A Mild and Simple Enzymatic Conversion of Aldono- and Alduronitriles into the Corresponding Amides and/or Carboxylic Acids

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Nitriles are frequently employed in carbohydrate chemistry as a means of carbon chain extension, this being well exemplified by the classical Kiliani–Fischer synthesis[1] recently utilized in the synthesis of ^{13}C- and ^{14}C-labeled carbohydrates.[2] The nitrile moiety can be easily introduced using a variety of methods. Depending on the synthetic target, the conversion of this functional group into an amide or carboxylic acid is of preparative importance. The chemical hydrolysis of nitriles generally requires rather harsh acidic or basic conditions, and as such, examples can be found in the carbohydrate literature.[1,3,4] BeMiller et al.[4] recently employed a mixture of titanium tetrachloride and glacial acetic acid to hydrolyze per-O-acetylated β-D-galactopyranosyl cyanide into the corresponding amide, a competitive inhibitor of *E. coli* lacZ β-galactosidase, in good yields. These workers also synthesized N-substituted *C*-β-D-galactosylformamides and -methylamines, substrates used in the study of the above-mentioned β-galactosidase, using the carboxylic acid analogue of the previously mentioned carbohydrate nitrile as the key intermediate. An extra synthetic step, deacetylation, was required to prepare this compound in order to avoid glycal formation under the basic conditions employed. Although the chemical hydrolysis of simple organic nitriles has been reported using neutral media, albeit with varying amounts of success,[5] there is room for improvement especially when dealing with such highly functionalized substrates containing additional acid or base sensitive groups.

In this context, we had become interested[6,7] in an immobilized enzyme preparation, "SP409", derived from a *Rhodococcus* sp.[8] This preparation consists of a two-enzyme system, namely a nitrile hydratase and an amidase (Scheme I). Employing this biocatalyst, successful hydrolysis of nonchiral or racemic aliphatic[6] and heteroaromatic[7] nitriles, which contained other base- and/or acid-sensitive groups, was achieved under neutral conditions. The immobilized enzyme was simply shaken with the respective substrate in aqueous phosphate buffer (0.1 M) at room temperature. The application of this enzyme system to sensitive carbohydrate substrates and related multifunctional chirons seemed to be a novel and attractive possibility.

Scheme I

$$\mathrm{RCN} \xrightarrow[\text{hydratase}]{\text{nitrile}} \mathrm{RCONH_2} \xrightarrow{\text{amidase}} \mathrm{RCOOH}$$

Initially, attention was focused on *C*-glycosyl compounds and related structures as these substances are popular synthetic targets and building blocks of natural products.[9] Crystalline 2,3-unsaturated cyano derivatives **1** (73%) and **2** (25%, mp 51–53 °C, $[\alpha]^{20}_D$ +204.9° (c = 2)) were readily obtained[10] via the carbon–Ferrier rearrangement,[11] although it is interesting to note that the previously unreported glycal derivative **3** (0.7%) was also afforded as an additional minor product (Scheme II). These substances, by virtue of the relatively acidic anomeric proton, are all prone to decomposition in basic media.

Treatment of α-configurated nitrile **1** with SP409 resulted in concurrent aglycon hydrolysis and deacetylation affording, as the major product, amide **5** (34%) as well as nitrile **4**[12] (17%). This undesired deprotection of acetylated compounds has been previously encountered with aliphatic nitrile substrates[6] and can be attributed to an ester hydrolase activity present in the enzyme preparation. Repeating the procedure with substrate **4**, obtained from **1** by chemical means, improved the yield of amide **5** (Table I, entry 1). Prolonged reaction times were still required under these conditions. Evidence for the formation of the corresponding carboxylic acid could not be found on the basis of TLC. This was also the case for saturated α-anomer **6**,[13] prepared from compound **1** by hydrogenation[14] (23%) and subsequent deprotection (64%), which also gave the corresponding amide derivative **7** in fair yield (entry 2).

In contrast, β-configurated nitrile **8**, synthesized from **2**, was rapidly converted into carboxylic acid **9** in high

(1) Fischer, E. *Ber. Dtsch. Chem. Ges.* **1889**, *22*, 2204.
(2) (a) Schaffer, R.; Isbell, H. S. *Methods Carbohydr. Chem.* **1962**, *1*, 281. (b) Barker, R.; Serianni, A. S. *Acc. Chem. Res.* **1986**, *19*, 307. (c) Serianni, A. S.; Vuorinen, T.; Bondo, P. B. *J. Carbohydr. Chem.* **1990**, *9*, 513.
(3) Baer, H. H.; Breton, R. L.; Shen, Y. *Carbohydr. Res.* **1990**, *200*, 377.
(4) BeMiller, J. N.; Yadav, M. P.; Kalabokis, V. N.; Myers, R. W. *Carbohydr. Res.* **1990**, *200*, 111.
(5) (a) Bennett, M. A.; Yoshida, T. *J. Am. Chem. Soc.* **1973**, *95*, 3030. (b) Paraskewas, S. *Synthesis*, **1974**, 574. (c) Chin, J.; Kim, J. H. *Angew. Chem., Int. Ed. Engl.* **1990**, *29*, 523. (d) Katritzky, A. R.; Pilarski, B.; Urogoli, L. *Synthesis* **1989**, 949.
(6) de Raadt, A.; Klempier, N.; Faber, K.; Griengl, H. *J. Chem. Soc., Perkin Trans. 1* **1992**, 137.
(7) Klempier, N.; de Raadt, A.; Griengl, H.; Heinisch, G. *J. Heterocycl. Chem.* **1992**, *29*, 93.
(8) Immobilized nitrilase complex (SP409) from Novo Industri A/S, Denmark. The batch used in this study had a given nitrile hydratase activity (one hydratase unit, HPU, is defined as the amount of enzyme which produces one mmol per minute of propionamide from propionitrile under a set of standard conditions) of 391 HPU g^{-1} and an amidase activity (one amidase unit, APU, is defined as the amount of enzyme which produces 1 mmol per minute of ammonium propionate from propionamide under a set of standard conditions) of 159 APU g^{-1}.
(9) The synthesis of palytoxin exemplifies the use of *C*-glycosides as structural subunits: Armstrong, R. W.; Beau, J.-M.; Cheon, S. H.; Christ, W. J.; Fujioka, H.; Ham, W.-H.; Hawkins, L. D.; Jin, H.; Kang, S. H.; Kishi, Y.; Martinelli, M. J.; McWhorter, W. W., Jr.; Mizuno, M.; Nakata, M.; Stütz, A. E.; Talamas, F. X.; Taniguchi, M.; Tino, J. A.; Ueda, K.; Uenishi, J.-i.; White, J. B.; Yonaga, M. *J. Am. Chem. Soc.* **1989**, *111*, 7525.
(10) De Las Heras, F. G.; San Felix, A.; Fernández-Resa, P. *Tetrahedron* **1983**, *39*, 1617.
(11) Danishefsky, S. J.; DeNinno, S.; Lartey, P. *J. Am. Chem. Soc.* **1987**, *109*, 2082.
(12) Tulshian, D. B.; Fraser-Reid, B. *J. Org. Chem.* **1984**, *49*, 518.
(13) Gubernator, K.; Kennard, C. H. L.; Schönholzer, P.; Weller, T. *J. Chem. Soc., Chem. Commun.* **1988**, 14.
(14) Grynkiewicz, G.; BeMiller, J. N. *Carbohydr. Res.* **1982**, *108*, 229.

