

Studies towards a Conjugate Vaccine for Anthrax: Synthesis of the Tetrasaccharide Side Chain of the *Bacillus anthracis* Exosporium

by Roberto Adamo, Rina Saksena, and Pavol Kováč^{*1)}

NIDDK, LMC, National Institutes of Health, 8 Center Drive, Bethesda, MD 20892-0815, USA
(phone: +1 301-496-3569; fax: +1 301-402-0589; e-mail: kpn@helix.nih.gov)

The first synthesis of β -L-glycoside **17** of the tetrasaccharide β -Ant-(1 $\rightarrow$ 3)- α -L-Rhap-(1 $\rightarrow$ 3)- α -L-Rhap-(1 $\rightarrow$ 2)-L-Rhap is described (*Schemes 1–3*). Its spacer can be functionalized to make it amenable to conjugation to proteins by different conjugation methods. The synthesis was performed in a stepwise manner starting from the aglycon-bearing terminal saccharide with thioglycosides as glycosyl donors. To attach the upstream terminal anthrose residue, the assembled linker-equipped trisaccharide was glycosylated with ethyl 4-azido-3-*O*-benzyl-2-*O*-(bromoacetyl)-4,6-dideoxy-1-thio- β -D-glucopyranoside (**11**). Further functionalization of the tetrasaccharide thus obtained, followed by deprotection gave the target substance **17**. Synthesis of substructures of **17** equipped with the same spacer, namely β -L-Rhap-1-*O*-(CH₂)₅COOMe (**21**), α -L-Rhap-(1 $\rightarrow$ 2)- β -L-Rhap-1-*O*-(CH₂)₅COOMe (**22**), and α -L-Rhap-(1 $\rightarrow$ 3)- α -L-Rhap-(1 $\rightarrow$ 2)- β -L-Rhap-1-*O*-(CH₂)₅COOMe (**23**), is also described (*Scheme 4*).

Introduction. – *Bacillus anthracis*, the causative agent of anthrax, is a spore-forming pathogen. One of the components of its exosporium is a collagen-like protein [1] whose carbohydrate portion is formed by the tetrasaccharide β -Ant-(1 $\rightarrow$ 3)- α -L-Rhap-(1 $\rightarrow$ 3)- α -L-Rhap-(1 $\rightarrow$ 2)-L-Rhap (*Fig.*) [2]. Anthrose (= 4,6-dideoxy-4-[(3-hydroxy-3-methyl-1-oxobutyl)amino]-2-*O*-methyl-D-glucopyranose), which forms the upstream terminal residue of the tetrasaccharide, was first discovered as a component of the exosporium glycoprotein. In connection with our attempts to develop a conjugate vaccine for anthrax, we have recently synthesized this rare sugar, as well as its methyl glycosides [3]. Our approach to the vaccine is based on the premise that spores could be destroyed by antibodies specific for components on their surface. Such antibodies should be possible to elicit by immunization with, for example, immunogens prepared from the above-mentioned tetrasaccharide. Within a project aimed at immunogenic neoglycoconjugates, we have prepared tetrasaccharide glycoside **17**, whose aglycone makes it amenable to conjugation with suitable carriers. Here we report full details of the synthesis of **17**, together with complete assignment of ¹H- and ¹³C-NMR spectra, that were not included in our preliminary report [4].

It has been proposed [2] that the tetrasaccharide is most probably attached to the glycoprotein through an *N*-acetyl-D-galactosamine linker, but the configuration of the glycosidic linkage providing that attachment to the linker is unknown. A prerequisite for obtaining a potent immunogen for anti exosporium antibodies from the spacer-

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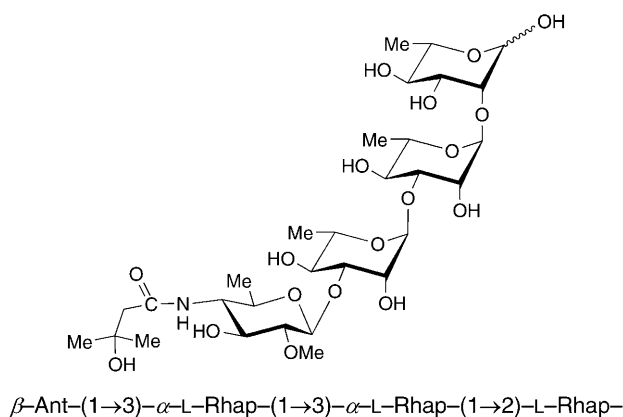
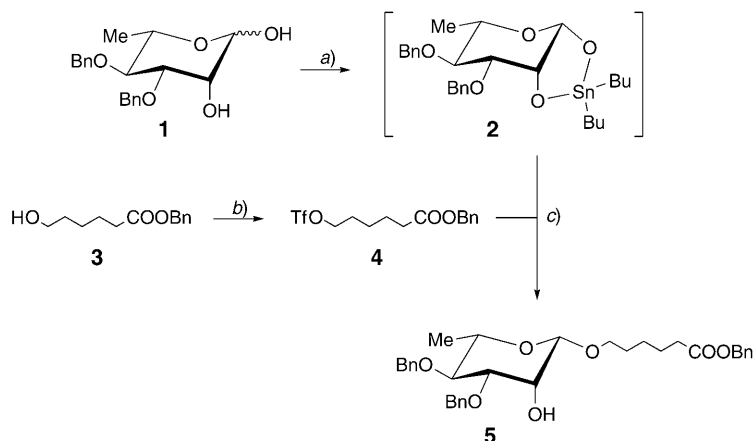


Figure. Structure of the tetrasaccharide side chain of the collagen-like glycoprotein of the exosporium of *Bacillus anthracis*

equipped tetrasaccharide is that its anomeric configuration in the glycoconjugate is the same as in the exosporium glycoprotein. With that information unavailable, conjugates made from both α - and β -linked tetrasaccharides will have to be prepared and tested for their immunogenicity. Described below is the first synthesis of tetrasaccharide **17** whose spacer is attached to the downstream β -L-rhamnosyl terminus (*Schemes 1–3*). Preparation of three structural fragments of **17**, namely the linker-equipped β -glycosides **21–23** (*Scheme 4*) is also described. These compounds will be used in binding and inhibition studies with anti exosporium antibodies. An independent synthesis of the tetrasaccharide equipped with a different, α -linked spacer has been published [5].

Results and Discussion. – Immune response to antigens is controlled by interaction of antigenic determinants with receptors of the complex immune system. With short-chain antigens, such as synthetic oligosaccharides, that interaction can be, presumably, facilitated when the oligosaccharide has sufficient translational and rotational freedom to access active sites on immunologically active cells. That can be achieved by linking the oligosaccharide antigen to a carrier through a flexible spacer moiety. Tetrasaccharide **17**, whose stepwise synthesis is presented here, is equipped with a β -L-linked C_6 -chain spacer attached to the downstream terminal L-rhamnosyl residue. Depending on chemistry of conjugation, that spacer may become further extended when it is functionalized for conjugation. The stepwise synthesis of **17** started from the downstream rhamnose residue. In this way, fragments of **17**, which will be needed in connection with the development of the vaccine, were easily accessible. The initial glycosyl acceptor **5** was synthesized from the triflate **4** of benzyl 6-hydroxyhexanoate (**3**) [6] and the stannylidene acetal **2** of 3,4-di-*O*-benzyl-L-rhamnose (**1**) [7] (*Scheme 1*). The formation of the β -L-rhamnopyranosyl linkage was stereospecific [8] [9], as manifested [10] in the ^1H -coupled ^{13}C -NMR spectrum of the sole glycoside formed ($J(\text{C}(1), \text{H}-\text{C}(1)) = 156.4 \text{ Hz}$). To introduce the spacer, we preferred the use of benzyl ester **3** to the more commonly employed analogous methyl ester [11] since triflation of **3** ($\rightarrow$ **4**) could be monitored by TLC with detection by UV light. The required, known [6] benzyl

Scheme 1



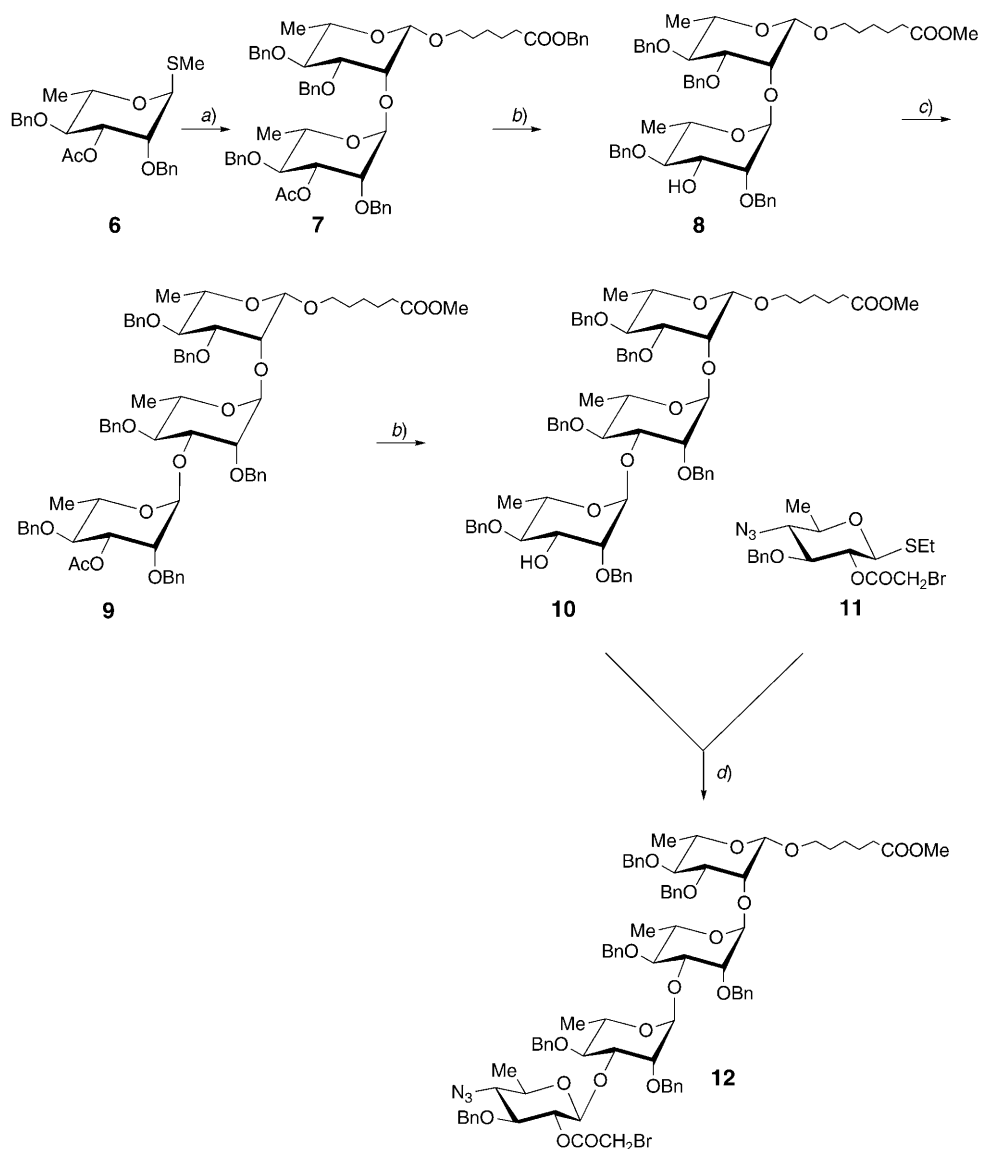
a) Bu_2SnO , toluene. b) Tf_2O , K_2CO_3 , CH_2Cl_2 . c) CsF , MeCN .

ester **3** was more conveniently made from commercially available ϵ -caprolactone (=oxepan-2-one) by transesterification with NaOBn in BnOH . The fully assigned ^1H -NMR data of **3** agreed with those reported [6], albeit not analyzed completely. Side reactions that occurred during subsequent triflation under standard conditions (Tf_2O , pyridine, not reported in the *Exper. Part*) were eliminated by the use of solid K_2CO_3 as a base [12]. In this way, pure (NMR) [4] triflate **4** was obtained from **3** in virtually theoretical yield. The compound is unstable, and should be freshly prepared before use.

