^{1}H NMR (CDCl$_3$, 200 MHz) δ 2.07, 2.10, 2.12 (3×s, 9H, COC*H*$_3$), 2.29 (s, 3H, H-1), 4.18 (dd, $J_{8a,7}$ = 6.3 Hz and $J_{8a,8b}$ = 12.1 Hz, 1H, H-8a), 4.28 (dd, $J_{8b,7}$ = 3.9 Hz and $J_{8b,8a}$ = 12.2 Hz, 1H, H-8b), 5.26 (td, $J_{7,8b}$ = 4.1 Hz and $J_{7,6}$ = $J_{7,8a}$ = 6.2 Hz, 1H, H-7), 5.62 (t, $J_{6,5}$ = $J_{6,7}$ = 6.2 Hz, 1H, H-6), 6.19 (dd, $J_{5,6}$ = 6.9 Hz and $J_{5,4}$ = 15.3 Hz, 1H, H-5), 6.49 (dd, $J_{4,3}$ = 11.3 Hz and $J_{4,5}$ = 15.2 Hz, 1H, H-4), 7.52 (d, $J_{3,4}$ = 11.2 Hz, 1H, H-3). ^{13}C NMR (CDCl$_3$, 50 MHz) δ 12.88 (C-1), 20.56, 20.7 (CO*C*H$_3$), 61.52 (C-8), 71.23 (C-7), 71.59 (C-6), 127.23 (C-5), 130.86 (C-4), 137.65 (C-3), 148.29 (C-2), 169.42, 169.84, 170.39 (*C*OCH$_3$). FAB MS *m*/*z* 329 [M]$^+$, 270 [M−OAc]$^+$, 256 [M−(CH$_2$–CHNO$_2$)]$^+$. Elemental analysis calculated for $C_{14}H_{19}NO_8$ (329.291) C: 51.07%, H: 5.82%, N: 4.25%. Found C: 51.81%, H: 6.05%, N: 3.70%.

(2*E*,4*E*)-6,7,8-Tri-*O*-acetyl-1,2,3,4,5-pentadeoxy-2-nitro-D-*threo*-oct-2,4-dienitol (14). Pale yellow oil (32.1%). Eluent for column chromatography: benzene/diethylether = 23:2, v/v. R_f 0.42 (benzene/diethylether = 4:1, v/v; KMnO$_4$ used as spraying agent). [α]$_D$ +20° (*c* 0.31, methanol). IR (neat, cm^{-1}) 1744 (>C=O), 1653 (> = <), 1518, 1435 (–NO$_2$). ^{1}H NMR (CDCl$_3$, 200 MHz) δ 2.07, 2.11, 2.14 (3×s, 9H, COC*H*$_3$), 2.28 (s, 3H, H-1), 4.04 (dd, $J_{8a,7}$ = 6.4 Hz and $J_{8a,8b}$ = 11.9 Hz, 1H, H-8a), 4.34 (dd, $J_{8b,7}$ = 4.3 Hz and $J_{8b,8a}$ = 11.9 Hz, 1H, H-8b), 5.29 (td, $J_{7,6}$ = $J_{7,8b}$ = 4.7 Hz and $J_{7,8a}$ = 5.5 Hz, 1H, H-7), 5.63 (t, $J_{6,5}$ = $J_{6,7}$ = 5.6 Hz, 1H, H-6), 6.16 (dd, $J_{5,6}$ = 6.4 Hz and $J_{5,4}$ = 15.3 Hz, 1H, H-5), 6.47 (dd, $J_{4,3}$ = 11.3 Hz and $J_{4,5}$ = 15 Hz, 1H, H-4), 7.49 (d, $J_{3,4}$ = 11.3 Hz, 1H, H-3). ^{13}C NMR (CDCl$_3$, 50 MHz) δ 12.91 (C-1), 20.59, 20.66, 20.73 (CO*C*H$_3$), 61.77 (C-8), 70.74 (C-7), 71.36 (C-6), 126.88 (C-5), 130.87 (C-4), 137.73 (C-3), 148.29 (C-2), 169.50, 169.79, 170.51 (*C*OCH$_3$). FAB MS *m*/*z* 329 [M]$^+$, 314 [M−CH$_3$]$^+$. Elemental analysis calculated for $C_{14}H_{19}NO_8.H_2O$ (347.305) C: 48.41%, H: 6.09%, N: 4.03%. Found C: 48.14%, H: 5.59%, N: 3.62%.

