156. Deoxy-nitrosugars

16th Communication[1])

Synthesis of *N*-Acetyl-4-deoxyneuraminic Acid

by **Franz Baumberger** and **Andrea Vasella***

Organisch-Chemisches Institut der Universität Zürich, Winterthurerstrasse 190, CH-8057 Zürich

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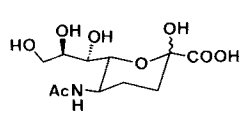
The synthesis of 5-acetamido-4-deoxyneuraminic acid (**1**) is described. Acetylation of a mixture of the epimeric triols **4** and **5** gave the tetraacetates **7** and **8** *(Scheme 1)*. Ozonolysis of a mixture of these acetates followed by base-promoted β-elimination led to the *(E)*-configurated α,β-unsaturated keto ester **10**, which was hydrogenated to give the saturated keto ester **11**. Saponification of **11** and hydrolytic removal of the benzylidene group followed by anion-exchange chromatography gave the 5-acetamido-4-deoxyneuraminic acid (**1**, *Scheme 1* and *2*). De-*O*-acetylation (NaOMe/MeOH) of the keto ester **11** gave a mixture of the *tert*-butyl ester **12** and the methyl ester **13**, which were converted to *tert*-butyl *N*-acetyl-4-deoxyneuraminate (**14**) and to methyl *N*-acetyl-4-deoxyneuraminate (**15**), respectively. Hydrogenolysis of the benzylidene acetal **11** followed by de-*O*-acetylation gave the pentahydroxy ester **16**.

Introduction. – Although the function of neuraminidases and the effect of sialic acids, their derivatives, and their glycosides upon these enzymes have been extensively studied [2–7], little is known about the detailed mechanism of their action. *Schauer et al.* [8] have shown that glycosides of *N*,4-*O*-diacetylneuraminic acid and *N*-acetyl-4-*O*-methylneuraminic acid were strongly resistant to mammalian and bacterial sialidases, and *Flashner et al.* [9] have shown that *N*-acetyl-2-deoxy-4-epineuraminic acid, *N*-acetyl-2,3-dehydro-4-epineuraminic acid, and *N*-acetyl-2,3-dehydro-4-oxoneuraminic acid are competitive inhibitors of *Arthrobacter sialophilus* neuraminidase and of influenza virus neuraminidase. These results indicate an important role for the C(4) OH group.

We, therefore, wished to prepare the *N*-acetyl-4-deoxyneuraminic acid (**1**) exploiting our synthesis of *N*-acetylneuraminic acid (**2**) and *N*-acetyl-4-epineuraminic acid (**3**) [1]. Intermediates of this synthesis, which appear to be suitable candidates for a deoxygenation at C(4) are the epimeric alcohols **4** and **5** *(Scheme 1)*.



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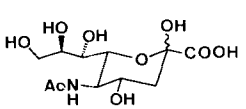

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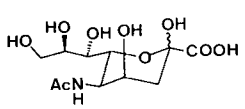

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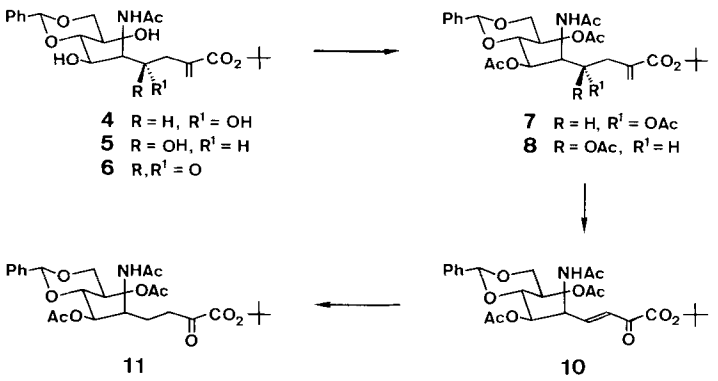
[1]) 15th Communication: [1].

Scheme 1



Results. – Acetylation (Ac_2O/pyridine 1:2) of a crude 1:3 mixture of the triols **4** and **5** [1], obtained by $NaBH_4$ reduction of the ketone **6**, gave the tetraacetates **7** and **8**[2]) *(Scheme 1)*. Pure samples of the tetraacetates **7** and **8** were obtained by chromatography and also by acetylation of the individual triols **4** and **5**. Ozonolysis of a mixture of the tetraacetates **7** and **8** in $CHCl_3$ at −60° and in the presence of $NaHCO_3$ followed by addition of $(i\text{-}Pr)_2EtN$ gave the *(E)*-configurated α,β-unsaturated keto ester **10**. Its IR spectrum is characterized by absorptions at 1704 (C=O), 1678 (NHAc), and 1630 (C=C), and its ^{1}H-NMR spectrum by $J(3,4) = 16$ Hz. Hydrogenation of **10** (10% PdC, AcOEt) gave the saturated keto ester **11**[3]).