Stepwise extension of the oligosaccharide chain, to give rhamnotrioside **9** was effected by repeated NIS (*N*-iodosuccinimide)/ AgOTf -mediated glycosylation [13] with methyl 3-*O*-acetyl-2,4-di-*O*-benzyl-1-thio- α -L-rhamnopyranoside (**6**) [7], first of **5** ($\rightarrow$ **7**) and then of the product of deacetylation of **7**, alcohol **8** ($\rightarrow$ **9**) (Scheme 2). The α -L-linked products were obtained in high yield, as shown by the typical [10] large $J(\text{C}(1), \text{H}-\text{C}(1))$. Complete, unambiguous assignments of the NMR spectra of **6** were now made by 2-D correlation spectroscopy (see *Exper. Part*).

Positioning anthrose at the upstream terminus of the desired tetrasaccharide **17** required formation of the β -D-glucosidic linkage. Because of the presence of the 2-*O*-methyl group in the β -D-glucosyl residue of **17**, formation of such a linkage in a highly stereoselective manner could be problematic with a glycosyl donor derived from anthrose. Therefore, after deacetylation of **9**, the glycosyl acceptor **10** formed was glycosylated with the glucosyl donor **11** [14], obtained by 2-*O*-bromoacetylation of methyl 4-azido-3-*O*-benzyl-4,6-dideoxy- β -D-glucopyranoside [3] to give the fully protected tetrasaccharide **12** (Scheme 2). The general utility of glycosyl donor **11** is due to the presence of a protecting acyl group at *O*-2 that can be selectively removed in the presence of other acyl groups. As such, it can also be used in alternate strategies towards analogs of **17** that involve acyl-protected intermediates.

Scheme 2

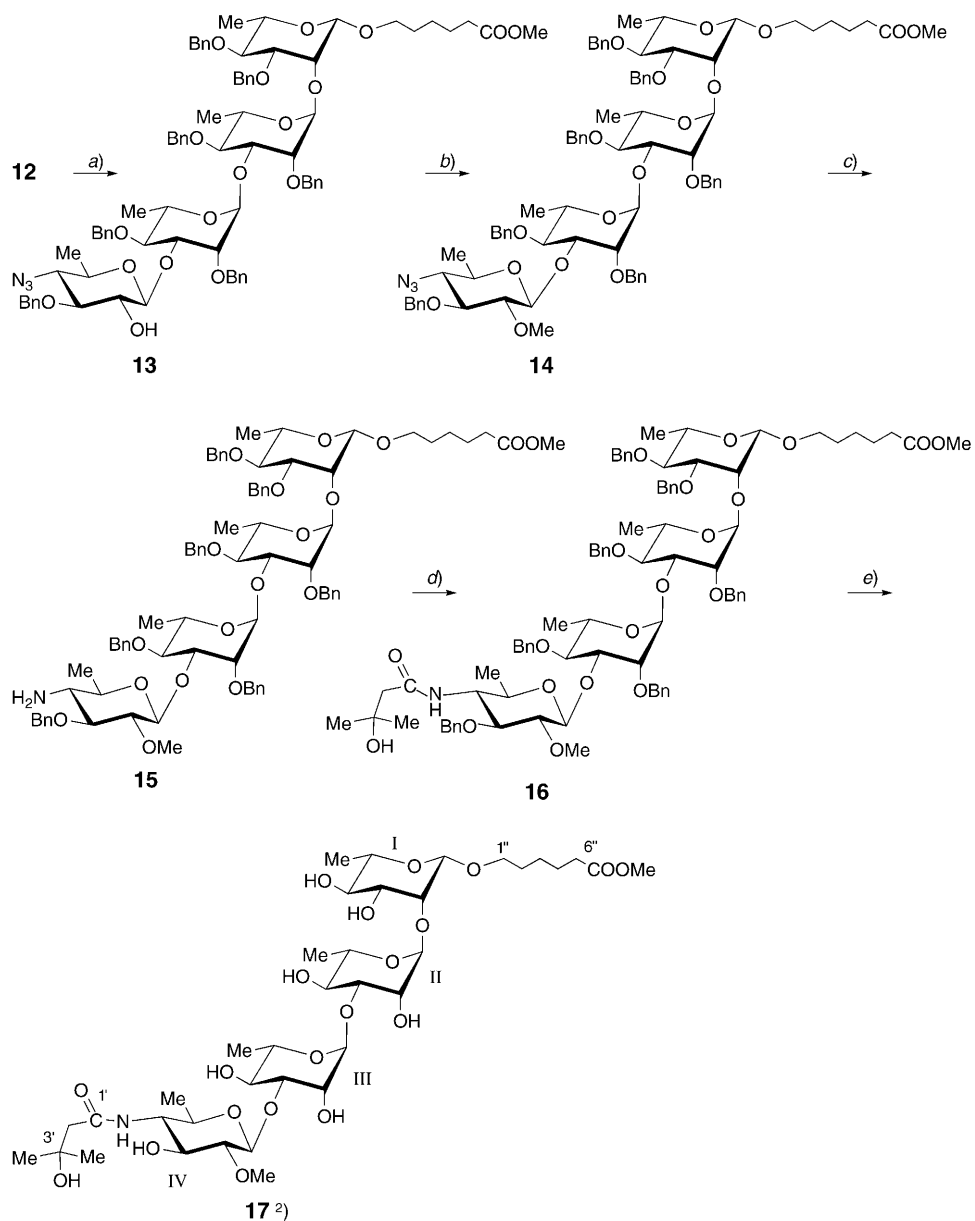


a) **5**, NIS, AgOTf, CH₂Cl₂. *b*) MeONa, MeOH. *c*) **6**, NIS, AgOTf, CH₂Cl₂. *d*) NIS, AgOTf, CH₂Cl₂.

Transformation of **12** into the target tetrasaccharide **17** was accomplished as shown in Scheme 3²⁾. Accordingly, the protected tetrasaccharide **12** was debromoacetylated,

²⁾ The aglycon spacer (CH₂)₅COOMe is numbered with doubly primed locants. The sugar residue bearing the aglycon spacer is denoted by the Roman numeral I and the remaining sugar residues sequentially by II–IV, as required. The terminal butanamido group is numbered with primed locants.

Scheme 3



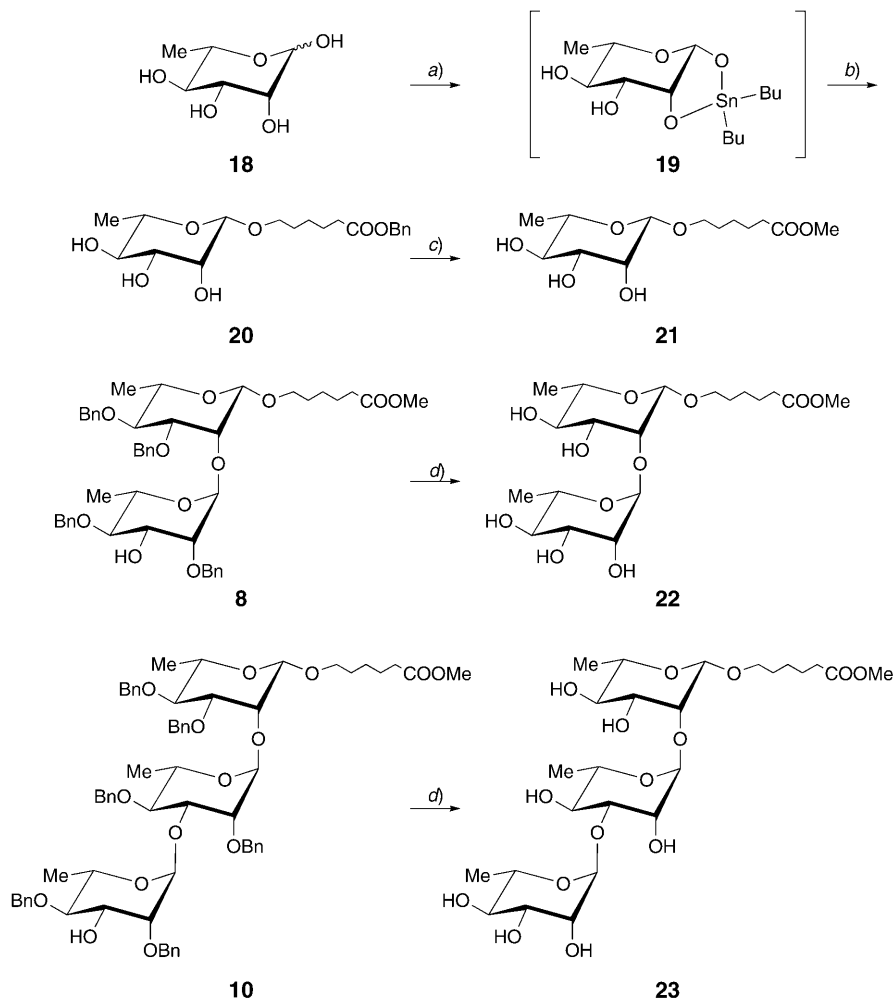
a) MeONa, MeOH. b) MeI, Me₂S, Ag₂O, CH₂Cl₂. c) H₂S, Py, H₂O. d) 3-Hydroxy-3-methylbutanoic acid, HATU, CH₂Cl₂. e) H₂, Pd/C, 2-methoxyethanol²⁾.

and the formed alcohol **13** was methylated [**15**] ($\rightarrow$ **14**). Selective reduction of the azido function in **14** with hydrogen sulfide [**16**] gave amine **15**, whose treatment with 3-hy-

droxy-3-methylbutanoic acid in the presence of HATU (= *O*-(1*H*-7-azabenzotriazol-1-yl)-1,1,3,3-tetramethyluronium hexafluorophosphate) yielded butanamido derivative **16**. Finally, hydrogenolytic debenzylation over 5% Pd/C catalyst gave the target tetrasaccharide **17** equipped with a spacer that makes it amenable to conjugation to proteins.

Future characterization of immunogenic epitopes in tetrasaccharide **17** and its α -L-glycoside analog [14] by carbohydrate microarray analysis will require fragments that mimic partial structures of both tetrasaccharides. Two such compounds, glycosides **22** and **23**, were obtained by catalytic debenzylation of intermediates **8** and **10** of the synthesis of **17**, respectively (Scheme 4).

Scheme 4



a) Bu₂SnO, MeOH. *b)* **4**, CsF, DMF. *c)* MeONa, MeOH. *d)* H₂, Pd/C, 2-methoxyethanol.

The β -L-rhamnopyranoside **21** was conveniently synthesized from benzyl ester **20**, obtained in turn from L-rhamnose (**18**) and triflate **4** by β -rhamnosylation *via* 1,2-*O*-stannylation [8][9], a method which does not require protection of the OH groups (*Scheme 4*). To insure an efficient use of the unstable triflate **4**, the reaction leading to **21** was carried out with excess stannylene acetal **19**. In view of the experimental simplicity of this method, we deem the relatively low yield (38%) of **20** satisfactory. Compound **20** was the only glycoside formed, and its β -L-configuration was confirmed by ^1H -coupled ^{13}C -NMR spectrum ($J(\text{C}(1), \text{H}-\text{C}(1)) = 155.6$ Hz). The desired methyl ester **21** was readily obtained from **20** by base-catalyzed transesterification.

