Synthesis, structure and epitope mapping of well-defined Staphylococcus aureus capsular polysaccharides

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Chapter 4

Investigation of trisaccharide repeating unit frameshifts of *Staphylococcus aureus* capsular polysaccharide type 5 and 8 to define the minimal binding epitope for antibody recognition

Introduction

The bacterial cell-wall of both Gram-positive and Gram-negative bacteria can consist of capsular polysaccharides (CPs), which generally are long, complex molecules built up of different monosaccharides, interconnected through various types of stereoisomeric and regioisomeric linkages, carrying different functional groups.[1] The repeating units (RUs), that make up the polymers, can contain several different monosaccharides. Because of the complexity of bacterial glycans that often contain rare monosaccharides, carrying multiple functional groups (uronic acids, free amines, acetamides), their synthesis presents a significant challenge. In the search for active epitopes for vaccine development, pressed by the synthetic hurdles, often the synthesis of a single RU is undertaken. However, if a CP is built up from RUs that contain multiple different monosaccharides, different frameshifted RUs may be defined. To ensure coverage of all possible RUs in a single molecule, larger molecules spanning at least 2 RU would have to be synthesized. Alternatively, different frameshifts of the minimal RU can be synthesized. For example, for a trisaccharide repeating unit, three different repeats "ABC", "BCA" and "CAB" can be defined.

Staphylococcus aureus (*S. aureus*) is a Gram-positive bacterium, which can be encapsulated by 13 different types of CPs, thereby defining their serotype. Of these, serotypes 5 and 8 are amongst the most isolated strains from clinical sources.[2–5] The *S. aureus* CP type 5 (CP5) and type 8 (CP8) are both built up from a D-ManNAcA – L-FucNAc – D-FucNAc (DM-LF-DF) trisaccharide repeating unit, as depicted in Figure 1A. Several syntheses have been reported of these trisaccharide repeats,[6–14] of which all have the same monosaccharide order (DM-LF-DF), which perhaps can be explained to originate from the biosynthesis route.[15] As described in Chapter 1, the polysaccharide is built up from DM-LF-DF trisaccharides, that are first generated on a phospholipid anchor in the cytoplasm lipid membrane, and then polymerized on the outer cell membrane in a [3+3n] matter. So far, the DM-LF-DF trisaccharides of CP5 and CP8 have shown limited antibody recognition, as demonstrated by the work of the groups of Adamo[6] and Hu,[13] which could indicate that these are too short to present the minimal binding epitope. However, it could also be that the wrong frameshifts have been investigated.

To date, only limited work has been published regarding the synthesis of frameshifted CP5/8 trisaccharides, other than the DM-LF-DF frameshift. In 2022 Demchenko and co-workers reported the assembly of the protected CP8 DF-DM-LF frameshift (Figure 1B).[16] By installing a picoloyl ester (Pico) group on the C-

3-OH of a $ManN_3A$ donor building block they took advantage of the H-bond-mediated aglycone delivery methodology they previously introduced,[17–19] to form the desired β-product in good yield and selectivity. The generated trisaccharide has not been deprotected and with the chosen protecting group pattern, conjugation to a carrier protein would be impossible.

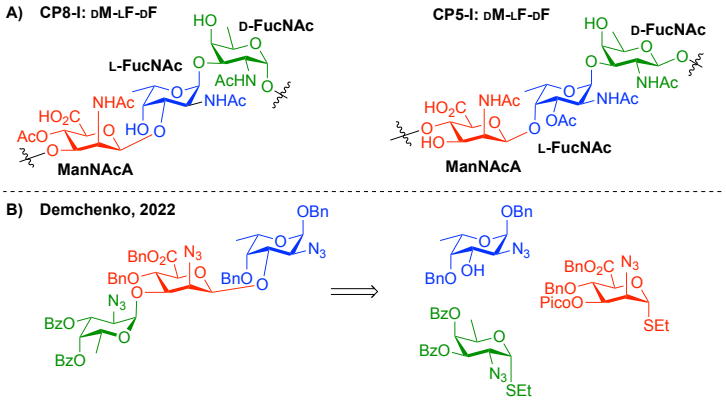
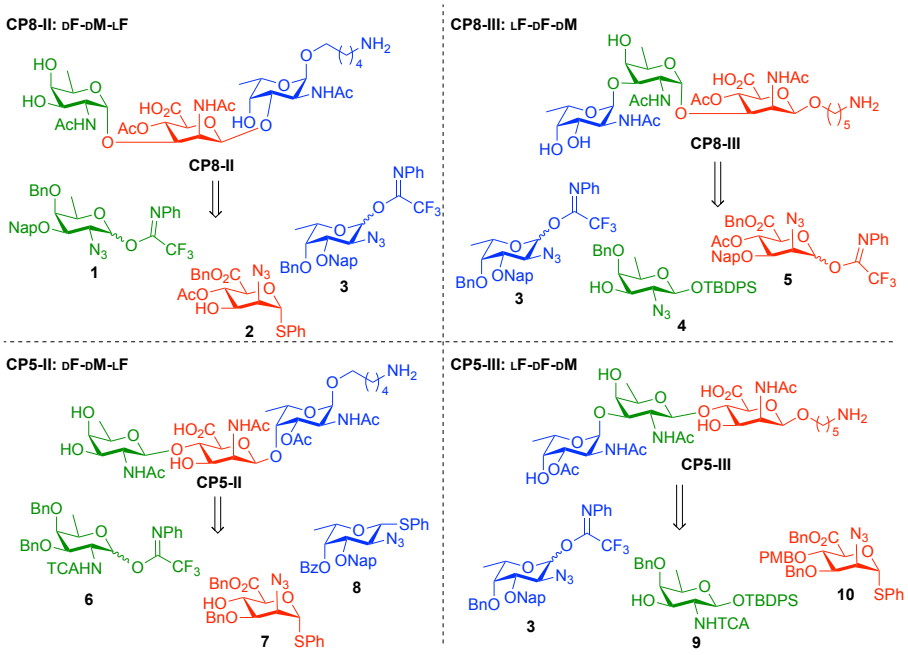


Figure 1: A) A schematic representation of the repeating unit of CP8 (left) and CP5 (right), B) Previously synthesis of a frameshifted CP8 trisaccharide.

In Chapter 2 and 3 the DM-LF-DF trisaccharides of CP5 and CP8 have been synthesized together with longer saccharides, and these were evaluated for antibody recognition. For both CP types, the DM-LF-DF trisaccharide did not show any or limited antibody recognition, where the longer oligosaccharides showed adequate binding, pointing to the oligosaccharide length as a crucial factor for binding. However, different trisaccharide frameshifts have not been investigated. Therefore, this Chapter presents the synthesis of all possible CP5 and CP8 RUs and their binding to anti-CP5 and anti-CP8 monoclonal antibodies. Besides the previously generated trisaccharides, two other CP5 and CP8 frameshifts are possible, one with the LF-DF-DM sequence and one having the DF-DM-LF structure. The same synthetic principles as implemented for the CP5 and CP8 oligosaccharides, *i.e.*, the use of a pre-glycosylation oxidation strategy and *O*-acetylation of the ManNAcA residue, the use of azides for the construction of the 1,2-*cis* linkages and trichloroacetamide protection for the β-D-FucNAc residue and the use of permanent benzyl-type protecting groups.

Results and discussion

The retrosynthesis of the two CP5/CP8-trisaccharides frameshifts is shown in Scheme 1. The same building blocks synthesized for the assembly of CP5 and CP8 oligosaccharides in Chapter 2 and 3 were applied, sometimes with minor modifications. As mentioned above, the same protection strategies were implemented for the acetamides and the installation of the *O*-acetyl esters. Different from the previous syntheses was the timing of the installation of the linker, as it will be installed on the monosaccharide level, to save steps at the more precious trisaccharide stage. Global deprotection should be facilitated by hydrogenation of the permanent benzyl-type protecting groups.

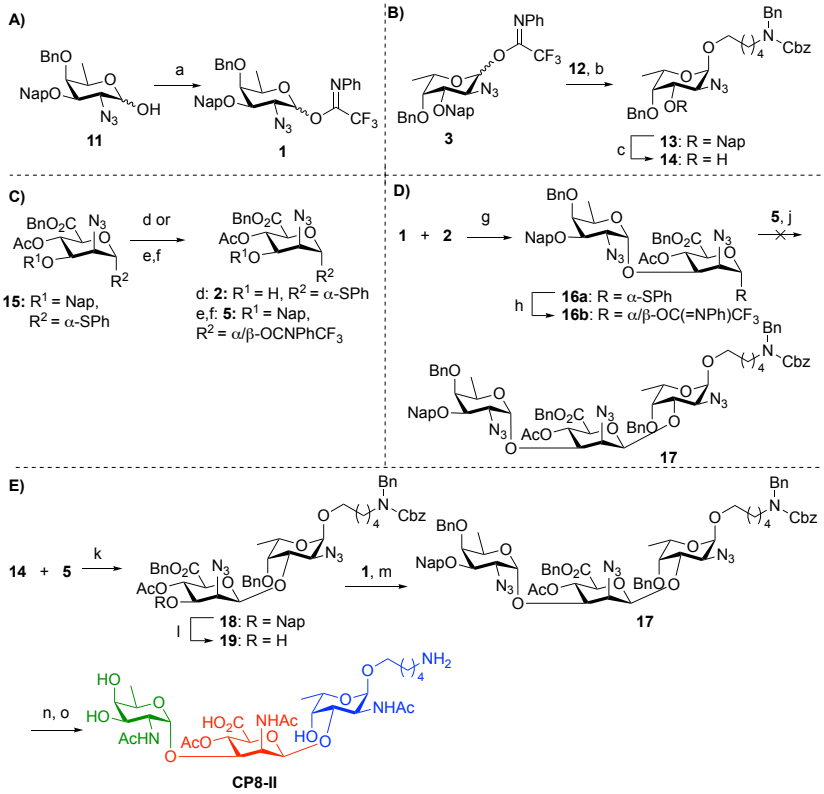


Scheme 1: Retrosynthetic analysis of the four different frameshifted trisaccharide of CP5/8

Synthesis of the frameshifted CP8 trisaccharides

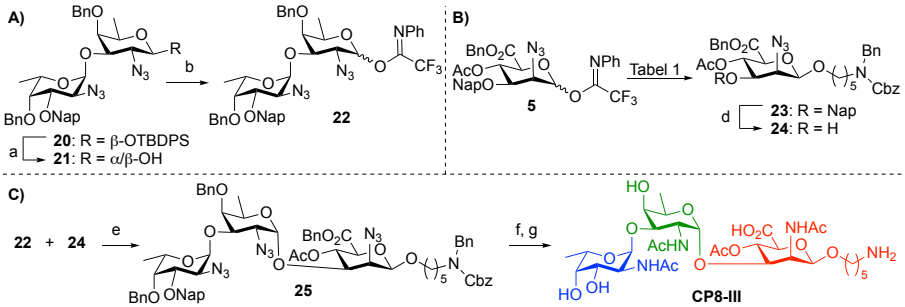
First, the CP8 DF-DM-LF trisaccharide (**CP8-II**) was constructed, and the synthesis of the required building blocks **1**, **2** and **3** (see retrosynthesis in Scheme 1) started from building blocks **11**, **3** and **15**, respectively, prepared in Chapter 2. The D-FucN_3 building block **11** was transformed into donor **1**, by installation of the *N*-phenyl trifluoroacetimidate leaving group (Scheme 2A). Next, the L-FucN_3

building block **3** was equipped with linker **12** to obtain **13** in 90% yield. Triphenylphosphine oxide (Ph_3PO) and trimethylsilyl iodide (TMSI)[20] were used to ensure the α-selectivity of the glycosylation delivering the product as a 70:30 α/β-mixture, from which the α-product could be purified by column chromatography (Scheme 2B). The newly formed α-bond was confirmed by ^{1}H-NMR with a doublet at 4.87 ppm with a coupling constant $J_{H1,H2}$ of 3.0 Hz. Acceptor **14** was then obtained by oxidative cleavage of the 2-methylnaphthyl (Nap) ether in good yields. For the mannosazide building block, **15** was converted into an acceptor by oxidative cleavage of the Nap ether giving **2** in 65% yield (Scheme 2C). With the three building blocks in hand, the construction of the trisaccharide started with investigation of a [2+1] strategy (Scheme 2D). Glycosylation between donor **1** and acceptor **2** in the presence of *tert*-butyldimethylsilyl trifluoromethanesulfonate (TBSOTf) afforded **16a** with excellent α-selectivity. Disaccharide **16a** was used directly as a donor in the glycosylation with acceptor **5**. Unfortunately, the [2+1] glycosylation did not yield the desired trisaccharide **17**. Neither using the thiophenyl donor **16a** or the imidate donor **16b** gave the desired glycosylation product, likely as the result of the difficult activation of the donor.[21] Therefore, a [1+2] glycosylation strategy was explored, where first glycosylation of acceptor **14** and imidate donor **5** in the presence of TBSOTf produced disaccharide **18**. Formation of the new β-linkage was confirmed by ^{1}H-NMR and ^{13}C-NMR, with the β-ManN_3A anomeric proton and carbon having a CH-coupling constant of $J_{C1,H1}$ = 160.5 Hz. Disaccharide **18** was converted into acceptor **19** by removal of the Nap ether and the ensuing glycosylation with imidate donor **1** with TBSOTf now afforded trisaccharide **17** in 57% yield with excellent α-selectivity. The stereochemistry of the newly formed glycosidic bond was again confirmed by the CH-coupling of $J_{C1,H1}$ = 170.1 Hz, as the new anomeric proton appeared in a multiplet. The same deprotection strategy, devised in Chapter 2 was implemented. Thus, reduction of the azides and one pot acetylation followed by hydrogenation gave trisaccharide **CP8-II** in 77% yield.



Scheme 2: Synthesis of the **CP8-II** trisaccharide. *Reaction conditions:* A) a) $ClC(=NPh)CF_3$, K_2CO_3, acetone, 81%, B) b) Ph_3PO, TMSI, DCM/Et_2O, 90%, α/β=70:30, c) DDQ, DCM/H_2O, 93%, C) d) DDQ, DCM/H_2O, 65%, e) NIS, TFA, DCM, 0 °C then Et_3N, 80%, f) $ClC(=NPh)CF_3$, K_2CO_3, acetone, 95%, D) g) TBSOTf, DCM, rt, 84%, h) i) NIS, TFA, DCM the Et_3N, 0 ºC, 44%, ii) $ClC(=NPh)CF_3$, K_2CO_3, acetone, 75%, j) For 1**6a**: TfOH, NIS, DCM, -78 to -10°C, 0%, for **16b**: TBSOTf, DCM, -78 to -40°C, 0%, E) k) TBSOTf, DCM, 0 °C, 53%, l) DDQ, DCM/H_2O, 53%, m) TBSOTf, DCM, rt, 57%, n) zinc, AcOH, Ac_2O, THF, 50 °C, quant., o) $Pd(OH)_2$/C, AcOH, H_2, *t*-BuOH/H_2O, 77%.

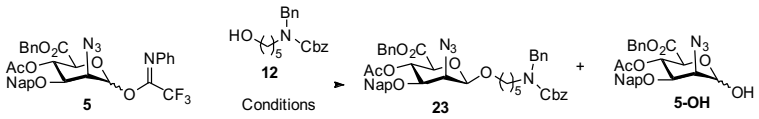
The construction of the CP8 LF-DF-DM (**CP8-III**) trisaccharide started with formation of disaccharide donor **22**, by removal of the *tert*-butyldiphenylsilyl (TBDPS) group in **20** followed by *N*-phenyl trifluoroacetimidate installation on the newly formed lactol giving donor **22**.