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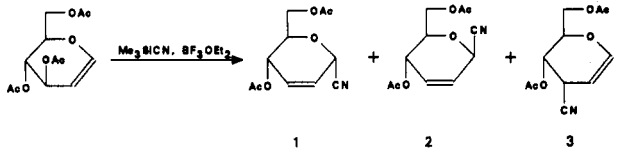
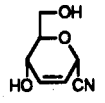
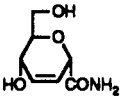
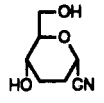
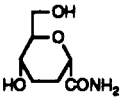
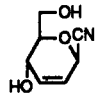
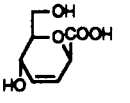
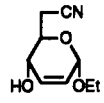
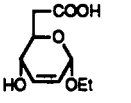
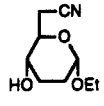
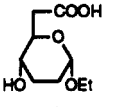
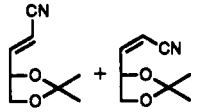
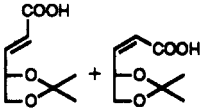
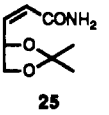
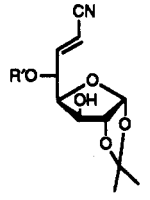
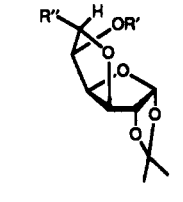
Scheme II



1 2 3

Table I. Hydrolysis of Carbohydrate Nitriles Using a Nitrilase Complex[a]

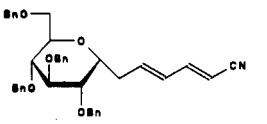
entry	starting material	reactn time, d	product	yield, %
1	 **4**	8	 **5**	54
2	 **6**	12	 **7**	56
3	 **8**	2	 **9**	75
4	 **14**	7	 **19**	90
5	 **18a**	11	 **20**	91
6	 **21** **22**	1	 **23** **24**  **25**	70 8
7	 **26a**, R′ = H	14	 **27a**, R′ = H, R″ = CN **27b**, R′ = H, R″ = $CONH_2$ **27c**, R′ = H, R″ = COOH	

[a] All reactions were carried out at room temperature with SP 409 according to the general procedure.

yield (entry 3). Due to the unusually low frequency[15] of the carbonyl stretch exhibited in the IR spectrum of **9** (1610 cm^{-1}), chemical confirmation of the structure of this compound was obtained upon derivatization into the corresponding methyl ester.[16] Unfortunately, the potentially interesting saturated analogue of **8** could not be obtained from **2**.[17]

Apparently, α-anomers **4** and **6** are not readily accepted by the nitrile hydratase, as indicated by the extended reaction times, and not at all converted by the amidase, leading to the slow formation of only the respective amide. Saturation of the double bond resulted in even longer reaction times. Conversely, the chemically more reactive β-configurated compound **8** was quickly and efficiently transformed by both enzymes affording **9**. This diastereomeric differentiation between compounds **4** and **8** is an interesting phenomenon and as such could warrant further investigation.

A limitation of SP409 is exemplified with *C*-glycosyl compound **10**. Apparently, owing to the lack of solubility of this compound in aqueous phosphate buffer, hydrolysis



10

of the nitrile group was not observed even after prolonged reaction times (28 days). In an effort to improve solubility, a number of cosolvents such as DMSO, DMF, and MeOH were added. This, in turn, caused the deactivation of the enzyme system.[6]

The second set of compounds to be investigated were heptopyranonononitriles **14** and **18a–c**. These substrates are base sensitive owing to the acidic hydrogen atoms α to the nitrile moiety. Their preparation was easily achieved as depicted in Schemes III and IV. A standard deacetylation, tritylation, acetylation, and detritylation[18] sequence was applied to glycosides **11** and **15** in order to obtain the 6-OH-deprotected sugars **12** and **16**, respectively. As the detritylation step was carried out under acidic conditions, acetyl migration from position 4 to 6 could be expected.[19] For saturated sugars **16a**[19] and **16b**, the 6-O-acetylated compound was afforded as a side product. In contrast, in the case of unsaturated sugar **12**[19] no migration product was observed. Introduction of the nitrile group was easily accomplished by using a modified method of Gross et al.[20] Trifluoromethylsulfonylation of the primary alcohol followed by nucleophilic displacement of the triflate with the aid of tetrabutylammonium cyanide afforded the corresponding cyanosugars **13** and **17**. Due to the mild

(15) Pouchert, C. J. *The Aldrich Library of Infrared Spectra*, 3rd ed.; Aldrich Chemical Company: Wisconsin, 1981.

(16) **Methyl (2,3-Dideoxy-β-D-*erythro*-hex-2-enopyranosyl)formate.** To a solution of compound **9** (70 mg, 0.40 mmol) in anhyd MeOH (8 mL) was added IR 120 [H^+], and the mixture was stirred at room temperature. After 5 h the mixture was filtered, the filtrate concentrated, and the resulting residue purified by silica gel chromatography (MeOH/CH_2Cl_2). This afforded the title compound (50 mg, 66%) as a syrup: $[\alpha]^{20}_D$ +175.6° (*c* = 0.4, MeOH); ^1H NMR (DMSO-d_6) spectrum poorly resolved apart from δ 5.15 (d, *J* = 6.5, 1 H, OH*), 4.71 (t, *J* = 5.5, 1 H, OH*) (*D_2O exchangeable); ^1H NMR (DMSO-d_6/D_2O) δ 5.82 (bs, 2 H, H2, H3), 4.79 (d, $J_{1,2}$ = 2.8, 1 H, H1), 3.87 (dd, $J_{3,4}$ = 3.1, $J_{4,5}$ = 9.1, 1 H, H4), 3.69 (bd, $J_{6,6'}$ = 12.4, 1 H, H6′), 3.65 (s, 3 H, $COCH_3$), 3.51 (dd, $J_{5,6}$ = 6.2, 1 H, H6), 3.29 (bdd, 1 H, H5); ^{13}C NMR (DMSO-d_6) δ 169.85 ($COOCH_3$), 132.97, 124.78 (C2, C3), 80.69 (C1), 74.11 (C4), 61.99, 61.44 (C5, C6), 52.24 ($COOCH_3$). Anal. Calcd for $C_8H_{12}O_5$: C, 51.06; H, 6.43. Found: C, 51.02; H, 6.46.

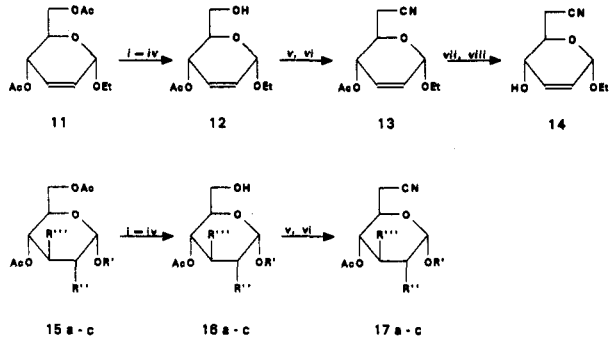
(17) Subjecting β-anomer **8** to the same reaction conditions as those described for α-anomer **1** [H_2 (1 atm), Pd/C, EtOH],[14] in addition to milder conditions [H_2 (0.3 atm), Pd/C, EtOAc], did not afford the desired saturated product.