The ^1H - and ^{13}C -NMR spectra of **17** (*Tables 1* and *2*), which were interpreted with the aid of two-dimensional techniques, fully confirmed the structure of the tetrasaccharide. Noteworthy in the ^1H -NMR spectrum ((D_6) DMSO) is the remarkable similarity of chemical shifts for signals associated with the anthrose residue of **17** with the corresponding resonances found in the spectrum of the reducing tetrasaccharide isolated from *Bacillus anthracis* [2] exosporium. Also, in addition to the resonances of $\text{H}-\text{C}(1^{\text{I}})$ to $\text{H}-\text{C}(1^{\text{IV}})^2$, the ^1H -NMR spectrum ((D_6) DMSO) of **17** showed eight signals in the

Table 1. ^1H -NMR Chemical Shifts [ppm] and Coupling Constants [Hz] for Tetrasaccharide **17**^{a)}

Moiety ^{b)}	H-C(1) or CH ₂ (1'') ²⁾	H-C(2), CH ₂ (2'') or CH ₂ (2')	H-C(3) or CH ₂ (3'') ²⁾	H-C(4) or CH ₂ (4'') ²⁾	H-C(5) or CH ₂ (6'') ²⁾	H-C(6)
I ^{c)}	4.641 ($J(1,2)=0.9$) 4.393 ^{d)}	4.002 ($J(2,3)=3.1$) <i>ca.</i> 3.755 ^{d)}	3.691 ($J(3,4)=9.4$) 3.333 ^{d)}	3.400 ^{d)} 2.091 ^{d)}	3.400 2.091	<i>ca.</i> 1.301 ^{d)} 1.134 ^{d)}
II ^{c)}	5.005 ($J(1,2)=1.8$) 4.906 ($J(1,2)=1.5$)	4.186 ($J(2,3)=3.3$) <i>ca.</i> 3.755 ^{d)}	3.902 ($J(3,4)=9.8$) 3.615 ^{d)}	3.508 ($J=10.0$) 3.281 ^{d)}	4.213 3.939	1.249 ($J(5,6)=6.3$) 1.028 ($J(5,6)=6.2$)
III ^{f)}	5.028 ($J(1,2)=1.7$) 4.894 ($J(1,2)=1.4$)	4.285 ($J(2,3)=3.3$) 3.879 ^{d)}	4.002 ^{d)} <i>ca.</i> 3.755 ^{d)}	3.620 ($J=10.0$) 3.404 ^{d)}	3.876 3.628	<i>ca.</i> 1.315 ^{d)} 1.129 ^{d)}
IV ^{g)}	4.740 ($J(1,2)=8.0$) 4.576 ($J(1,2)=7.9$)	3.135 ($J(2,3)=9.2$) 2.850 ($J(2,3)=8.9$)	3.543 ($J=10.0$) 3.275 ^{d)}	3.636 ($J=10.0$) <i>ca.</i> 3.400 ^{d)}	3.561 3.270	1.223 ($J(5,6)=6.1$) 1.067 ($J(5,6)=6.2$)
Spacer ^{h)}	3.840, 3.670 3.695, 3.333	1.612 1.480	1.422 1.313	1.612 1.480	2.400 ($J=7.5$) 2.287 ($J=7.5$)	
Amido group ⁱ⁾		2.472, 2.446 2.208				

^{a)} Measured at 600 MHz at 20°. Data in the first and second row for each moiety refer to measurements in D₂O and (D_6) DMSO, respectively. ^{b)} For notation of the sugar residues, see *Footnote 2*. ^{c)} δ ((D_6) DMSO) 5.030 (*d*, $J=4.2$, OH-C(3)); 4.864 (*br. d.*, $J=4.7$, OH-C(4)). ^{d)} Coupling constant not determined. ^{e)} δ ((D_6) DMSO) 4.844 (*d*, $J=6.4$, OH-C(4)); 4.643 (*d*, $J=4.7$, OH-C(2)). ^{f)} δ ((D_6) DMSO) 4.704 (*d*, $J=5.3$, OH-C(4)); 4.511 (*d*, $J=4.5$, OH-C(2)). ^{g)} δ (D₂O) 3.632 (*s*, MeO). δ ((D_6) DMSO) 7.728 (*d*, $J(\text{NH},4)=9.1$, NH); 4.997 (*d*, $J=6.1$, OH-CH(3)); 3.525 (*s*, MeO). ^{h)} δ (D₂O) 3.680 (*s*, COOMe). δ ((D_6) DMSO) 3.572 (*s*, COOMe). ⁱ⁾ δ (D₂O) *ca.* 1.309, *ca.* 1.300 (2*s*, 2 Me). δ ((D_6) DMSO) 4.831 (*s*, OH-C(3'')); 1.158, 1.146 (2*s*, 2 Me).

Table 2. ^{13}C -NMR Chemical Shifts [ppm] for Tetrasaccharide **17**^{a)}

Moiety ^{b)}	C(1), C(1''), or C(1'') ²⁾	C(2), C(2''), or C(2'') ²⁾	C(3), C(3''), or C(3'') ²⁾	C(4) or C(4'') ²⁾	C(5) or C(5'') ²⁾	C(6) or C(6'') ²⁾
I	102.336 99.733	79.615 75.304	76.053 73.803	75.150 ^{c)} 72.320 ^{c)}	74.979 ^{c)} 72.132 ^{c)}	19.455 ^{d)} 18.043
II	103.995 100.756	72.781 70.196	80.864 79.966	74.099 71.527	71.547 68.099	19.492 17.678
III	104.929 101.476	72.606 69.634	82.392 79.585	73.912 71.633	72.047 68.187	19.424 ^{d)} 17.678
IV ^{f)}	106.431 103.479	86.030 84.348	75.620 72.749	59.345 59.389	73.536 70.315	19.806 18.153
Spacer ^{g)}	72.486 68.414	26.858 29.024	27.728 25.224	31.451 24.424	36.393 33.261	176.830 171.401
Amido group ^{h)}	180.399 173.445	51.687 48.645	72.990 68.674			

^{a)} Measured at 150 MHz at 20°. Data in the first and second row for each moiety refer to measurements in D₂O and (D₆)DMSO, respectively. ^{b)} For the notation of the sugar residues, see Footnote 2. ^{c)} The assignment can be reversed. ^{d)} The assignment can be reversed. ^{e)} The assignment can be reversed. ^{f)} MeO–C(2): δ (D₂O) 62.811; δ ((D₆)DMSO) 59.989. ^{g)} COOMe: δ (D₂O) 54.855; δ ((D₆)DMSO) 51.213. ^{h)} Me: δ (D₂O) 31.040, 30.855; δ ((D₆)DMSO) 29.522, 29.418.

anomeric region whose presence in the spectrum of their material was not reported by Daubenspeck *et al.* [2]. Two-dimensional NMR spectra of **17** showed that these resonances, which did not produce cross-peaks in heteronuclear ^1H , ^{13}C contour maps, arose from the eight OH groups present in the molecule. The attachment to their respective C-atoms could be individually assigned by homonuclear ^1H , ^1H correlation spectroscopy.

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Experimental Part

General. Pd/C Catalyst (5%; ESCAT 103) was a product of Engelhard Industries. HATU (= *O*-1H-7-azabenzotriazol-1-yl)-1,1,3,3-tetramethyluronium hexafluorophosphate) was purchased from Applied Biosystems. Solns. in org. solvents were dried with anh. Na₂SO₄, and evaporated at 40°/2 kPa. TLC: for reaction monitoring, silica gel 60 coated glass slides. Column chromatography (CC): silica gel, CombiFlash Companion Chromatograph (Isco, Inc.); solvent mixtures less polar than those used for TLC were used at the onset of separation. Optical rotations: unless stated otherwise at r.t. in CHCl₃; Perkin-Elmer-341 automatic polarimeter. NMR Spectra²⁾: Varian-Gemini or Varian-Mercury spectrometers at 300 (^1H) and 75 MHz (^{13}C); Bruker-Avance-600 spectrometer at 600 (^1H) and 150 MHz (^{13}C); chemical shifts δ in ppm and coupling constants *J* in Hz; assignments by homonuclear and heteronuclear 2-D correlation spectroscopy, run with the software supplied with the spectrometers. MS: liquid chromatography electron-spray ionization (ESI-MS) with Hewlett-Packard-1100-MSD spectrometer; gas chromatography/electron impact ionization (GC/EI-MS) with Hewlett-Packard-5898A spectrometer. Microanalyses: some compounds tenaciously retained traces of solvents, despite exhaustive drying, and anal. figures for C could not be obtained within $\pm 0.4\%$; their structures were established from the mode of synthesis and NMR and MS data.

3,4-Di-O-benzyl-6-deoxy-L-mannopyranose (1). H₂O (0.1 ml) was added to a stirred soln. of methyl 3,4-di-O-benzyl-6-deoxy-1-thio- α -L-mannopyranoside [17] (4 g, 10.7 mmol) in acetone (15 ml), followed by NIS (2.9 g, 12.8 mmol). After 1 h (TLC (hexane/AcOEt 3:2) monitoring), the mixture was evaporated, and the residue subjected to CC (toluene/AcOEt 4:1): 2.61 g (85%) of **1**. ¹H-NMR: in accord with that of independently synthesized material [7]. ¹³C-NMR (75 MHz, CDCl₃): 93.68 (C(1) (α)); 93.55 (C(1) (β)); 81.50 (C(3) (β)); 79.89 (C(4) (α)); 79.53 (C(3) (α)); 79.43 (C(4) (β)); 75.38 (PhCH₂ (β)); 75.36 (PhCH₂ (β)); 72.14 (PhCH₂ (α)); 72.05 (PhCH₂ (β)); 71.16 (C(5) (α)); 68.88 (C(2) (β)); 68.60 (C(2) (α)); 67.48 (C(5) (α)); 17.94 (C(6) (β)); 17.89 (C(6) (α)).

Benzyl 6-Hydroxyhexanoate (3). ϵ -Caprolactone (29 g, 20 ml) was added to a soln. of Na (1 g) in BnOH (250 ml), and the mixture was stirred at r.t. overnight. After neutralization with *Amberlite IR-120*, the filtrate was concentrated, and the residue subjected to CC (CH₂Cl₂/MeOH 100:1). The product thus obtained was distilled at 145°/1.3·10⁻² Torr: pure (NMR, TLC) **3** (22 g, 39%). ¹H-NMR (300 MHz, CDCl₃): 5.10 (s, PhCH₂); 3.57 (t, *J*=6.5, CH₂(6)); 2.50 (br. s, OH-C(6)); 2.35 (t, *J*=7.5, CH₂(2)); 1.71–1.61 (m, CH₂(3)); 1.59–1.50 (m, CH₂(5)); 1.42–1.34 (m, CH₂(4)). ¹³C-NMR (75 MHz, CDCl₃): 173.44 (C=O); 65.94 (PhCH₂); 62.10 (C(6)); 34.00 (C(2)); 32.02 (C(5)); 25.06 (C(4)); 24.43 (C(3)). HR-ESI-MS: 245.1154 ([*M*+Na]⁺; calc. 245.1151).

6-(Benzyloxy)-6-oxohexyl 3,4-Di-O-benzyl-6-deoxy- β -L-mannopyranoside (5). Trifluoromethanesulfonic acid anhydride (1.95 ml, 11.52 mmol) was added at –20° to a stirred mixture of **3** (2.13 g, 9.6 mmol) and finely powdered K₂CO₃ (1.6 g, 11.52 mmol) in CH₂Cl₂ (20 ml). After 1 h at –20° (TLC (hexane/AcOEt 4:1) monitoring), the mixture was filtered and washed quickly with ice-cold aq. NaHCO₃ soln. and the org. phase dried and concentrated at 15–20°: unstable *benzyl 6-[(trifluoromethyl)sulfonyl]oxyhexanoate (4)*; quant.), sufficiently pure for the next step. ¹H-NMR (600 MHz, CDCl₃): 5.13 (s, PhCH₂); 4.51 (t, *J*=7.3, CH₂(6)); 2.39 (t, *J*=7.3, CH₂(2)); 1.85–1.81 (m, CH₂(5)); 1.73–1.67 (m, CH₂(3)); 1.48–1.44 (m, CH₂(4)). ¹³C-NMR (150 MHz, CDCl₃): 173.15 (C=O); 118.61 (q, CF₃SO₂); 77.16 (C(6)); 66.33 (PhCH₂); 33.84 (C(2)); 28.93 (C(5)); 24.60 (C(4)); 24.11 (C(3)). GC/EI-MS: 354 (*M*⁺; calc. 354).