Scheme 3: Synthesis of the **CP8-III** trisaccharide. *Reaction conditions:* A) a) TBAF, AcOH, THF, 0 °C to rt, 65%, b) $ClC(=NPh)CF_3$, K_2CO_3, acetone, 70%, B) c) TBSOTf, DCM, -78 to -30 °C, 24%, d) DDQ, DCM/H_2O, 51%, C) e) TBSOTf, DCM, rt, 65%, f) zinc, AcOH, Ac_2O, THF, 50 °C, 78%, g) $Pd(OH)_2$/C, AcOH, H_2, *t*-BuOH/H_2O, 60%.

Next, the mannosaziduronic acid **5** was equipped with the linker by glycosylation between donor **5** and linker **12**. Unfortunately, the yield was quite low, and while solely the β-anomer was formed (as confirmed by the CH-coupling constant of $J_{C1,H1}$ = 161.6 Hz), product **23** was isolated in only 28% yield. Different attempts to improve the low yields were investigated (see Table 1). Switching the promoter from triflic acid (TfOH, Entry 1) to TBSOTf (Entry 2) only afforded **23** in 20% yield together with 6% of lactol **5-OH**, and neither changing the concentration to 0.3 M (Entry 3), prolonging the reaction time (Entry 4) or using stoichiometric amount of promoter were found to improve the yield of **23**, and only more lactol **5-OH** was formed. As enough material could be procured, the synthesis was continued without further optimization. Cleavage of the Nap ether afforded acceptor **24**, which was glycosylated with disaccharide donor **22** to give trisaccharide **25** in 65% yield and excellent α-selectivity (again confirmed by the CH-coupling constant of $J_{C1,H1}$ = 170.4 Hz, as the new anomeric proton appeared in a multiplet). Deprotection by reduction of the azides and concomitant acetylation followed by hydrogenation then gave trisaccharide **CP8-III** in 60% yield.

Table 1: Optimization of the linker installation using donor **5**.

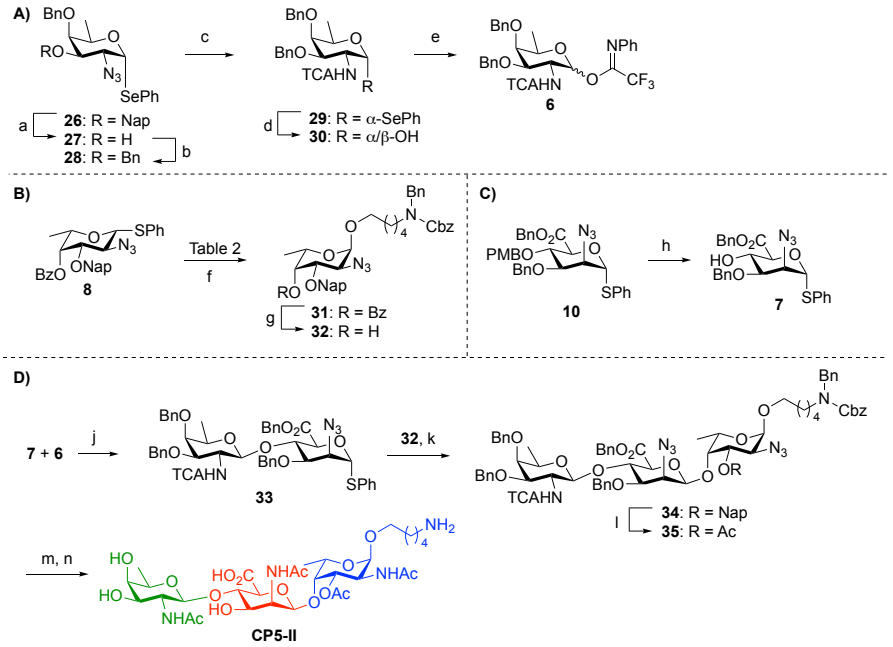


Entry	Promoter	Concentration (M)	Time (h)	Yield 23 (%)	Yield 5-OH (%)	α/β[a]
1	TfOH (0.2 equiv.)	0.1	1.5	27[b]	0	1:99[c]
2	TBSOTf (0.2 equiv.)	0.1	3	20[b]	6	1:99[c]
3	TBSOTf (0.2 equiv.)	0.3	1.5	28	33	1:99[c]
4	TBSOTf (0.2 equiv.)	0.1	19	28	45	1:99[c]
5	TBSOTf (1 equiv.)	0.1	2	25	43	1:99[c]

General conditions: 3 Å molecular sieves, -78 to -40 ºC, 1.3 equiv. acceptor for entry 1-3, 3 equiv. acceptor for entry 4-5, in DCM. [a] The α/β ratio was determined by NMR after purification. [b] Based on recovered donor. [c] No α-product was isolated.

Synthesis of the frameshifted CP5 trisaccharides

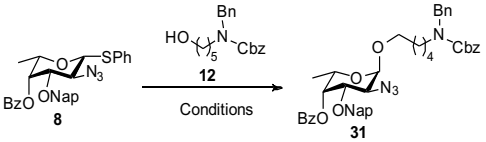
Turning to the CP5 trisaccharide frameshifts, the first trisaccharide to be investigated was the CP5 DF-DM-LF (**CP5-II**), which was synthesized from building blocks **26**, **8** and **10** (see Scheme 4) that were generated as described in Chapter 2 and 3. For the D-FucN_3 synthon, it was necessary to exchange the Nap ether with a permanent benzyl protection group to ensure orthogonality with respect to the Nap ether on the L-FucN_3 building block, that was used as precursor for the *O*-acetyl ester. Therefore, oxidative cleavage of the Nap ether yielded **26** and subsequent benzylation of the newly released alcohol gave **28** (Scheme 4A). To ensure the 1,2-*trans* linkages in the upcoming glycosylation reactions, the azide was changed to a trichloroacetamide by reduction and trichloroacetyl (TCA) acetylation to give **29** in excellent yield. Hydrolysis of the phenylselenyl group and installation of an *N*-phenyl trifluoroacetimidate on the newly formed lactol gave donor **6**.



Scheme 4: Synthesis of the **CP5-II** trisaccharide. *Reaction conditions:* A) a) DDQ, DCM/H_2O, 91%, b) BnBr, NaH, DMF, 0 °C to rt, 99%, c) i) zinc, AcOH, THF, ii) TCA-Cl, THF, 0 °C, 93% over two steps, d) NIS, acetone/H_2O, 0 °C, 75%, e) ClC(=NPh)CF_3, K_2CO_3, acetone, 99%, B) f) NIS, TBSOTf, DCM, 99%, α/β=43:57, g) NaOMe, MeOH, 87%, C) h) DDQ, DCM/H_2O, 95%, D) j) TMSOTf, DCM/MeCN, -78 °C, 51%, α/β=29:71, k) NIS, TBSOTf, DCM, -30 to 10 °C, 52%, α/β=15:85, %, l) i) DDQ, DCM/H_2O, ii) Ac_2O, DMAP, pyridine, 81% over two steps, m) zinc, AcOH, Ac_2O, THF, 50 °C, 21%, n) $Pd(OH)_2$/C, AcOH, H_2, *t*-BuOH/H_2O, 64%.

The L-FucN_3 was equipped with a linker by glycosylation of β-thiophenyl-donor **8** and linker **12** to give **31** (Scheme 4B). However, it was found difficult to obtain good α-selectivity, due to the high reactivity of the acceptor alcohol. When using *N*-iodosuccinimide (NIS) and trimethylsilyl trifluoromethanesulfonate (TMSOTf) as promoter at low temperature (-60 to -30 °C), only β-product was obtained (Table 2, Entry 1). Increasing the temperature to 0 °C (Entry 2) or room temperature (Entry 3) influenced the stereoselectivity, but still favored the β-product (α/β=35:65 and α/β=34:66, respectively). By switching the promoter to NIS and TBSOTf the α/β-selectivity increased to 43:57. Also the use of NIS in combination with TMSI and Ph_3PO was investigated, as similar conditions have shown to provide α-selective glycosylation reactions with reactive primary alcohol acceptors previously,[22] but these conditions did not work with the thiophenyl donor **8**. As sufficient material was available, no further optimization has been undertaken. Acceptor **32** was obtained by saponification of the benzoyl ester under Zemplén condition.

Table 2: Optimization of the linker installation using donor **8**.



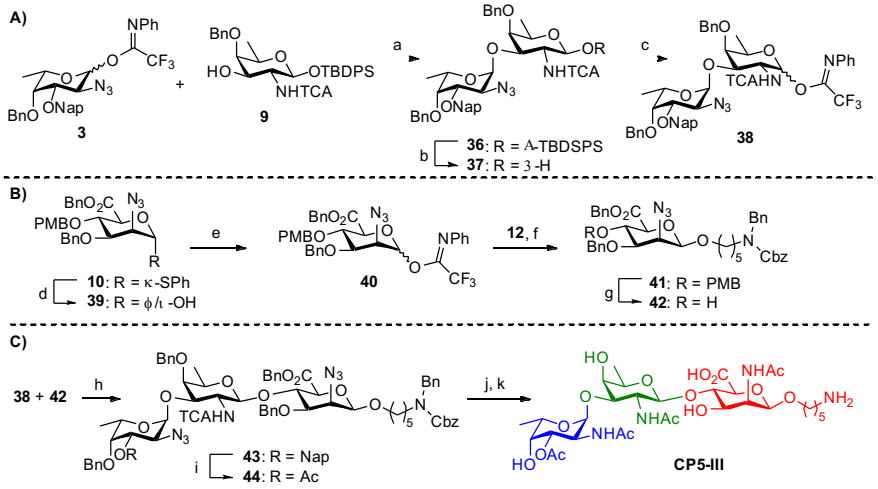
Entry	Conditions	Temp (°C)	Yield (%)	α/β [(a)]
1	NIS, TMSOTf	-60 to -30	77	0:100
2	NIS, TMSOTf	0	94	35:65
3	NIS, TMSOTf	rt	94	34:66
4	NIS, TBSOTf	rt	99	43:57
5	NIS, Ph_3PO, TMSI	rt	--	--

General conditions: 3 Å molecular sieves, 1.3 equiv. acceptor, 1.5 equiv. NIS and 0.2 equiv. TMSOTf /TBSOTf or 6 equiv. Ph_3PO and 1 equiv. TMSI, 0.1 M DCM. [(a)] The α/β ratio was determined by NMR after purification.

The D-ManAN_3 acceptor **7** was synthesized by cleavage of the *p*-methoxybenzyl (PMB) ether from **10**, to set the stage for the assembly trisaccharide (Scheme 4C). First, the glycosylation of donor **6** and acceptor **7** afforded disaccharide **33** in 51% yield with an α/β-ratio of 29:71 (Scheme 4D). The β-product could be readily purified by column chromatography and the stereochemistry was confirmed by ^{1}H-NMR with the D-FucN anomeric proton appeared as a doublet at 5.67 ppm with a coupling constant $J_{H1,H2}$ of 10.3 Hz. Both the use of low temperature and acetonitrile (MeCN) as a co-solvent were found to be necessary to obtain sufficient β-selectivity. Disaccharide **33** was used directly as donor in a glycosylation with acceptor **32** to give trisaccharide **34** in 52% yield (Scheme 4D). The trisaccharide was obtained as an inseparable α/β-mixture (α/β = 15:85), and the desired β-product was separated after the introduction of the *O*-acetyl. The structure of the main β-product was confirmed by the C1'-H1'-coupling constant of 159.6 Hz. The deprotection strategy for trisaccharide **34** followed the same strategy as described in Chapter 3. First the Nap ether was exchanged for an *O*-acetyl ester. Next, the azides and the TCA group were reduced and acetylated, which was followed by a purification method employing silica gel column chromatography and subsequent HPLC purification to obtain the pure product in 21% yield. Lastly, hydrogenation afforded the wanted trisaccharide **CP5-II** in 64% yield.

Finally, the last CP5 LF-DF-DM (**CP5-III**) trisaccharide was created by a glycosylation of acceptor **9** and donor **3** to provide disaccharide **36** in 87% yield and excellent α-selectivity, as confirmed by ^{1}H-NMR with the L-FucN H1 doublet

resonating at 4.98 ppm with a coupling constant $J_{H1,H2}$ of 3.7 Hz (Scheme 5A). Disaccharide **36** was converted into a donor by removal of the anomeric TBDPS group followed by installation of a *N*-phenyl trifluoroacetimidate on the newly formed lactol giving disaccharide donor **38**. The linker was installed on the D-ManAN$_3$ building block **10**, by first hydrolysis of the thiophenyl group providing lactol **36** followed by installation of a *N*-phenyl trifluoroacetimidate to give donor **40**. Glycosylation with linker acceptor **12** gave **41** in 65% yield and an α/β ratio of 24:76. The β-product was separable by column chromatography and the newly formed β-linkage was confirmed by the CH-coupling constant $J_{C1,H1}$ of 161.3 Hz. The use of low temperature (-78 °C) was found to increase the β-selectivity of the reaction, and it was noted that the glycosylation of thio-donor **10** did not lead to any glycosylation product. Continuing the assembly, the PMB ether in **41** was cleaved yielding acceptor **42**. Trisaccharide **43** was then obtained in 81% yield by glycosylation of donor **38** and acceptor **42** at -78 °C (Scheme 3) as an inseparable 10:90 α/β mixture. The newly generated β-linkage was again confirmed by the CH-coupling constant $J_{C1,H1}$ of 159.2 Hz. The desired stereoisomer was separated on a later stage, namely after silica gel column and the reduction from azides/TCA to acetamides, however before the HPLC purification.



Scheme 5: Synthesis of the **CP5-III** trisaccharide. *Reaction conditions*: A) a) TBSOTf, DCM, 87%, α/β=99:1, b) TBAF, AcOH, THF, 0 °C to rt, 90%, c) ClC(=NPh)CF$_3$, K$_2$CO$_3$, acetone, 99%, B) d) NIS, TFA, DCM, 0 °C, 93% yield, e) ClC(=NPh)CF$_3$, K$_2$CO$_3$, acetone, 87%, f) TBSOTf, DCM, -78 °C, 65%, α/β=24:76, g) DDQ, DCM/H$_2$O, 92%, C) h) TBSOTf, DCM, -78 °C, 81%, α/β=10:90, i) i) DDQ, DCM/H$_2$O, ii) Ac$_2$O, DMAP, pyridine, 81% over two steps, j) zinc, AcOH, Ac$_2$O, THF, 50 °C, 35%, k) Pd(OH)$_2$/C, AcOH, H$_2$, *t*-BuOH/H$_2$O, 57%.

The final steps involved exchanging the Nap ether to an *O*-acetyl and reduction of the azides and TCA group and concomitant acetylation, which was followed by silica gel column chromatography and subsequent HPLC purification. Hydrogenation afforded trisaccharide **CP5-III**. It was observed however that migration of the C''-3-*O*-acetyl to the neighboring C''-4-OH took place after purification, leading to a mixture of the two regioisomeric acetylated trisaccharides.

Antibody binding

Having the CP5/8 trisaccharide frameshifts in hand, the binding properties to monoclonal antibodies (mAb) and polyclonal antibodies (pAb) was investigated. First the binding properties of the CP8-fragments was explored. **CP8-II** and **CP8-III** were included in a competitive ELISA experiment using both anti-CP8-mAb and anti-CP8-pAb sera, however no inhibition was observed for either of the trisaccharides even at high concentrations. These findings were also confirmed by Saturation-Transfer Difference (STD) NMR, were low to no STD was found for either of the trisaccharides. These findings are in line with the results from Chapter 2, where the length of the CP8 oligosaccharides was found to be important for antibody recognition, and the binding epitope was found to consist of at least 3 RUs.