General procedure for hydrogenation

To a solution of the compound to be hydrogenated (3 mmol) in methanol (20 mL) was added 10% Pd/C (2 g). The mixture was hydrogenated at 379 KPa for 7 h. It was filtered through a celite pad and the residue obtained after solvent evaporation in vacuo was acetylated as per general procedure stated earlier to furnish crude amino alkanol which was chromatographed over silica gel to yield pure amino alkanol.

Mixture of 1-acetamido-2,5,6,7-tetra-*O*-acetyl-1,3,4-trideoxy-D-(*lyxo* and *xylo*)-heptitol (16). Pale yellow oil (45%). Eluent for column chromatography: benzene/methanol = 24:1, v/v. R_f 0.45 (benzene/methanol = 9:1, v/v; iodine vapors used as developing agent). IR (neat, cm^{-1}) 3021 (alkane), 1602 (>C=O). ^{1}H NMR (CDCl$_3$, 200 MHz) δ 1.60 (brs, 4H, H-3 and H-4), 1.98 (s, 3H, NHCOCH_3), 2.05, 2.07, 2.09, 2.11 (4×s, 12H, COCH_3), 3.38 (m, 2H, H-1), 4.00 (dd, $J_{7a,6}$ = 3.2 Hz 1H, H$_A$-7a), 4.03 (dd$_e$, $J_{7a,6}$ = 3.2 Hz, 1H, H$_B$-7a), 4.27 (dd, $J_{7b,6}$ = 4.1 Hz, $J_{7b,7a}$ = 11.9 Hz, 1H, H-7b), 4.85 (m, 1H, H-6), 5.09 (m, 1H, H-5), 5.19 (m, 1H, H-2), 5.75 (brs, N*H*Ac, exchangeable with deuterium). ^{13}C NMR (CDCl$_3$, 50 MHz) δ 20.54, 20.62, 20.70, 20.94 (CO*C*H$_3$), 23.00 (NHCO*C*H$_3$), 25.97, 26.05 (C-3), 27.07, 27.26 (C-4), 42.29, 42.44 (C-1), 62.13 (C-7), 70.75, 70.99 (C-6), 70.65 (C-5), 72.39, 72.65 (C-2), 170.16, 170.40, 170.84 (*C*OCH$_3$). FAB MS *m*/*z* 390 [M + 1]$^+$, 330 [M−OAc]$^+$. Elemental analysis calculated for $C_{17}H_{27}NO_9$ (389.40) C: 52.44%, H: 6.99%, N: 3.60%. Found C: 51.46%, H: 7.09%, N: 3.86%.

Mixture of 2-acetamido-3,6,7,8-tetra-*O*-acetyl-1,2,4,5-tetradeoxy-D-(*allo, altro, gluco* and *manno*)-octitol (17). Pale yellow oil (41.3%). Eluent for column chromatography: benzene/methanol = 24:1, v/v. R_f 0.48 (benzene/methanol = 9:1, v/v; iodine vapors used as developing agent). IR (neat, cm^{-1}) 3197 (alkane), 1741 (>C=O), 1659, 1545 (–NHAc). ^{1}H NMR (CDCl$_3$, 200 MHz) δ 1.09 (d, $J_{1,2}$ = 6.3 Hz, 3H, H-1), 1.61 (brs, 4H, H-4 and H-5), 1.95 (s, 3H, NHCOCH_3), 1.99, 2.07, 2.08, 2.17 (4×s, 12H, COCH_3), 4.14–4.30 (m$_e$, 3H, H-2, H-8a and H-8b), 4.80 (m, 1H, H-3), 5.10 (m$_e$, 2H, H-6 and H-7), 5.76 (brs, N*H*Ac, exchangeable with deuterium). ^{13}C NMR (CDCl$_3$, 50 MHz) δ 14.82, 15.24, 17.95, 18.04 (C-1), 20.50, 20.67, 20.81 (CO*C*H$_3$), 23.10 (NHCO*C*H$_3$), 25.66, 25.84, 25.99, 26.13, 26.48, 26.75 (C-4 and C-5), 46.51, 46.98 (C-2), 61.66 (C-8), 70.48, 70.57, 70.64, 70.77, 70.92, 70.98, 74.83, 75.25, 75.35, 75.85, 76.49, 76.79, 77.13 (C-3, C-6 and C-7), 169.24, 169.28, 169.35, 169.40, 169.89, 169.97, 170.05, 170.11, 170.39, 170.42, 170.47, 170.88, 170.94 (*C*OCH$_3$). FAB MS *m*/*z* 404 [M + 1]$^+$, 343 [M−AcOH]$^+$, 302 [M−COCH$_2$]$^+$, 260 [M−(CH$_2$=CH-NHAc + AcOH)]$^+$. Elemental analysis calculated for $C_{18}H_{29}NO_9.2H_2O$ (439.44) C: 49.19%, H: 7.11%, N: 3.19%. Found C: 49.42%, H: 6.81%, N: 2.12%.