Saponification of the keto ester **11** with K_2CO_3 in aq. MeOH *(Scheme 2)* followed by acid hydrolysis (*Dowex 50* (H^+), aq. dioxane), and purification of the resulting crude *N*-acetyl-4-deoxyneuraminic acid (**1**) by anionexchange chromatography (*Dowex 1* ($HCOO^-$)) gave **1** as a microcrystalline solid (53% from **7**/**8**).

The ^{1}H-NMR spectrum of **1** showed only signals of the β-D-anomer. Comparison of the ^{1}H-NMR spectrum of **1** with the one of *N*-acetylneuraminic acid (**2**) [1] [12] showed similar chemical shifts for H–C(6) to CH_2(9) ($\Delta\delta = \pm 0.1$ ppm) and the same coupling constants with the exception of $J(6,7)$, where a difference of 1 Hz is noted.

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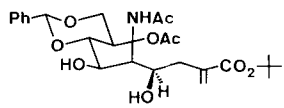
[2]) Besides **7** and **8**, we obtained 10–20% of the mono-*O*-acetate **9**, which was not converted to the tetraacetate **8** even upon addition of a large excess of Ac_2O/pyridine or in the presence of 4-(dimethylamino)pyridine



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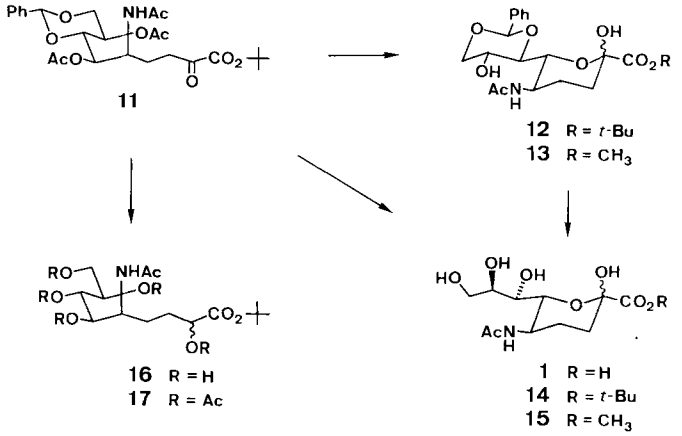
(formation of borate esters?), while acetylation (Ac_2O/pyridine) of a pure sample of **9** gave the tetraacetate **8** under mild conditions.

[3]) Hydrogenolysis of the benzylidene group was not observed under these conditions. Two-dimensional TLC of **11** revealed the presence of (at least) three interconverting compounds. The ^{1}H-NMR spectrum showed 4 signals for the *t*-Bu group and two NH signals. These observations may be rationalized by assuming the presence of two anomeric 'furanoses' [10], the ketone **11** and the enol corresponding to it (none of the products described in this or the preceding paper [1] showed multiple signals of the NHAc group due to rotational isomerism).

Scheme 2



The signal of H–C(5) in the spectrum of **1** is shifted downfield and overlaps with the one of H–C(6) at 4.13–3.93 ppm. A similar comparison of the ^{13}C-NMR spectra (see [13]) shows an upfield shift of 0.5–1.0 ppm for the signals of C(7), C(8), and C(9) of **1**, one of 8.2 and 8.8 ppm, respectively, for the signals of C(5) and C(3), and a downfield shift of 0.7 ppm for the one of C(6).

De-*O*-acetylation of the keto ester **11** with (NaOMe/MeOH) at 0° gave, after flash chromatography, the crystalline *tert*-butyl ester **12** and the crystalline methyl ester **13** (54% from **7**/**8**; *Scheme 2*). Both compounds did not mutarotate in MeOH solution, and their ^{1}H-NMR and ^{13}C-NMR spectra ((D$_6$)DMSO) showed signals of the β-D-anomer only.

A comparison of the ^{1}H-NMR spectra of the *N*-acetyl-4-deoxyneuraminic-acid derivative **12** with the one of the corresponding *N*-acetylneuraminic-acid derivative (see [1]) showed approximately the same chemical shifts ($\Delta\delta = \pm 0.01$ ppm) and the same coupling constants for the H-atoms at C(7), C(8), and C(9), while the signals of H–C(6) and H–C(5) were slightly shifted to lower fields (0.05 ppm), and J(5,6) and J(6,7) were larger by 1 Hz for the *N*-acetyl-4-deoxyneuraminic-acid derivative **12**. A similar comparison of the ^{13}C-NMR spectra showed an upfield shift of 1–2 ppm for C(6), C(7), C(8), and C(9) and one of *ca.* 10 ppm for C(3) and C(5) for **12**.

Hydrogenolytic removal of the benzylidene group of **12** and **13**, respectively, gave the crystalline *tert*-butyl *N*-acetyl-4-deoxyneuraminate (**14**) and the crystalline methyl *N*-acetyl-4-acetylneuraminate (**15**), which was also obtained by treating **1** with CH_2N_2 or CF_3COOH in MeOH[4]).