For the CP5 trisaccharide frameshifts a different picture emerged. Only **CP5-II** was assessed together with **CP5-I**, as **CP5-III** showed *O*-acetyl migration, leading to a non-natural trisaccharide. The two trisaccharides together with the hexa- and nonasaccharide from Chapter 3 were evaluated for binding to a rat anti-CP5 mAb using competitive surface plasmon resonance (SPR). To this end, the natural CP5 polysaccharide (CP5 PS) was immobilized on the SA-chip. A large difference in binding was discovered between the **CP5-II** trisaccharide and the **CP5-I** fragment as shown in Figure 2. No significant inhibition for **CP5-I** was observed, but **CP5-II** was found to inhibit binding of the rat anti-CP5 mAb at low concentration with an IC50 value close to the IC50 value obtained for the hexasaccharide, although the latter bound slightly better. In 2012 Adamo and co-workers tested **CP5-I** for its ability to induce an antibody response, but no to minimal binding levels were found. They concluded that longer fragments were needed for a model vaccine candidate,[6] however the findings in this Chapter indicate that the LF-DM-DF trisaccharide **CP5-II** could be used as a minimal epitope in future conjugate vaccine design, as it seems to hold the minimal binding epitope for antibody binding.

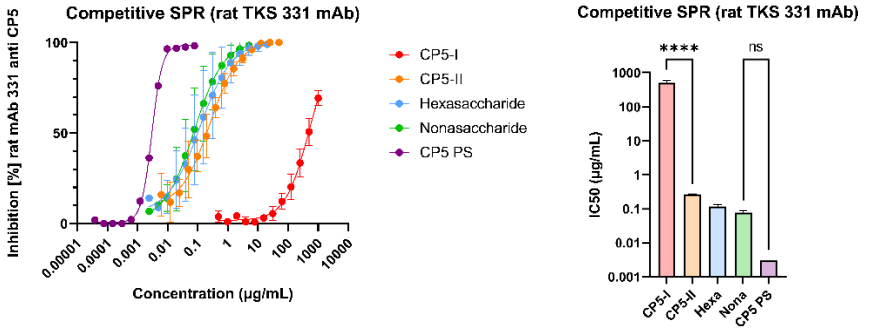


Figure 2: Competitive SPR results using the synthetic oligosaccharides and rat TKS 331 mAb showed that **CP5-I** is barely recognized while the recognition of **CP5-II** is similar to the hexasaccharide and nonasaccharide. **** Identifies a significant difference; ns identifies no significant difference.

Conclusion

In this Chapter, the frameshifted trisaccharides of CP5 and CP8 have been synthesized. The syntheses were found to be more complicated than the syntheses used in the previous chapters to access the other repeating units, as lower yields and poorer anomeric selectivity were obtained. For CP8, two frameshifted trisaccharides were synthesized and tested for their ability to recognize mAb and pAb. Both trisaccharides failed to show binding indicating that longer fragments of CP8 are necessary for good interaction. Also, two CP5 frameshift trisaccharides were generated, but only one was useful for biological evaluation as *O*-acetyl migration from the C''-3-*O*-acetyl to the C''-4-OH occurred in the **CP5-III** trisaccharide. Interestingly, the generated **CP5-II** frameshift showed good binding of the rat-anti-CP5 mAb, with binding levels being comparable to the hexasaccharide.

Overall, this Chapter has shown that in the search for an optimal minimal epitope, it may be required to generate longer oligosaccharides encompassing multiple RUs. At the same time, scanning different frameshift RUs may reveal shorter oligosaccharides to be adequate antigens. It is difficult to predict upfront what the optimal synthetic antigen will be for any given bacterial polysaccharide. Binding studies using systematic sets of oligosaccharides, differing in length and nature of the RU, in combination with structural studies such as reported in the preceding chapters are imperative to deepen the insights into which structural features are most important in shaping oligosaccharide-antibody interactions.

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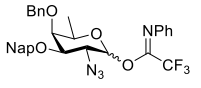
Experimental

General experimental procedures

All reagents were of commercial grade and used as received unless otherwise noted. All moisture sensitive reactions were performed under an argon or nitrogen (N_2) atmosphere. Dried solvents (DCM, DMF, THF, toluene, Et_2O) were stored over flame-dried 3 or 4Å molecular sieves. Reactions were monitored by thin layer chromatography (TLC) analysis conducted with Merck aluminum sheets with 0.20 mm of silica gel 60. The plates were detected by UV (254 nm) and were applicable by spraying with 20% sulfuric acid in EtOH or with a solution of $(NH_4)_6Mo_7O_{24}{\cdot}4H_2O$ (25 g/L) and $(NH_4)_4Ce(SO_4)_4{\cdot}2H_2O$ (10 g/L) in 10% sulfuric acid (aq.) followed by charring at ~150 °C. Flash column chromatography was performed with silica gel (40-63µm). Size-exclusion chromatography was carried out using SephadexTM (LH-20, GE Healthcare Life Sciences) by isocratic elution with DCM/MeOH (1:1, v:v). High-resolution mass spectra were recorded on a Thermo Finigan LTQ Orbitrap mass spectrometer equipped with an electrospray ion source in positive mode (source voltage 3.5 kV, sheath gas flow 10, capillary temperature 275 ºC) with resolution R=60.000 at m/z=400 (mass range 150-4000). ^{1}H and ^{13}C spectra were recorded on a Bruker AV-400 (400 and 101 MHz respectively), Bruker AV-500 (500 and 126 MHz respectively), Bruker AV-600 (600 and 151 MHz respectively), Bruker AV-850 (800 and 214 MHz respectively) or a Bruker AV-1200 (1200 and 302 MHz respectively). Chemical shifts (δ) are given in ppm relative to the residual signal of the deuterated solvent (^{1}H-NMR: 7.26 ppm for $CDCl_3$, 3.31 ppm for MeOD, 1.94 for $CNCD_3$ or 4.79 for D_2O. ^{13}C-NMR: 77.16 ppm for $CDCl_3$, 49.00 ppm for MeOD, 1.32 for $CNCD_3$). Coupling constants (*J*) are given in Hz. All ^{13}C spectra are proton decoupled. NMR peak assignments were made using COSY and HSQC experiments, where applicable, HMBC and GATED experiments were used to further elucidate the structure. The anomeric product ratios were analyzed through integration of proton NMR signals.

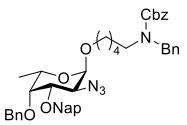
CP8-II: DF–DM–LF

2-azido-4-*O*-benzyl-2-deoxy-3-*O*-(2-naphtylmethyl)-1-*O*-(*N*-phenyl-2,2,2-trifluoroacetimidoyl)-α/β-D-fucopyranoside (1)



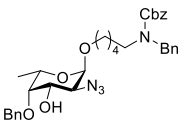
11 (300 mg, 0.72 mmol) was co-evaporated with toluene (x3) and dissolved in dry acetone (4 mL, 0.2 M). K_2CO_3 (200 mg, 1.44 mmol, 2 equiv.) and $ClC(=NPh)CF_3$ (0.24 mL, 1.44 mmol, 2 equiv.) were added and the mixture was stirred under N_2 for 18 h until TLC analysis (pentane/EtOAc, 7:3) showed full conversion of starting material. The mixture was filtered and concentrated *in vacuo*. The crude product was purified by column chromatography (pentane/EtOAc 99:1 → 85:15) to give **1** in 98% yield (416.7 mg, 0.70 mmol) **^{1}H NMR (400 MHz; CD_3CN)** δ : 7.96 – 7.78 (4H, m), 7.69 – 7.05 (11H, m), 6.91 – 6.81 (2H, m), 6.30 (1H, s), 5.07 – 4.78 (3H, m), 4.65 (1H, dd, J = 11.1, 7.2 Hz), 4.16 – 3.70 (4H, m), 1.98 (3H, s). **^{13}C NMR (100.65 MHz; CD_3CN)** δ : 139.69, 139.66, 136.56, 134.25, 134.21, 134.00, 130.09, 129.88, 129.85, 129.26, 129.08, 128.81, 128.68, 128.62, 127.74, 127.64, 127.28, 127.13, 127.08, 125.46, 122.06, 120.12, 118.30, 81.28, 78.13, 76.71, 75.49, 72.81, 72.64, 72.40, 70.21, 63.44, 59.82, 16.98, 16.89.

5-(Benzyl(benzyloxycarbonyl)amino)pentyl 2-azido-2-deoxy-4-*O*-benzyl-3-*O*-(2-naphtylmethyl)-α-L-fucopyranoside (13)



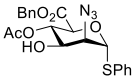
Donor **3** (59 mg, 0.10 mmol, 1 equiv.) and acceptor **12** (42 mg, 0.13 mmol, 1.3 equiv.) were co-evaporated with toluene (x3) and dissolved in dry DCM/Et_2O (1 mL, 1:1, 0.1 M). Triphenylphosphine oxide (169 mg, 0.6 mmol, 6 equiv.) was added and the mixture was stirred with 3 Å molecular sieve under argon for 1 h before TMSI (14 µL, 0.10 mmol, 1 equiv.) was added. The mixture was stirred for 22 h at rt until TLC analysis (pentane/EtOAc, 8:2) showed full conversion of the donor. The reaction was quenched with Et_3N, dissolved in EtOAc, washed with $Na_2S_2O_3$ (sat. aq.; 1x), $NaHCO_3$ (sat. aq.; 1x) and brine (1x), dried over Na_2SO_4, filtered and concentrated *in vacuo*. The crude product was purified by column chromatography (pentane/EtOAc 9:1 → 8:2) to give **13** in 90% yield and with α/β = 70:30 (65.3 mg, 0.09 mmol). NMR data are reported for the pure α-isomer. **^{1}H NMR (400 MHz; $CDCl_3$)** δ : 7.88 – 7.78 (m, 4H), 7.56 – 7.45 (m, 3H), 7.39 – 7.24 (m, 15H), 5.18 (2H, d, J = 13.3 Hz, Ar-CH_2), 4.96 (1H, d, J = 11.5 Hz, Ar-CH_2), 4.88 (3H, m, H-1, Ar-CH_2), 4.66 (1H, d, J = 11.5 Hz, Ar-CH_2), 4.50 (2H, d, J = 8.8 Hz, Ar-CH_2), 3.99 (1H, q, J = 12.6, 11.6 Hz, H-3), 3.87 (2H, dd, J = 10.7, 3.6 Hz, H-2, H-5), 3.74 (1H, s, H-4), 3.68 – 3.14 (4H, m, CH_2-Linker), 1.66 – 1.47 (6H, m, CH_2-Linker), 1.17 (3H, d, J = 6.5 Hz, CH_3). **^{1}H NMR (400 MHz; $CDCl_3$)** δ : 138.39 (Ar-C_q), 137.83 (Ar-C_q), 135.09 (Ar-C_q), 133.42 (Ar-C_q), 133.18 (Ar-C_q), 128.67 (Ar-*C*), 128.58 (Ar-*C*), 128.46 (Ar-*C*), 128.42 (Ar-*C*), 128.09 (Ar-*C*), 127.94 (Ar-*C*), 127.88 (Ar-*C*), 127.85 (Ar-*C*), 127.41 (Ar-*C*), 126.65 (Ar-*C*), 126.31 (Ar-*C*), 126.14 (Ar-*C*), 125.83 (Ar-*C*), 97.37 (C-1), 77.89 (C-3), 76.34 (CH_2), 75.67 (CH_2), 72.52 (CH_2), 68.16, 67.29 (C-4), 66.69 (C-2, C-5), 59.82 (C-2, C-5), 29.84 (*C*H_2-Linker), 16.91(CH_3). **HRMS**: $[M+Na]^+$ calculated for $C_{44}H_{46}N_4O_6Na$: 751.34715; found 751.34661

5-(Benzyl(benzyloxycarbonyl)amino)pentyl 2-azido-2-deoxy-4-*O*-benzyl-α-L-fucopyranoside (14)



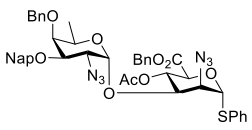
13 (91 mg, 0.14 mmol, 1 equiv.) was dissolved in DCM (1.3 mL , 0.1 M). DDQ (64 mg, 0.28 mmol, 2 equiv.) and H_2O (0.1 mL) were added, and the mixture was stirred for 3 h until TLC analysis (pentane/EtOAc, 8:2) showed full conversion of starting material. The reaction was quenched with $Na_2S_2O_3$ (sat. aq) and extracted with EtOAc (3x). The combined organic layers were washed with $NaHCO_3$ (sat. aq.; 4x) and brine (1x), dried over Na_2SO_4, filtered and concentrated *in vacuo*. The crude product was purified by column chromatography (pentane/EtOAc 9:1 → 7:3) to give **14** in 50% yield (39 mg, 0.07 mmol). **^{1}H NMR (400 MHz; $CDCl_3$)** δ : 7.39 – 7.24 (m, 15H), 4.96 (1H, d, J = 11.5 Hz, CH_2), 4.88 (3H, m, H-1, Ar-CH_2), 4.66 (1H, d, J = 11.5 Hz, Ar-CH_2), 4.51 (2H, d, J = 8.8 Hz, Ar-CH_2), 3.99 (1H, q, J = 12.6, 11.6 Hz, H-3), 3.87 (2H, dd, J = 10.7, 3.6 Hz, H-2, H-5), 3.74 (1H, s, H-4), 3.68 – 3.14 (4H, m, CH_2-Linker), 1.66 – 1.47 (6H, m, CH_2-Linker), 1.17 (3H, d, J = 6.5 Hz, H-6). **^{13}C NMR (101 MHz, $CDCl_3$)** δ 137.95 (Ar-C_q), 128.74 (Ar-*C*), 128.62 (Ar-*C*), 128.54 (Ar-*C*), 128.24 (Ar-*C*), 128.01 (Ar-*C*), 127.89 (Ar-*C*), 127.37 (Ar-*C*), 127.28 (Ar-*C*), 98.22 (C-1), 80.22 (C-4), 76.21 (Ar-*C*H_2), 68.78 (C-3), 68.15 (Ar-*C*H_2), 67.24 (Ar-*C*H_2), 66.52 (C-5), 60.98 (C-2), 50.57 (*C*H_2-Linker), 50.29 (*C*H_2-Linker), 47.16 (*C*H_2-Linker), 46.21 (*C*H_2-Linker), 29.18 (*C*H_2-Linker), 28.03 (*C*H_2-Linker), 27.94 (*C*H_2-Linker), 27.52 (*C*H_2-Linker), 23.46 (*C*H_2-Linker), 16.87 (C-6). **HRMS**: $[M+Na]^+$ calculated for $C_{33}H_{40}N_4O_6Na$: 611.28455; found 611.28401

Benzyl 4-*O*-acetyl-2-azido-2-deoxy-1-thio-α-D-mannopyranosiduronate (2)



15 (500 mg, 0.86 mmol, 1 equiv.) was dissolved in DCM (9.50 mL). DDQ (778 mg, 3.43 mmol, 4 equiv.) and H_2O (0.5 mL) were added, and the mixture was stirred for 6 h until TLC analysis (pentane/EtOAc, 8:2) showed full conversion of starting material. The reaction was quenched with $Na_2S_2O_3$ (sat. aq.) and extracted with EtOAc (x3). The combined organic layers were washed with $NaHCO_3$ (sat. aq.; x4) and brine (x1), dried over Na_2SO_4, filtered and concentrated *in vacuo*. The crude product was purified by column chromatography (pentane/EtOAc 95:5 → 80:20) to give **2** in 65% yield (248 mg, 0.56 mmol). **^{1}H NMR (400 MHz; $CDCl_3$)** δ : 7.84 – 7.79 (3H, m, Ar-*H*), 7.76 (1H, s, Ar-*H*), 7.62 – 7.58 (2H, m, Ar-*H*), 7.50 – 7.46 (2H, m, Ar-*H*), 7.43 (1H, dd, *J* = 8.5, 1.7 Hz, Ar-*H*), 7.27 – 7.22 (6H, m, Ar-*H*), 7.13 – 7.09 (2H, m, Ar-*H*), 5.78 (1H, d, *J* = 9.3 Hz, H-1), 5.62 (1H, dd, *J* = 4.8, 2.9 Hz, H-4), 5.02 (1H, d, *J* = 12.1 Hz, Ar-CH_2), 4.83 (1H, d, *J* = 12.2 Hz, Ar-CH_2), 4.68 (2H, s, Ar-CH_2), 4.63 (1H, d, *J* = 2.9 Hz, H-5), 3.98 (1H, dd, *J* = 4.7, 2.9 Hz, H-3), 3.45 (1H, dd, *J* = 9.4, 2.9 Hz, H-2), 2.03 (3H, s, COCH_3). **^{13}C NMR (100.65 MHz; $CDCl_3$)** δ : 169.75 (C=O), 167.88 (C=O), 134.86 (Ar-C_q), 133.98 (Ar-C_q), 133.28 (Ar-C_q), 133.23 (Ar-C_q), 132.47 (Ar-*C*), 131.93 (Ar-C_q), 129.01 (Ar-*C*), 128.67 (Ar-*C*), 128.60 (Ar-*C*), 128.58 (Ar-*C*), 128.49 (Ar-*C*), 128.13 (Ar-*C*), 128.03 (Ar-*C*), 127.84 (Ar-*C*), 127.38 (Ar-*C*), 126.40 (Ar-*C*), 126.36 (Ar-*C*), 125.95 (Ar-*C*), 81.06 (C-1), 74.64 (C-3), 73.69 (C-5), 73.04 (Ar-CH_2), 68.50 (C-4), 67.56 (Ar-CH_2), 57.93 (CH-2), 21.02 (CO*C*H_3). **HRMS**: $[M+Na]^+$ calculated for $C_{21}H_{21}N_3O_6Na$: 466.10488; found 466.10433