Mixture of 2-acetamido-3,6,7,8-tetra-*O*-acetyl-1,2,4,5-tetradeoxy-D-(*gulo, ido, galacto* and *talo*)-octitol (18). Pale yellow oil (42.4%). Eluent for column chromatography: benzene/methanol = 24:1, v/v. R_f 0.47 (benzene/methanol = 9:1, v/v; iodine vapours used as developing agent). IR (neat, cm^{-1}) 3769 (–NHAc), 1739 (>C=O). ^{1}H NMR (CDCl$_3$, 200 MHz) δ 1.09 (d, $J_{1,2}$ = 6.8 Hz, 3H, H$_A$-1), 1.11 (d, $J_{1,2}$ = 6.7 Hz, 3H, H$_B$-1), 1.58 (brs, 4H, H-4 and H-5), 1.96 (s, 3H, NHCOCH_{3A}), 1.99 (s, 3H, NHCOCH_{3B}), 2.05, 2.09, 2.11 (3×s, 12H, COCH_3), 3.94–4.31 (m$_e$, 3H, H-2, H-8a and H-8b), 4.83 (brs, 1H, H-3), 5.08–5.22 (m$_e$, 2H, H-6 and H-7), 5.51, 5.53, 5.72, 5.74 (4×brs, N*H*Ac, exchangeable with deuterium). ^{13}C NMR (CDCl$_3$, 50 MHz) δ 14.92, 15. 34, 18.06, 18.07 (C-1), 20.34, 20.51, 20.58, 20.67, 20.73, 20.84, 20.95, 21.01, 21.10, 21.16, 21.22 (CO*C*H$_3$), 23.15 (NHCO*C*H$_3$), 25.94, 26.14, 26.24, 26.38, 26.65, 26.74, 26.90 (C-4 and C-5), 46.43, 46.70, 47.00, 47.10 (C-2), 62.11 (C-8) 70.54, 70.62, 70.71, 70.77, 70.81, 70.92, 71.12 (C-6 and C-7), 75.29, 75.44, 75.66, 75.83 (C-3), 169.30, 169.33, 169.44, 170.01, 170.11, 170.17, 170.19, 170.30, 170.39, 170.53, 171.00 (*C*OCH$_3$). FAB MS *m*/*z* 404 [M + 1]$^+$, 344 [M−OAc]$^+$, 302 [M−(OAc + COCH$_2$)]$^+$, 284 [M−(OAc + AcOH)]$^+$. Elemental analysis calculated for $C_{18}H_{29}NO_9$ (403.41) C: 53.59%, H: 7.25%, N: 3.47%. Found C: 53.56%, H: 7.61%, N: 2.77%.

1-Acetamido-5,6,7-tri-*O*-acetyl-1,2,3,4-tetradeoxy-D-*erythro*-heptitol (19). Elemental analysis calculated for $C_{15}H_{25}NO_7$ (331.350) C: 54.37%, H: 7.61%, N: 4.23%. Found C: 54.52%, H: 8.00%, N: 4.28%.