Note Added in Proof. – *N*-Acetyl-4-deoxyneuraminic acid (**1**) has also been prepared by Prof. Dr. *R. Brossmer*, Universität Heidelberg (personal communication).

We thank the *Swiss National Science Foundation* and *Sandoz AG*, Basel, for generous support.

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[4]) Hydrogenolysis of the keto ester **11** in the presence of 20% Pd(OH)$_2$ (MeOH) [11] followed by de-*O*-acetylation of the intermediate (K_2CO_3 in anh. MeOH) gave the overreduction products **16** (68% from **7** and **8**) instead of the desired **14**. Acetylation of **16** gave a mixture of the acetates **17**, which were characterized by the presence of 5 *O*-acetyl groups in its ^{1}H-NMR spectrum. The ^{13}C-NMR spectrum of **17** showed an additional *d* at *ca.* 70 ppm, but no carbonyl resonance at *ca.* 210 ppm. The NMR spectra showed the presence of two diastereoisomers in a ratio of *ca.* 2:3.

Experimental Part

General. See [1] and [14]. All solvents were distilled before use. THF was distilled from NaH, pyridine from CaH_2, anh. MeOH from Mg. Solvents were removed in a rotary evaporator at or below 40°. *Dowex 1 × 8* ($HCOO^-$) and *Dowex 50 × 4* (H^+) were washed and activated according to [15]. Flash chromatography (FC) [16]: silica gel *Merck 60*, 40–60 μm.

tert-*Butyl 5-Acetamido-4,6,8-tri-*O*-acetyl-7,9-*O*-benzylidene-2,3,5-trideoxy-2-methylidene-*D-glycero-D-galacto-*nononate* (**7**) *and* tert-*Butyl 5-Acetamido-4,6,8-tri-*O*-acetyl-7,9-*O*-benzylidene-2,3,5-trideoxy-2-methylidene-*D-glycero-D-talo-*nononate* (**8**). A soln. of **6** (1.50 g, 3.34 mmol) in THF (25 ml) was treated with NH_4Cl (180 mg, 3.51 mmol) and heated to 50°. $NaBH_4$ (120 mg) was added in one portion. The soln. changed to yellow. After 30 min, TLC (CH_2Cl_2/EtOH 9:1) of the colourless soln. indicated the disappearance of **6**. Excess $NaBH_4$ was destroyed with phosphate buffer pH 6.6 (15 ml). The mixture was diluted with AcOEt (100 ml) and extracted with H_2O and brine to give **4**/**5** as a colourless foam (D-*glycero*-D-*galacto*/D-*glycero*-D-*talo* = 1:3, as indicated by HPLC [1]). An ice-cold soln. of **4**/**5** in pyridine (10 ml) and Ac_2O (5 ml) was stirred at 0° (5 h), then at r.t. (over night). The mixture was concentrated and dried by co-evaporation with 3 × 10 ml toluene. FC of the residue (50 g of SiO_2, AcOEt/hexane 7:3) gave **7**/**8** (1.61 g, 83%) and the mono-*O*-acetate **9** (240 mg). Acetylation of the mono-*O*-acetate **9** (Py/Ac_2O) gave further **8** (266 mg, 14%). Anal. samples of **7** and **8** were obtained by FC (AcOEt/hexane 1:1) and crystallization from Et_2O/hexane. *Data of* **7**: M.p. 159–161°. $[\alpha]_D^{25} = -28.5°$ (*c* = 1.0, $CHCl_3$). IR: 3435*m*, 3035*w*, 3005*m*, 2985*m*, 2935*m*, 2870*w*, 1745*s* (br.), 1686*s*, 1635*w*, 1507*m*, 1370*s*, 1413*w*, 1225*s* (br.), 1156*s*, 1088*s*, 1048*s*, 1030*s*. ^{1}H-NMR (200 MHz): 7.69–7.32 (*m*, 5 arom. H); 6.10 (*d* = 1.0, 1 olef. H); 5.89 (*d*, *J* = 10.0, NH); 5.51 (br. *s*, ArCH, 1 olef. H); 5.36 (*ddd*, *J* = 9.0, 3.5, 3.5, H–C(4)); 5.05 (*dd*, *J* = 8.5, 1.8, H–C(6)); 4.89–4.69 (*m*, H–C(5), H–C(8)); 4.42 (*dd*, *J* = 10.2, 5.2, H–C(9)); 4.03 (*dd*, *J* = 10.0, 1.8, H–C(7)); 3.60 (*dd*, *J* = 10.2, 10.0, H–C(9)); 2.60 (*dd*, *J* = 14.0, 3.5, H–C(3)); 2.42 (*dd*, *J* = 14.0, 9.0 H–C(3)); 2.05 (*s*, 2 CH_3); 2.03 (*s*, CH_3); 1.97 (*s*, CH_3); 1.47 (*s*, *t*-Bu). ^{13}C-NMR (25.2 MHz): 169.97 (*s*); 169.50 (*s*); 169.48 (*s*); 169.33 (*s*); 165.17 (*s*); 137.16 (*s*); 136.82 (*s*); 128.68 (*d*); 127.97 (*d*); 126.94 (*t*); 125.89 (*d*); 101.13 (*d*); 80.79 (*s*); 77.00 (*d*); 70.08 (*d*); 67.77 (*t*); 67.47 (*d*); 62.16 (*d*); 49.90 (*d*); 35.24 (*t*); 27.91 (*q*); 23.30 (*q*); 20.95 (*q*); 20.80 (*q*); 20.63 (*q*). EI-MS: *cf.* **8**. Anal. calc. for $C_{29}H_{39}NO_{11}$ (577.63): C 60.30, H 6.81, N 2.42; found: C 60.02, H 6.90, 2.69.