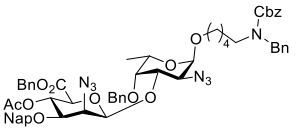
Phenyl 2-azido-4-*O*-benzyl-2-deoxy-3-*O*-(2-naphtylmethyl)-α-D-fucopyranosyl-(1→3)-benzyl(4-*O*-acetyl-2-azido-2-deoxy-1-thio)-α-D-mannopyranosiduronate (16)



Acceptor **2** (120 mg, 0.27 mmol, 1 equiv.) and donor **1** (245 mg, 0.41 mmol, 1.5 equiv.) were co-evaporated with toluene (3x), dissolved in dry DCM (3 mL), added 3 Å molecular sieve and stirred under argon for 30 min. TBSOTf (12 µL, 0.05 mmol, 0.2 equiv.) was added at rt and the mixture was stirred at rt under argon for 30 min until TLC analysis (pentane/EtOAc, 8:2) showed full conversion of the acceptor. The reaction was quenched with Et_3N, dissolved in EtOAc, washed with $NaHCO_3$ (sat. aq.; 1x) and brine (1x), dried over Na_2SO_4, filtered and concentrated *in vacuo*. The crude product was purified by column chromatography (pentane/EtOAc 9:1 → 8:2) to give **16** in 84% yield (191.1 mg, 0.23 mmol). **^{1}H NMR (400 MHz; $CDCl_3$)** δ : 7.88 – 7.81 (5H, m, Ar-*H*), 7.55 – 7.46 (6H, m, Ar-*H*), 7.40 – 7.24 (11H, m, Ar-*H*), 5.66 (1H, d, *J* = 4.1 Hz, H-1'), 5.53 (1H, t, *J* = 7.8 Hz, H-4'), 5.24 – 5.13 (2H, m, Ar-CH_2), 5.07 (1H, d, *J* = 3.6 Hz, H-1), 4.94 (1H, d, *J* = 11.4 Hz, Ar-CH_2), 4.90 – 4.85 (2H, m, Ar-CH_2), 4.72 (1H, d, *J* = 7.6 Hz, H-3), 4.62 (1H, d, *J* = 11.5 Hz, Ar-CH_2), 4.17 (1H, dd, *J* = 7.9, 3.5 Hz, H-5'), 4.08 – 3.99 (3H, m, H-2', H-3, H-5), 3.94 (1H, dd, *J* = 10.8, 3.6 Hz, H-2), 3.72 (1H, m, H-4), 1.93 (3H, s, COCH_3), 1.20 (3H, d, *J* = 6.5 Hz, CH_3). **^{13}C NMR (100.65 MHz; $CDCl_3$)** δ : 170.04 (C=O), 167.30 (C=O), 138.69 (Ar-C_q), 135.98 (Ar-C_q), 134.94 (Ar-C_q), 133.40 (Ar-C_q), 133.19 (Ar-C_q), 129.36 (Ar-C_q), 128.74 (Ar-*C*), 128.66 (Ar-*C*), 128.45 (Ar-*C*), 128.35 (Ar-*C*), 128.31 (Ar-*C*), 128.15 (Ar-*C*), 127.94 (Ar-*C*), 127.81 (Ar-*C*), 126.96 (Ar-*C*), 126.31 (Ar-*C*), 126.18 (Ar-*C*), 125.97 (Ar-*C*), 99.69 (C-1'), 84.68 (C-1) , 77.50 (C-2', C-3, C-5), 76.09 (C-4, C-5'), 75.06 (Ar-*C*H_2), 72.73 (Ar-*C*H_2), 71.55 (C-3'), 68.10 (C-2', C-3, C-5), 67.90 (C-4'), 67.87 (Ar-*C*H_2), 59.49 (C-2, C-2', C-3, C-5), 20.71

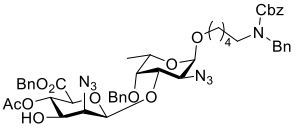
(COCH_3), 16.96 (CH_3). **HRMS**: $[M+Na]^+$ calculated for $C_{45}H_{44}N_6O_9Na$: 867.27036; found 867.27827

5-(Benzyl(benzyloxycarbonyl)amino)pentyl benzyl(4-*O*-acetyl-2-azido-2-deoxy-3-*O*-(2-naphthylmethyl))-β-D-mannopyranosiduronsyl-(1→3)-2-azido-4-*O*-benzyl-2-deoxy-α-L-fucopyranoside (18)



Donor **5** (106 mg, 0.16 mmol, 2 equiv.) and acceptor **14** (50 mg, 0.08 mmol, 1 equiv.) were co-evaporated with toluene (3x), dissolved in dry DCM (1 mL, 0.1 M), added 3 Å molecular sieve and stirred under argon for 30 min. The mixture was cooled to 0°C and added TBSOTf (3.7 µL, 0.016 mmol, 0.2 equiv.). The mixture was stirred for 4 h until TLC analysis (pentane/EtOAc, 8:2) showed full conversion of the acceptor. The reaction was quenched with Et_3N at 0°C, warmed to rt, dissolved in EtOAc, washed with $NaHCO_3$ (sat. aq.; 1x) and brine (1x), dried over Na_2SO_4, filtered and concentrated *in vacuo*. The crude product was purified by column chromatography (pentane/EtOAc 9:1 → 6:4) to give **18** in 53% yield (45.3 mg, 0.043 mmol). **^{1}H NMR (400 MHz; $CDCl_3$)** δ : 7.85 – 7.75 (4H, m, Ar-*H*), 7.75 – 7.71 (1H, m, Ar-*H*), 7.51 – 7.40 (4H, m, Ar-*H*), 7.37 – 7.11 (18H, m, Ar-*H*), 5.44 (1H, t, *J* = 9.4 Hz, H-4), 5.15 (3H, d, *J* = 11.6 Hz, Ar-CH_2), 5.08 (2H, d, *J* = 3.9 Hz, Ar-CH_2), 4.86 (1H, s, H-1'), 4.82 – 4.69 (3H, m, Ar-CH_2), 4.59 (2H, d, *J* = 3.6 Hz, H-5), 4.54 (1H, s, H-1'), 4.47 (2H, d, *J* = 7.6 Hz, Ar-CH_2), 4.26 (1H, d, *J* = 11.1 Hz, H-3'), 3.89 – 3.77 (2H, m, H-3', H-3), 3.70 – 3.66 (6H, m, H-2', H-5', H-2, CH_2-Linker), 3.48 (3H, t, *J* = 6.7 Hz, H-4', CH_2-Linker), 3.20 (2H, m, CH_2-Linker), 1.83 (3H, s, COCH_3), 1.62 – 1.47 (6H, m, CH_2-Linker), 1.29 – 1.21 (3H, m, CH_3). **^{13}C NMR (100.65 MHz; $CDCl_3$)** δ : 169.89 (C=O), 168.69 (C=O), 166.50 (C=O), 134.81 (Ar-C_q), 132.83 (Ar-C_q), 128.68 (Ar-*C*), 128.61 (Ar-*C*), 128.57 (Ar-*C*), 128.49 (Ar-*C*), 128.28 (Ar-*C*), 128.07 (Ar-*C*), 127.94 (Ar-*C*), 126.85 (Ar-*C*), 126.57 (Ar-*C*), 126.42 (Ar-*C*), 125.66 (Ar-*C*), 99.17 (C-1'), 96.97 (C-1), 75.75 (CH_2), 73.84 (CH-3), 72.63 (CH_2), 72.23 (CH_2), 71.43 (CH_2), 70.64 (CH_2), 70.32 (CH_2), 68.76 (CH_2), 68.14 (C-4), 67.70 (CH_2), 67.16 (C-4'), 66.43 (C-3'), 62.02 (CH_2), 61.47 (C-2, C-5'), 58.27 (C-2', C-5), 31.78 (CH_2-Linker), 29.84 (CH_2-Linker), 27.44 (CH_2-Linker), 20.43 (COCH_3), 18.96 (CH_2-Linker), 16.60 (C-6). **HRMS**: $[M+Na]^+$ calculated for $C_{59}H_{63}N_7O_{12}Na$: 775.27036; found 775.26981

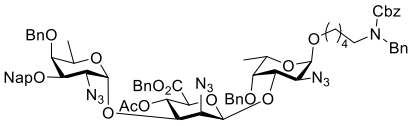
5-(Benzyl(benzyloxycarbonyl)amino)pentyl benzyl(4-*O*-acetyl-2-azido-2-deoxy)-β-D-mannopyranosiduronsyl-(1→3)-2-azido-4-*O*-benzyl-2-deoxy-α-L-fucopyranoside (19)



18 (58 mg, 0.0546 mmol) was dissolved in DCM/H_2O (20:1, 1 mL, 0.05 M). DDQ (25 mg, 0.109 mmol, 2 equiv.) was added and the mixture was stirred for 4 h at rt until TLC analysis (pentane/EtOAc, 6:4) showed full conversion of starting material. The reaction was neutralized with $Na_2S_2O_3$ (sat. aq.) and extracted with EtOAc (x3). The combined organic layers were washed with $NaHCO_3$ (sat. aq.; x4) and brine (x1), dried over Na_2SO_4, filtered and concentrated *in vacuo*. The crude product was purified by column chromatography (pentane/EtOAc 8:2 → 5:5) to give **19** in 53% yield (27 mg, 0.0289 mmol). **^{1}H NMR (400 MHz, $CDCl_3$)** δ 7.41 – 7.27 (m, 15H, Ar-*H*), 7.26 – 7.13 (m, 3H, Ar-*H*), 5.22 – 5.04 (m, 5H, Ar-CH_2, H-4'), 4.88 (d, *J* = 5.1 Hz, 1H, H-1), 4.71 (s, 2H, Ar-CH_2), 4.67 (s, 1H, H-1'), 4.53 – 4.44 (m, 2H, CH_2-Linker), 4.37 – 4.26 (m, 1H, H-

3), 3.92 (q, *J* = 9.1, 7.8 Hz, 1H, H-5), 3.85 (d, *J* = 9.7 Hz, 1H, H-5'), 3.71 – 3.68 (m, 1H, H-4), 3.62 – 3.55 (m, 4H, H-2, H-3', H-2', CH_2-Linker), 3.45 – 3.34 (m, 1H, CH_2-Linker), 3.34 – 3.11 (m, 2H, CH_2-Linker), 2.69 (s, 1H, OH), 1.86 (s, 3H, COCH_3), 1.65 – 1.46 (m, 6H, CH_2-Linker), 1.46 – 1.21 (m, 12H, H-6, CH_2-Linker). **^{13}C NMR (101 MHz, CDCl$_3$)** δ 170.62 (C=O), 166.40 (C=O), 138.00 (Ar-C_q), 135.16 (Ar-C_q), 128.67 (Ar-*C*), 128.57 (Ar-*C*), 128.33 (Ar-*C*), 128.26 (Ar-*C*), 128.18 (Ar-*C*), 128.06 (Ar-*C*), 127.92 (Ar-*C*), 127.31 (Ar-*C*), 98.69 (C-1), 97.76 (C-1'), 77.48 (C-4), 76.84 (C-3), 75.12 (Ar-*C*H$_2$), 73.71 (C-5'), 71.17 (C-3'), 69.79 (C-4'), 68.32 (*C*H$_2$-Linker), 67.65 (Ar-*C*H$_2$), 67.27 (Ar-*C*H$_2$), 66.43 (C-5), 64.42 (C-2/C-2'), 58.09 (C-2/C-2'), 50.36 (*C*H$_2$-Linker), 47.51 (*C*H$_2$-Linker), 46.27 (*C*H$_2$-Linker), 31.75 (*C*H$_2$-Linker), 29.82 (*C*H$_2$-Linker), 29.16 (*C*H$_2$-Linker), 20.66 (CO*C*H$_3$), 16.95 (C-6). **HRMS:** [M+Na]$^+$ calculated for $C_{48}H_{55}N_7O_{12}Na$: 944.38064; found 944.38009

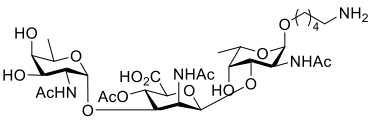
5-(Benzyl(benzyloxycarbonyl)amino)pentyl 2-azido -4-*O*-benzyl-2-deoxy -3-*O*-(2-naphtylmethyl)- α-D-fucopyranosyl-(1→3)-benzyl(4-*O*-acetyl-2-azido-2-deoxy)-β-D-mannopyranosiduronsyl-(1→3)-2-azido-4-*O*-benzyl-2-deoxy-α-L-fucopyranoside (17)