(18) Dax, K.; Wolflehner, W.; Weidmann, H. *Carbohydr. Res.* **1978**, *65*, 132. For ethyl glycosides **11**, **15a**, and **15b**, an equivalent amount of ethanol was used in place of methanol.

(19) Yunker, M. B.; Fraser-Reid, B. *J. Chem. Soc., Chem. Commun.* **1975**, 61.

(20) Gross, B.; Sekily, M. A. E.; Mancy, S. *Carbohydr. Res.* **1982**, *100*, C31–C32.

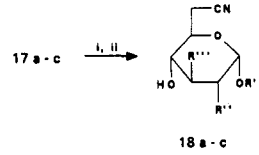
Scheme III[a]



a: R' = Et, R'' = R''' = H
b: R' = Et, R'' = H, R''' = OAc
c: R' = Me; R'' = R''' = OAc

[a] Reagents and conditions: (i) NaOMe, MeOH, rt; IR 120 [H+]; (ii) Ph_3CCl, py, rt; (iii) Ac_2O, py, rt; (iv) BF_3-OEt_2, EtOH, CH_2Cl_2, rt; for 15c MeOH was used in place of EtOH; (v) Tf_2O, py, CH_2Cl_2, 0 °C; (vi) Bu_4NCN, CH_3CN, 0 °C; (vii) NaOMe, MeOH, rt; (viii) IR 120 [H+].

Scheme IV[a]



18 a: R' = Et, R'' = R''' = H
18 b: R' = Et, R'' = H, R''' = OH
18 c: R' = Me, R'' = R''' = OH

[a] Reagents and conditions: (i) NaOMe, MeOH, rt; (ii) IR 120 [H+].

reaction conditions in this step, any sulfonated acetyl migration product present in the reaction mixture remained unreactive. Zemplen deacetylation of compounds **13** and **17** led to the desired urononitriles **14** and **18**, respectively.

Treatment of the new carbohydrate nitriles **14** and **18a** with SP409, the former on gram scale, rapidly gave the respective novel carboxylic acids **19** and **20** in excellent yields (Table I, entries 4 and 5). However, the formal introduction of a hydroxyl group at C-3 or at both C-2 and C-3 (nitriles **18b** and **18c**, respectively) led to completely unreactive compounds. Owing to the fact that the enzyme employed was still active after exposure to these substances, the possibility of irreversible inhibition of the enzyme complex can be ruled out. In addition, solubility problems cannot be a contributing factor. In the study of aliphatic dinitrile hydrolysis it has been found that transformations using SP409 cease at the cyanocarboxylic acid stage.[21a] These very polar compounds are evidently hampered from being converted by, at least, the nitrile hydratase. The nonacceptance of urononitriles **18b** and **18c** by SP409 could also reflect polar as well as steric restriction at the active site. This class of carbohydrates could therefore prove useful for probing the structure of the active site.

The hydrolysis of nitriles in molecules containing both acid- and base-sensitive groups would certainly be the most interesting application of this method. In this context, examples are shown in Table I (entries 6 and 7).

Carbohydrate nitriles **21**, **22**, and **26** are base sensitive by virtue of the relatively acidic allylic proton and the α,β-unsaturated nitrile system being prone to Michael addition. In particular, for D-glyceraldehyde derivatives **21** and **22**, removal of this proton would lead to epimerization at the chiral center with a concomitant reduction or loss of optical activity. Sensitivity to acidic media arises from the isopropylidene protecting group as well as possible addition to the unsaturated nitrile system.

The (*E*)- and (*Z*)-derivatives **21** and **22** (3:1),[22] prepared by the Horner–Emmons reaction of diethyl cyanomethylphosphonate with 2,3-*O*-isopropylidene-D-glyceraldehyde, were easily converted by SP409 to produce a mixture of (*E*)- and (*Z*)-carboxylic acids **23** and **24**[23] in good yields and preparative quantities. As (*Z*)-amide **25** was also afforded, this suggests that (*E*)-isomer **21** was accepted more readily by SP409 than the corresponding (*Z*)-isomer **22**. The optical rotation of **25** pointed to the fact that the chiral integrity of this product had remained intact.[24]

Substrate **26a** only produced, surprisingly, a mixture of bicyclic products **27a–c** after treatment with SP409. On the basis of TLC evidence, conversion of the nitrile moiety had occurred after the nucleophilic addition of O-3 to the unsaturated system, the α,β-unsaturated nitrile **26a** apparently being less reactive than **27a** toward the enzyme complex. In an attempt to prevent this intramolecular reaction from taking place, a bulky protecting group was placed adjacent to the addition center (**26b**, R' = TBDMS).[25] However, partially water-soluble compound **26b** was completely unreactive toward the nitrilase complex. A rationale to explain this interesting behavior has not yet been provided, although steric effects are suspected to be a contributing factor to this outcome.[21b]

In conclusion, SP409 can be successfully applied to a number of acid- and/or base-sensitive carbohydrate-derived nitriles to obtain the corresponding amides or carboxylic acids on a gram scale (**14** and **21**/**22**) in good to excellent yields. Clearly carbohydrate nitriles may also prove useful as "active site probes" due to the variety of structures and stereoisomers accessible in this series of compounds.

Experimental Section

General Methods. Proton and carbon nuclear magnetic resonance spectra were recorded at 300 and 75.47 MHz, respectively, in $CDCl_3$ unless otherwise stated. *J* values are given in Hz. Melting points are uncorrected. Column chromatography was performed on 230–400-mesh silica gel. TLC and HPTLC were performed on TLC and HPTLC silica gel Merck 60 F_{254} plates, respectively. Detection was by UV (254 nm), followed by heating after spraying with a 5% vanillin in concd sulfuric acid solution or a 10% aqueous sulfuric acid solution containing ammonium molybdate (10%) and cerium sulfate hydrate (0.8%). Optical rotations were measured on a digital polarimeter in $CHCl_3$, unless otherwise stated, using a 1-dm cell. Where indicated, reaction mixtures were processed as follows. Procedure 1: diluted

(21) (a) Cohen, M. A.; Sawden, J.; Turner, N. J. *Tetrahedron Lett.* **1990**, *31*, 7223. (b) Ingvorsen, K.; Yde, B.; Godtfredsen, S. E.; Tsuchiya, R. *Ciba Foundation Symposium No. 140*: Cyanide Compounds in Biology, London, 15–17 March 1988.

(22) Buchanan, J. G.; Craven, D. A.; Wightman, R. H.; Harnden, M. R. *J. Chem. Soc., Perkin Trans. 1* **1991**, 195.

(23) Sanchez Ballesteros, J. *Bol. Soc. Quim. Peru* **1980**, *46*, 234; *Chem. Abstr.* **1982**, *97*, 216588.

(24) Leonard, J.; Mohialdin, S.; Swain, P. A. *Synth. Commun.* **1989**, *19*, 3529.

(25) Berger, A.; Dax, K.; Gradnig, G.; Grassberger, V.; Stütz, A. E. *J. Carbohydr. Chem.* **1992**, *11*, 217.

with CH_2Cl_2 (300 mL), washed with 5% aqueous HCl (1 × 250 mL), freshly prepared saturated aqueous $NaHCO_3$ (1 × 250 mL), and water (1 × 250 mL). After filtration, the filtrate was concentrated down under reduced pressure. Procedure 2: as for procedure 1 except the water extraction step was omitted. Organic solutions were dried using Na_2SO_4.