Data of **8**: M.p. 101–103°. $[\alpha]_D^{25} = +26.0°$ (*c* = 1.0, $CHCl_3$). IR: 3430*m*, 3035*w*, 3005*m*, 2985*m*, 2940*w*, 2870*w*, 1746*s*(br.), 1682*s*, 1631*w*, 1510*m*, 1370*s*, 1310*m*, 1225*s* (br.), 1156*s*, 1090*s*, 1050*s*, 1028*s*, 1015*s*. ^{1}H-NMR (200 MHz): 7.62–7.36 (*m*, 5 arom. H); 6.53 (*d*, *J* = 10.0, NH); 6.09 (*d*, *J* = 1.5, 1 olef. H); 5.56 (*s*, ArCH); 5.54 (*d*, *J* = 1.5, 1 olef. H); 5.25 (*dd*, *J* = 5.5, 2.0, H–C(6)); 5.23 (*ddd*, *J* = 9.5, 9.0, 3.0, H–C(4)); 4.95 (*ddd*, *J* = 10.5, 10.0, 5.5, H–C(8)); 4.62 (*ddd*, *J* = 10.0, 9.0, 5.5, H–C(5)); 4.38 (*dd*, *J* = 10.5, 5.5, H–C(9)); 4.17 (*dd*, *J* = 10.0, 2.0, H–C(7)); 3.63 (*dd*, *J* = 10.5, 10.5, H–C(9)); 2.82 (*dd*, *J* = 14.0, 3.0, H–C(3)); 2.30 (*dd*, *J* = 14.0, 9.5, H–C(3)); 2.08 (*s*, CH_3)); 2.05 (*s*, CH_3); 2.03 (*s*, CH_3); 1.83 (*s*, CH_3); 1.44 (*s*, *t*-Bu). ^{13}C-NMR (25.2 MHz): 170.18 (*s*); 170.02 (*s*); 169.68 (*s*); 169.19 (*s*); 165.14 (*s*); 137.01 (*s*); 136.64 (*s*); 129.41 (*d*); 128.35 (*d*); 127.18 (*t*); 125.83 (*d*); 101.46 (*d*); 80.63 (*s*); 78.48 (*d*); 71.71 (*d*); 67.92 (*t*); 65.99 (*d*); 61.38 (*d*); 52.08 (*d*); 35.56 (*t*); 27.95 (*q*); 23.24 (*q*); 20.90 (*q*); 20.60 (*q*); 20.53 (*q*). EI-MS: 577 (0.3, M^+), 522 (0.5), 520 (0.8), 517 (0.4), 471 (0.7), 464 (2), 462 (2), 415 (1), 364 (6), 356 (4), 302 (11), 258 (33), 228 (25), 186 (11), 168 (48), 115 (44), 57 (56), 43 (100). Anal. calc. for $C_{29}H_{39}NO_{11}$ (577.63): C 60.30, H 6.81, N 2.42; found: 60.13, H 6.79, N 2.35.

tert-*Butyl (*E*)-5-Acetamido-6,8-di-*O*-acetyl-7,9-*O*-benzylidene-3,4,5-trideoxy-*D-manno-*non-3-en-2-ulosonate* (**10**). A mixture **7**/**8** (58 mg, 0.10 mmol) was ozonized and treated with (i-Pr)$_2$EtN as described for **1**. FC (5 g of SiO_2, AcOEt/hexane 1:1) gave **10** (47 mg, 91%)[5]). IR: 3430*m*, 3030*w*, 2985*m*, 2930*w*, 2870*w*, 1743*s*, 1704 (sh), 1678*s*, 1630*m*, 1502*m*, 1370*s*, 1220*s* (br.), 1086*s*. ^{1}H-NMR (200 MHz): 7.56–7.32 (*m*, 5 arom. H); 7.12 (*dd*, *J* = 16.0, 4.5, H–C(4)); 6.77 (*dd*, *J* = 16.0, 2.0, H–C(3)); 6.66 (*d*, *J* = 8.5, NH); 5.49 (*s*, ArCH); 5.32–5.20 (*m*, H–C(5)); 5.18 (*dd*, *J* = 4.5, 2.2, H–C(6)); 5.04 (*ddd*, *J* = 10.5, 9.8, 5.2, H–C(8)); 4.40 (*dd*, *J* = 10.5, 5.2, H–C(9)); 4.07 (*dd*, *J* = 9.8, 2.2, H–C(7)); 3.65 (*dd*, *J* = 10.5, 10.5, H–C(9)); 2.11 (*s*, CH_3); 2.07 (*s*, CH_3); 1.97 (*s*, CH_3); 1.56 (*s*, *t*-Bu).