1-Acetamido-5,6,7-tri-*O*-acetyl-1,2,3,4-tetradeoxy-D-*threo*-heptitol (20). Pale yellow oil (50.2%). Eluent for column chromatography: benzene/methanol = 47:3, v/v. R_f 0.39 (benzene/methanol = 9:1, v/v; iodine vapours used as developing agent). $[\alpha]_D$ + 33.68° (*c* 0.19, methanol). IR (neat, cm^{-1}) 3020 (–NHAc), 1741 (>C=O). ^{1}H NMR (CDCl$_3$, 200 MHz) δ 1.33–1.63 (m$_e$, 6H, H-2, H-3 and H-4), 1.97 (s, 3H, NHCOCH_3), 2.05, 2.09, 2.11 (s, 9H, COCH_3), 3.22 (q, $J_{1,2}$ = $J_{1,NHAc}$ = 6.4H, 2H, H-1),[18] 4.02 (dd, $J_{7a,6}$ = 4.3 Hz and $J_{7a,7b}$ = 11.8 Hz, 1H, H-7a), 4.28 (dd, $J_{7b,6}$ = 6.7 Hz and $J_{7b,7a}$ = 11.8 Hz, 1H, H-7b), 5.10 (m, 1H, H-6), 5.22 (dt, $J_{5,4}$ = 4.1 Hz and $J_{5,6}$ = 6.8 Hz, 1H, H-5), 5.54 (brs, N*H*Ac, exchangeable with deuterium). ^{13}C NMR (CDCl$_3$, 50 MHz) δ 20.54, 20.62, 20.71 (CO*C*H$_3$), 23.04 (NHCO*C*H$_3$), 22.10, 28.92, 29.84 (C-2, C-3, C-4), 38.99 (C-1), 62.31 (C-7), 70.9, 71.15 (C-5, and C-6), 170.16, 170.33, 170.46 (*C*OCH$_3$). FAB MS *m*/*z* 331 [M]$^+$, 289 [M−COCH$_2$]$^+$. Elemental analysis calculated for $C_{15}H_{25}NO_7$. 1/2H_2O (340.3575) C: 52.92%, H: 7.70%, N: 4.12%. Found C: 53.36%, H: 7.97%, N: 3.86%.

Mixture of 2-acetamido-6,7,8-tri-*O*-acetyl-1,2,3,4,5-pentadeoxy-D-(*arabino* and *ribo*)-octitol (21). Pale yellow oil (51.3%). Eluent for column chromatography: benzene/methanol = 24:1, v/v. R_f 0.4 (benzene/methanol = 9:1, v/v; iodine vapours used as developing agent). IR (neat, cm^{-1}) 3020 (–NHAc), 1740 (>C=O). ^{1}H NMR (CDCl$_3$, 200 MHz) δ 1.11 (d, $J_{1,2}$ = 6.5 Hz, 3H, H$_A$-1), 1.12 (d, $J_{1,2}$ = 6.4 Hz, 3H, H$_B$-1), 1.42–1.62 (m$_e$, 6H, H-3, H-4 and H-5), 1.95 (s, 3H, NHCOCH_3), 2.07, 2.08, 2.083 (3×s, 9H, COCH_3), 3.9 (m$_e$, 1H, H-2), 4.16 (dd, $J_{8a,7}$ = 6.5 Hz and $J_{8a,8b}$ = 12.1 Hz, 1H, H-8a), 4.28 (dd, $J_{8b,7}$ = 2.4 Hz and $J_{8b,8a}$ = 12.1 Hz, 1H, H-8b), 5.12 (m$_e$, 2H, H-6 and H-7), 5.3 (brs, N*H*Ac, exchangeable with deuterium). ^{13}C NMR (CDCl$_3$, 50 MHz) δ 20.72, 20.88, 21.00, 21.40 (C-1 and CO*C*H$_3$), 23.35, 23.40 (NHCO*C*H$_3$), 21.53, 29.92, 36.04, 36.20 (C-3, C-4, C-5), 44.85, 44.92 (C-2), 61.87 (C-8), 66.83, 71.33, 71.61, 71.88 (C-6 and C-7), 169.35, 170.14, 170.25, 170.42, 170.61 (*C*OCH$_3$). FAB MS *m*/*z* 346 [M + 1]$^+$, 304 [M$^+$−COCH$_2$ + 1]$^+$. Elemental analysis calculated for $C_{16}H_{27}NO_7$ (345.376) C: 55.64%, H: 7.88%, N: 4.06%. Found C: 55.54%, H: 8.16%, N: 4.41%.