Donor **1** (26 mg, 0.0446 mmol, 1.5 equiv.) and Acceptor **19** (27 mg, 0.0297 mmol, 1 equiv.) were co-evaporated with toluene (3x), dissolved in dry DCM (1 mL, 0.03 M), added 3 Å molecular sieve and stirred under argon for 30 min. The mixture was added TBSOTf (1.3 µL, 0.0059 mmol, 0.2 equiv.) at rt and stirred for 25 min until TLC analysis (pentane/EtOAc, 7:3) showed full conversion of the acceptor. The reaction was quenched with Et_3N dissolved in EtOAc, washed with $NaHCO_3$ (sat. aq.; 1x) and brine (1x), dried over Na_2SO_4, filtered and concentrated *in vacuo*. The crude product was purified by column chromatography (pentane/EtOAc 8:2 → 6:4) to give **17** in 57% yield (22.5 mg, 0.017 mmol). **^{1}H NMR (400 MHz, CDCl$_3$)** δ 7.88 – 7.80 (m, 4H, Ar-*H*), 7.55 – 7.46 (m, 3H, Ar-*H*), 7.39 – 7.27 (m, 22H, Ar-*H*), 7.17 (d, *J* = 7.3 Hz, 1H, Ar-*H*), 5.38 (t, *J* = 9.8 Hz, 1H, H-4'), 5.17 (d, *J* = 12.8 Hz, 2H, Ar-CH_2), 5.11 (d, *J* = 4.2 Hz, 2H, Ar-CH_2), 4.96 – 4.87 (m, 4H, Ar-CH_2, H-1''), 4.92 – 4.82 (m, 4H, Ar-CH_2, H-1), 4.71 (s, 2H, Ar-CH_2), 4.60 (d, *J* = 11.4 Hz, 1H, Ar-CH_2), 4.57 (d, *J* = 1.5 Hz, 1H, H-1'), 4.49 (d, *J* = 7.9 Hz, 2H, CH_2-Linker), 4.34 (m, 1H, H-3), 4.10 (q, *J* = 6.3 Hz, 1H, H-5''), 4.03 (dd, *J* = 10.8, 2.6 Hz, 1H, H-3), 3.98 – 3.87 (m, 2H, H-2'', H-5), 3.87 – 3.78 (m, 2H, H-5', H-2'), 3.72 (dd, *J* = 2.7, 1.2 Hz, 1H, H-4''), 3.71 – 3.67 (m, 1H, H-4), 3.65 – 3.56 (m, 3H, H-3', H-2, CH_2-Linker), 3.49 – 3.32 (m, 1H, CH_2-Linker), 3.32 – 3.11 (m, 2H, CH_2-Linker), 1.88 (s, 3H, COCH_3), 1.60 – 1.45 (m, 4H, CH$_2$-Linker), 1.29 – 1.23 (m, 7H, CH$_2$-Linker, H-6), 1.12 (d, *J* = 6.5 Hz, 3H, H-6''). **^{13}C NMR (101 MHz, CDCl$_3$)** δ 169.36 (C=O), 166.38 (C=O), 138.12 (Ar-C_q), 137.96 (Ar-C_q), 135.06 (Ar-C_q), 133.42 (Ar-C_q), 133.19 (Ar-C_q), 128.73 (Ar-*C*), 128.67 (Ar-*C*), 128.65 (Ar-*C*), 128.63 (Ar-*C*), 128.58 (Ar-*C*), 128.52 (Ar-*C*), 128.46 (Ar-*C*), 128.26 (Ar-*C*), 128.18 (Ar-*C*), 128.06 (Ar-*C*), 127.99 (Ar-*C*), 127.93 (Ar-*C*), 127.81 (Ar-*C*), 127.33 (Ar-*C*), 126.90 (Ar-*C*), 126.31 (Ar-*C*), 126.17 (Ar-*C*), 125.95 (Ar-*C*), 101.12 (C-1''), 98.70 (C-1), 97.03 (C-1'), 79.32 (C-3'), 77.78 (C-3''), 77.16 (C-4), 76.14 (C-4''), 75.65 (Ar-*C*H$_2$), 75.17 (Ar-*C*H$_2$), 74.92 (C-3), 73.87 (C-5'), 72.79 (Ar-*C*H$_2$), 68.33 (Ar-*C*H$_2$), 68.18 (C-5''), 67.76 (Ar-*C*H$_2$), 67.34 (C-4'), 67.26 (Ar-*C*H$_2$), 66.48 (C-5), 63.80 (C-2'), 59.84 (C-2''), 58.09 (C-2), 50.62 (*C*H$_2$-Linker), 50.37 (*C*H$_2$-Linker), 47.27 (*C*H$_2$-Linker), 46.23 (*C*H$_2$-Linker), 29.83 (*C*H$_2$-Linker), 29.16 (*C*H$_2$-Linker), 20.60 (CO*C*H$_3$), 17.02

(C-6''/C-6), 16.98 (C-6''/C-6). **HRMS**: $[M+Na]^+$ calculated for $C_{72}H_{78}N_{10}O_{15}Na$: 1345.55458; found 1345.55403

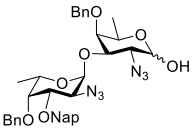
5-aminopentyl 2-*N*-acetamide-2-deoxy-α-D-fucopyranosyl-(1→3)-4-*O*-acetyl-2-*N*-acetamide-2-deoxy-β-D-mannopyranosiduronsyl-(1→3)-2-*N*-acetamide-2-deoxy-α-L-fucopyranoside (CP8-II)



17 (17 mg, 0.0128 mmol) was dissolved in THF (3 mL) and added zinc powder (252 mg, 3.85 mmol, 300 equiv.), AcOH (1 mL) and Ac_2O (0.5 mL). The mixture was heated to 50 °C and stirred for 18 h until TLC analysis (DCM/MeOH, 95:5) showed full conversion of the starting material. The solution was cooled to rt, filtered over a path of Celite and concentrated *in vacuo*. The crude product was purified by column chromatography (DCM/MeOH, 98:1 → 95:5). The product (18 mg, 0.0135 mmol) was dissolved in *t*-Bu-OH (2.5 mL). AcOH (0.1 mL in 100 mL MilliQ, 1 mL) was added and the mixture was birched under argon for 20 min. $Pd(OH)_2$ (catalytic amount) was added and the mixture was birched under argon for 5 min, then with H_2 for 2 min, before to be stirred for 3 days. The mixture was birched with argon for 20 min, filtered over a Whatman filter and lyophilized. Purification by a HW40 column with NH_4OAc followed by lyophilization gave **CP8-II** in 77% yield (7.6 mg, 0.0104 mmol). **^{1}H NMR (600 MHz, D_2O)** δ 5.18 (t, *J* = 9.9 Hz, 1H, H'-4), 5.00 (d, *J* = 3.9 Hz, 1H, H''-1), 4.98 (d, *J* = 1.5 Hz, 1H, H'-1), 4.89 (d, *J* = 1.8 Hz, 1H, H-1), 4.51 (dd, *J* = 4.5, 1.4 Hz, 1H, H'-2), 4.26 (q, *J* = 7.2, 6.7 Hz, 1H, H-5), 4.15 (d, *J* = 4.5 Hz, 1H, H-2), 4.15 – 4.11 (m, 2H, H-4, H'-3), 4.08 – 4.05 (m, 1H, H''5), 4.06 – 4.04 (m, 1H, H''-2), 4.03 – 4.00 (m, 1H,H-3), 3.81 (d, *J* = 10.1 Hz, 1H, H'-5), 3.77 (dd, *J* = 3.3, 1.1 Hz, 1H, H''-4), 3.75 – 3.71 (m, 1H, H''-3), 3.69 (dt, *J* = 10.2, 6.3 Hz, 1H, CH_2-Linker), 3.45 (dt, *J* = 10.1, 6.2 Hz, 1H, CH_2-Linker), 2.99 (dd, *J* = 8.6, 6.8 Hz, 2H, CH_2-Linker), 2.08 (s, 3H, COCH_3), 2.07 (s, 6H, COCH_3), 2.01 (s, 3H, COCH_3), 1.72 – 1.58 (m, 4H, CH_2-Linker), 1.48 – 1.36 (m, 2H, CH_2-Linker), 1.25 (dd, *J* = 6.7, 3.0 Hz, 6H, H-6, H''-6). **^{13}C NMR (151 MHz, D_2O)** δ 175.97 (C=O), 175.48 (C=O), 175.27 (C=O), 175.03 (C=O), 173.46 (C=O), 99.49 (C''-1), 97.78 (C'-1), 95.60 (C-1), 75.08 (C-4), 74.98 (C'-5), 73.88 (C'-3), 71.95 (C''-4), 71.17 (C'-4), 68.77 (C-3), 68.76 (CH_2-Linker), 68.40 (C''-3), 67.93 (C-5), 67.30 (C''-5), 53.44 (C'-2), 50.35 (C''-2), 48.91 (C-2), 40.28 (CH_2-Linker), 28.83 (CH_2-Linker), 27.38 (CH_2-Linker), 23.24 (CH_2-Linker), 22.96 (COCH_3), 22.92 (COCH_3), 22.67 (COCH_3), 21.20 (COCH_3), 16.42 (C''-6/ C-6), 16.30 (C''-6/ C-6). **HRMS**: $[M+H]^+$ calculated for $C_{31}H_{52}N_4O_{16}H$: 737.34566; found 737.34510

CP8-III: LF–DF–DM

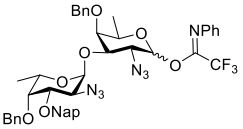
2-azido-4-*O*-benzyl-2-deoxy-3-*O*-(2-naphthylmethyl)-α-L-fucopyranosyl-(1→3)-2-azido-4-*O*-benzyl-2-deoxy-α/β-D-fucopyranose (21)



20 (279 mg, 0.304 mmol) was dissolved in dry THF (2.5 mL, 0.1 M) and cooled to 0°C. AcOH (26 µL, 0.456 mmol, 1.5 equiv.) and TBAF (1M in THF, 0.46 mL, 0.456 mmol, 1.5 equiv.) were added subsequently and the mixture was stirred for 19 h until TLC analysis (pentane/EtOAc, 8:2) showed full conversion of starting material. The reaction was

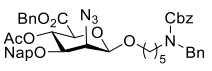
quenched with NH_4Cl (sat. aq.). The mixture was extracted with EtOAc (x3), and the combined organic phases were wash with H_2O (x3), brine (x1), dried over Na_2SO_4, filtered and concentrated *in vacuo*. The crude product was purified by column chromatography (pentane/EtOAc 9:1 → 7:3) to give **21** in 65% yield (135 mg, 0.198 mmol). **^{1}H NMR (400 MHz; $CDCl_3$)** δ : 7.89 – 7.78 (8H, m), 7.76 – 7.69 (1H, m), 7.57 – 7.51 (3H, m), 7.50 – 7.42 (4H, m), 7.38 – 7.27 (21H, m), 5.36 (1H, s), 5.30 (1H, d, J = 3.3 Hz), 5.24 (1H, d, J = 3.5 Hz), 4.97 (2H, d, J = 11.5 Hz), 4.93 – 4.73 (6H, m), 4.67 – 4.60 (4H, m), 4.53 – 4.47 (1H, m), 4.19 – 4.12 (1H, m), 4.09 (1H, dd, J = 10.6, 2.7 Hz), 4.04 – 3.86 (8H, m), 3.84 – 3.76 (2H, m), 3.61 – 3.47 (5H, m), 3.40 (1H, dd, J = 2.8, 1.0 Hz), 3.25 (1H, s), 1.12 (6H, d, J = 6.4 Hz). **^{13}C NMR (100.65 MHz; $CDCl_3$)** δ : 138.35, 138.17, 138.13, 135.19, 135.13, 133.34, 133.16, 128.53, 128.50, 128.48, 128.44, 128.05, 128.03, 127.97, 127.94, 127.92, 127.82, 127.80, 126.81, 126.78, 126.34, 126.31, 126.19, 126.17, 125.90, 125.88, 99.96, 99.80, 96.87, 92.44, 79.86, 78.88, 78.82, 77.52, 76.41, 76.35, 75.43, 75.39, 75.09, 75.05, 72.75, 72.70, 71.09, 67.75, 67.63, 67.10, 65.39, 61.22, 59.98, 59.71, 17.01, 16.85. **HRMS**: $[M+Na]^+$ calculated for $C_{37}H_{40}N_6O_7Na$: 703.28562; found 703.28507

2-Azido-4-*O*-benzyl-2-deoxy-3-*O*-(2-naphthylmethyl)-α-L-fucopyranosyl-(1→3)-2-azido-4-*O*-benzyl-2-deoxy-1-*O*-(*N*-phenyl-2,2,2-trifluoroacetimidoyl)-α/β-D-fucopyranose (22)



21 (135 mg, 0.198 mmol, 1 equiv.) was co-evaporated with toluene (x3) and dissolved in dry acetone (1 mL, 0.2 M). K_2CO_3 (54 mg, 0.396 mmol, 2 equiv.) and ClC(=NPh)CF_3 (64 µL, 0.396 mmol, 2 equiv.) were added and the mixture was stirred under N_2 for 18 h until TLC analysis (pentane/EtOAc, 8:2) showed full conversion of starting material. The mixture was filtered and concentrated *in vacuo*. The crude product was purified by column chromatography (pentane/EtOAc 99:1 → 85:15) to give **22** in 70% yield (116 mg, 0.136 mmol) **^{1}H NMR (400 MHz; CD_3CN)** δ : 7.93 – 7.83 (4H, m), 7.79 – 7.72 (1H, m), 7.65 – 7.53 (1H, m), 7.39 – 7.27 (11H, m), 7.19 – 7.09 (1H, m), 6.88 (2H, d, J = 7.7 Hz), 5.25 (1H, d, J = 3.7 Hz), 4.91 (2H, dd, J = 11.5, 9.1 Hz), 4.86 – 4.73 (2H, m), 4.62 (2H, dd, J = 17.6, 11.1 Hz), 4.01 – 3.81 (4H, m), 3.77 (1H, dd, J = 10.9, 3.7 Hz), 3.63 (1H, d, J = 24.0 Hz), 2.16 (1H, s), 1.97 – 1.93 (2H, m), 1.24 – 1.14 (8H, m). **^{13}C NMR (100.65 MHz; CD_3CN)** δ : 139.83, 139.68, 136.74, 134.69, 134.23, 129.86, 129.35, 129.24, 129.09, 129.03, 128.79, 128.62, 128.61, 127.49, 127.23, 127.07, 127.03, 125.48, 120.09, 118.30, 100.69, 79.42, 78.45, 77.49, 76.49, 75.58, 73.24, 72.81, 72.31, 71.66, 68.48, 67.84, 60.42, 60.40, 17.01, 16.72.

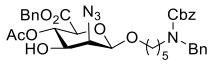
5-(Benzyl(benzyloxycarbonyl)amino)pentyl benzyl(4-*O*-acetyl-2-azido-2-deoxy-3-*O*-(2-naphtylmethyl))-β-D-mannopyranoside (23)



Donor **5** (472 mg, 0.71 mmol, 1 equiv.) and acceptor **12** (278 mg, 0.93 mmol, 1.3 equiv.) were co-evaporated with toluene (3x), dissolved in dry DCM (2.5 mL, 0.3 M) added 3 Å molecular sieve and stirred under argon for 30 min. The mixture was cooled to -78°C and added TBSOTf (34 µL, 0.15 mmol, 0.2 equiv.). The mixture was stirred for 2 h during which it was allowed to warm to -30°C until TLC analysis (pentane/EtOAc, 7:3) showed full conversion of the donor. The reaction was quenched with Et_3N and warmed to rt. The mixture was dissolved in EtOAc, washed with Na-HCO_3 (sat. aq.; 1x) and brine (1x), dried over Na_2SO_4, filtered and concentrated *in vacuo*. The crude product was purified by column chromatography (pentane/EtOAc 8:2 → 7:3) to give **23**

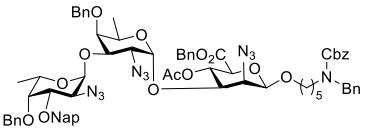
in 24% yield (139 mg, 0.17 mmol). **^{1}H NMR (400 MHz; $CDCl_3$)** δ : 7.86 – 7.79 (3H, m, Ar-*H*), 7.78 – 7.74 (1H, m, Ar-*H*), 7.52 – 7.47 (2H, m, Ar-*H*), 7.46 – 7.43 (1H, m, Ar-*H*), 7.37 – 7.24 (15H, m, Ar-*H*), 5.43 (1H, t, *J* = 9.0 Hz, H-4), 5.21 – 5.02 (2H, m, Ar-CH_2), 4.91 – 4.67 (2H, m, Ar-CH_2), 4.60 – 4.44 (4H, m, CH_2, H-1, H-2), 3.87 (1H, d, *J* = 9.0 Hz, H-3), 3.72 – 3.63 (1H, m, H-5), 3.47 – 3.12 (4H, m, CH_2-Linker), 1.83 (3H, s, COCH_3), 1.66 – 1.45 (4H, m, CH_2-Linker), 1.37 – 1.27 (2H, m, CH_2-Linker). **^{13}C NMR (100.65 MHz; $CDCl_3$)** δ : 169.47 (C=O), 168.24 (C=O), 167.11 (C=O), 137.99 (Ar-C_q), 135.02 (Ar-C_q), 134.88 (Ar-C_q), 134.72 (Ar-C_q), 134.48 (Ar-C_q), 133.20 (Ar-C_q), 128.76 (Ar-*C*), 128.63 (Ar-*C*), 128.58 (Ar-*C*), 128.56 (Ar-*C*), 128.36 (Ar-*C*), 128.05 (Ar-*C*), 128.01 (Ar-*C*), 127.96 (Ar-*C*), 127.91 (Ar-*C*), 127.86 (Ar-*C*), 127.80 (Ar-*C*), 127.41 (Ar-*C*), 127.34 (Ar-*C*), 126.74 (Ar-*C*), 126.47 (Ar-*C*), 126.32 (Ar-*C*), 126.22 (Ar-*C*), 125.65 (Ar-*C*), 125.60 (Ar-*C*), 99.81 (C-1), 76.72 (C-5), 73.08 (C-3), 72.92 (Ar-*C*H_2), 72.16 (Ar-*C*H_2), 70.12 (Ar-*C*H_2), 70.07 (Ar-*C*H_2), 68.54 (C-2), 68.22 (C-4), 67.71 (*C*H_2-Linker), 67.59 (*C*H_2-Linker), 67.24 (*C*H_2-Linker), 50.58 (*C*H_2-Linker), 50.27 (*C*H_2-Linker), 47.12 (*C*H_2-Linker), 46.20 (*C*H_2-Linker), 29.80 (*C*H_2-Linker), 29.12 (*C*H_2-Linker), 27.89 (*C*H_2-Linker), 27.42 (*C*H_2-Linker), 23.13 (*C*H_2-Linker), 20.74 (CO*C*H_3). **HRMS**: $[M+Na]^+$ calculated for $C_{46}H_{48}N_4O_9Na$: 823.33190; found 823.33135