*5-Acetamido-3,4,5-trideoxy-*D-manno-*2-nonulosonic Acid* (= N-*Acetyl-4-deoxyneuraminic Acid*; **1**). A mixture **7**/**8** (6.00 g, 10.4 mmol) and $NaHCO_3$ (600 mg, 7.1 mmol) in $CHCl_3$ (350 ml) was cooled to −60° and ozonized until the soln. turned blue[6]). It was purged with N_2 (10 min), treated with Me_2S (1.15 ml), slowly warmed to −40° (2 h), treated with (i-Pr)$_2$EtN (3.5 ml), warmed to 0° (6 h), and stirred over night (TLC: AcOEt/hexane 4:1). Extractive workup (0.1M HCl, 10% $NaHCO_3$ soln., brine) gave **10** (5.9 g) as a slightly yellow foam. To a prehydrogenated suspension of 10% PdC (600 mg) in AcOEt (85 ml) was added a soln. of **10** in AcOEt (20 ml) and hydrogenated at

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[5]) Addn. of 0.1% Et_3N to the eluent or using SiO_2 impregnated with 2% $NaHCO_3$ led to faster moving, strongly UV-active by-products.

[6]) Ozonolysis in AcOEt at −70° gave approximately the same results.

r.t. for 24 h. TLC (AcOEt/hexane 4:1, $KMnO_4$) indicated then complete hydrogenation of the olefinic double bond[7]). The mixture was filtered through *Celite*, and the residue was carefully washed with a total of 100 ml AcOEt. Evaporation of the filtrates gave crude **11** as a colourless foam, which was dissolved in anh. MeOH (25 ml) and added (30 min) to a stirred, ice-cold soln. of K_2CO_3 (2.9 g) in MeOH/H_2O 4:1 (200 ml). Stirring was continued at 0° (14 h) and then at r.t. (10 h). H_2O was added (40 ml), MeOH was removed and replaced by dioxane (40 ml). After addn. of *Dowex 50* × *4* (H^+; 30 g), the mixture was stirred at r.t. over night. The resin was filtered off and washed twice with 30 ml of H_2O. The combined filtrate and washings were extracted with $CHCl_3$ (100 ml). The aq. layer was freeze-dried to give crude **1** (brown residue, 3.5 g). A soln. of the residue in H_2O (5 ml) was brought to pH 9–10 by addn. of 1.0M NaOH (*ca.* 10.5 ml) and the mixture was kept in the refrigerator (24 h). Compound **1** was purified by anion-exchange chromatography (160 ml *Dowex 1X8* ($HCOO^-$); elution with 0.0–0.3N aq. HCOOH). Fractions containing **1** were combined, concentrated, freeze-dried, and finally dried over P_2O_5 (10^{-5} mbar, 2 d) to give **1** (1.60 g, 53%) as a microcrystalline solid, which decomposed at 162°. $[\alpha]_D^{25} = -47.6°$ (c = 1.1, H_2O). pK_a = 3.08 (H_2O, 22°). IR (KBr): 3700–2300*s*, 2925*m*, 1723*s*, 1658*s*, 1529*m*. ^{1}H-NMR (400 MHz, D_2O): 4.13–3.93 (*m*, H–C(5), H–C(6)); 3.83 (*dd*, J = 11.8, 2.7, H–C(9)); 3.76 (*dd*, J = 9.0, 6.3, 2.7, H–C(8)); 3.61 (*dd*, J = 11.8, 6.3, H–C(9)); 3.57 (*d*, J = 9.0, H–C(7)); 2.22–1.79 (*m*, 2 H–C(4), 2 H–C(3)); 2.00 (*s*, CH_3). ^{13}C-NMR (50 MHz, D_2O): 174.57 (*s*); 174.44 (*s*); 94.88 (*s*); 72.02 (*d*); 70.71 (*d*); 69.04 (*d*); 63.82 (*t*); 45.02 (*d*); 31.17 (*t*); 24.65 (*t*); 22.54 (*q*). FAB-MS: 294 ([M + 1]$^+$). Anal. calc. for $C_{11}H_{19}NO_8$ (293.28): C 45.05, H 6.53, N 4.78; found: C 45.17, H 6.56, N 4.65.