General Procedure for the Enzymatic Hydrolysis of Nitriles. Substrate (100–300 mg, substrates 14 and 21/22 were scaled up to 1.1 g) and immobilized enzyme (1:10, w/w) were suspended in aqueous phosphate buffer (0.1 M, pH = 7.0, 20–50 mL) and the mixture shaken at 200 rpm at rt. After TLC indicated that all starting material had been converted to, in most cases, one major more polar product (CH_3Ph/dioxane/CH_3OH/NH_3 concd (2:5:2:1) was found to be a most suitable solvent system to detect the afforded product/s), the biocatalyst was removed by filtration. The filtrate was evaporated to dryness, resuspended in anhyd toluene, and again evaporated under reduced pressure. This was repeated twice. The resulting residue was stirred with anhyd MeOH (20–40 mL) overnight at rt. The suspension was filtered and the filtrate evaporated in vacuo. The afforded residue was then purified by column chromatography using MeOH/CH_2Cl_2 as eluant unless otherwise stated.

General Procedure for Glycopyranosyl Cyanide Deacetylation. The respective C-1 cyano sugar was dissolved in 0.05 M methanolic hydrochloric acid and the solution refluxed into TLC indicated that all starting material had reacted to give one more polar product. The solution was then cooled to rt, and the volatile components were removed in vacuo. The resulting residue was purified by column chromatography, if necessary, employing MeOH/CH_2Cl_2 as eluant.

4,6-Di-*O*-acetyl-1,5-anhydro-2,3-dideoxy-3-*C*-cyano-D-*ribo*-hex-1-enitol (3): syrup; $[\alpha]^{20}_D$ +72.6° (c = 1); ^1H NMR δ 6.71 (d, $J_{1,2}$ = 6.0 1 H, H1), 5.22 (dd, $J_{2,3}$ = 4.8, 1 H, H2), 4.71 (dd, $J_{3,4}$ = 5.7, 1 H, H3), 4.5 (dd, $J_{5,6'}$ = 2.2, $J_{6,6'}$ = 12.5, 1 H, H6'), 4.25 (dd, $J_{5,6}$ = 5.5, 1 H, H6), 4.19 (dd, $J_{4,5}$ = 10.1, 1 H, H4), 3.43 (ddd, 1 H, H5), 2.10 (s, 3 H, $COCH_3$), 1.8 (s, 3 H, $COCH_3$); ^{13}C NMR δ 170.9 ($COCH_3$), 149.59 (C1), 117.48 (CN), 97.67 (C2), 73.72 (C5), 70.68 (C4), 62.37 (C6), 25.77 (C3), 20.94 ($COCH_3$). Anal. Calcd for $C_{11}H_{13}NO_5$: C, 55.23; H, 5.48; N, 5.85. Found: C, 55.19; H, 5.51; N, 5.90.

(2,3-Dideoxy-α-D-*erythro*-hex-2-enopyranosyl)formamide (5): syrup; $[\alpha]^{20}_D$ −7.7° (c = 3.4, MeOH); IR (film) 1670 cm^{-1}; ^1H NMR (DMSO-d_6) δ 7.46 (s, 1 H, HN*H**), 7.34 (s, 1 H, *H**NH), 5.97 (ddd, $J_{2,4}$ = 1.9, $J_{1,2}$ = 3.0, $J_{2,3}$ = 10.4, 1 H, H2), 5.83 (dd, $J_{3,4}$ = 1.8, 1 H, H3), 5.15 (d, J = 6.2, 1 H, OH*), 4.98 (bs, 1 H, OH*), 4.57 (d, 1 H, H1), 3.78 (bs, 2 H, H4, H6'), 3.47 (bs, 1 H, H6), 3.33 (m, 1 H, H5)(*D_2O exchangeable); ^{13}C NMR (DMSO-d_6) δ 172.46 ($CONH_2$), 131.62, 125.52 (C2, C3), 78.44 (C1), 72.92 (C4), 62.32, 61.49 (C5, C6). Anal. Calcd for $C_7H_{11}NO_4$: C, 48.55; H, 6.41; N, 8.09. Found: C, 48.70; H, 6.50; N, 8.12.

(2,3-Dideoxy-α-D-*erythro*-hexopyranosyl)formamide (7): syrup; $[\alpha]^{20}_D$ +33.5° (c = 0.3, MeOH); IR (film) 1670 cm^{-1}; ^1H NMR (DMSO-d_6) δ 7.35 (s, 1 H, HN*H**), 7.21 (s, 1 H, *H**NH), 4.78 (s, 1 H, OH*), 4.59 (s, 1 H, OH*), 4.15 (dd, $J_{1,2}$ = 2.8, $J_{1,2'}$ = 5.4, 1 H, H1), 3.71 (bd, 1 H, H6), 3.47 (bd, 1 H, H6'), 3.25 (m, 2 H, H4, H5), 2.13 (m, 1 H), 1.79 (m, 1 H), 1.59 (m, 1 H), 1.32 (m, 1 H) (*D_2O exchangeable); ^{13}C NMR (DMSO-d_6) δ 173.39 ($CONH_2$), 80.04 (C1), 72.43 (C4), 65.16, 61.84 (C5, C6), 29.16, 23.85 (C2, C3). Anal. Calcd for $C_7H_{13}NO_4$: C, 47.99; H, 7.49; N, 7.99. Found: C, 47.82; H, 7.50; N, 8.03.

2,3-Dideoxy-β-D-*erythro*-hex-2-enopyranosyl Cyanide (8). Deacetylation of substance 2 (531 mg, 2.2 mmol) using the standard procedure gave glycoside 8 (337 mg, 98%) as a white solid: mp 108–110 °C; $[\alpha]^{20}_D$ +197.6° (c = 1.3, MeOH); ^1H NMR (CD_3OD) δ 6.06 (ddd, $J_{1,2}$ = 2.3, $J_{2,4}$ = 2.3, $J_{2,3}$ = 10.3, 1 H, H2), 5.86 (ddd, $J_{1,3}$ = $J_{3,4}$ = 1.9, 1 H, H3), 5.26 (ddd, $J_{1,4}$ = 2.4, 1 H, H1), 4.09 (dddd, $J_{4,5}$ = 8.6, 1 H, H4), 3.90 (dd, $J_{5,6'}$ = 2.4, $J_{6,6'}$ = 12.2, 1 H, H6'), 3.71 (dd, $J_{5,6}$ = 6.2, 1 H, H6), 3.39 (ddd, 1 H, H5); ^{13}C NMR (CD_3OD) δ 134.74, 123.99 (C2, C3), 118.36 (CN), 81.97 (C1), 64.95, 63.13, 62.88 (C4, C5, C6). Anal. Calcd for $C_7H_9NO_3$: C, 54.19; H, 5.85; N, 9.03. Found: C, 54.25; H, 5.91; N, 9.06.