A mixture of **1** (1.1 g, 3.2 mmol) and dibutyloxostannane (0.95 g, 3.8 mmol) in dry toluene (5 ml) was stirred for 2 h under reflux in a *Soxhlet* apparatus. After addition of CsF (0.57 g, 3.8 mmol), the mixture was concentrated at 50°/0.1 Torr. Molecular sieves (3 Å, 1 g) and MeCN (15 ml) were added to the residue containing stannylene acetal **2**, the suspension was cooled to 0°, and triflate **4** (3.4 g, 9.6 mmol) was added slowly with vigorous stirring, which was continued for 2 h at r.t. (TLC (toluene/acetone 4:1) monitoring). The mixture was filtered through a *Celite* pad, the filtrate concentrated, and the residue chromatographed (toluene/acetone 6:1): **5** (0.82 g, 52%). [α]_D = +11.0 (*c*=0.6, CHCl₃). ¹H-NMR (600 MHz, CDCl₃): 5.11 (s, PhCH₂OCO); 4.94 (*d*, *J*=10.9, 1 H, PhCH); 4.76 (*d*, *J*=11.9, 1 H, PhCH); 4.67 (*d*, *J*=11.9, 1 H, PhCH); 4.64 (*d*, *J*=10.9, 1 H, PhCH); 4.35 (*d*, *J*(1,2)=1.0, H-C(1)); 4.09 (br. s, H-C(2)); 3.89, 3.88 (2*t*, *J*=6.5, 1 H, H_a-C(1'')); 3.55–3.50 (m, H-C(4), H-C(3)); 3.47, 3.45 (2*t*, *J*=6.5, 1 H, H_b-C(1'')); 3.33–3.28 (m, H-C(5)); 2.41 (br. s, OH-C(2)); 2.36 (t, *J*=7.5, CH₂(5'')); 1.69–1.60 (m, CH₂(4''), CH₂(2'')); 1.41–1.36 (m, CH₂(3'')); 1.33 (*d*, *J*(5,6)=6.2, Me(6)). ¹³C-NMR (150 MHz, CDCl₃): 173.40 (C=O); 95.53 (*J*(C,H)=156.4, C(1)); 81.35 (C(3)); 79.61 (C(4)); 75.43 (PhCH₂); 71.37 (C(5)); 71.27 (PhCH₂); 69.30 (C(1'')); 68.44 (C(2)); 66.05 (PhCH₂OCO); 34.10 (C(5'')); 29.10 (C(2'')); 25.51 (C(3'')); 24.60 (C(4'')); 17.81 (C(6)). HR-ESI-MS: 571.2676 ([*M*+Na]⁺; calc. 571.2672). Anal. calc. for C₃₃H₄₀O₇ (548.67): C 72.24, H 7.35; found: C 72.13, H 7.38.

Methyl 3-O-Acetyl-2,4-di-O-benzyl-6-deoxy-1-thio- α -L-mannopyranoside (6) was prepared as described [18]. ¹H-NMR (300 MHz, CDCl₃): 5.16–5.12 (m, 2 H; incl. br. s at 5.15, H-C(1)); 5.13 (*dd*, *J*(2,3)=3.3, *J*(3,4)=9.6, H-C(3)); 4.68 (*d*, *J*=12.2, 1 H, PhCH); 4.56 (*d*, *J*=11.4, 1 H, PhCH); 4.53 (*d*, *J*=11.4, 1 H, PhCH); 4.50 (*d*, *J*=12.2, 1 H, PhCH); 4.13–4.04 (m, H-C(5)); 3.95 (*dd*, *J*(1,2)=1.9, H-C(2)); 3.66 (t, *J*=9.5, H-C(4)); 2.10 (s, MeS); 1.95 (s, MeCO); 1.34 (*d*, *J*(5,6)=6.3, Me(6)). ¹³C-NMR (75 MHz, CDCl₃): 170.04 (C=O); 82.94 (C(1)); 79.21 (C(4)); 77.30 (C(2)); 74.84 (PhCH₂); 73.78 (C(3)); 72.42 (PhCH₂); 68.04 (C(5)); 20.97 (MeCO); 17.96 (C(6)); 13.64 (MeS).

6-(Benzyloxy)-6-oxohexyl 2-O-[3-O-Acetyl-2,4-di-O-benzyl-6-deoxy- α -L-mannopyranosyl]-3,4-di-O-benzyl-6-deoxy- β -L-mannopyranoside (7). A mixture of **5** (2.36 g, 4.37 mmol), **6** (2.2 g, 5.25 mmol), and 4-Å molecular sieves (1.35 g) in CH₂Cl₂ (30 ml) was stirred for 15 min at r.t. The mixture was cooled to 0°, and NIS (1.39 g, 6.16 mmol) was added, followed by solid AgOTf (0.45 g, 1.76 mmol). The mixture

turned immediately red, and stirring was continued for 30 min at r.t. when TLC (hexane/AcOEt 3:2) showed that the reaction was complete. The mixture was filtered through a *Celite* pad into a separatory funnel containing aq. NaHCO_3 and $\text{Na}_2\text{S}_2\text{O}_3$ soln., and the product was extracted with CH_2Cl_2 . The combined org. phase was dried and concentrated and the residue subjected to CC (hexane/AcOEt 20:1 $\rightarrow$ 7:1): 3.23 g (82%) of **7**. $[\alpha]_{\text{D}}^{25} = +38.5$ ($c = 0.8$, CHCl_3). $^1\text{H-NMR}$ (600 MHz, CDCl_3): 5.27 (*dd*, $J(2,3) = 3.5$, $J(3,4) = 9.5$, $\text{H-C}(3^{\text{II}})$); 5.14 (*d*, $J(1,2) = 1.6$, $\text{H-C}(1^{\text{II}})$); 5.09 (*s*, PhCH_2OCO); 4.90–4.61 (*m*, 6 H, PhCH); 4.41 (*d*, $J = 12.2$, 1 H, PhCH); 4.35–4.32 (*m*, 2 H, $\text{H-C}(5^{\text{II}})$, incl. *d* at 4.32, $J(1,2) = 0.8$, $\text{H-C}(1^{\text{I}})$); 4.18–4.16 (*m*, 2 H, PhCH ; incl. signal at ca. 4.16, $\text{H-C}(2^{\text{I}})$); 3.96 (*dd*, $\text{H-C}(2^{\text{II}})$); 3.86, 3.84 (*2t*, $J = 6.5$, 1 H, $\text{H}_a\text{-C}(1'')$); 3.57 (*t*, $J = 9.4$, $\text{H-C}(4^{\text{II}})$); 3.50–3.47 (*m*, $\text{H-C}(3^{\text{I}})$, $\text{H-C}(4^{\text{I}})$); 3.37, 3.36 (*2t*, $J = 6.5$, 1 H, $\text{H}_b\text{-C}(1'')$); 3.30–3.27 (*m*, $\text{H-C}(5^{\text{I}})$); 2.36 (*t*, $J = 6.8$, $\text{CH}_2(5'')$); 1.90 (*s*, MeCO); 1.65–1.56 (*m*, $\text{CH}_2(4'')$, $\text{CH}_2(2'')$); 1.40–1.34 (*m*, $\text{CH}_2(3'')$); 1.33 (*d*, $J(5,6) = 5.8$, $\text{Me}(6^{\text{II}})$); 1.27 (*d*, $J(5,6) = 6.2$, $\text{Me}(6^{\text{I}})$). $^{13}\text{C-NMR}$ (150 MHz, CDCl_3): 173.47, 170.06 (C=O); 99.74 ($J(\text{C,H}) = 155.6$, $\text{C}(1^{\text{I}})$); 98.40 ($J(\text{C,H}) = 173.2$, $\text{C}(1^{\text{II}})$); 82.79 ($\text{C}(3^{\text{I}})$); 80.37 ($\text{C}(4^{\text{I}})$); 79.26 ($\text{C}(4^{\text{II}})$); 76.05 ($\text{C}(2^{\text{II}})$); 75.49 (PhCH_2); 74.49 (PhCH_2); 73.62 ($\text{C}(3^{\text{II}})$); 73.60 ($\text{C}(2^{\text{I}})$); 72.72 (PhCH_2); 72.24 (PhCH_2); 71.75 ($\text{C}(5^{\text{I}})$); 69.29 ($\text{C}(1'')$); 67.32 ($\text{C}(5^{\text{II}})$); 66.02 (PhCH_2OCO); 34.11 ($\text{C}(5'')$); 29.30 ($\text{C}(2'')$); 25.62 ($\text{C}(3'')$); 24.70 ($\text{C}(4'')$); 21.07 (MeCO); 17.85 ($\text{C}(6^{\text{II}})$); 17.78 ($\text{C}(6^{\text{I}})$). HR-ESI-MS: 939.4394 ($[\text{M} + \text{Na}]^+$; calc. 939.4295). Anal. calc. for $\text{C}_{55}\text{H}_{64}\text{O}_{12}$ (917.09): C 72.03, H 7.03; found: C 72.10, H 7.08.

6-Methoxy-6-oxohexyl 3,4-Di-O-benzyl-2-O-(2,4-di-O-benzyl-6-deoxy- α -L-mannopyranosyl)-6-deoxy- β -L-mannopyranoside (8). Methanolic 0.1M MeONa was added to a soln. of **7** (3.35 g, 3.65 mmol) in MeOH (50 ml) until the soln. became strongly basic. After ca. 5 h (TLC (hexane/AcOEt 3:2) monitoring), the mixture was neutralized with *Amberlite IR-120* and filtered, the filtrate concentrated, and the residue subjected to CC (toluene/acetone 100:1 $\rightarrow$ 20:1): 2.73 g (94%) of pure **8**. Colorless oil. $[\alpha]_{\text{D}}^{25} = +13$ ($c = 0.6$, CHCl_3). $^1\text{H-NMR}$ (600 MHz, CDCl_3): 5.26 (*d*, $J(1,2) = 1.2$, $\text{H-C}(1^{\text{II}})$); 4.92–4.63 (*m*, 6 H, PhCH); 4.42 (*d*, $J = 11.8$, 1 H, PhCH); 4.33 (*d*, $J(1,2) = 0.9$, $\text{H-C}(1^{\text{I}})$); 4.25–4.22 (*m*, 2 H, $\text{H-C}(5^{\text{II}})$, incl. *d* at 4.23, $J = 11.8$, PhCH); 4.20 (*br. d*, $\text{H-C}(2^{\text{I}})$); 4.07 (*ddd*, $J(2,3) = 3.7$, $J(3,4) = 9.1$, $\text{H-C}(3^{\text{II}})$); 3.86, 3.85 (*2t*, $J = 6.7$, 1 H, $\text{H}_a\text{-C}(1'')$); 3.79 (*dd*, $\text{H-C}(2^{\text{II}})$); 3.62 (*s*, MeOCO); 3.51 (*dd*, $J(2,3) = 2.7$, $J(3,4) = 9.3$, $\text{H-C}(3^{\text{I}})$); 3.44 (*t*, $J = 9.2$, $\text{H-C}(4^{\text{I}})$); 3.39–3.37 (*2t*, $J = 6.9$, 1 H, $\text{H}_b\text{-C}(1'')$); 3.31–3.27 (*m*, 2 H, $\text{H-C}(5^{\text{I}})$, incl. *t* at 3.30, $J = 9.3$, $\text{H-C}(4^{\text{I}})$); 2.33 (*d*, $\text{OH-C}(3^{\text{II}})$); 2.23 (*t*, $J = 7.4$, $\text{CH}_2(5'')$); 1.62–1.56 (*m*, $\text{CH}_2(4'')$, $\text{CH}_2(2'')$); 1.39–1.35 (*m*, $\text{CH}_2(3'')$); 1.33 (*d*, $J(5,6) = 6.1$, $\text{Me}(6^{\text{I}})$); 1.28 (*d*, $J(5,6) = 6.3$, $\text{Me}(6^{\text{II}})$). $^{13}\text{C-NMR}$ (150 MHz, CDCl_3): 173.98 (C=O); 99.77 ($\text{C}(1^{\text{II}})$); 97.23 ($\text{C}(1^{\text{I}})$); 82.87 ($\text{C}(3^{\text{I}})$); 82.15 ($\text{C}(4^{\text{II}})$); 80.17 ($\text{C}(4^{\text{I}})$); 78.28 ($\text{C}(2^{\text{II}})$); 75.32 (PhCH_2); 74.38, 72.55 (PhCH_2); 72.44 ($\text{C}(2^{\text{I}})$); 72.04 (PhCH_2); 71.61 ($\text{C}(5^{\text{I}})$); 71.08 ($\text{C}(3^{\text{II}})$); 69.27 ($\text{C}(1'')$); 66.88 ($\text{C}(5^{\text{II}})$); 51.25 (MeOCO); 33.78 ($\text{C}(5'')$); 29.24 ($\text{C}(2'')$); 25.57 ($\text{C}(3'')$); 24.60 ($\text{C}(4'')$); 17.79, 17.78 ($\text{C}(6^{\text{I}})$, $\text{C}(6^{\text{II}})$). HR-ESI-MS: 799.4048 ($[\text{M} + 1]^+$; calc. 799.4057). Anal. calc. for $\text{C}_{47}\text{H}_{50}\text{O}_{11}$ (798.96): C 70.65, H 7.32; found: C 70.52, H 7.41.