tert-*Butyl 5-Acetamido-7,9*-O-*benzylidene-3,4,5-trideoxy*-D-manno-*2-nonulosonate* (**12**) *and Methyl 5-Acetamido-7,9*-O-*benzylidene-3,4,5-trideoxy*-D-manno-*2-nonulosonate* (**13**). A mixture **7**/**8** (500 mg, 0.86 mmol) was ozonized, treated with $(i\text{-Pr})_2$EtN and hydrogenated as described for **1**. To an ice-cold soln. of the residue **11** in anh. MeOH (20 ml) was added dropwise a 0.5M NaOMe/MeOH soln. (200 µl). The mixture was stirred at 0° (24 h). Additional NaOMe/MeOH (200 µl) was added and stirring was continued. After 48 h, the soln. was neutralized with 1.0M AcOH in anh. MeOH. The solvent was evaporated. FC of the residual foam (40 g of SiO_2, CH_2Cl_2/EtOH 95:5) gave **12** (67 mg, 18%), **13** (81 mg, 24%), and 45 mg (12%) of a mixture of both. For analysis, **12** was crystallized from MeOH/Et_2O/hexane; **13** was crystallized from anh. MeOH/Et_2O. *Data of* **12**: M.p. 153–155°. $[\alpha]_D^{25} = -53.0°$ (c = 1.0, MeOH). IR: 3435*m*(br.), 3350(sh), 2980*m*, 2935*w*, 2865*w*, 1730*s*, 1668*s*, 1510*m*, 1453*m*, 1390*m*, 1370*s*, 1147*s*, 1080*s*, 1028*s*, 928*m*.^{1}H-NMR (400 MHz, (D_6)DMSO): 7.80 (*d*, J = 9.0, NH); 7.51–7.27 (*m*, 5 arom. H); 6.16 (br.*s*, OH–C(2)); 5.32 (*s*, ArCH); 5.01 (br. *s*, OH–C(8)); 4.16 (*dd*, J = 10.4, 5.3, H–C(9)); 4.06 (*dd*, J = 10.5, 1.5, H–C(6)); 4.02–3.91 (*m*, H–C(5)); 3.79–3.70 (*m*, H–C(8); addn. of D_2O: 3.73, *ddd*, J = 10.0, 9.2, 5.3); 3.52 (*dd*, J = 9.2, 1.5, H–C(7)); 3.47 (*dd*, J = 10.5, 10.0, H–C(9)); 1.86–1.64 (*m*, 2 H–C(4), 2 H–C(3)); 1.81 (*s*, CH_3); 1.40 (*s*, *t*-Bu). ^{13}C-NMR (50 MHz, (D_6)DMSO): 169.08 (*s*); 168.19 (*s*); 138.26 (*s*); 128.04 (*d*); 127.53 (*d*); 125.97 (*d*); 99.79 (*d*); 93.59 (*s*); 80.64 (*s*); 79.50 (*d*); 70.54 (*t*); 68.62 (*d*); 59.10 (*d*); 42.65 (*d*); 30.29 (*t*); 27.43 (*q*); 25.36 (*t*); 22.65 (*q*). CI-MS: 438 ([M + 1]$^+$), 420, 384, 380, 366, 320. Anal. calc. for $C_{22}H_{31}NO_8 \cdot 2H_2O$ (473.53): C 55.80, H 7.45, N 2.97; found: C 55.64, H 7.62, N 2.78.

Data of **13**: M.p. 215° (dec). $[\alpha]_D^{25} = -73.4°$ (c = 1.0, MeOH). IR (KBr): 3480*m* (br.), 3280*m* (br.), 2980*w*, 2905*w*, 2870*w*, 2840*w*, 1748*s*, 1646*s*, 1563*m*, 1388*s*, 1092*s*, 1080*s*, 1028*m*.^{1}H-NMR (400 MHz, (D_6)DMSO): 7.82 (*d*, J = 8.9, NH); 7.51–7.27 (*m*, 5 arom. H); 6.51 (br. *s*, OH–C(2)); 5.33 (*s*, ArCH); 5.13 (br. *s*, OH–C(8)); 4.16 (*dd*, J = 10.5, 5.5, H–C(9)); 4.08 (*dd*, J = 10.5, 1.2, H–C(6)); 4.06–3.94 (*m*, H–C(5)); 3.77–3.66 (*m*, H–C(8); addn. of D_2O: 3.70 *ddd*, J = 10.0, 9.0, 5.5); 3.65 (*s*, CH_3); 3.53 (*dd*, J = 9.0, 1.2, H–C(7)); 3.47 (*dd*, J = 10.5, 10.0, H–C(9)); 1.82 (*s*, CH_3); 1.87–1.66 (*m*, 2 H–C(4), 2 H–C(3)). ^{13}C-NMR (25.2 MHz, (D_6)DMSO): 170.42 (*s*); 168.13 (*s*); 138.08 (*s*); 128.01 (*d*); 127.54 (*d*); 125.89 (*d*); 99.66 (*d*); 93.71 (*s*); 79.49 (*d*); 70.54 (*t*); 68.73 (*d*); 59.01 (*d*); 52.02 (*q*); 42.64 (*d*); 30.54 (*t*); 25.43 (*t*); 22.81 (*q*). CI-MS: 396 ([M + 1]$^+$), 378, 336, 320, 290, 272. Anal. calc. for $C_{19}H_{25}NO_8$ (395.42): C 57.71, H 6.37, N 3.54; found: C 57.91, H 6.62, N 3.40.