(2,3-Dideoxy-β-D-*erythro*-hex-2-enopyranosyl)formic acid (9): mp 138–141 °C; $[\alpha]^{20}_D$ +192.9°, (c = 1.0, MeOH); IR (film) 1610 cm^{-1}; ^1H NMR (DMSO-d_6) spectrum poorly resolved apart from δ 5.3 (bs, 1 H, COOH, D_2O exchangeable), 4.34 (bd, 1 H, H1); ^1H NMR (DMSO-d_6/D_2O) δ 5.88 (ddd, $J_{1,2}$ = $J_{2,4}$ = 1.8, $J_{2,3}$ = 10.1, 1 H, H2), 5.66 (ddd, $J_{1,3}$ = $J_{3,4}$ = 2.3, 1 H, H3), 3.96 (m, 1 H, H4), 3.67 (dd, $J_{5,6'}$ = 2.5, $J_{6,6'}$ = 12.0, 1 H, H6'), 3.56 (dd, $J_{5,6}$ = 4.7, 1 H, H6), 3.26 (ddd, $J_{4,5}$ = 8.5, 1 H, H5); ^{13}C NMR (DMSO-d_6) δ 173.01 (COOH), 129.59, 128.81 (C2, C3), 79.03 (C1), 76.69 (C4), 61.71, 60.16 (C5, C6). Anal. Calcd for $C_7H_{10}O_5$: C, 48.28; H, 5.79. Found: C, 48.22; H, 5.83.

6-(2',3',4',6'-Tetra-*O*-benzyl-α-D-glucopyranosyl)hexa-2-(*E*),4(*E*)-dienonitrile (10). Under nitrogen, NaH (78 mg, 3.3 mmol) was added to a clear, colorless solution of diethyl cyanomethylphosphonate (640 mg, 3.6 mmol) in anhyd THF (8 mL) at rt. After being stirred for 1 h, this clear yellow solution was added dropwise over 10 min to a stirred mixture of 4-(2',3',4',6'-tetra-*O*-benzyl-α-D-glucopyranosyl)but-2(*E*)-enal[26] (1.07 g, 1.8 mmol) and anhyd THF (20 mL) at 0 °C under nitrogen. After 10 min the reaction mixture was treated according to workup procedure 1. Purification by chromatography (hexane/EtOAc) gave a mixture of (*E*)-isomer 10 and (*Z*)-isomer (3:1, 710 mg, 64%). Careful separation on HPTLC plates afforded the individual isomers. (*E*)-Isomer 10: white crystalline solid; mp 102–104 °C; $[\alpha]^{20}_D$ +78.2° (c = 1.1, $CHCl_3$); IR (film) 2215 cm^{-1}; ^1H NMR δ 7.37–7.14 (m, 20 H, $OCH_2C_6H_5$), 6.92 (dd, J = 16.1, J = 10.1, 1 H), 6.14 (m, 2 H), 5.17 (d, 1 H), 4.96–4.47 (m, 8 H, $OCH_2C_6H_5$), 4.16 (m, 1 H), 3.79–3.62 (m, 6 H), 2.61 (m, 2 H); ^{13}C NMR δ 150.51, 141.39, 138.85, 138.32, 138.25, 130.13, 128.73, 128.64, 128.15, 128.07, 127.90, 118.38 (C1), 97.52, 82.45, 80.09, 78.34, 77.69, 75.62, 75.27, 73.80, 73.64, 71.93, 69.48 (C1', C2', C3', C4', C5', C6', $OCH_2C_6H_5$), 29.40 (C6). Anal. Calcd for $C_{40}H_{41}NO_5$: C, 78.02; H, 6.72; N, 2.27. Found: C, 78.00; H, 6.80; N, 2.21.

(*Z*)-Isomer: syrup; $[\alpha]^{20}_D$ +46.9° (c = 0.3, $CHCl_3$); IR (film) 2215 cm^{-1}; ^1H NMR δ 7.33–7.13 (m, 20 H, $OCH_2C_6H_5$), 6.75 (dd, $J_{2,3}$ = $J_{3,4}$ = 10.9, 1 H), 6.61 (dd, $J_{4,5}$ = 14.7, 1 H, H4), 6.16 (ddd, $J_{5,6}$a = $J_{5,6}$b = 7.1, 1 H, H5), 5.11 (d, 1 H, H2), 4.96–4.45 (m, 8 H), 4.12 (m, 1 H), 3.78–3.57 (m, 6 H), 2.64 (m, 2 H); ^{13}C NMR δ 149.30, 141.83, 138.24, 128.76, 128.62, 128.49, 127.97, 127.72, 116.66 (C1), 95.63, 82.33, 80.01, 78.19, 76.68, 75.51, 75.08, 73.79, 73.57, 71.77, 69.23, 29.44 (C6). Anal. Calcd for $C_{40}H_{41}NO_5$: C, 78.02; H, 6.72; N, 2.27. Found: C, 77.95; H, 6.77; N, 2.30.

Ethyl 3,4-Di-*O*-acetyl-2-deoxy-α-D-*arabino*-hexopyranoside (16b). Derivative 15b[27] (260 mg, 0.82 mmol) yielded, after chromatography, glycoside 16b (130 mg, 58%, based on ^1H NMR 16b/acetyl migration product (6:1)): syrup; $[\alpha]^{20}_D$ +111.8° (c = 1.0, CH_2Cl_2); ^1H NMR δ 5.37 (ddd, $J_{2a,3}$ = 5.3, $J_{2b,3}$ = 11.6, $J_{3,4}$ = 9.6, 1 H, H3), 4.92 (m, 2 H, H1, H4), 3.77–3.38 (m, 5 H, H5, H6, H6', OCH_2CH_3), 2.45 (bs, 1 H, OH, exchangeable with D_2O), 2.22 (dd, $J_{2a,2b}$ = 12.8, 1 H, H2a), 2.06 (s, 3 H, $COCH_3$), 2.00 (s, 3 H, $COCH_3$), 1.76 (ddd, $J_{1,2b}$ = 3.6, 1 H, H2b), 1.19 (t, J = 7.1, OCH_2CH_3); ^{13}C NMR δ 171.08, 170.34 ($COCH_3$), 96.90 (C1), 70.38, 70.15, 69.22, (C3, C4, C5), 63.27 (C6), 61.71 (OCH_2CH_3), 35.48 (C2), 21.15 ($COCH_3$), 20.93 ($COCH_3$), 15.16 (OCH_2CH_3).

General Procedure. Preparation of Ethyl 4-*O*-Acetyl-2,3,6-trideoxy-α-D-*erythro*-hept-2-enopyranurononitrile (13). Step 1: Trifluoromethyl Sulfonylation. To a clear, colorless, ice-cooled solution of the 6-OH deprotected *O*-glycoside 12 (1.36 g, 6.3 mmol) in anhyd CH_2Cl_2 (70mL)/pyridine(7 mL) was added triflic anhydride (9.4 mmol, 1.5 mL) and the mixture stirred under an atmosphere of nitrogen. After 5 min, TLC showed that all starting material had been converted to one less polar product. The orange reaction mixture was treated according to workup procedure 2. The organic phase was then dried, concentrated down to ~15 mL, and used immediately for the next step.

Step 2: Introduction of Nitrile Moiety. To the crude triflated product was added anhyd CH_3CN (90 mL) and the solution cooled in an ice bath, with stirring, under nitrogen. Subsequently, tetrabutylammonium cyanide (3.4 g, 12.6 mmol) was added. TLC indicated that after 5 min all triflated product had been converted in to a single more polar product. The reaction mixture was diluted with CH_2Cl_2 (300 mL) and washed with water (4 × 200 mL) until the aqueous phase was no longer

(26) 4-(2',3',4',6'-Tetra-*O*-benzyl-α-D-glucopyranosyl)but-2(*E*)-enal was kindly donated by Mr. A. Berger.