6-Methoxy-6-oxohexyl O-3-O-Acetyl-2,4-di-O-benzyl-6-deoxy- α -L-mannopyranosyl-(1 $\rightarrow$ 3)-O-2,4-di-O-benzyl-6-deoxy- α -L-mannopyranosyl-(1 $\rightarrow$ 2)-3,4-di-O-benzyl-6-deoxy- β -L-mannopyranoside (9). As described for **7**, with **8** (2.65 g, 3.31 mmol), **6** (1.66 g, 3.98 mmol), 4-Å molecular sieves (1 g), AgOTf (0.34 g, 1.32 mmol), and NIS (1.05 g, 4.63 mmol). After ca. 30 min (TLC (toluene/AcOEt 9:1) monitoring), the mixture was worked up as described for **7**: 3.6 g (92%) of amorphous **9**. $[\alpha]_{\text{D}}^{25} = +21.9$ ($c = 1.2$, CHCl_3). $^1\text{H-NMR}$ (600 MHz, CDCl_3): 5.28 (*dd*, $J(2,3) = 3.4$, $J(3,4) = 9.7$, $\text{H-C}(3^{\text{III}})$); 5.18 (*d*, $J(1,2) = 1.7$, $\text{H-C}(1^{\text{II}})$); 5.17 (*d*, $J(1,2) = 1.6$, $\text{H-C}(1^{\text{III}})$); 4.88 (*d*, $J = 10.9$, 1 H, PhCH); 4.81 (*d*, $J = 11.9$, 1 H, PhCH); 4.68–4.65 (*m*, 3 H, PhCH); 4.61 (*d*, $J = 10.9$, 1 H, PhCH); 4.58 (*d*, $J = 11.2$, 1 H, PhCH); 4.48 (*d*, $J = 12.2$, 1 H, PhCH); 4.43 (*d*, $J = 12.2$, 1 H, PhCH); 4.36 (*d*, $J = 12.2$, 1 H, PhCH); 4.34 (*br. s*, $\text{H-C}(1^{\text{I}})$); 4.30–4.28 (*m*, $\text{H-C}(5^{\text{II}})$); 4.23 (*dd*, $J(2,3) = 3.0$, $J(3,4) = 9.5$, $\text{H-C}(3^{\text{II}})$); 4.18 (*dd*, $J(1,2) = 0.8$, $J(2,3) = 2.6$, $\text{H-C}(2^{\text{I}})$); 4.13 (*d*, $J = 12.2$, 1 H, PhCH); 3.90, 3.88 (*2t*, $J = 6.2$, 1 H, $\text{H}_a\text{-C}(1'')$); 3.83 (*dd*, $J(2,3) = 2.9$, $\text{H-C}(2^{\text{II}})$); 3.82 (*dd*, $J(2,3) = 3.2$, $\text{H-C}(2^{\text{III}})$); 3.81–3.77 (*m*, $\text{H-C}(5^{\text{III}})$); 3.63 (*s*, MeOCO); 3.59 (*t*, $J \approx 9.8$, $\text{H-C}(4^{\text{II}})$, $\text{H-C}(4^{\text{III}})$); 3.50 (*dd*, $J(3,4) = 9.3$, $\text{H-C}(3^{\text{I}})$); 3.41 (*t*, $J = 9.3$, $\text{H-C}(4^{\text{I}})$); 3.41, 3.39 (*2t*, $J = 6.2$, 1 H, $\text{H}_b\text{-C}(1'')$); 3.32–3.27 (*m*, $\text{H-C}(5^{\text{I}})$); 2.27 (*t*, $J = 7.5$, $\text{CH}_2(5'')$); 1.91 (*s*, MeCO); 1.65–1.59 (*m*, $\text{CH}_2(4'')$, $\text{CH}_2(2'')$); 1.42–1.38 (*m*, $\text{CH}_2(3'')$); 1.32 (*d*, $J(5,6) = 6.1$, $\text{Me}(6^{\text{I}})$); 1.24 (*d*, $J(5,6) = 6.4$, $\text{Me}(6^{\text{II}})$); 1.21 (*d*, $J(5,6) = 6.1$, $\text{Me}(6^{\text{III}})$). $^{13}\text{C-NMR}$ (150 MHz, CDCl_3): 174.07, 170.07 (C=O); 99.90 ($\text{C}(1^{\text{I}})$); 99.17 ($\text{C}(1^{\text{II}})$); 98.07 ($\text{C}(1^{\text{III}})$); 82.72 ($\text{C}(3^{\text{I}})$); 80.68 ($\text{C}(4^{\text{II}})$); 80.29 ($\text{C}(4^{\text{I}})$); 79.00 ($\text{C}(4^{\text{III}})$); 77.96 ($\text{C}(2^{\text{II}})$); 77.84 ($\text{C}(3^{\text{II}})$); 76.86 ($\text{C}(2^{\text{III}})$); 75.41, 74.59, 74.36 (PhCH_2); 73.53 ($\text{C}(3^{\text{III}})$);

73.21 (C(2^l)); 72.57, 72.51, 71.97 (PhCH₂); 71.64 (C(5^l)); 69.44 (C(1^{''})); 67.96 (C(5^{ll})); 67.93 (C(5^{lll})); 51.39 (MeOCO); 33.89 (C(5^{''})); 29.33 (C(2^{''})); 25.63 (C(3^{''})); 24.69 (C(4^{''})); 21.03 (MeCO); 17.91 (C(6^{lll})); 17.87 (C(6^{ll})); 17.71 (C(6^l)). HR-ESI-MS: 1189.5505 ([M+Na]⁺; calc. 1189.5501). Anal. calc. for C₆₉H₈₂O₁₆ (1167.38): C 70.99, H 7.08; found: C 70.96, H 7.24.

6-Methoxy-6-oxohexyl O-2,4-Di-O-benzyl-6-deoxy- α -L-mannopyranosyl-(1 $\rightarrow$ 3)-O-2,4-di-O-benzyl-6-deoxy- α -L-mannopyranosyl-(1 $\rightarrow$ 2)-3,4-di-O-benzyl-6-deoxy- β -L-mannopyranoside (10). As described for **8**, with **9** (3.73 g, 3.31 mmol) and 0.1M MeONa. The mixture was neutralized with dry ice and concentrated and the residue subjected to CC (hexane/AcOEt 10 : 1 $\rightarrow$ 5 : 1): 3.45 g (96%) of syrupy **10**. [α]_D = +13.7 (*c* = 0.4, CHCl₃). ¹H-NMR (600 MHz, CDCl₃): 5.22 (br. s, H-C(1^{lll})); 5.18 (*d*, *J*(1, 2) = 1.6, H-C(1^{ll})); 4.89–4.39 (*m*, 10 H, PhCH); 4.34–4.25 (*m*, 3 H, H-C(5^{ll}), PhCH; incl. *d* at 4.34, *J*(1, 2) = 0.8, H-C(1^l)); 4.24 (*dd*, *J*(2, 3) = 2.9, *J*(3, 4) = 9.5, H-C(3^{ll})); 4.17 (*d*, *J*(2, 3) = 2.6, H-C(2^l)); 4.03 (*d*, *J* = 11.8, 1 H, PhCH); 3.93 (*ddd*, H-C(3^{lll})); 3.90, 3.89 (*2t*, *J* = 6.4, 1 H, H_a-C(1^{''})); 3.80 (*dd*, *J*(1, 2) = 1.9, H-C(2^{ll})); 3.72–3.68 (*m*, H-C(5^{lll})); 3.67 (*dd*, *J*(1, 2) = 1.4, *J*(2, 3) = 3.5, H-C(2^{lll})); 3.64 (*s*, MeOCO); 3.57 (*t*, *J* = 9.5, H-C(4^{ll})); 3.50 (*dd*, *J*(3, 4) = 9.2, H-C(3^l)); 3.41 (*t*, *J* = 9.2, H-C(4^l)); 3.42, 3.39 (*2t*, *J* = 6.4, 1 H, H_b-C(1^{''})); 3.31–3.28 (*m*, H-C(5^l)); 3.26 (*t*, *J* = 9.3, H-C(4^{lll})); 2.28 (*t*, *J* = 7.4, CH₂(5^{''})); 2.25 (*d*, *J*(3, OH) = 10.1, OH-C(3^{lll})); 1.65–1.60 (*m*, CH₂(4^{''}), CH₂(2^{''})); 1.42–1.38 (*m*, CH₂(3^{''})); 1.32 (*d*, *J*(5, 6) = 6.1, Me(6^l)); 1.25 (*d*, *J*(5, 6) = 6.2, Me(6^{ll})); 1.21 (*d*, *J*(5, 6) = 6.3, Me(6^{lll})). ¹³C-NMR (150 MHz, CDCl₃): 174.07 (C=O); 99.93 (C(1^l)); 98.30 (C(1^{lll})); 98.15 (C(1^{ll})); 82.69 (C(3^l)); 82.17 (C(4^{lll})); 80.99 (C(4^{ll})); 80.41 (C(4^l)); 79.18 (C(2^{lll})); 78.06 (C(2^{ll})); 77.00 (C(3^{ll})); 75.46, 74.74, 74.35 (PhCH₂); 73.31 (C(2^l)); 72.61, 72.32, 72.00 (PhCH₂); 71.68 (C(3^{lll})); 71.62 (C(5^l)); 69.50 (C(1^{''})); 67.98 (C(5^{ll})); 67.35 (C(5^{lll})); 51.42 (MeOCO); 33.91 (C(5^{''})); 29.33 (C(2^{''})); 25.63 (C(3^{''})); 24.71 (C(4^{''})); 17.93 (C(6^{ll})); 17.90, (C(6^l)); 17.79 (C(6^{lll})). HR-ESI-MS: 1147.5372 ([M+Na]⁺; calc. 1147.5395). Anal. calc. for C₆₇H₈₀O₁₅ (1125.34): C 71.51, H 7.17; found: C 71.56, H 7.16.