tert-*Butyl 5-Acetamido-3,4,5-trideoxy*-D-manno-*2-nonulosonate* (**14**). Similarly to **15**, a soln. of **12** (115 mg, 0.26 mmol) in MeOH (5 ml) was hydrogenated in the presence of 10% PdC (25 mg). The crude product was crystallized from MeOH/Et_2O to give **14** (70 mg, 77%). M.p. 168–169° (dec.). $[\alpha]_D^{25} = -23.4°$ (c = 0.9, H_2O). IR(KBr): 3480*s*(br.), 2985*m*, 2945*m*, 2885*w*, 1735*s*, 1662*s*, 1552*m*, 1374*m*, 1320*m*, 1300*m*, 1156*m*, 1085*m*, 1035*m*, 1024*m*. ^{1}H-NMR (400 MHz, D_2O): 4.04 (*d*, J = 10.3, H–C(6)); 4.04–3.94 (*m*, H–C(5)); 3.86 (*dd*, J = 11.7, 2.6, H–C(9)); 3.77 (*ddd*, J = 8.8, 6.4, 2.6, H–C(8)); 3.63 (*dd*, J = 11.7, 6.4, H–C(9)); 3.59 (*d*, J = 8.8, H–C(7)); 2.01 (*s*, CH_3); 2.08–1.83 (*m*, 2 H–C(3), 2 H–C(4)); 1.51 (*s*, *t*-Bu); 2 *s* at 2.00 and 1.54 hint to the α-D-anomer. ^{13}C-NMR (50 MHz, D_2O): β-D-anomer: 174.39 (*s*); 171.47 (*s*); 94.74 (*s*); 85.21 (*s*); 71.83 (*d*); 70.68 (*d*); 68.88 (*d*); 63.61 (*t*); 44.91 (*d*); 30.79 (*t*); 27.42 (*q*); 24.65 (*t*); 22.42 (*q*); α-D-anomer: 174.65 (*s*); 85.42 (*d*); 71.15 (*d*); 69.80 (*d*); 67.42

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[7]) Shaking the suspension instead of stirring it shortens the reaction time. Cleavage of the benzylidene acetal was not observed.

(*d*); 27.66 (*q*). CI-MS: 350 ([*M* + 1]$^+$), 294, 276. Anal. calc. for $C_{15}H_{27}NO_8$ (349.39): C 51.57, H 7.79, N 4.01; found: C 51.31, H 8.01, N 3.84.

Methyl 5-Acetamido-3,4,5-trideoxy-D-manno-*2-nonulosonate* (**15**). A suspension of **13** (85 mg, 0.215 mmol) and 10% PdC (20 mg) in MeOH (5 ml) was hydrogenated at r.t. until TLC (CH_2Cl_2/MeOH 9:1) indicated the disappearance of **13**. The catalyst was filtered off and washed with anh. MeOH. A soln. of the residue of the filtrates in H_2O (5 ml) was extracted with AcOEt. The aq. layer was freeze-dried to give **15** (65 mg, 98%), which was crystallized from MeOH/AcOEt/Et_2O. M.p. 163–164° (dec.). $[\alpha]_D^{25} = -38.0°$ (*c* = 1.0, MeOH). IR (KBr): 3340*s*(br.), 2935*w*, 1740*s*, 1650*s*, 1535*m*, 1440*w*, 1375*m*, 1315*m*, 1288*s*, 1202*w*, 1152*m*, 1122*m*, 1075*s*, 1054*m*, 1022*m*. ^{1}H-NMR (400 MHz, D_2O): 4.05 (*d*, *J* = 10.5, H–C(6)); 4.00 (*ddd*, *J* = 10.5, 10.5, 4.2, H–C(5)); 3.85 (*dd*, *J* = 11.8, 2.7, H–C(9)); 3.83 (*s*, CH_3); 3.76 (*ddd*, *J* = 9.7, 6.4, 2.7, H–C(8)); 3.62 (*dd*, *J* = 11.8, 6.4, H–C(9)); 3.57 (*d*, *J* = 9.7, H–C(7)); 2.06–1.85 (*m*, 2 H–C(3), 2 H–C(4)); 2.01 (*s*, CH_3). ^{13}C-NMR (50 MHz, (D_6)DMSO): 170.57 (*s*); 170.40 (*s*); 93.60 (*s*); 71.34 (*d*); 69.54 (*d*); 68.99 (*d*); 63.59 (*t*); 52.05 (*q*); 44.51 (*d*); 30.67 (*t*); 24.71 (*t*); 22.46 (*q*). CI-MS: 308 ([*M* + 1]$^+$), 290, 248, 232. Anal. calc. for $C_{12}H_{21}NO_8$ (307.31): C 46.90, H 6.89, N 4.56; found: C 46.62, H 6.77, N 4.48.