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basic (pH paper). The organic phase was dried (Na_2SO_4), concentrated, and purified by chromatography to give 0.72 g (51%) of **13** as a white solid: mp 38–39 °C; $[\alpha]^{20}_D$ +124.7° (c = 1.5, CH_2Cl_2); IR (film) 2255 cm^{-1}; ^{1}H NMR δ 5.84 (s, 2 H, H2, H3), 5.16 (d, $J_{4,5}$ = 9.3, 1 H, H4), 5.04 (s, 1 H, H1), 4.15 (ddd, $J_{5,6}$ = 8.0, $J_{5,6'}$ = 4.2, 1 H, H5), 3.91 (m, 1 H, OCH_2CH$_3$) 3.58 (m, 1 H, OCH_2CH$_3$), 2.67 (dd, $J_{6,6'}$ = 16.8, 1 H, H6'), 2.56 (dd, 1 H, H6), 2.09 (s, 3 H, COCH$_3$), 1.25 (t, J = 7.1, 3 H, OCH$_2$CH_3); ^{13}C NMR δ 170.45 (*C*OCH$_3$), 128.67, 128.45 (C2, C3), 116.83 (CN), 94.47 (C1), 68.74, 65.27, 64.75 (C4, C5, OCH_2CH$_3$), 21.76, 21.13 (C6, CO*C*H$_3$), 15.41 (OCH$_2$*C*H$_3$). Anal. Calcd for $C_{11}H_{15}NO_4$: C, 58.65; H, 6.72; N, 6.22. Found: C, 58.60; H, 6.72; N, 6.29.

Ethyl 4-*O*-Acetyl-2,3,6-trideoxy-α-D-*erythro*-heptopyranurononitrile (17a). Compound **16a** (1.08g, 4.9 mmol) was submitted to the general procedure above to give nitrile **17a** (0.62 g, 55%): syrup; $[\alpha]^{20}_D$ +124.8° (c = 0.7, CH_2Cl_2); IR (film) 2254 cm^{-1}; ^{1}H NMR δ 4.84 (s, 1 H, H1), 4.58 (m, 1 H, H4), 3.99 (ddd, $J_{4,5}$ = 9.9, $J_{5,6'}$ = 4.0, $J_{5,6}$ = 7.8, 1 H, H5), 3.77 (m, 1 H, OCH_2CH$_3$), 3.51 (m, 1 H, OCH_2CH$_3$), 2.62 (dd, $J_{6,6'}$ = 16.7, 1 H, H6'), 2.49 (dd, 1 H, H6), 2.08 (s, 3 H, COCH$_3$), 2.01–1.79 (m, 4 H, H2, H2', H3, H3'), 1.24 (t, J = 7.0, OCH$_2$CH_3); ^{13}C NMR δ 170.18 (*C*OCH$_3$), 117.16 (CN), 96.45 (C1), 71.64, 66.92, 63.20 (C4, C5, OCH$_2$CH$_3$), 29.10, 24.15 (C2, C3), 21.67, 21.25 (C6, CO*C*H$_3$), 15.26 (OCH$_2$*C*H$_3$). Anal. Calcd for $C_{11}H_{17}NO_4$: C, 58.13; H, 7.55; N, 6.16. Found: C, 58.10; H, 7.51; N, 6.10.

Ethyl 3,4-Di-*O*-acetyl-2,6-dideoxy-α-D-*arabino*-heptopyranurononitrile (17b). Following the standard procedure above, compound **16b** (90 mg, 0.33 mmol) afforded nitrile **17b** (50 mg, 54%): mp 87–89 °C; $[\alpha]^{20}_D$ +110.5° (c = 2.1, CH_2Cl_2); IR (film) 2255 cm^{-1}; ^{1}H NMR δ 5.30 (ddd, $J_{2\alpha,3}$ = 5.3, $J_{2\beta,3}$ = 11.7, $J_{3,4}$ = 9.5, 1 H, H3), 4.94 (d, $J_{1,2\alpha}$ = 3.2, 1 H, H1), 4.81 (dd, $J_{4,5}$ = 9.6, 1 H, H4), 4.00 (ddd, $J_{5,6}$ = 7.2, $J_{5,6'}$ = 4.8, 1 H, H5), 3.72 (m, 1 H, OCH_2CH$_3$), 3.48 (m, 1 H, OCH_2CH$_3$), 2.59 (dd, $J_{6,6'}$ = 16.7, 1 H, H6'), 2.51 (dd, 1 H, H6), 2.22 (dd, $J_{2\alpha,2\beta}$ = 13.5, 1 H, H2α), 2.06 (s, 3 H, COCH$_3$), 1.99 (s, 3 H, COCH$_3$), 1.79 (ddd, 1 H, H2β), 1.21 (t, J = 7.1 Hz, 3 H, OCH$_2$CH_3); ^{13}C NMR δ 170.27, 170.18 (*C*OCH$_3$), 116.61 (CN), 96.91 (C1), 73.07, 68.73, 66.01 (C3, C4, C5), 63.66 (OCH_2CH$_3$), 35.34 (C2), 21.51, 21.05, 20.89 (C6, CO*C*H$_3$), 15.11 (OCH$_2$*C*H$_3$). Anal. Calcd for $C_{13}H_{19}NO_6$: C, 54.73; H, 6.72; N, 4.91. Found: C, 54.79; H, 6.78; N, 4.85.

Ethyl 2,3,6-Trideoxy-α-D-*erythro*-hept-2-enopyranurononitrile (14). Zemplen deacetylation of substance **13** (0.60 g, 2.7 mmol) gave substrate **14** (0.47 g, 96%): syrup; $[\alpha]^{20}_D$ +57.9° (c = 4.5, CH_2Cl_2); IR (film) 2252 cm^{-1}; ^{1}H NMR δ 5.87 (d, $J_{2,3}$ = 10.2, 1 H, H2$^+$), 5.71 (dd, J = 1.9, 1 H, H3$^+$), 4.96 (s, 1 H, H1), 3.96–3.77 (m, 3 H, H4, H5, OCH_2CH$_3$), 3.53 (m, 1 H, OCH_2CH$_3$), 3.26 (d, J = 7.8, 1 H, OH, exchangeable with D_2O), 2.81 (dd, $J_{5,6'}$ = 3.3, $J_{6,6'}$ = 16.9, 1 H, H6'), 2.63 (dd, $J_{5,6}$ = 7.2, 1 H, H6), 1.19 (t, J = 7.1, 3 H, OCH$_2$CH_3) (*could be interchanged); ^{13}C NMR δ 132.86, 126.70 (C2, C3), 117.67 (CN), 94.32 (C1), 67.86, 66.66, 64.43 (C4, C5, OCH$_2$CH$_3$), 21.21 (C6), 15.30 (OCH$_2$*C*H$_3$). Anal. Calcd for $C_9H_{13}NO_3$: C, 59.00; H, 7.16; N, 7.64. Found: C, 58.96; H, 7.18; N, 7.70.