Mixture of 2-acetamido-6,7,8-tri-*O*-acetyl-1,2,3,4,5-pentadeoxy-D-(*lyxo* and *xylo*)-octitol (22). Pale yellow oil (51.8%). Eluent for column chromatography: benzene/

methanol = 24:1, v/v. R_f 0.39 (benzene/methanol = 9:1, v/v; iodine vapours used as developing agent). IR (neat, cm^{-1}) 3022 (–NHAc), 1739 (>C=O). ^{1}H NMR (CDCl$_3$, 200 MHz) δ 1.11 (d, $J_{1,2}$ = 6.6 Hz, 3H, H$_A$-1), 1.12 (d, $J_{1,2}$ = 6.6 Hz, 3H, H$_B$-1), 1.22–1.70 (m$_e$, 6H, H-3, H-4 and H-5), 1.95 (s, 3H, NHCOC*H*$_{3A}$), 1.96 (s, 3H, NHCOC*H*$_{3B}$), 2.05, 2.07, 2.09, 2.11 (s, COC*H*$_3$), 3.92 (m$_e$, 1H, H-2), 3.93 (dd$^{19}_{e}$, $J_{8a,7}$ = 7 Hz, 1H, H$_A$-8a), 4.01 (dd, $J_{8a,7}$ = 6.7 Hz and $J_{8a,8b}$ = 11.8 Hz, 1H, H-8a), 4.27 (dd, $J_{8b,7}$ = 4.3 Hz and $J_{8b,8a}$ = 11.7 Hz, 1H, H$_A$-8b), 4.28 (dd, $J_{8b,7}$ = 4.3 Hz and $J_{8b,8a}$ = 11.8 Hz, 1H, H$_B$-8b), 5.03–5.25 (m$_e$, 2H, H-6 and H-7), 5.29, 5.33 (2×brs, N*H*Ac, exchangeable with deuterium). ^{13}C NMR (CDCl$_3$, 50 MHz) δ 20.62, 20.69, 20.79, 21.09 (C-1 and CO*C*H$_3$), 23.35, 23.40 (NHCO*C*H$_3$), 21.20, 21.28, 30.06, 36.09, 36.32 (C-3, C-4 and C-5), 44.71, 44.93 (C-2), 62.33, 62.41 (C-8), 70.88, 70.94, 71.09, 71.27 (C-6 and C-7), 169.30, 169.38, 170.15, 170.33, 170.51 (*C*OCH$_3$). FAB MS *m*/*z* 346 [M + 1]$^+$. Elemental analysis calculated for $C_{16}H_{27}NO_7.1/2H_2O$ (354.3835) C: 54.22%, H: 7.96%, N: 3.95% Found C: 54.21%, H: 8.04%, N: 3.25%.

General procedure for sodium borohydride reduction

To a stirred solution of the compound to be reduced (**11–14**) (0.15 mmol) in aqueous acetonitrile (1:1, v/v; 1.5 mL) was added sodium borohydride (50 mg) and the mixture was stirred for 30 min at room temperature. Aqueous acetic acid (9:1, v/v) was added to decompose the excess of reducing agent. Evaporation of the solvent, extraction of the residue with acetone and the evaporation of the solvent was followed by acetylation as per general method stated earlier, furnished **23–26** after column chromatographic purification over silica gel or as specified.

(3*E*)-5,6,7-Tri-*O*-acetyl-1,2,3,4-tetradeoxy-1-nitro-D-*threo*-hept-3-enitol (24). Pale yellow oil (69%). Eluent for column chromatography: benzene/diethylether = 47:3, v/v. R_f 0.33 (benzene/diethylether 4:1, v/v; KMnO$_4$ used as spraying agent). [α]$_D$ + 10.5° (*c* 0.19, methanol). IR (neat, cm^{-1}) 3024 (>=<), 1743 (>C=O), 1519, 1404 (–NO$_2$). ^{1}H NMR (CDCl$_3$, 200 MHz) δ 2.06, 2.07, 2.08 (3×s, 9H, COC*H*$_3$), 2.75 (q, $J_{2,1} = J_{2,3}$ = 6.7 Hz, 2H, H-2), 4.01 (dd, $J_{7a,6}$ = 6.3 Hz and $J_{7a,7b}$ = 12 Hz, 1H, H-7a), 4.30 (dd, $J_{7b,6}$ = 4 Hz and $J_{7b,7a}$ = 12 Hz, 1H, H-7b), 4.43 (t, $J_{1,2}$ = 6.8 Hz, 2H, H-1), 5.19 (td, $J_{6,7b} = J_{6,5}$ = 4 Hz and $J_{6,7a}$ = 6.1 Hz, 1H, H-6), 5.45 (t$_e$,^{20} 1H, H-5), 5.56 (dd$_e$,^{20} $J_{4,5}$ = 5.7 Hz, 1H, H-4), 5.78 (dt, $J_{3,2}$ = 6.6 Hz and $J_{3,4}$ = 14.9 Hz, 1H, H-3). ^{13}C NMR (CDCl$_3$, 50 MHz) δ 20.64, 20.79, 20.80 (CO*C*H$_3$), 29.81 (C-2), 61.96 (C-7), 71.06 (C-6), 71.57 (C-5), 74.27 (C-1), 127.97 (C-4), 129.54 (C-3), 169.57, 169.98, 170.37 (*C*OCH$_3$). FAB MS *m*/*z* 318 [M + 1]$^+$, 258 [M–OAc]$^+$. Elemental analysis calculated for $C_{13}H_{19}NO_8$ (317.29) C: 49.21%, H: 6.03%, N: 4.41%. Found C: 50.18%, H: 6.47%, N: 4.09%.