6-Methoxy-6-oxohexyl O-4-Azido-3-O-benzyl-4,6-dideoxy- β -D-glucopyranosyl-(1 $\rightarrow$ 3)-O-2,4-di-O-benzyl-6-deoxy- α -L-mannopyranosyl-(1 $\rightarrow$ 3)-O-2,4-di-O-benzyl-6-deoxy- α -L-mannopyranosyl-(1 $\rightarrow$ 2)-3,4-di-O-benzyl-6-deoxy- β -L-mannopyranoside (13). As described for **9**, with ethyl 4-azido-3-O-benzyl-2-O-(bromoacetyl)-4,6-dideoxy-1-thio- β -D-glucopyranoside (**11**) [14] (0.72 g, 1.6 mmol), **10** (2 g, 1.78 mmol), NIS (0.5 g, 2.24 mmol), and AgOTf (0.16 g, 0.64 mmol). After the usual workup, chromatography gave 1.72 g of **13** (66%). [α]_D = +17.9 (*c* = 1, CHCl₃). ¹H-NMR (600 MHz, CDCl₃): 5.17 (*d*, *J*(1, 2) = 1.8, H-C(1^{ll})); 5.11 (br. s, H-C(1^{lll})); 5.04 (*dd*, *J*(1, 2) = 8.0, *J*(2, 3) = 9.6, H-C(2^{IV})); 4.89–4.32 (*m*, 16 H, 14 PhCH; incl. *d* at 4.64, H-C(1^{IV}) and *s* at 4.34, H-C(1^l)); 4.32–4.28 (*m*, H-C(5^{ll})); 4.19 (*dd*, *J*(2, 3) = 3.1, *J*(3, 4) = 9.4, H-C(3^{ll})); 4.16 (*d*, *J*(2, 3) = 2.5, H-C(2^l)); 4.08 (*dd*, *J*(2, 3) = 3.0, *J*(3, 4) = 9.6, H-C(3^{lll})); 3.88, 3.87 (*2t*, *J* = 6.5, 1 H, H_a-C(1^{''})); 3.86 (*dd*, H-C(2^{ll})); 3.83–3.79 (*m*, 2 H, H-C(5^{lll}); incl. *dd* at 3.82, *J*(1, 2) = 1.8, H-C(2^{lll})); 3.63 (*s*, MeOCO); 3.55 (*t*, *J* = 9.5, H-C(4^{lll})); 3.52–3.49 (*m*, H-C(4^{ll}), H-C(3^l)); 3.43 (*t*, *J* = 9.2, H-C(4^l)); 3.41, 3.39 (*2t*, *J* = 6.5, 1 H, H_b-C(1^{''})); 3.37 (*t*, *J* = 9.4, H-C(3^{IV})); 3.32, 3.30 (*2d*, *J* = 12.6, CH₂Br); 3.31–3.28 (*m*, H-C(5^l)); 3.12 (*t*, *J* = 9.5, H-C(4^{IV})); 3.02–2.90 (*m*, H-C(5^{IV})); 2.25 (*t*, *J* = 7.8, CH₂(5^{''})); 1.62–1.58 (*m*, CH₂(4^{''}), CH₂(2^{''})); 1.40–1.36 (*m*, CH₂(3^{''})); 1.31 (*d*, *J*(5, 6) = 6.1, Me(6^l)); 1.23 (*d*, *J*(5, 6) = 6.2, Me(6^{ll})); 1.21 (*d*, *J*(5, 6) = 6.2, Me(6^{lll})); 1.10 (*d*, *J*(5, 6) = 6.1, Me(6^{IV})). ¹³C-NMR (150 MHz, CDCl₃): 174.00, 165.53 (C=O); 100.59 (*J*(C, H) = 160.9, C(1^{IV})); 99.97 (br. s, *J*(C, H) = 171.2, C(1^{lll})); 98.84 (*J*(C, H) = 152.3, C(1^l)); 97.90 (br. s, *J*(C, H) = 169.9, C(1^{ll})); 82.69 (C(3^l)); 80.98 (C(3^{IV})); 82.42 (C(4^{lll})); 80.37 (C(4^{ll})); 80.29 (C(4^l)); 79.14 (C(3^{lll})); 78.59 (C(2^{lll}), C(3^{ll})); 78.29 (C(2^{ll})); 75.40, 75.04, 74.93, 74.51, 74.42 (PhCH₂); 73.48 (br. s, C(2^l)); 73.15, 72.57, 71.70 (PhCH₂); 71.63 (C(5^l)); 70.51 (C(5^{IV})); 69.42 (C(1^{''})); 68.14 (C(5^{lll})); 67.72 (C(5^{ll})); 67.63 (C(4^{IV})); 51.40 (MeOCO); 33.87 (C(5^{''})); 29.31 (C(2^{''})); 25.63 (C(3^{''})); 25.22 (CH₂Br); 24.70 (C(4^{''})); 18.15 (C(6^{IV})); 17.86, 17.83, 17.79 (C(6^l), C(6^{ll}), C(6^{lll})). HR-ESI-MS: 1528.5813 ([M+Na]⁺; calc. 1528.5719), 1506.5923 ([M+1]⁺; calc. 1506.5900).

As described for **6**, the fully protected **13** (1.47 g, 1.39 mmol) was treated with 0.1M MeONa for 1 h (TLC (toluene/acetone 9 : 1) monitoring). CC (toluene/acetone 30 : 1) gave amorphous **13** (1.2 g, 91%). [α]_D = +40 (*c* = 0.4, CHCl₃). ¹H-NMR (600 MHz, CDCl₃): 5.18 (*d*, *J*(1, 2) = 1.8, H-C(1^{ll})); 5.10 (br. s, H-C(1^{lll})); 4.90–4.40 (*m*, 13 H, PhCH); 4.34–4.32 (*m*, 3 H, PhCH; incl. *d* at 4.34, *J*(1, 2) = 0.8, H-C(1^l), and *d* at 4.33, *J*(1, 2) = 8.0, H-C(1^{IV})); 4.27–4.22 (*m*, 1 H, H-C(5^{ll})); 4.19–4.16 (*m*, H-C(3^{ll}), H-C(2^l)); 4.04 (*dd*, *J*(2, 3) = 3.1, *J*(3, 4) = 9.5, H-C(3^{lll})); 3.89, 3.87 (*2t*, *J* = 6.3, 1 H, H_a-C(1^{''})); 3.84–3.83 (*m*, H-C(2^{ll}), H-C(2^{lll})); 3.81–3.76 (*m*, H-C(5^{lll})); 3.63 (*s*, MeOCO); 3.61 (*t*, *J* = 9.6, H-

C(4^{III}); 3.52–3.48 (*m*, H–C(3^I), H–C(2^{IV}), H–C(4^{II})); 3.43 (*t*, *J* = 9.2, H–C(4^I)); 3.41, 3.39 (2*t*, *J* = 6.3, 1 H, H_b–C(1^{IV})); 3.32–3.27 (*m*, 2 H, H–C(5^I); incl. *t* at 3.30, *J* = 9.2, H–C(3^{IV})); 3.04 (*t*, *J* = 9.2, H–C(4^{IV})); 3.03–2.98 (*m*, H–C(5^{IV})); 2.70 (*br. s.*, OH–C(2^{IV})); 2.25 (*t*, *J* = 7.8, H–CH₂(5^{IV})); 1.61–1.58 (*m*, 4 H, CH₂(4^{IV}), CH₂(2^{IV})); 1.38–1.33 (*m*, CH₂(3^{IV})); 1.32 (*d*, *J*(5,6) = 6.1, Me(6^I)); 1.24 (*d*, *J*(5,6) = 6.3, Me(6^{III})); 1.20 (*d*, *J*(5,6) = 6.2, Me(6^{II})); 1.14 (*d*, *J*(5,6) = 5.8, Me(6^{IV})). ¹³C-NMR (150 MHz, CDCl₃): 174.07 (C=O); 103.67 (C(1^{IV})); 99.89 (C(1^I)); 99.77 (*br. s.*, C(1^{III})); 97.88 (*br. s.*, C(1^{II})); 82.72 (C(3^I)); 82.01 (C(3^{IV})); 80.65 (C(4^{III})); 80.44 (C(4^{II})); 80.40 (C(4^I)); 80.08 (C(3^{III})); 78.31 (C(2^{II}), C(2^{III})); 78.15 (C(3^{II})); 75.73 (C(2^{IV})); 75.47, 75.16, 74.75, 74.41 (PhCH₂); 73.40 (*br. s.*, C(2^I)); 73.04, 72.63, 71.78 (PhCH₂); 71.68 (C(5^I)); 70.61 (C(5^{IV})); 69.49 (C(1^{IV})); 68.28 (C(5^{III})); 67.76 (C(5^{II})); 67.00 (C(4^{IV})); 51.40 (MeOCO); 33.87 (C(5^{IV})); 29.30 (C(2^{IV})); 25.61 (C(3^{IV})); 24.68 (C(4^{IV})); 18.46 (C(6^{IV})); 18.04 (C(6^{III})); 17.87 (C(6^{II})); 17.80 (C(6^I)). HR-ESI-MS: 1408.6567 ([*M* + Na]⁺; calc. 1408.6508). Anal. calc. for C₈₀H₉₅N₃O₁₈ (1386.62): C 69.29, H 6.91, N 3.03; found: C 69.44, H 7.04, N 3.05.

6-Methoxy-6-oxohexyl O-4-Azido-3-O-benzyl-4,6-dideoxy-2-O-methyl-β-D-glucopyranosyl-(1 → 3)-O-2,4-di-O-benzyl-6-deoxy-α-L-mannopyranosyl-(1 → 3)-O-2,4-di-O-benzyl-6-deoxy-α-L-mannopyranosyl-(1 → 2)-3,4-di-O-benzyl-6-deoxy-β-L-mannopyranoside (14). A soln. of **13** (440 mg, 0.32 mmol) in 1,2-dimethoxyethane (10 ml) was treated with MeI (18 ml, 288 mmol) and Ag₂O (16.9 g, 65.8 mmol) in the presence of a cat. amount of Me₂S [15]. After stirring for 24 h at r.t. (TLC (toluene/acetone 10 : 1) monitoring), the mixture was filtered, the solids were washed with toluene, the filtrate was concentrated, and the residue subjected to CC (toluene/acetone 1 : 1): syrupy **14** (400 mg, 90%). [*a*]_D = +28.9 (*c* = 0.4, CHCl₃). ¹H-NMR (600 MHz, CDCl₃): 5.16 (*d*, *J*(1,2) = 1.7, H–C(1^{II})); 5.12 (*br. s.*, H–C(1^{III})); 4.96–4.36 (*m*, 15 H, 14 PhCH; incl. *d* at 4.58, *J*(1,2) = 8.0, H–C(1^{IV})); 4.33 (*d*, *J*(1,2) = 0.8, H–C(1^I)); 4.28–4.24 (*m*, H–C(5^{II})); 4.20 (*dd*, *J*(2,3) = 3.1, *J*(3,4) = 9.4, H–C(3^{II})); 4.16 (*d*, *J*(2,3) = 2.3, H–C(2^I)); 4.10 (*dd*, *J*(2,3) = 3.1, *J*(3,4) = 9.5, H–C(3^{III})); 3.89, 3.88 (2*t*, *J* = 6.9, 1 H, H_a–C(1^{IV})); 3.87–3.80 (*m*, H–C(2^{II}), H–C(2^{III}), H–C(5^{III})); 3.63 (*s*, 2 MeO); 3.61 (*t*, *J* = 9.5, H–C(4^{III})); 3.54–3.49 (*m*, 2 H, H–C(4^{II}); incl. *dd* at 3.50, *J*(3,4) = 9.3, H–C(3^I)); 3.43 (*t*, *J* = 9.2, H–C(4^I)); 3.40, 3.38 (2*t*, *J* = 6.9, 1 H, H_b–C(1^{IV})); 3.33–3.28 (*m*, 2 H, H–C(5^I); incl. *t* at 3.31, *J* = 9.2, H–C(3^{IV})); 3.11 (*dd*, *J*(2,3) = 9.0, H–C(2^{IV})); 3.02 (*t*, *J* = 9.8, H–C(4^{IV})); 2.95–2.99 (*m*, H–C(5^{IV})); 2.26 (*t*, *J* = 7.8, H–CH₂(5^{IV})); 1.64–1.57 (*m*, CH₂(4^{IV}), CH₂(2^{IV})); 1.41–1.36 (*m*, CH₂(3^{IV})); 1.33 (*d*, *J*(5,6) = 6.2, Me(6^I)); 1.27 (*d*, *J*(5,6) = 6.0, Me(6^{III})); 1.21 (*d*, *J*(5,6) = 6.2, Me(6^{II})); 1.07 (*d*, *J*(5,6) = 5.9, Me(6^{IV})). ¹³C-NMR (150 MHz, CDCl₃): 174.03 (C=O); 103.44 (C(1^{IV})); 100.03 (*br. s.*, C(1^{III})); 99.84 (C(1^I)); 98.04 (C(1^{II})); 84.66 (C(2^{IV})); 82.68 (C(3^I)); 82.59 (C(3^{IV})); 80.85 (C(4^{III})); 80.44 (C(4^{II})); 80.33 (C(4^I)); 79.28 (C(2^{III})); 78.55 (C(3^{III})); 78.25 (C(2^{II})); 78.21 (C(3^{II})); 75.42, 75.32, 74.45 (PhCH₂); 73.44 (*br. s.*, C(2^I)); 73.15, 72.53, 71.81 (PhCH₂); 71.64 (C(5^I)); 70.08 (C(5^{IV})); 69.41 (C(1^{IV})); 68.12 (C(5^{III})); 67.79 (C(5^{II})); 67.50 (C(4^{IV})); 60.04 (MeO); 51.37 (MeOCO); 33.87 (C(5^{IV})); 29.28 (C(2^{IV})); 25.60 (C(3^{IV})); 24.67 (C(4^{IV})); 17.98 (C(6^{IV})); 17.92 (C(6^{III})); 17.83 (C(6^{II})); 17.76 (C(6^I)). HR-ESI-MS: 1422.6680 ([*M* + Na]⁺; calc. 1422.6665). Anal. calc. for C₈₁H₉₇N₃O₁₈ (1400.65): C 69.46, H 6.98, N 3.00; found: C 69.74, H 7.04, N 2.94.