5-(Benzyl(benzyloxycarbonyl)amino)pentyl benzyl(4-*O*-acetyl-2-azido-2-deoxy)-β-D-mannopyranoside (24)



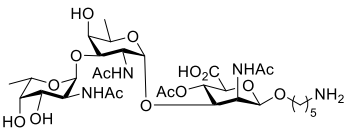
23 (94 mg, 0.117 mmol) was dissolved in DCM (1.1 mL, 0.1 M). DDQ (53 mg, 0.234 mmol, 2 equiv.) and H_2O (50 µL) were added and the mixture was stirred for 3 h until TLC analysis (pentane/EtOAc, 1:1) showed full conversion of the starting material. The reaction was quenched with $Na_2S_2O_3$ (sat. aq.) and extracted with EtOAc (x3). The combined organic layers were washed with $NaHCO_3$ (sat. aq.; x4) and brine (x1), dried over Na_2SO_4, filtered and concentrated *in vacuo*. The crude product was purified by column chromatography (pentane/EtOAc 7:3 → 5:5) to give **24** in 51% yield (39 mg, 0.06 mmol). **^{1}H NMR (400 MHz; $CDCl_3$)** δ : 7.37 – 7.24 (15H, m, Ar-*H*), 5.43 (1H, t, *J* = 9.0 Hz, H-4), 5.21 – 5.02 (3H, m, CH_2), 4.91 – 4.67 (3H, m, CH_2), 4.60 – 4.44 (4H, m, CH_2, H-1, H-2), 3.87 (1H, d, *J* = 9.0 Hz, H-3), 3.72 – 3.63 (1H, m, H-5), 3.47 – 3.12 (4H, m, CH_2-Linker), 1.83 (3H, s, COCH_3), 1.66 – 1.45 (4H, m, CH_2-Linker), 1.37 – 1.27 (2H, m, CH_2-Linker). **^{13}C NMR (100.65 MHz; $CDCl_3$)** δ : 169.47 (C=O), 168.24 (C=O), 167.11 (C=O), 137.99 (Ar-C_q), , 134.88 (Ar-C_q), 134.48 (Ar-C_q), 128.76 (Ar-*C*), 128.63 (Ar-*C*), 128.58 (Ar-*C*), 128.56 (Ar-*C*), 128.36 (Ar-*C*), 128.05 (Ar-*C*), 128.01 (Ar-*C*), 127.96 (Ar-*C*), 127.91 (Ar-*C*), 127.86 (Ar-*C*), 127.80 (Ar-*C*), 127.41 (Ar-*C*), 126.47 (Ar-*C*), 125.65 (Ar-*C*), 125.60 (Ar-*C*), 99.81 (C-1), 76.72 (C-5), 73.08 (C-3), 72.92 (CH_2), 72.16 (CH_2), 70.12 (CH_2), 70.07 (CH_2), 68.54 (C-2), 68.22 (C-4), 67.59 (CH_2), 67.24 (CH_2), 50.58 (*C*H_2-Linker), 47.12 (*C*H_2-Linker), 46.20 (*C*H_2-Linker), 29.80 (*C*H_2-Linker), 29.12 (*C*H_2-Linker), 27.89 (*C*H_2-Linker), 27.42 (*C*H_2-Linker), 23.13 (*C*H_2-Linker), 20.74 (CO*C*H_3). **HRMS**: $[M+Na]^+$ calculated for $C_{35}H_{40}N_4O_9Na$: 683.26930; found 683.26875

5-(Benzyl(benzyloxycarbonyl)amino)pentyl 2-Azido-4-*O*-benzyl-2-deoxy-3-*O*-(2-naphthylmethyl)-α-L-fucopyranosyl-(1→3)-2-azido-4-*O*-benzyl-2-deoxy-α-D-fucopyranosyl-(1→3)-benzyl(4-*O*-acetyl-2-azido-2-deoxy)-β-D-mannopyranoside (25)



Acceptor **24** (30 mg, 0.045 mmol, 1 equiv.) and Donor **22** (58 mg, 0.068 mmol, 1.5 equiv.) were co-evaporated with toluene (x3), dissolved in DCM (1 mL), added 3 Å molecular sieve and stirred under argon for 30 min. TBSOTf (3.1 µL, 0.014 mmol, 0.2 equiv.) was added and the mixture was stirred at rt for 30 min until TLC analysis (pentane/EtOAc, 7:3) showed full conversion of the acceptor. The reaction was quenched with Et_3N, dissolved in EtOAc, washed with $NaHCO_3$ (sat. aq.; 1x) and brine (1x), dried over Na_2SO_4, filtered and concentrated *in vacuo*. The crude product was purified by column chromatography (pentane/EtOAc 8:2 → 7:3) to give **25** in 65% yield (38 mg, 0.029 mmol). **^{1}H NMR (400 MHz; $CDCl_3$)** δ : 7.85 – 7.77 (4H, m, Ar-*H*), 7.75 – 7.70 (1H, m, Ar-*H*), 7.54 – 7.50 (1H, m, Ar-*H*), 7.48 – 7.44 (3H, m, Ar-*H*), 7.38 – 7.26 (23H, m, Ar-*H*), 5.31 (1H, t, *J* = 9.8 Hz, H-4''), 5.22 – 5.11 (5H, m, H-1, H-3', CH_2), 5.01 – 4.72 (5H, m, H-1', CH_2), 4.66 – 4.58 (3H, m, H-2'', H-1'', CH_2), 4.50 (2H, d, *J* = 6.8 Hz, CH_2), 4.06 – 3.84 (8H, m, H-2, H-2', H-3, H-3'', H-4, H-5', C*H*$_2$-Linker), 3.76 – 3.68 (1H, m, H-5''), 3.58 (2H, s, H-5, H-4'), 3.50 – 3.15 (4H, m, C*H*$_2$-Linker), 1.85 (3H, s, COC*H*$_3$), 1.71 – 1.45 (6H, m, C*H*$_2$-Linker), 1.25 – 1.10 (6H, m, H-6, H-6'). **^{13}C NMR (100.65 MHz; $CDCl_3$)** δ : 169.30 (C=O), 166.81 (C=O), 138.29 (Ar-C_q), 138.23 (Ar-C_q), 138.17 (Ar-C_q), 138.03 (Ar-C_q), 135.22 (Ar-C_q), 135.00 (Ar-C_q), 133.36 (Ar-C_q), 133.16 (Ar-C_q), 128.88 (Ar-*C*), 128.70 (Ar-*C*), 128.67 (Ar-*C*), 128.65 (Ar-*C*), 128.56 (Ar-*C*), 128.53 (Ar-*C*), 128.49 (Ar-*C*), 128.43 (Ar-*C*), 128.38 (Ar-*C*), 128.05 (Ar-*C*), 127.98 (Ar-*C*), 127.93 (Ar-*C*), 127.90 (Ar-*C*), 127.81 (Ar-*C*), 127.42 (Ar-*C*), 127.37 (Ar-*C*), 127.34 (Ar-*C*), 126.73 (Ar-*C*), 126.32 (Ar-*C*), 126.16 (Ar-*C*), 125.87 (Ar-*C*), 100.91 (C-1'), 100.07 (C-1''), 99.80 (C-1), 79.81 (CH), 79.48 (CH), 77.58 (CH), 76.28 (CH), 76.20 (CH), 75.57 (CH_2), 74.99 (CH_2), 73.37 (CH), 72.61 (CH_2), 70.08 (CH_2), 70.06 (CH_2), 68.44 (CH), 67.84 (CH_2), 67.80 (CH), 67.25 (CH_2), 63.55 (CH), 61.05 (CH), 59.98 (CH), 29.82 (*C*H_2-Linker), 29.18 (*C*H_2-Linker), 23.09 (*C*H_2-Linker), 20.47 (CO*C*H_3), 17.07 (CH_3), 16.89 (CH_3). **HRMS:** $[M+Na]^+$ calculated for $C_{72}H_{78}N_{10}O_{15}Na$: 1345.55458; found 1345.55403

5-aminopentyl 2-*N*-acetamide-2-deoxy-α-L-fucopyranosyl-(1→3)-2-*N*-acetamide-2-deoxy-α-D-fucopyranosyl-(1→3)- 4-*O*-acetyl-2-*N*-acetamide-2-deoxy-β-D-mannopyranoside (CP8-III)

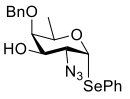


25 (38 mg, 0.029 mmol, 1 equiv.) was dissolved in THF (3 mL) and added zinc powder (0.575 g, 8.73 mmol, 300 equiv.), AcOH (1 mL) and Ac_2O (0.5 mL). The mixture was heated to 50°C and stirred for 18 h until TLC analysis (DCM/MeOH, 95:5) showed full conversion of the starting material. The solution was cooled to rt, filtered over a path of Celite and concentrated *in vacuo*. The crude product was purified by column chromatography (DCM/MeOH, 99:1 → 95:5) to give the acetamide intermediate (31 mg, 0.0227 mmol) in 78%. The product was dissolved in *t*-BuOH (2.5 mL). AcOH (0.1 mL in 100 mL MilliQ, 1 mL) was added and the mixture was birched under argon for 20 min. $Pd(OH)_2$/C (catalytic amount) was added and the mixture was birched under argon for 5 min, then with H_2 for 2 min, before to be stirred for 3 days. The

mixture was birched with argon for 20 min, filtered over a Whatman filter and lyophilized. Purification by a HW40 column with NH_4OAc followed by lyophilization gave **CP8-III** in 60% yield (3 mg, 0.00421 mmol). **^{1}H NMR (600 MHz, D_2O)** δ 5.14 (t, *J* = 10.0 Hz, 1H, H-4), 4.97 (d, *J* = 4.1 Hz, 1H, H''-1), 4.95 (d, *J* = 4.0 Hz, 1H, H'-1), 4.86 (d, *J* = 1.5 Hz, 1H, H-1), 4.50 (dd, *J* = 4.6, 1.5 Hz, 1H, H-2), 4.36 (q, *J* = 6.5 Hz, 1H, H''-5), 4.24 (dd, *J* = 10.9, 4.0 Hz, 1H, H''-2), 4.16 (dd, *J* = 9.8, 4.6 Hz, 1H, H-3), 4.12 (dd, *J* = 11.1, 4.0 Hz, 1H, H'-2), 4.08 (q, *J* = 6.5 Hz, 1H, H'-5), 3.92 (dd, *J* = 11.1, 3.2 Hz, 1H, H'-3), 3.86 (dt, *J* = 10.2, 6.4 Hz, 1H, CH_2-Linker), 3.83 – 3.82 (m, 1H, H'-4), 3.80 (d, *J* = 10.2 Hz, 1H, H-5), 3.79 – 3.78 (m, 1H, H''-4), 3.74 (dd, *J* = 11.0, 3.2 Hz, 1H, H''3), 3.67 (dt, *J* = 10.2, 6.5 Hz, 1H, CH_2-Linker), 3.00 (t, *J* = 7.5 Hz, 2H, CH_2-Linker), 2.11 (s, 3H, COCH_3), 2.05 (s, 3H, COCH_3), 2.03 (s, 3H, COCH_3), 1.96 (s, 3H, COCH_3), 1.72 – 1.60 (m, 4H, CH_2-Linker), 1.48 – 1.38 (m, 2H, CH_2-Linker), 1.25 (dd, *J* = 8.7, 6.6 Hz, 6H, H'-6, H''-6). **^{13}C NMR (151 MHz, D_2O)** δ 175.78 (C=O), 175.47 (C=O), 175.18 (C=O), 174.62 (C=O), 173.34 (C=O), 99.53 (C'-1), 99.51 (C-1), 99.25 (C''-1), 75.17 (C''-5), 74.82 (C-3), 74.11 (C''-3), 71.91 (C'-5/C''-5), 71.88 (C'-5/C''-5), 71.39 (C-4), 70.78 (*C*H_2-Linker), 68.59 (C'-3), 68.06 (C'-5), 67.68 (C''-5), 53.37 (C-2), 50.38 (C'-2), 49.44 (C''-2), 40.30 (*C*H_2-Linker), 28.92 (*C*H_2-Linker), 27.22 (*C*H_2-Linker), 23.18 (CO*C*H_3), 22.98 (CO*C*H_3), 22.95 (*C*H_2-Linker), 21.13 (CO*C*H_3), 16.34 (C'-6/ C''-6), 16.20 (C'-6/ C''-6). **HRMS**: $[M+H]^+$ calculated for $C_{31}H_{52}N_4O_{16}H$: 737.34566; found 737.34511

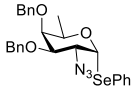
CP5-II: DF–DM–LF

Phenyl 2-azido-4-*O*-benzyl-2-deoxy-1-seleno-α-D-fucopyranoside (27)



26 (727 mg, 1.3 mmol) was dissolved in DCM/H_2O (20:1, 13 mL, 0.1 M) and added DDQ (590 mg, 2.6 mmol, 2 equiv.). The reaction was stirred at rt under N_2 for 2 h until TLC (pentane, EtOAc, 9:1) showed full conversion. The solution was quenched with $Na_2S_2O_3$ (sat. aq.) and diluted/extracted with EtOAc (x3). The combined organic phases were washed with $NaHCO_3$ (sat. aq.; x4) and brine (x1), dried over Na_2SO_4, filtered and concentrated *in vacuo*. Column chromatography (pentane/EtOAc, 95:5 → 80:20) yielded **27** in 89% (483 mg, 1.15 mmol). **^{1}H NMR (400 MHz, $CDCl_3$)** δ 7.61 – 7.54 (m, 2H, Ar-*H*), 7.41 – 7.31 (m, 5H, Ar-*H*), 7.31 – 7.26 (m, 3H, Ar-*H*), 5.91 (d, *J* = 5.2 Hz, 1H, H-1), 4.82 (d, *J* = 11.4 Hz, 1H, Ar-CH_2), 4.71 (d, *J* = 11.4 Hz, 1H, Ar-CH_2), 4.34 (q, *J* = 6.5 Hz, 1H, H-4), 4.03 (dd, *J* = 10.3, 5.2 Hz, 1H, H-2), 3.82 (ddd, *J* = 10.3, 8.7, 3.4 Hz, 1H, H-3), 3.70 (dd, *J* = 3.5, 1.3 Hz, 1H, H-5), 2.23 (d, *J* = 8.7 Hz, 1H, O*H*), 1.26 (d, *J* = 6.6 Hz, 3H, H-6). **^{13}C NMR (101 MHz, $CDCl_3$)** δ 138.07 (Ar-C_q), 134.51 (Ar-*C*), 129.23 (Ar-*C*), 128.86 (Ar-*C*), 128.42 (Ar-*C*), 128.24 (Ar-*C*), 127.90 (Ar-*C*), 85.33 (C-1), 79.89 (C-5), 76.32 (Ar-*C*H_2), 72.07 (C-3), 69.49 (C-4), 62.72 (C-2), 16.09 (C-6). **HRMS**: $[M+Na]^+$ calculated for $C_{19}H_{21}N_3O_3SeNa$: 442.06458; found 442.06405