tert-*Butyl 5-Acetamido-3,4,5-trideoxy*-D-glycero-D-talo- *and* -D-glycero-D-galacto-*nononates* **16**. Similarly to **1**, a soln. of **7**/**8** (250 mg, 0,433 mmol) was ozonized treated with (i-Pr)$_2$EtN and hydrogenated to give crude **11** (200 mg). A soln. of the residue in MeOH (5 ml) was hydrogenated in the presence of 20% $Pd(OH)_2$ (30 mg) [11]. After 24 h, TLC (AcOEt/hexane 4:1) indicated the disappearance of the intermediate **11**. The catalyst was filtered off and washed with MeOH. A soln. of the residue (170 mg) of the filtrates in anh. MeOH (4.5 ml) was treated with K_2CO_3 (4.5 mg) and stirred at r.t.; after 8 h, additional K_2CO_3 (3 mg) was added. Stirring was continued over night, TLC (CH_2Cl_2/MeOH 4:1) indicated then the disappearance of the intermediate. The soln. was neutralized (0.1M AcOH/MeOH, 550 µl) and evaporated. FC of the residue (6 g of SiO_2, CH_2Cl_2/MeOH 9:1) on a preconditioned column (CH_2Cl_2/MeOH 95:5) gave **16** (104 mg, 68%; 36 h P_2O_5 at 10^{-1} mbar), which was crystallized from MeOH/Et_2O 1:1 and hexane. M.p. 97–105°. IR (KBr): 3370*s* (br.), 1733*s*, 1649*s*, 1556*m*, 1372*m*, 1170*m*, 1085*m*, 1035*m*. ^{1}H-NMR (200 MHz, CD_3OD): 4.07–3.38 (*m*, H–C(2), H–C(5), H–C(6), H–C(7), H–C(8), CH_2(9)); 2.14–1.50 (*m*, CH_2(3), CH_2(4)); 1.98 (*s*, CH_3); 1.48 (*s*, *t*-Bu).

For analysis, **16** was acetylated (Ac_2O/Py 1:1, 0°) to give, after FC ($CHCl_3$/MeOH 200:1), **17** as a colourless foam. IR: 3430*w*, 3030*w*, 2980*w*, 2935*w*, 1740*s*, 1680*s*, 1500*m*, 1367*s*, 1216*s*(br.), 1152*m*, 1042*m*. ^{1}H-NMR (400 MHz): 5.41 (*d*, *J* = 5.3, NH); 5.39 (*dd*, *J* = 8.0, 3.0, H–C(7)); 5.13 (*dd*, *J* = 8.0, 3.0, H–C(6)); 5.08 (*ddd*, *J* = 8.0, 5.8, 3.0, H–C(8)); 4.84–4.79 (*m*, H–C(2)); 4.25 (*dd*, *J* = 12.5, 3.0, H–C(9)); 4.27–4.19 (*m*, H–C(5)); 4.05 (*dd*, *J* = 12.5, 5.8, H–C(9)); 2.13–2.05 (5 CH_3); 1.97 (*s*, CH_3); 1.90–1.57 (*m*, CH_2(3), CH_2(4)); 1.45 (*s*, *t*-Bu). ^{13}C-NMR (50 MHz): major product: 170.37 (*s*); 170.19 (*s*); 170.13 (*s*); 169.83 (*s*); 169.68 (*s*); 168.86 (*s*); 168.72 (*s*); 82.12 (*s*); 72.38 (*d*); 71.59 (*d*); 68.59 (*d*); 68.23 (*d*); 61.93 (*t*); 48.01 (*d*); 27.73 (*q*); 27.30 (*t*); 26.64 (*t*); 23.00 (*q*); 20.61 (*q*); 20.57 (*q*); 20.51 (*q*); 20.46 (*q*); 20.41 (*q*); minor product: 72.19 (*d*); 71.73 (*d*); 47.85 (*d*); 27.24 (*t*); 26.81 (*t*). CI-MS: 562 ([*M* + 1]$^+$), 506. Anal. calc. for $C_{25}H_{39}NO_{13}$ (561.60): C 53.47, H 7.00, N 2.49; found: C 53.25, H 7.04, N 2.70.

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