6-Methoxy-6-oxohexyl O-3-O-Benzyl-4,6-dideoxy-4-[(3-hydroxy-3-methyl-1-oxobutyl)amino]-2-O-methyl-β-D-glucopyranosyl-(1 → 3)-O-2,4-di-O-benzyl-6-deoxy-α-L-mannopyranosyl-(1 → 3)-O-2,4-di-O-benzyl-6-deoxy-α-L-mannopyranosyl-(1 → 2)-3,4-di-O-benzyl-6-deoxy-β-L-mannopyranoside (16). H₂O (*ca.* 4 ml) was added to a mixture of **14** (440 mg, 0.31 mmol) and pyridine (16 ml) until slight turbidity, followed by a few drops of pyridine until a clear soln. was formed. A slow stream of H₂S gas was passed through this soln. for 0.5 h. The mixture was stirred overnight at 40° (TLC (hexane/AcOEt 2 : 1) monitoring). After concentration, the residue was subjected to CC (hexane/AcOEt 3 : 1 → 2 : 5): pure (TLC, NMR) **15** (310 mg, 72%). ¹H-NMR (600 MHz, CDCl₃): 5.15 (*d*, *J*(1,2) = 1.7, H–C(1^{II})); 5.13 (*br. s.*, H–C(1^{III})); 4.98 (*d*, *J* = 10.5, 1 H, PhCH); 4.97 (*d*, *J* = 11.3, 1 H, PhCH); 4.88 (*d*, *J* = 10.8, 1 H, PhCH); 4.71 (*d*, *J* = 11.5, 1 H, PhCH); 4.68–4.37 (*m*, 11 H, 10 PhCH; incl. *d* at 4.67, *J*(1,2) ≈ 9.8, H–C(1^{IV})); 4.32 (*br. s.*, H–C(1^I)); 4.27–4.23 (*m*, H–C(5^{II})); 4.19 (*dd*, *J*(2,3) = 3.1, *J*(3,4) = 9.2, H–C(3^{II})); 4.15 (*dd*, *J*(2,3) = 2.5, H–C(3^{III})); 4.12 (*br. s.*, H–C(2^I)); 3.90 (*dd*, *J*(1,2) = 1.7, H–C(2^{III})); 3.88, 3.86 (2*t*, *J* = 6.5, 1 H, H_a–C(1^{IV})); 3.84 (*dd*, H–C(2^{II})); 3.83–3.79 (*m*, H–C(5^{III})); 3.65 (*s*, MeO); 3.63 (*s*, MeOCO); 3.62 (*t*, *J* = 9.5, H–C(4^{III})); 3.54–3.51 (*m*, H–C(4^{II})); 3.49 (*dd*, *J*(2,3) = 2.5, *J*(3,4) = 9.4, H–C(3^I)); 3.42 (*t*, *J* = 9.3, H–C(4^I)); 3.39–3.37 (2*t*, *J* = 6.5, 1 H, H_b–C(1^{IV})); 3.30–3.26 (*m*, H–C(5^I)); 3.14–3.11 (*m*, H–C(2^{IV}), H–C(3^{IV})); 3.05–3.01 (*m*, H–C(5^{IV})); 2.48–2.45 (*m*, H–C(4^{IV})); 2.26 (*t*, *J* = 7.8, CH₂(5^{IV})); 1.65–1.58 (*m*, CH₂(4^{IV}), CH₂(2^{IV})); 1.40–1.36 (*m*, CH₂(3^{IV})); 1.31 (*d*, *J*(5,6) = 6.1,

Me(6^I)); 1.26 (*d*, *J*(5,6)=6.2, Me(6^{III})); 1.20 (*d*, *J*(5,6)=6.2, Me(6^{II})); 1.03 (*d*, *J*(5,6)=6.1, Me(6^{IV})). ¹³C-NMR (150 MHz, CDCl₃): 174.12 (C=O); 103.75 (C(1^{IV})); 100.17 (br. *s*, C(1^{III})); 99.87 (C(1^I)); 98.13 (C(1^{II})); 85.28 (C(3^{IV})); 83.95 (C(2^{II})); 82.70 (C(3^I)); 80.99 (C(4^{III})); 80.50 (C(4^{II})); 80.38 (C(4^I)); 79.44 (C(2^{III})); 78.25 (C(2^{II})); 78.17 (C(3^{III})); 78.10 (C(3^{II})); 75.46, 74.95, 74.52, 74.43 (PhCH₂); 73.52 (br. *s*, C(2^I)); 73.20, 72.55 (PhCH₂); 72.36 (C(5^{IV})); 71.86 (PhCH₂); 71.66 (C(5^I)); 69.42 (C(1^{II})); 68.16 (C(5^{III})); 67.86 (C(5^{II})); 60.41 (MeO); 58.04 (C(4^{IV})); 51.43 (MeOCO); 33.90 (C(5^{II})); 29.31 (C(2^{II})); 25.63 (C(3^{II})); 24.68 (C(4^{II})); 17.99 (C(6^{IV})); 17.94 (C(6^{III})); 17.86 (C(6^I)); 17.75 (C(6^{II})). HR-ESI-MS: 1374.6906 ([*M*+1]⁺; calc. 1374.6900).

HATU (83 mg, 0.22 mmol) followed by ⁱPr₂NEt (0.05 ml, 0.27 mmol) was added to a soln. of **15** (205 mg, 0.15 mmol) and 3-hydroxy-3-methylbutanoic acid (23 mg, 0.22 mmol) in CH₂Cl₂ (6 ml). The mixture was stirred for 1 h at r.t., when the soln. became clear (TLC (CH₂Cl₂/AcOEt 4:1) monitoring). After concentration, the residue was subjected to CC (CH₂Cl₂/MeOH 100:1): syrupy **16** (195 mg, 89%). [*α*]_D = +14 (*c*=0.3, CHCl₃). ¹H-NMR (600 MHz, CDCl₃): 5.35 (*d*, *J*(4,NH)=8.9, NH); 5.15 (*d*, *J*(1,2)=1.7, H-C(1^{II})); 5.11 (br. *s*, H-C(1^{III})); 5.00–4.37 (*m*, 15 H, 14 PhCH; incl. *d* at 4.58, *J*(1,2)=8.0, H-C(1^{IV})); 4.32 (*d*, *J*(1,2)=0.8, H-C(1^I)); 4.27–4.23 (*m*, H-C(5^{II})); 4.19 (*dd*, *J*(2,3)=3.1, *J*(3,4)=9.4, H-C(3^{II})); 4.15 (br. *d*, H-C(2^I)); 4.11 (*dd*, *J*(2,3)=3.1, *J*(3,4)=9.5, H-C(3^{III})); 3.89 (*dd*, *J*(1,2)=1.8, H-C(2^{III})); 3.88, 3.87 (*2t*, *J*=6.8, 1 H, H_a-C(1^{II})); 3.86–3.81 (*m*, 2 H, H-C(5^{III})); incl. *dd* at 3.85, *J*(1,2)=2.0, H-C(2^{II})); 3.65 (*t*, *J*=9.9, H-C(4^{IV})); 3.63 (*s*, MeOCO); 3.62 (*s*, MeO); 3.52–3.48 (*m*, 2 H, H-C(4^{II})); incl. *t* at 3.62, *J*=9.5, H-C(4^{III})); 3.49 (*dd*, *J*(2,3)=2.6, *J*(3,4)=9.3, H-C(3^I)); 3.42 (*t*, *J*=9.2, H-C(4^I)); 3.39, 3.38 (*2t*, *J*=6.8, 1 H, H_b-C(1^{II})); 3.31–3.26 (*m*, 2 H, H-C(5^I)); incl. *dd* at *ca.* 3.28, H-C(3^{IV})); 3.16 (*dd*, *J*(2,3)=8.9, H-C(2^{IV})); 3.12–3.08 (*m*, H-C(5^{IV})); 2.26 (*t*, *J*=7.8, CH₂(5^{II})); 2.19 (*d*, *J*=15.0, H_a-C(1^I)); 2.08 (*d*, *J*=15.0, H_b-C(1^I)); 1.63–1.57 (*m*, CH₂(4^{II}), CH₂(2^{II})); 1.40–1.36 (*m*, CH₂(3^{II})); 1.32 (*d*, *J*(5,6)=6.1, Me(6^I)); 1.27 (*d*, *J*(5,6)=6.3, Me(6^{III})); 1.21 (*s*, Me-C(3^I)); 1.20 (*d*, *J*(5,6)=6.3, Me(6^{II})); 1.18 (*s*, Me-C(3^{II})); 1.00 (*d*, *J*(5,6)=6.2, Me(6^{IV})). ¹³C-NMR (150 MHz, CDCl₃): 174.10, 172.31 (C=O); 103.68 (C(1^{IV})); 100.15 (br. *s*, C(1^{III})); 99.86 (C(1^I)); 98.08 (C(1^{II})); 84.82 (C(2^{IV})); 82.69 (C(3^I)); 80.82 (C(4^{III})); 80.44 (C(4^{II})); 80.36 (C(4^I)); 80.03 (C(3^{IV})); 79.24 (C(2^{III})); 78.91 (C(3^{III})); 78.23 (C(2^{II}), C(3^{II})); 75.44 (PhCH₂); 74.43 (2 PhCH₂); 73.65, (PhCH₂); 73.46 (br. *s*, C(2^I)); 73.27, 72.52, 71.83 (PhCH₂); 71.66 (C(5^I)); 70.78 (C(5^I)); 69.42 (C(3^I)); 69.39 (C(1^I)); 68.15 (C(5^{III})); 67.80 (C(5^{II})); 60.54 (MeO); 55.47 (C(4^{IV})); 51.43 (MeOCO); 47.68 (C(2^I)); 33.92 (C(5^{II})); 29.29 (C(2^{II})); 29.28, 29.26 (Me₂C(3^I)); 25.64 (C(3^{II})); 24.71 (C(4^{II})); 18.04 (C(6^{IV})); 17.96 (C(6^{III})); 17.84 (C(6^I)); 17.78 (C(6^{II})). HR-ESI-MS: 1496.7227 ([*M*+Na]⁺; calc. 1496.7284). Anal. calc. for C₈₆H₁₀₇NO₂₀ (1474.76): C 70.04, H 7.31, N 0.95; found: C 69.83, H 7.31, N 0.85.