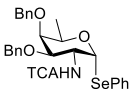
Phenyl 2-azido-3,4-di-*O*-benzyl-2-deoxy-1-seleno-α-D-fucopyranoside (28)



27 (475 mg, 1.14 mmol) was co-evaporated with toluene (x3), dissolved in DMF (11 mL, 0.1 M) and cooled to 0 °C. NaH (60% dispersion in mineral oil, 59 mg, 1.48 mmol, 1.3 equiv.) and BnBr (0.17 mL, 1.48 mmol, 1.3 equiv.) were added and the reaction was allowed to warm to rt and stirred under N_2 overnight until TLC (pentane/EtOAc, 9:1) showed full conversion. The reaction was quenched

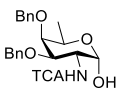
with H_2O and extracted with Et_2O (x3). The combined organic phases were washed with brine (x1), dried over Na_2SO_4, filtered and concentrated *in vacuo*. Column chromatography (pentane/EtOAc, 100:0 → 90:10) yielded **28** in 99 % (577 mg, 1.13 mmol). **^{1}H NMR (400 MHz, $CDCl_3$)** δ 7.60 – 7.54 (m, 2H, Ar-*H*), 7.47 – 7.27 (m, 12H, Ar-*H*), 7.26 – 7.22 (m, 1H, Ar-*H*), 5.93 (d, *J* = 5.3 Hz, 1H, H-1), 4.94 (d, *J* = 11.4 Hz, 1H, Ar-CH_2), 4.77 (d, *J* = 2.2 Hz, 2H, Ar-CH_2), 4.61 (d, *J* = 11.5 Hz, 1H, Ar-CH_2), 4.36 (dd, *J* = 9.7, 5.3 Hz, 1H, H-2), 4.23 (q, *J* = 6.5 Hz, 1H, H-5), 3.76 – 3.70 (m, 2H, *H*-4, H-3), 1.13 (d, *J* = 6.4 Hz, 3H, H-6). **^{13}C NMR (101 MHz, $CDCl_3$)** δ 138.25 (Ar-C_q), 137.57 (Ar-C_q), 134.48 (Ar-*C*), 129.17 (Ar-*C*), 128.84 (Ar-C_q), 128.74 (Ar-*C*), 128.45 (Ar-*C*), 128.31 (Ar-*C*), 128.18 (Ar-*C*), 127.99 (Ar-*C*), 127.92 (Ar-*C*), 127.79 (Ar-*C*), 85.71 (C-1), 80.78 (C-4), 75.87 (C-3), 75.12 (Ar-*C*H$_2$), 72.66 (Ar-*C*H$_2$), 69.53 (C-5), 61.05 (C-2), 16.24 (C-6). **HRMS**: $[M+Na]^+$ calculated for $C_{26}H_{27}N_3O_3SeNa$: 532.11153; found 532.11115

Phenyl 3,4-di-*O*-benzyl-2-deoxy-2-*N*-trichloroacetamide-1-seleno-α-D-fucopyranoside (29)



28 (566 mg, 1.11 mmol) was dissolved in distilled, dry THF (11 mL, 0.1 M) and added zinc powder (800 mg, 12.24 mmol, 11 equiv.) and AcOH (0.6 mL, 10.02 mmol, 9 equiv.). The reaction was stirred under N_2 at rt overnight until TLC (pentane/EtOAc, 90:10) showed full conversion. The solution was filtered over a path of Celite and concentrated *in vacuo*. The crude was co-evaporated with toluene (x3) and dissolved in distilled, dry THF (7.5 mL, 0.15 M). Activated 3Å molecular sieves were added to the solution and the mixture was stirred under N_2 for 30 min. The solution was cooled to 0 °C and trichloroacetyl chloride (0.25 mL, 2.26 mmol, 2 equiv.) was added and stirred for 30 min at 0 °C under N_2 until TLC (pentane/EtOAc, 9:1) showed full conversion. The reaction mixture was diluted in DCM, washed with brine (x1), dried over Na_2SO_4, filtered and concentrated *in vacuo*. Column chromatography (pentane/EtOAc, 95:15 → 80:20) yielded **29** in 93% yield (657 mg, 1.05 mmol). **^{1}H NMR (400 MHz, $CDCl_3$)** δ 7.54 – 7.47 (m, 2H, Ar-*H*), 7.42 – 7.26 (m, 11H, Ar-*H*), 6.84 (d, *J* = 7.5 Hz, 1H, N*H*), 6.04 (d, *J* = 4.7 Hz, 1H, H-1), 4.99 (d, *J* = 11.6 Hz, 1H, Ar-CH_2), 4.79 – 4.70 (m, 2H, Ar-CH_2, H-2), 4.67 (d, *J* = 11.5 Hz, 1H, Ar-CH_2), 4.54 (d, *J* = 11.9 Hz, 1H, Ar-CH_2), 4.21 (q, *J* = 6.3 Hz, 1H, H-5), 3.82 (dd, *J* = 2.7, 1.3 Hz, 1H, H-4), 3.58 (dd, *J* = 11.0, 2.5 Hz, 1H, H-3), 1.29 – 1.22 (m, 5H, H-6). **^{13}C NMR (101 MHz, $CDCl_3$)** δ 134.16 (Ar-*C*), 129.38 (Ar-*C*), 128.91 (Ar-*C*), 128.46 (Ar-*C*), 128.38 (Ar-*C*), 128.05 (Ar-*C*), 127.95 (Ar-*C*), 88.93 (C-1), 78.57 (C-3), 74.88 (Ar-*C*H$_2$), 74.48 (C-4), 71.54 (Ar-*C*H$_2$), 70.56 (C-5), 51.97 (C-2), 16.75 (C-6). **HRMS**: $[M+Na]^+$ calculated for $C_{28}H_{28}Cl_3NO_4SeNa$: 650.01468; found 650.01367

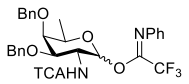
3,4-di-*O*-benzyl-2-deoxy-2-*N*-trichloroacetamide-α-D-fucopyranose (30)



29 (586 mg, 0.934 mmol) was dissolved in acetone/H_2O (10:1, 18 mL, 0.05 M) and cooled to 0 °C. NIS (420 mg, 1.87 mmol, 2 equiv.) was added and the reaction was stirred at 0 °C for 20 min until TLC (pentane/EtOAc, 9:1) showed full conversion. The reaction was quenched with $Na_2S_2O_3$ and the acetone was evaporated. The residue was dissolved in EtOAc and washed with $Na_2S_2O_3$ (sat. aq.; x1), $NaHCO_3$ (sat. aq.; x1) and brine (x1), dried over Na_2SO_4, filtered and concentrated *in vacuo*. Column chromatography (pentane/EtOAc, 80:20 → 60:40) yielded **30** in 75% (340 mg, 0.695 mmol). **^{1}H NMR (400 MHz, $CDCl_3$)** δ 7.41 – 7.25 (m, 13H, Ar-*H*), 6.79 (d, *J* = 8.9 Hz, 1H, N*H*), 5.38

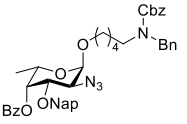
(t, *J* = 3.6 Hz, 1H, H-1), 5.00 (d, *J* = 11.6 Hz, 1H, Ar-C*H*$_2$), 4.76 – 4.67 (m, 1H, Ar-C*H*$_2$), 4.70 – 4.61 (m, Ar-C*H*$_2$, 2H, H-2), 4.56 (d, *J* = 12.0 Hz, 1H, Ar-C*H*$_2$), 4.10 (q, *J* = 6.3 Hz, 1H, H-5), 3.86 – 3.77 (m, 1H, H-3), 3.78 – 3.71 (m, 1H, H-4), 2.88 (dd, *J* = 3.5, 1.5 Hz, 1H, O*H*), 1.20 (d, *J* = 6.5 Hz, 3H, H-6). **^{13}C NMR (101 MHz, CDCl$_3$)** δ 138.28 (Ar-*C*$_q$), 137.76 (Ar-*C*$_q$), 128.99 (Ar-*C*), 128.75 (Ar-*C*), 128.57 (Ar-*C*), 128.52 (Ar-*C*), 128.45 (Ar-*C*), 128.32 (Ar-*C*), 128.08 (Ar-*C*), 127.90 (Ar-*C*), 127.81 (Ar-*C*), 91.80 (C-1), 77.16 (C-3), 75.02 (C-6), 74.73 (Ar-*C*H$_2$), 71.80 (Ar-*C*H$_2$), 67.10 (C-5), 51.22 (C-2), 17.11 (C-6). **HRMS**: [M+Na]$^+$ calculated for $C_{22}H_{24}Cl_3NO_5Na$: 510.01678; found 510.016123

3,4-di-*O*-benzyl-2-deoxy-2-*N*-trichloroacetamide-1-*O*-(*N*-phenyl-2,2,2-trifluoroacetimidoyl)-α/β-D-fucopyranose (6)



30 (340 mg, 0.695 mmol) was co-evaporated with toluene (x3) and dissolved in dry acetone (3.3 mL, 0.2 M). K_2CO_3 (183 mg, 1.324 mmol, 2 equiv.) and ClC(=NPh)CF_3 (0.2 mL, 1.324 mmol, 2 equiv.) were added to the reaction and it was stirred overnight under N_2 at rt until TLC (pentane/EtOAc, 8:2) showed full conversion. The mixture was filtered and concentrated *in vacuo*. Column chromatography (pentane/EtOAc, 95:5 → 80:20) furnished imidate donor **6** in 97% (446 mg, 0.676 mmol). **^{1}H NMR (400 MHz, CD$_3$CN)** δ 7.68 – 7.22 (m, 38H), 7.17 – 7.08 (m, 2H), 6.82 (d, *J* = 7.8 Hz, 3H), 6.26 (d, *J* = 6.8 Hz, 1H), 4.92 (d, *J* = 11.2 Hz, 1H), 4.84 (dd, *J* = 24.1, 11.1 Hz, 3H), 4.76 (s, 2H), 4.70 – 4.59 (m, 4H), 4.44 (m, 1H), 4.26 (dd, *J* = 7.9, 6.8 Hz, 1H), 4.20 – 4.02 (m, 4H), 4.02 – 3.95 (m, 1H), 3.82 (dd, *J* = 2.9, 1.6 Hz, 1H), 3.51 (dd, *J* = 7.9, 2.7 Hz, 1H), 1.25 (s, 3H). **^{13}C NMR (101 MHz, CD$_3$CN)** δ 139.76, 139.64, 139.13, 130.09, 129.91, 129.40, 129.34, 129.30, 129.27, 129.11, 129.04, 129.01, 128.78, 128.71, 128.64, 128.59, 128.55, 126.98, 122.08, 108.93, 81.30, 76.61, 76.20, 75.97, 75.36, 74.78, 72.09, 72.05, 71.69, 70.66, 66.45, 51.69, 29.72, 17.33, 17.10.

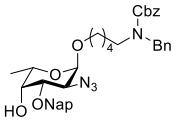
5-(Benzyl(benzyloxycarbonyl)amino)pentyl 2-azido-4-*O*-benzoyl-2-deoxy-3-*O*-(2-naphthylmethyl)-L-fucopyranoside (31)



Donor **8** (201 mg, 0.383 mmol, 1 equiv.) and acceptor **12** (181 mg, 0.575 mmol, 1.5 equiv.) were co-evaporated with toluene (x3) before being dissolved in DCM (3.8 mL, 0.1 M). Activated 3Å molecular sieves were added and the solution was stirred for 30 min under argon at rt. NIS (129 mg, 0.575 mmol, 1.5 equiv.) and TBSOTf (18 µL, 0.0766 mmol, 0.2 equiv.) were added at rt and the reaction was stirred at rt for 15 min until TLC (pentane/EtOAc, 8:2) showed full conversion. The reaction was quenched with Et_3N, diluted in EtOAc, washed with $Na_2S_2O_3$ (sat. aq.; x1), $NaHCO_3$ (sat. aq.; x1) and brine (x1), dried over Na_2SO_4, filtered and concentrated *in vacuo*. Column chromatography (pentane/EtOAc, 90:10 → 70:30) yielded **31** in 98% (α: 103 mg, 0.139 mmol; β: 175 mg, 0.235 mmol) in a α/β = 37:63. **^{1}H NMR (400 MHz, CDCl$_3$)** δ 8.13 – 8.06 (m, 2H, Ar-*H*), 7.83 – 7.64 (m, 6H, Ar-*H*), 7.64 – 7.26 (m, 22H, Ar-*H*), 5.77 – 5.65 (m, 1H, H-4), 5.24 – 5.12 (m, 2H, Ar-C*H*$_2$), 4.99 (d, *J* = 11.0 Hz, 1H, Ar-C*H*$_2$), 4.94 (d, *J* = 8.9 Hz, 1H, H-1), 4.71 (d, *J* = 11.1 Hz, 1H, Ar-C*H*$_2$), 4.49 (t, *J* = 9.4 Hz, 4H, C*H*$_2$-Linker), 4.18 – 4.03 (m, 2H, H-3, H-5), 4.06 – 3.90 (m, 1H, C*H*$_2$-Linker), 3.75 (dd, *J* = 10.6, 3.5 Hz, 1H, H-2), 3.71 – 3.36 (m, 2H, C*H*$_2$-Linker), 3.32 – 3.08 (m, 2H, C*H*$_2$-Linker), 1.68 – 1.42 (m, 3H, C*H*$_2$-Linker), 1.40 – 1.13 (m, 6H, C*H*$_2$-Linker, H-6). **^{13}C NMR (101 MHz, CDCl$_3$)** δ 166.32 (C=O), 134.84 (Ar-*C*$_q$), 133.42 (Ar-*C*), 133.34 (Ar-*C*$_q$), 133.16 (Ar-*C*$_q$),

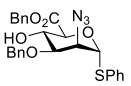
132.21 (Ar-*C*), 130.02 (Ar-*C*), 129.17 (Ar-*C*), 128.67 (Ar-*C*), 128.61 (Ar-*C*), 128.27 (Ar-*C*), 128.08 (Ar-*C*), 127.98 (Ar-*C*), 127.96 (Ar-*C*), 127.74 (Ar-*C*), 127.44 (Ar-*C*), 127.31 (Ar-*C*), 127.14 (Ar-*C*), 126.14 (Ar-*C*), 126.06 (Ar-*C*), 125.98 (Ar-*C*), 125.35 (Ar-*C*), 98.30 (C-1), 74.40 (C-5/C-3), 71.59 (Ar-*C*H_2), 70.12 (C-4), 68.45 (*C*H_2-Linker), 67.30 (Ar-*C*H_2), 65.29 (C-3/C-5), 59.52 (C-2), 50.33 (*C*H_2-Linker), 47.22 (*C*H_2-Linker), 29.47 (*C*H_2-Linker), 23.09 (*C*H_2-Linker), 16.50 (C-6). **HRMS**: $[M+Na]^+$ calculated for $C_{44}H_{46}N_4O_7Na$: 765.32642; found 765.32587

5-(Benzyl(benzyloxycarbonyl)amino)pentyl 2-azido-2-deoxy-3-*O*-(2-naphthylmethyl)-L-fucopyranoside (32)