Mixture of (4*E*)-6,7,8-tri-*O*-acetyl-1,2,3,4,5-pentadeoxy-2-nitro-D-(*arabino* and *ribo*)-oct-4-enitol (25). Yellow oil (55.2%). Eluent for column chromatography: benzene/diethylether = 47:3, v/v. R_f 0.39 (benzene/diethylether = 4:1, v/v; KMnO$_4$ used as spraying agent). IR (neat, cm^{-1}) 1743 (>C=O), 1552, 1371 (–NO$_2$). ^{1}H NMR (CDCl$_3$, 200 MHz) δ 1.53 (d, $J_{1,2}$ = 6.6 Hz, 3H, H$_A$-1), 1.54 (d, $J_{1,2}$ = 6.7 Hz, 3H, H$_B$-1), 2.06, 2.09 (2×s, 9H, COC*H*$_3$), 2.56 (m, 1H, H-3a), 2.68 (m, 1H, H-3b), 4.13 (dd, $J_{8a,7}$ = 6.7 Hz and $J_{8a,8b}$ = 12.1 Hz, 1H, H-8a), 4.22 (dd, $J_{8b,7}$ = 3.8 Hz and $J_{8b,8a}$ = 12.1 Hz, 1H, H-8b), 4.59 (m, 1H, H-2), 5.18 (td, $J_{7,8b} = J_{7,6}$ = 4.5 Hz and $J_{7,8a}$ = 5.2 Hz, 1H, H-7), 5.40 (m, 1H, H-6), 5.54 (dd, $J_{5,6}$ = 3.5 Hz and $J_{5,4}$ = 14.7 Hz, 1H, H$_A$-5), 5.58 (dd, $J_{5,6}$ = 3.1 Hz and $J_{5,4}$ = 14.7 Hz, 1H, H$_B$-5), 5.75 (ddd, $J_{4,3a}$ = 6.5 Hz, $J_{4,3b}$ = 7 Hz and $J_{4,5}$ = 14.7 Hz, 1H, H-4). ^{13}C NMR (CDCl$_3$, 50 MHz) δ 18.64 (C-1), 20.67, 20.80, 20.92 (CO*C*H$_3$), 37.51, 37.64 (C-3), 61.75 (C-8), 66.75, 66.98 (C-7), 71.29, 71.79 (C-6), 83.35 (C-2), 128.64 (C-5), 129.50, 129.84 (C-4), 169.44, 170.05, 170.53 (*C*OCH$_3$). FAB MS *m*/*z* 332 [M + 1]$^+$, 272 [M–OAc]$^+$. Elemental analysis calculated for $C_{14}H_{21}NO_8$ (331.316) C: 50.75%, H: 6.39%, N: 3.86%. Found C: 50.52%, H: 6.08%, N: 3.52%.