Ethyl 2,3,6-Trideoxy-α-D-*erythro*-heptopyranurononitrile (18a). Deacetylation of compound **17a** (570 mg, 2.5 mmol) under Zemplen conditions yielded substrate **18a** (390 mg, 84%): syrup; $[\alpha]^{20}_D$ +134.0° (c = 1.1, CH_2Cl_2); IR (film) 2257 cm^{-1}; ^{1}H NMR δ 4.77 (d, $J_{1,2}$ = 2.2, 1 H, H1), 3.77–3.63 (m, 2 H, H4, OCH_2CH$_3$), 3.50–3.35 (m, 2 H, H5, OCH_2CH$_3$), 2.77 (dd, $J_{5,6'}$ = 3.5, $J_{6,6'}$ = 16.9, 1 H, H6'), 2.71 (bs, 1 H, OH, exchangeable with D_2O), 2.62 (dd, $J_{5,6}$ = 6.9, 1 H, H6), 1.89–1.71 (m, 4 H, H2, H2', H3, H3'), 1.19 (t, J = 7.2, 3 H, OCH$_2$CH_3); ^{13}C NMR δ 117.93 (CN), 96.26 (C1), 69.38, 69.27 (C4, C5), 62.87 (OCH_2CH$_3$), 29.44, 27.62 (C2, C3), 21.21 (C6), 15.16 (OCH$_2$*C*H$_3$). Anal. Calcd for $C_9H_{15}NO_3$: C, 58.36; H, 8.17; N, 7.56. Found: C, 58.31; H, 8.25; N, 7.52.

Ethyl 2,6-Dideoxy-α-D-*arabino*-heptopyranurononitrile (18b). Zemplen deacetylation of glycoside **17b** (45 mg, 0.15 mmol) furnished compound **18b** (30 mg, 95%): syrup; $[\alpha]^{20}_D$ +119.5° (c = 1, MeOH); IR (film) 2257 cm^{-1}; ^{1}H NMR δ 4.25 (bs, 2 H, OH, exchangeable with D_2O); ^{1}H NMR ($CDCl_3/D_2O$) δ 4.92 (d, $J_{1,2\beta}$ = 3.4, 1 H, H1), 3.95 (ddd, $J_{2\alpha,3}$ = 5.0, $J_{2\beta,3}$ = 11.6, $J_{3,4}$ = 9.0, 1 H, H3), 3.75 (m, 2 H, H5, OCH_2CH$_3$), 3.45 (m, 1 H, OCH_2CH$_3$), 3.29 (dd, $J_{4,5}$ = 9.2, 1 H, H4), 2.82 (dd, $J_{5,6'}$ = 3.6, $J_{6,6'}$ = 16.8, 1 H, H6'), 2.68 (dd, $J_{5,6}$ = 6.8, 1 H, H6), 2.13 (dd, $J_{2\alpha,2\beta}$ = 12.5, 1 H, H2α), 1.71 (ddd, 1 H, H2β), 1.20 (t, J = 7.0, 3 H, OCH$_2$CH_3); ^{13}C NMR δ 117.87 (CN), 97.55 (C1), 75.23, 69.25, 67.69 (C3, C4, C5), 63.43 (OCH_2CH$_3$), 37.96 (C2), 21.27 (C6), 15.22 (OCH$_2$*C*H$_3$). Anal. Calcd for $C_9H_{15}NO_4$: C, 53.72; H, 7.52; N, 6.96. Found: C, 53.68; H, 7.56; N, 7.00.

Methyl 6-Deoxy-α-D-*gluco*-heptopyranurononitrile (18c). Deacetylation of glucoside **17c** (100 mg, 0.30 mmol) under Zemplen conditions afforded nitrile **18c** (58 mg, 94%): syrup; $[\alpha]^{20}_D$ +138.1° (c = 1.4, MeOH); IR (film) 2253 cm^{-1}; ^{1}H NMR (CD_3OD) δ 4.73 (d, $J_{1,2}$ = 3.8, 1 H, H1), 3.73 (m, 1 H, H5), 3.62 (dd, $J_{3,4}$ = $J_{4,5}$ = 9.3, 1 H, H4$^+$), 3.44 (m, 4 H, H2, OCH$_3$), 3.20 (dd, $J_{2,3}$ = 9.3, 1 H, H3$^+$), 2.93 (dd, $J_{5,6'}$ = 3.2, $J_{6,6'}$ = 17.0, 1 H, H6'), 2.74 (dd, $J_{5,6}$ = 7.0, 1 H, H6); ^{13}C NMR (CD_3OD) δ 119.08 (CN), 101.78 (C1), 74.95, 74.83, 73.68, 69.17 (C2, C3, C4, C5), 56.06 (OCH$_3$), 21.51 (C6). Anal. Calcd for $C_8H_{13}NO_5$: C, 47.29; H, 6.45; N, 6.89. Found: C, 47.18; H, 6.50; N, 6.95.

Ethyl 2,3,6-trideoxy-α-D-*erythro*-hept-2-enopyranuronic acid (19): syrup; $[\alpha]^{20}_D$ +62.4° (c = 1.4, MeOH); IR (film) 1715 cm^{-1}; ^{1}H NMR (DMSO-d_6) δ 5.85 (d, $J_{2,3}$ = 10.1, 1 H, H2$^+$), 5.70 (bs, 2 H, OH, COOH, D_2O exchangeable) 5.66 (d, 1 H, H3$^+$), 4.89 (s, 1 H, H1), 3.91 and 3.77 (m, 3 H, H4, H5, OCH_2CH$_3$), 3.42 (m, 1 H, OCH_2CH$_3$), 2.71 (dd, $J_{5,6'}$ = 3.4, $J_{6,6'}$ = 15.5, 1 H, H6'), 2.25 (dd, $J_{5,6}$ = 8.7, 1 H, H6), 1.14 (t, J = 7.0, 3 H, OCH$_2$CH_3) (*could be interchanged); ^{13}C NMR (DMSO-d_6) δ 174.61 (C7), 134.11, 125.88 (C2, C3), 93.44 (C1), 68.91, 66.66, 62.96 (C4, C5, OCH$_2$CH$_3$), 39.48 (C6), 15.35 (OCH$_2$*C*H$_3$). Anal. Calcd for $C_9H_{14}O_5$: C, 53.46; H, 6.98. Found: C, 53.41; H, 7.02.

Ethyl 2,3,6-trideoxy-α-D-*erythro*-heptopyranuronic acid (20): syrup; $[\alpha]^{20}_D$ +115.1 (c = 2.6, CH_2Cl_2); IR (film) 1715 cm^{-1}; ^{1}H NMR δ 6.60 (bs, 2 H, OH, COOH, D_2O exchangeable), 4.71 (s, 1 H, H1), 3.93 (ddd, $J_{4,5}$ = $J_{5,6}$ = 8.9, $J_{5,6'}$ = 3.5, 1 H, H5), 3.74 (m, 1 H, OCH_2CH$_3$), 3.37 (m, 2 H, H4, OCH_2CH$_3$), 2.85 (dd, $J_{6,6'}$ = 15.5, 1 H, H6'), 2.40 (dd, 1 H, H6), 1.76 (m, 4 H, H2, H2', H3, H3'), 1.17 (t, 3 H, J = 7.1, OCH$_2$CH_3); ^{13}C NMR δ 176.74 (C7), 95.87 (C1), 70.27, 70.07 (C4, C5), 62.55 (OCH$_2$CH$_3$), 37.99 (C6), 29.59, 27.57 (C2, C3), 15.06 (OCH$_2$*C*H$_3$). Anal. Calcd for $C_9H_{16}O_5$: C, 52.93; H, 7.90. Found: C, 52.87; H, 7.95.