6-Methoxy-6-oxohexyl O-4,6-Dideoxy-4-[(3-hydroxy-3-methyl-1-oxobutyl)amino]-2-O-methyl-β-D-glucopyranosyl-(1 → 3)-O-6-deoxy-α-L-mannopyranosyl-(1 → 3)-O-6-deoxy-α-L-mannopyranosyl-(1 → 2)-6-deoxy-β-L-mannopyranoside (17). A soln. of **16** (140 mg, 0.1 mmol) in 2-methoxyethanol (5 ml) was added to a suspension of 5% Pd/C catalyst (140 mg) in the same solvent (5 ml), and the mixture was stirred overnight under H₂ (TLC (CH₂Cl₂/MeOH 4:1) monitoring: 1 product). After filtration through a Celite pad, the filtrate was concentrated, and the residue subjected to CC (CH₂Cl₂/MeOH 4:1). After freeze-drying, **17** (75 mg, 94%) was obtained as a white amorphous solid. [*α*]_D = –31 (*c*=0.4, H₂O). HR-ESI-MS: 866.3970 ([*M*+Na]⁺; calc. 866.3998). NMR: *Tables 1 and 2.*

6-Methoxy-6-oxohexyl 6-Deoxy-β-L-mannopyranoside (21). A mixture of 6-deoxy-L-mannose monohydrate (1 g, 5.39 mmol) and dibutylxostannane (1.3 g, 5.31 mmol) in dry MeOH (25 ml) was stirred for 2 h at 60° until a clear soln. was obtained. Toluene (5 ml) and CsF (1 g, 6.62 mmol) were added, and the mixture was concentrated. The residue was dried at 50°/0.3 kPa for 2 h, then dissolved in DMF (8 ml), molecular sieves (3 Å, 0.75 g) were added, and the mixture was cooled to 0°. Freshly prepared benzyl 6-[[[(trifluoromethyl)sulfonyl]oxy]hexanoate (**4**; 1.3 g, 3.66 mmol) was added, and the mixture was stirred at 0° for 2 h (TLC (CH₂Cl₂/MeOH 4:1) monitoring of **4**). The mixture was filtered and concentrated, and the residue subjected to CC (CH₂Cl₂/acetone 4:1): 0.52 g (38%) of 6-(benzyloxy)-6-oxohexyl 6-deoxy-β-L-mannopyranoside (**20**). ¹H-NMR (300 MHz, CHCl₃): 5.11 (*s*, PhCH₂OCO); 4.45 (*s*, H-C(1)); 3.98 (br. *s*, H-C(2)); 3.93, 3.90 (*2t*, *J*=6.4, 1 H, H_a-C(1^{II})); 3.53, 3.50 (*2t*, *J*=6.4, 1 H, H_b-C(1^{II})); 3.44–3.41 (*m*, H-C(3), H-C(4)); 3.29–3.25 (*m*, H-C(5)); 2.52 (*d*, *J*(3,OH)=8.6, OH-C(3)); 2.48 (*d*, *J*(2,OH)=2.5, OH-C(2)); 2.37 (*t*, *J*=7.2, CH₂(5^{II})); 2.33 (br. *s*, OH-C(4)); 1.73–1.58 (*m*, CH₂(4^{II}), CH₂(2^{II})); 1.45–1.35 (*m*, 5 H, CH₂(3^{II})); incl. *d* at 1.36, *J*(5,6)=6.1, Me(6)). ¹³C-NMR (75 MHz, CHCl₃): 175.82

(C=O); 99.46 (C(1)); 74.40 (C(3)); 73.62 (C(4)); 71.51 (C(5)); 70.83 (C(2)); 69.34 (PhCH₂OCO); 66.18 (C(1'')); 34.11 (C(5'')); 29.03 (C(2'')); 27.40 (C(3'')); 24.62 (C(4'')); 18.02 (C(6)). HR-ESI-MS: 391.1741 ([M+Na]⁺; calc. 391.1733).

Ester **20** (0.5 g, 1.35 mmol) was dissolved in MeOH (25 ml), and the soln. was made strongly basic by addition of 0.1M MeONa/MeOH. After stirring overnight at r.t. (TLC (CH₂Cl₂/acetone 4:1) monitoring of the UV-positive **20**, formation of a single charring product) and usual workup, chromatography gave **21** (370 mg, 93%). M.p. 87–88° (from AcOEt). [α]_D = +42.0 (*c* = 0.3, CHCl₃). ¹H-NMR (600 MHz, CD₃OD): 4.47 (*d*, *J*(1,2) = 1.0, H–C(1)); 3.88–3.85 (*m*, H_a–C(1'')); 3.87 (*br. d*, H–C(2)); 3.67 (*s*, MeOCO); 3.55, 3.53 (*2t*, *J* = 6.7, 1 H, H_b–C(1'')); 3.42 (*dd*, *J*(2,3) = 3.2, *J*(3,4) = 9.4, H–C(3)); 3.39 (*t*, *J* = 9.3, H–C(4)); 3.26–3.22 (*m*, H–C(5)); 2.36 (*t*, *J* = 7.4, CH₂(5'')); 1.68–1.62 (*m*, CH₂(4''), CH₂(2'')); 1.46–1.41 (*m*, CH₂(3'')); 1.32 (*d*, *J*(5,6) = 6.2, Me(6)). ¹³C-NMR (150 MHz, CD₃OD): 175.80 (C=O); 100.58 (*J*(C,H) = 155.6); 74.95 (C(3)); 73.84 (C(4)); 73.51 (C(5)); 72.49 (C(2)); 70.16 (C(1'')); 34.66 (C(5'')); 30.29 (C(2'')); 26.59 (C(3'')); 25.73 (C(4'')); 17.98 (C(6)). HR-ESI-MS: 315.1411 ([M+Na]⁺; calc. 315.1420). Anal. calc. for C₁₃H₂₄NO₇ (292.33): C 53.41, H 8.28; found: C 53.11, H 8.27.

6-Methoxy-6-oxohexyl 6-Deoxy-2-O-[6-deoxy- α -L-mannopyranosyl]- β -L-mannopyranoside (**22**). As described for **17**, **8** (200 mg, 0.25 mmol) was treated with H₂. The product was eluted from a small silica gel column, mainly to remove residual catalyst. After freeze-drying, amorphous **22** (105 mg, 95%) was obtained. White solid. [α]_D = +66.6 (*c* = 0.5, H₂O). ¹H-NMR (600 MHz, D₂O): 5.03 (*d*, *J*(1,2) = 1.7, H–C(1'')); 4.62 (*d*, *J*(1,2) = 0.9, H–C(1'')); 4.17–4.13 (*m*, H–C(5'')); 4.08 (*dd*, *J*(2,3) = 3.4, H–C(2'')); 4.00 (*dd*, *J*(2,3) = 3.1, H–C(2'')); 3.85–3.81 (*m*, 2 H, H_a–C(1'')); incl. *dd* at 3.84, *J*(3,4) = 9.8, H–C(3'')); 3.68–3.66 (*m*, 4 H, H–C(3'')); incl. *s* at 3.68, MeOCO); 3.57, 3.55 (*2t*, *J* = 6.5, 1 H, H_b–C(1'')); 3.42–3.37 (*m*, 3 H, H–C(5'')); incl. *t* at 3.41, *J* = 9.8, H–C(4'') and *t* at 3.40, *J* = 9.3, H–C(4'')); 2.40 (*t*, *J* = 7.5, CH₂(5'')); 1.64–1.57 (*m*, CH₂(4''), CH₂(2'')); 1.40–1.36 (*m*, CH₂(3'')); 1.29 (*d*, *J*(5,6) = 5.9, Me(6'')); 1.23 (*d*, *J*(5,6) = 6.3, Me(6'')). ¹³C-NMR (150 MHz, D₂O): 181.75 (C=O); 105.55 (*J*(C,H) = 170.8, C(1'')); 103.94 (*J*(C,H) = 158.4, C(1'')); 80.86 (C(2'')); 77.68 (C(3'')); 76.66 (C(5'')); 76.42, 76.38 (C(4'')); 74.49 (C(2'')); 74.28 (C(3'')); 73.97 (C(1'')); 72.80 (C(5'')); 56.39 (MeOCO); 37.92 (C(5'')); 32.99 (C(2'')); 29.73 (C(3'')); 28.42 (C(4'')); 21.03 (C(6'')); 21.01 (C(6'')). HR-ESI-MS: 461.1979 ([M+Na]⁺; calc. 461.1999).

6-Methoxy-6-oxohexyl O-6-Deoxy- α -L-mannopyranosyl-(1 $\rightarrow$ 3)-O-6-deoxy- α -L-mannopyranosyl-(1 $\rightarrow$ 2)-6-deoxy- β -L-mannopyranoside (**23**). As described for **22**, from **10** (110 mg, 0.1 mmol). Elution of the crude product from a small silica gel column (CH₂Cl₂/MeOH 4:1) and freeze-drying yielded **23** (55 mg, 96%). White amorphous solid. [α]_D = +3.3 (*c* = 0.4, H₂O). ¹H-NMR (600 MHz, D₂O): 5.02 (*d*, *J*(1,2) = 1.7, H–C(1'')); 5.00 (*d*, *J*(1,2) = 1.8, H–C(1'')); 4.63 (*d*, *J*(1,2) = 0.9, H–C(1'')); 4.23–4.19 (*m*, H–C(5'')); 4.18 (*dd*, *J*(2,3) = 3.3, H–C(2'')); 4.07 (*dd*, *J*(2,3) = 3.4, H–C(2'')); 4.00 (*dd*, *J*(2,3) = 2.9, H–C(2'')); 3.90 (*dd*, *J*(3,4) = 9.8, H–C(3'')); 3.91–3.82 (*m*, H–C(3''), H–C(5''), H_a–C(1'')); 3.70–3.67 (*m*, 4 H, H–C(3'')); incl. *s* at 3.68, MeOCO); 3.58, 3.56 (*2t*, *J* = 6.5, 1 H, H_b–C(1'')); 3.50 (*t*, *J* = 9.8, H–C(4'')); 3.46 (*t*, *J* = 9.8, H–C(4'')); 3.42–3.39 (*m*, 2 H, H–C(5'')); incl. *t* at 3.40, *J* = 9.4, H–C(4'')); 2.40 (*t*, *J* = 7.5, CH₂(5'')); 1.65–1.58 (*m*, CH₂(4''), CH₂(2'')); 1.40–1.35 (*m*, CH₂(3'')); 1.31 (*d*, *J*(5,6) = 6.3, Me(6'')); 1.30 (*d*, *J*(5,6) = 5.8, Me(6'')); 1.24 (*d*, *J*(5,6) = 6.2, Me(6'')). ¹³C-NMR (150 MHz, D₂O): 180.33 (C=O); 105.19 (*J*(C,H) = 172.7, C(1'')); 103.94 (*J*(C,H) = 173.8, C(1'')); 102.34 (*J*(C,H) = 155.0, C(1'')); 80.93 (C(3'')); 79.52 (C(2'')); 76.07 (C(3'')); 75.14 (C(5'')); 74.97 (C(4'')); 74.73 (C(4'')); 74.08 (C(4'')); 72.91 (C(2'')); 72.87 (C(2'')); 72.79 (C(3'')); 72.46 (C(1'')); 71.84 (C(5'')); 71.51 (C(5'')); 54.85 (MeOCO); 36.38 (C(5'')); 31.44 (C(2'')); 27.72 (C(3'')); 26.85 (C(4'')); 19.49 (C(6'')); 19.46 (C(6'')); 19.43 (C(6'')). HR-ESI-MS: 607.2599 ([M+Na]⁺; calc. 607.2578).

REFERENCES

- [1] P. Sylvestre, E. Couture-Tosi, M. Mock, *Mol. Microbiol.* **2002**, *45*, 169.
- [2] J. M. Daubenspeck, H. Zeng, P. Chen, S. Dong, C. T. Steichen, N. R. Krishna, D. G. Pritchard, C. L. Turnbough, *J. Biol. Chem.* **2004**, *279*, 30945.
- [3] R. Saksena, R. Adamo, P. Kováč, *Carbohydr. Res.* **2005**, *340*, 1591.
- [4] R. Adamo, R. Saksena, P. Kováč, *Carbohydr. Res.* **2005**, *340*, 2579.
- [5] D. B. Werz, P. H. Seeberger, *Angew. Chem., Int. Ed.* **2005**, *44*, 6315.

- [6] F. Niderberg, T. Bowden, J. Hilborn, *Macromolecules* **2004**, *37*, 954.
- [7] V. K. Srivastava, C. Schuerch, *J. Org. Chem.* **1981**, *46*, 1121.
- [8] G. Hodosi, P. Kováč, *J. Am. Chem. Soc.* **1997**, *119*, 2335.
- [9] G. Hodosi, P. Kováč, *Carbohydr. Res.* **1998**, *308*, 63.
- [10] K. Bock, C. Pedersen, *J. Chem. Soc., Perkin Trans. 2* **1974**, 293.
- [11] G. Solladie, C. Ziani-Cherif, *J. Org. Chem.* **1993**, *58*, 2181.
- [12] N. Ranganathan, B. T. Storey, *J. Heterocycl. Chem.* **1980**, *17*, 1069.
- [13] P. Konradson, U. E. Udodong, B. Fraser-Reid, *Tetrahedron Lett.* **1990**, *31*, 4313.
- [14] R. Saksena, R. Adamo, P. Kováč, *Bioorg. Med. Chem. Lett.* **2006**, *16*, 615.
- [15] P. Kováč, in 'Handbook of Derivatives for Chromatography', Eds. K. Blau, J. M. Halket, John Wiley & Sons, Ltd., Chichester, 1993, p. 109.
- [16] T. Adachi, Y. Yamada, I. Inoue, M. Saneyoshi, *Synthesis* **1977**, 45.
- [17] V. Pozsgay, H. J. Jennings, *J. Org. Chem.* **1988**, *53*, 4042.
- [18] A. Liptak, L. Szabo, J. Harangi, *J. Carbohydr. Chem.* **1988**, *7*, 687.

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