Mixture of (4*E*)-6,7,8-tri-*O*-acetyl-1,2,3,4,5-pentadeoxy-2-nitro-D-(*lyxo* and *xylo*)-oct-4-enitol (26). Pale yellow oil (50%). Eluent for column chromatography: benzene/diethylether = 47:3, v/v. R_f 0.38 (benzene/diethylether = 4:1, v/v; KMnO$_4$ used as spraying agent). IR (neat, cm^{-1}) 3024 (> = <), 1743 (>C=O), 1552, 1371 (–NO$_2$). ^{1}H NMR (CDCl$_3$, 200 MHz) δ 1.52 (d, $J_{1,2}$ = 6.7 Hz, 3H, H$_A$-1), 1.524 (d, $J_{1,2}$ = 6.6 Hz, 3H, H$_B$-1), 2.06, 2.07, 2.08, 2.09 (4×s, 12H, COC*H*$_3$), 2.54 (m$_e$, 1H, H-3a), 2.68 (m$_e$, 1H, H-3b), 3.99 (dd, $J_{8a,7}$ = 6.2 Hz and $J_{8a,8b}$ = 12.0 Hz, 1H, H$_A$-8a), 4.00 (dd, $J_{8a,7}$ = 6.3 Hz and $J_{8a,8b}$ = 11.9 Hz, 1H, H$_B$-8a), 4.30 (dd, $J_{8b,7}$ = 3.9 Hz and $J_{8b,8a}$ = 12 Hz, 1H, H$_A$-8b), 4.31 (dd, $J_{8b,7}$ = 3.9 Hz and $J_{8b,8a}$ = 12 Hz, 1H, H$_B$-8b), 4.58 (sex, $J_{2,1} = J_{2,3a} = J_{2,3b}$ = 6.7 Hz, 1H, H-2), 5.19 (m$_e$, 1H, H-6), 5.43 (t$_e$,^{21} 1H, H-6), 5.51 (dd$_e$,^{21} $J_{5,6}$ = 6.7 Hz, 1H, H-5), 5.71 (dt, $J_{4,3}$ = 6 Hz and $J_{4,5}$ = 14 Hz, 1H, H$_A$-4), 5.74 (dt, $J_{4,3}$ = 6 Hz and $J_{4,5}$ = 14 Hz, 1H, H$_B$-4). ^{13}C NMR (CDCl$_3$, 50 MHz) δ 18.53, 18.61 (C-1), 20.60, 20.67, 20.82 (CO*C*H$_3$), 37.49 (C-3), 61.98 (C-8), 71.05, 71.54, 71.69 (C-6 and C-7), 82.28, 82.35 (C-2), 128.51, 128.62 (C-5), 129.10, 129.30 (C-4), 169.58, 169.98, 170.40 (*C*OCH$_3$). FAB MS *m*/*z* 332 [M + 1]$^+$, 285 [M–NO$_2$]$^+$, 272 [M–OAc]$^+$. Elemental analysis calculated for $C_{14}H_{21}NO_8$ (331.316) C: 50.75%, H: 6.39%, N: 3.86%. Found C: 51.23%, H: 6.7%, N: 3.92%.

Biological Activity

Agar dilution method[13]

Briefly serial 2-fold dilutions of each test compound/drug were incorporated into 7H10 agar. Inoculum of *M. tuberculosis H$_{37}$Rv* was prepared from fresh Lowenstein–Jensen slants adjusted to 1 mg/mL (wet weight) in Tween 80 (0.05%) saline and diluted to 10^{-2} to give a concentration of approximately 10^7 cfu/mL. The 5 μL of bacterial suspension was spotted into 7H10 agar tubes containing serial 2-fold dilution of drugs per mL. The tubes were incubated at 37 °C and final readings were recorded on 30 days. The MICs were read as the minimum concentration of drugs/compounds that completely inhibited the growth of *M. tuberculosis* per spot. Oxofloxacin was used as the standard drug for comparison.

Microwell plate Alamar blue assay[15]

Antimycobacterial activity was determined by microwell plate based Alamar blue assay (MABA) using *M. tuberculosis* $H_{37}Ra$ as a surrogate for the virulent $H_{37}Rv$ strain. Alamar blue^{TM} is an oxidation-reduction dye and serves as general indicator of cellular growth and/or viability. Its blue, non fluorescent oxidised form, becomes pink and fluorescent upon reduction. The results of MABA have been found comparable to the standard BACTEC 460 system based assay.[15] The standard antitubercular drugs Rifamycin, Isoniazid, *p*-Amino salicylic acid, Ethambutol and Ethionamide (MIC range $3<0.3\,\mu g/mL$) were taken as positive controls. A molecule showing $MIC<12.5\,\mu g/mL$ was considered active.

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16. In the PMR spectrum, only some signals of two isomers are resolved in the diastereomeric mixture. They are named arbitrarily as A and B.
17. As in the PMR spectrum, only some CMR signals are resolved in the diastereomeric mixture.
18. In D_2O shake experiment, H-1 appeared as a triplet at δ 3.22.
19. dd for H-8a was merged with multiplet for H-2.
20. Triplet for H-5 was merged with double doublet for H-4.
21. Triplet for H-6 was merged with double doublet for H-5.