(*Z*)-2,3-Dideoxy-4,5-*O*-isopropylidene-D-*glycero*-pent-2-enonamide (25): mp 63.0–65.0 °C; $[\alpha]^{20}_D$ +140.4° (c = 0.3); IR (film) 1674 cm^{-1}; ^{1}H NMR (DMSO-d_6) δ 7.59 (s, 1 H, *H**NH), 7.12 (s, 1 H, HN*H**), 6.08 (dd, $J_{2,3}$ = 11.5, $J_{3,4}$ = 6.5, 1 H, H3), 5.92 (d, 1 H, H2), 5.50 (dd, $J_{4,5}$ = $J_{4,5'}$ = 6.7, 1 H, H4), 4.23 (dd, $J_{5,5'}$ = 7.0, 1 H, H5), 3.50 (dd, 1 H, H5'), 1.38 (s, 3 H, isop.), 1.32 (s, 3 H, isop.)(*exchangeable with D_2O); ^{13}C NMR (DMSO-d_6) δ 168.80 (C1), 144.97 (C3), 124.86 (C2), 110.25 (isop.), 73.79 (C4), 69.91 (C5), 27.37, 26.34 (isop.). Anal. Calcd for $C_8H_{13}NO_3$: C, 56.12; H, 7.66; N, 8.18. Found: C, 56.05; H, 7.70; N, 8.10.

(*E*)-6,7-Dideoxy-1,2-*O*-isopropylidene-α-D-*gluco*-oct-6-enofuranurononitrile (26a): white crystalline solid: mp 125–126 °C; $[\alpha]^{20}_D$ +23.1° (c = 0.7); IR (film) 2228 cm^{-1}; ^{1}H NMR δ 6.91 (dd, $J_{5,6}$ = 3.4, $J_{6,7}$ = 16.1, 1 H, H6), 5.99 (d, $J_{1,2}$ = 3.6, 1 H, H1), 5.86 (dd, $J_{5,7}$ = 2.1, 1 H, H7), 4.77 (m, 1 H, H5), 4.52 (d, 1 H, H2), 4.33 (d, $J_{3,4}$ = 2.7 Hz, 1 H, H3), 4.07 (dd, $J_{4,5}$ = 6.1, 1 H, H4), 3.30 (bs, 2 H, OH, exchangeable with D_2O), 1.49 (s, 3 H, isop.), 1.33 (s, 3 H, isop.); ^{13}C NMR δ 152.41 (C6), 117.19 (C8), 112.50 (isop.), 105.33 (C1), 100.98 (C7), 85.49 , 80.93, 75.78, 69.94 (C2, C3, C4, C5), 27.03, 26.41 (isop.). Anal. Calcd for $C_{11}H_{15}NO_5$: C, 54.76; H, 6.27; N, 5.81. Found: C, 54.70; H, 6.30; N, 5.75.

3,6-Anhydro-7-deoxy-1,2-*O*-isopropylidene-α-D-*glycero*-D-*gluco*-octofuranurononitrile (27a): mp 105–107 °C; $[\alpha]^{20}_D$ +56.0° (c = 1.6, CH_2Cl_2); IR (film) 2257 cm^{-1}; ^{1}H NMR (assignments confirmed with 2D NMR experiments) δ 5.94 (d, $J_{1,2}$ = 3.5, 1 H, H1), 4.81 (dd, $J_{3,4}$ = 3.8, $J_{4,5}$ = 3.8, 1 H, H4), 4.68 (d, 1 H, H3), 4.64 (d, 1 H, H2), 3.95 (dd, $J_{5,6}$ = 8.5, 1 H, H5), 3.75 (ddd, $J_{6,7}$ = $J_{6,7'}$ = 4.4, 1 H, H6), 2.82 (dd, $J_{7,7'}$ = 17.1, H7'), 2.70 (bs, 1 H, OH, exchangeable with D_2O), 2.63 (dd, 1 H, H7), 1.49 (s, 3 H, isop.), 1.34 (s, 3 H, isop.); ^{13}C NMR δ 116.67 (C8), 113.42 (isop.), 107.00 (C1), 85.68, 84.71, 82.48 (C2, C3, C4), 77.67, 76.50 (C5, C6), 27.58, 26.97 (isop.), 21.43 (C7). Anal. Calcd for $C_{11}H_{15}NO_5$: C, 54.76; H, 6.27; N, 5.81. Found: C, 54.72; H, 6.32; N, 5.75.

3,6-Anhydro-7-deoxy-1,2-*O*-isopropylidene-α-D-*glycero*-D-*gluco*-octofuranurononamide (27b): syrup; $[\alpha]^{20}_D$ +45.1° (c = 0.6, MeOH); IR (film) 1655 cm^{-1}; ^{1}H NMR δ 6.12 (s, 1 H, HN*H**), 5.97 (d, $J_{1,2}$ = 2.9, 1 H, H1), 5.83 (s, 1 H, *H**NH), 4.81 (s, 1 H, H4), 4.62 (m, 2 H, H2, H3), 3.86 (bs, 3 H, H5, H6, OH*), 2.66 (dd, $J_{6,7'}$ = 3.5, $J_{7,7'}$ = 15.0, 1 H, H7'), 2.48 (dd, $J_{6,7}$ = 6.9, 1 H, H7),

1.49 (s, 3 H, isop.), 1.34 (s, 3 H, isop.)(*exchangeable with D_2O); ^{13}C NMR (acetone-d_6) δ 173.70 (C8), 112.60 (isop.), 107.72 (C1), 86.40, 84.71, 83.62 (C2, C3, C4), 78.80, 77.76 (C5, C6), 39.89 (C7), 27.58, 26.99 (isop.). Anal. Calcd for $C_{11}H_{17}NO_6$: C, 50.96; H, 6.62; N, 5.40. Found: C, 51.00; H, 6.68; N, 5.39.

3,6-Anhydro-7-deoxy-1,2-*O*-isopropylidene-α-D-*glycero*-D-*gluco*-octofuranuronic acid (27c): syrup; $[\alpha]^{20}_D$ +24.4° (*c* = 2.1, MeOH); IR (film) 1720 cm^{-1}; ^1H NMR (DMSO-d_6) δ 3.61–3.50 (bs, 2 H, OH, COOH, D_2O exchangeable); ^1H NMR (DMSO-d6/D_2O) δ 5.92 (d, $J_{1,2}$ = 3.5, 1 H, H1), 4.62 (dd, $J_{3,4} = J_{4,5}$ = 3.4, 1 H, H4), 4.52 (d, 1 H, H3), 4.42 (d, 1 H, H2), 3.84 (m, 1 H, H6), 3.69 (m, 1 H, H5), 2.37 (dd, $J_{6,7'}$ = 5.3, $J_{7,7'}$ = 15.8, 1 H, H7'), 2.26 (dd, $J_{6,7}$ = 7.3, 1 H, H7), 1.40 (s, 3 H, isop.), 1.27 (s, 3 H, isop.); ^{13}C NMR (DMSO-d_6) δ 176.94 (C8), 111.61 (isop.), 106.66 (C1), 85.15, 83.47, 83.27 (C2, C3, C4), 78.28, 76.74 (C5, C6), 41.54 (C7), 27.54, 27.09 (isop.). Anal. Calcd for $C_{11}H_{16}O_7$: C, 50.77; H, 6.20. Found: C, 50.69; H, 6.23.

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Supplementary Material Available: ^1H and ^{13}C NMR spectra for compounds **4**, **6**, **12**, **16a**, **17c**, **21**, **22**, **23**, and **24** (4 pages). This material is contained in libraries on microfiche, immediately follows this article in the microfilm version of the journal, and can be ordered from the ACS; see any current masthead page for ordering information.