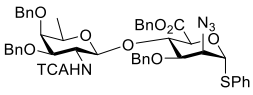
31 (264 mg, 0.355 mmol) was dissolved in MeOH (1.8 mL, 0.2 M) and added NaOMe (0.08 mL, 0.355 mmol, 1 equiv.) and stirred at rt for 2 days until TLC (pentane/EtOAc, 8:2) showed full conversion. The reaction was neutralized with Amberlite IR-120 H^+ until pH≈8-9, filtered and concentrated *in vacuo*. Column chromatography (pentane/EtOAc, 80:20 → 60:40) yielded **32** in 87% (201 mg, 0.315 mmol). **^{1}H NMR (400 MHz, $CDCl_3$)** δ 7.91 – 7.78 (m, 4H, Ar-*H*), 7.53 – 7.41 (m, 3H, Ar-*H*), 7.40 – 7.26 (m, 12H, Ar-*H*), 7.17 (d, *J* = 7.2 Hz, 2H, Ar-*H*), 5.18 (d, *J* = 12.5 Hz, 2H, Ar-CH_2), 4.94 – 4.78 (m, 3H, H-1, Ar-CH_2), 4.50 (d, *J* = 6.9 Hz, 2H, Ar-CH_2), 3.99 – 3.80 (m, 3H, H-3, H-4, H-5), 3.64 (dd, *J* = 10.4, 3.6 Hz, 1H, H-2), 3.59 (m, 1H, CH_2-Linker), 3.61 – 3.11 (m, 3H, CH_2-Linker), 2.37 (s, 1H, OH), 1.61 – 1.45 (m, 5H, CH_2-Linker), 1.46 – 1.18 (m, 5H, H-6, CH_2-Linker). **^{13}C NMR (101 MHz, $CDCl_3$)** δ 138.09 (Ar-C_q), 134.72 (Ar-C_q), 133.39 (Ar-C_q), 128.73 (Ar-*C*), 128.68 (Ar-*C*), 128.10 (Ar-*C*), 127.96 (Ar-*C*), 127.89 (Ar-*C*), 127.43 (Ar-*C*), 126.46 (Ar-*C*), 126.36 (Ar-*C*), 125.84 (Ar-*C*), 99.01 (C-1), 76.48 (C-3), 72.19 (Ar-*C*H_2), 69.06 (C-4/C-5), 68.21 (*C*H_2-Linker), 67.31 (Ar-*C*H_2), 65.60 (C-4/C-5), 59.07 (C-2), 50.24 (*C*H_2-Linker), 46.14 (*C*H_2-Linker), 29.21 (*C*H_2-Linker), 22.93 (*C*H_2-Linker), 16.37 (C-6). **HRMS**: $[M+Na]^+$ calculated for $C_{37}H_{42}N_4O_6Na$: 661.30020; found 661.2996

Benzyl (2-azido-3-*O*-benzyl-2-deoxy-1-thio)-α-D-mannopyranosiduronate (7)



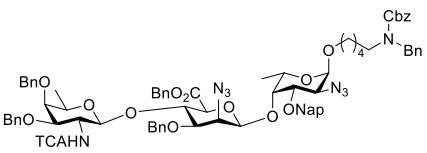
10 (405 mg, 0.663 mmol) was dissolved in DCM/H_2O (20:1, 6.6 mL, 0.1 M), added DDQ (451 mg, 1.98 mmol, 2 equiv.) and stirred at rt under N_2 for 2 h until TLC (pentane/EtOAc, 8:2) showed full conversion. The solution was quenched with $Na_2S_2O_3$ (sat. aq.) and diluted/extracted with EtOAc (x3). The combined organic phases were washed with $NaHCO_3$ (sat. aq.; x4) and brine (x1), dried over Na_2SO_4, filtrated and concentrated *in vacuo*. Column chromatography (pentane/EtOAc, 90:10 → 75:25) yielded **7** in 93% (304 mg, 0.618 mmol). **^{1}H NMR (400 MHz, $CDCl_3$)** δ 7.53 – 7.48 (m, 2H, Ar-*H*), 7.44 – 7.30 (m, 10H, Ar-*H*), 7.29 – 7.21 (m, 3H, Ar-*H*), 5.56 (d, *J* = 3.4 Hz, 1H, H-1), 5.18 (s, 2H, Ar-CH_2), 4.76 (dd, *J* = 11.6, 10.1 Hz, 2H, Ar-CH_2), 4.70 (d, *J* = 8.1 Hz, 1H, H-5), 4.39 (td, *J* = 8.0, 3.5 Hz, 1H, H-4), 3.98 (t, *J* = 3.5 Hz, 1H, H-2), 3.92 (dd, *J* = 8.0, 3.4 Hz, 1H, H-3), 2.90 (t, *J* = 3.0 Hz, 1H, O*H*). **^{13}C NMR (101 MHz, $CDCl_3$)** δ 169.39 (C=*O*), 137.35 (Ar-C_q), 135.07 (Ar-C_q), 132.50 (Ar-C_q), 132.28 (Ar-*C*), 129.25 (Ar-*C*), 128.76 (Ar-*C*), 128.61 (Ar-*C*), 128.33 (Ar-*C*), 128.23 (Ar-*C*), 128.18 (Ar-*C*), 85.55 (C-1), 78.08 (C-3), 73.45 (Ar-*C*H_2), 72.98 (C-5), 68.53 (C-4), 67.52 (Ar-*C*H_2), 61.20 (C-2). **HRMS**: $[M+Na]^+$ calculated for $C_{19}H_{19}N_3O_5SNa$: 492.15932; found 492.15877

Phenyl 3,4-di-*O*-benzyl-2-deoxy-2-*N*-trichloroacetamide-β-D-fucopyranosyl-(1→4) benzyl (2-azido-3-*O*-benzyl-2-deoxy-1-thio)-α-D-mannopyranosiduronate (33)



Acceptor **7** (195 mg, 0.396 mmol, 1 equiv.) and donor **6** (339 mg, 0.514 mmol, 1.3 equiv.) were co-evaporated with toluene (x3), dissolved in dry DCM/MeCN (2:1, 4 mL, 0.1 M), added activated 3Å molecular sieves and stirred for 30 min under argon at rt. The mixture was cooled to -78 °C, after which TBSOTf (18 µL, 0.0791 mmol, 0.2 equiv.) was added. The reaction mixture was stirred at -78 °C for 3 h until TLC (pentane/EtOAc, 7:3) showed full conversion. The reaction mixture was quenched with Et_3N and diluted in EtOAc. The organic phase was washed with $NaHCO_3$ (sat. aq.; x1) and brine (x1), dried over Na_2SO_4, filtered and concentrated *in vacuo*. Column chromatography (pentane/EtOAc, 85:15 → 70:30) and size exclusion chromatography furnished **33** in 51% yield (α: 56 mg, 0.058 mmol; β: 140 mg, 0.146 mmol) in a α/β = 29:71. **^{1}H NMR (400 MHz, $CDCl_3$)** δ 7.58 – 7.46 (m, 2H, Ar-*H*), 7.43 – 7.23 (m, 22H, Ar-*H*), 7.22 – 7.03 (m, 6H, Ar-*H*), 6.88 (d, *J* = 7.2 Hz, 1H, , N*H*), 5.67 (d, *J* = 10.3 Hz, 1H, H-1'), 5.02 (d, *J* = 8.3 Hz, 1H, Ar-CH_2), 4.94 (d, *J* = 11.9 Hz, 2H, Ar-CH_2), 4.79 (d, *J* = 12.1 Hz, 1H, Ar-CH_2), 4.70 – 4.63 (m, 2H, Ar-CH_2), 4.55 (d, *J* = 11.3 Hz, 1H, Ar-CH_2), 4.52 – 4.42 (m, 4H, Ar-CH_2, H-4, H-5), 4.21 (dd, *J* = 11.0, 2.8 Hz, 1H, H-3), 4.07 (t, *J* = 3.4 Hz, 1H, H-3'), 3.74 (dt, *J* = 11.1, 7.7 Hz, 1H, H-2), 3.66 (d, *J* = 2.7 Hz, 1H, H-4'), 3.60 – 3.46 (m, 2H, H-5', H-2'), 1.15 (d, *J* = 6.3 Hz, 3H, H-6'). **^{13}C NMR (101 MHz, $CDCl_3$)** δ 168.84 (*C*=O), 162.34 (*C*=O), 138.05 (Ar-C_q), 137.40 (Ar-C_q), 136.92 (Ar-C_q), 135.00 (Ar-C_q), 132.39 (Ar-C_q), 132.16 (Ar-*C*), 128.80 (Ar-*C*), 128.71 (Ar-*C*), 128.66 (Ar-*C*), 128.54 (Ar-*C*), 128.50 (Ar-*C*), 128.42 (Ar-*C*), 128.36 (Ar-*C*), 128.31 (Ar-*C*), 128.22 (Ar-*C*), 128.08 (Ar-*C*), 127.62 (Ar-*C*), 98.73 (C-1'), 80.56 (C-1), 77.59 (C-3), 75.95 (C-3'), 74.89 (Ar-*C*H$_2$), 74.68 (C-4, C-4', C-5), 74.56 (C-4, C-4', C-5), 74.41 (C-4, C-4', C-5), 73.05 (Ar-*C*H$_2$), 72.77 (Ar-*C*H$_2$), 71.12 (C-5'), 67.39 (Ar-*C*H$_2$), 57.39 (C-2), 55.71 (C-2'), 17.06 (C-6'). **HRMS**: $[M+Na]^+$ calculated for $C_{48}H_{47}Cl_3N_4O_9SNa$: 983.20270; found 983.20215

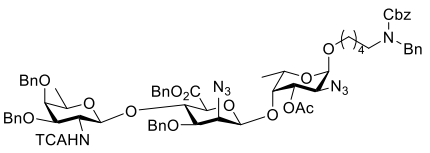
5-(Benzyl(benzyloxycarbonyl)amino)pentyl 3,4-di-*O*-benzyl-2-deoxy-2-*N*-trichloroacetamide-β-D-fucopyranosyl-(1→4) benzyl(2-azido-3-*O*-benzyl-2-deoxy-1-thio)-α-D-mannopyranosiduronsyl-(1→4)- 2-azido-2-deoxy-3-*O*-(2-naphthylmethyl)-L-fucopyranoside (34)



Acceptor **32** (68 mg, 0.107 mmol, 1 equiv.) and donor **33** (154 mg, 0.16 mmol, 1.5 equiv.) were co-evaporated with toluene (x3), dissolved in dry DCM (1 mL, 0.1 M), added activated 3Å molecular sieves and stirred for 30 min under argon at rt. The mixture was cooled to -30 °C and added NIS (48 mg, 0.213 mmol, 2 equiv.) and TBSOTf (5 µL, 0.0213 mmol, 0.2 equiv.). The reaction was allowed to warm to 10 °C and stirred for 5 h until TLC (pentane/EtOAc, 7:3) showed full conversion. The reaction mixture was quenched with Et_3N, diluted in EtOAc, washed with NaS_2O_3 (sat. aq.; x1), $NaHCO_3$ (sat. aq.; x1) and brine (x1), dried over Na_2SO_4, filtered and concentrated *in vacuo*. Column chromatography (pentane/EtOAc, 80:20 → 65:35) furnished **34** in 52% yield (83 mg, 0.0555 mmol). **^{1}H NMR (400 MHz, $CDCl_3$)** δ 7.90 – 7.68 (m, 5H, Ar-*H*), 7.65 – 7.27 (m, 34H, Ar-*H*), 7.26 – 6.89 (m, 19H, Ar-*H*), 6.56 (d, *J* = 8.6 Hz, 1H, Ar-*H*), 5.17 (d, *J* = 10.1 Hz, 2H, Ar-CH_2), 5.00 – 4.87 (m, 5H, Ar-CH_2), 4.78 (d, *J* = 11.9 Hz, 2H, H-1, Ar-CH_2), 4.72 – 4.64 (m, 4H, H-1', Ar-

CH_2), 4.59 (dd, *J* = 11.8, 6.8 Hz, 3H, Ar-CH_2), 4.52 – 4.43 (m, 5H, Ar-CH_2), 4.35 (d, *J* = 8.3 Hz, 1H, H-1''), 4.25 (t, *J* = 9.2 Hz, 1H, H-4'), 4.16 – 4.07 (m, 2H, H-3, H-2''), 4.03 (d, *J* = 3.8 Hz, 1H, H-2'), 3.86 – 3.79 (m, 2H, H-4, H-5), 3.76 (dd, *J* = 10.1, 4.3 Hz, 2H, H-2, H-5''), 3.55 (d, *J* = 2.8 Hz, 1H, H-4''), 3.49 (dd, *J* = 8.9, 3.7 Hz, 1H, H-3'), 3.39 (dt, *J* = 12.2, 4.0 Hz, 2H, H-3'', CH_2-Linker), 3.33 – 3.13 (m, 3H, CH_2-Linker), 3.10 (q, *J* = 6.3 Hz, 1H, H-5''), 1.62 – 1.41 (m, 6H, CH_2-Linker), 1.33 – 1.09 (m, 15H, CH_2-Linker, H-6'', H-6). **^{13}C NMR (101 MHz, $CDCl_3$)** δ 167.99 (C=O), 161.90 (C=O), 138.63 (Ar-C_q), 138.23 (Ar-C_q), 137.77 (Ar-C_q), 135.65 (Ar-C_q), 134.85 (Ar-C_q), 133.36 (Ar-C_q), 133.08 (Ar-C_q), 129.26 (Ar-*C*), 128.77 (Ar-*C*), 128.74 (Ar-*C*), 128.69 (Ar-*C*), 128.65 (Ar-*C*), 128.59 (Ar-*C*), 128.56 (Ar-*C*), 128.51 (Ar-*C*), 128.45 (Ar-*C*), 128.42 (Ar-*C*), 128.35 (Ar-*C*), 128.32 (Ar-*C*), 128.26 (Ar-*C*), 128.21 (Ar-*C*), 128.16 (Ar-*C*), 128.07 (Ar-*C*), 127.95 (Ar-*C*), 127.92 (Ar-*C*), 127.84 (Ar-*C*), 127.82 (Ar-*C*), 127.74 (Ar-*C*), 127.68 (Ar-*C*), 127.64 (Ar-*C*), 127.37 (Ar-*C*), 126.90 (Ar-*C*), 126.53 (Ar-*C*), 126.23 (Ar-*C*), 125.99 (Ar-*C*), 125.83 (Ar-*C*), 100.92 (C-1'), 99.53 (C-1''), 98.04 (C-1), 79.29 (C-3''), 77.89 (C-3'), 75.36 (C-5'), 74.98 (C-3), 74.71 (C-5), 74.68 (Ar-*C*H_2), 74.00 (C-4''), 73.62 (C-4'), 70.60 (Ar-*C*H_2), 71.72 (Ar-*C*H_2), 70.60 (C-5''), 68.25 (Ar-*C*H_2), 67.56 (Ar-*C*H_2), 67.26 (Ar-*C*H_2), 66.10 (Ar-*C*H_2), 65.89 (C-4), 62.36 (C-2'), 58.61 (C-2), 54.63 (C-2''), 50.60 (CH_2-Linker), 50.31 (CH_2-Linker), 47.20 (CH_2-Linker), 46.21 (CH_2-Linker), 29.82 (CH_2-Linker), 29.13 (CH_2-Linker), 27.95 (CH_2-Linker), 27.52 (CH_2-Linker), 23.40 (CH_2-Linker), 17.21 (C-6''/C-6), 17.07 (C-6''/C-6). **HRMS**: $[M+Na]^+$ calculated for $C_{79}H_{83}Cl_3N_8O_{15}Na$: 1511.49412; found 1511.49357

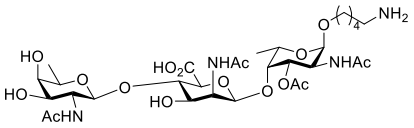
5-(Benzyl(benzyloxycarbonyl)amino)pentyl 3,4-di-*O*-benzyl-2-deoxy-2-*N*-trichloroacetamide-β-D-fucopyranosyl-(1→4) benzyl(2-azido-3-*O*-benzyl-2-deoxy-1-thio)-α-D-mannopyranosiduronsyl-(1→4)-2-*O*-acetyl-2-azido-2-deoxy-L-fucopyranoside (35)



34 (81 mg, 0.0545 mmol) was dissolved in DCM/H_2O (4:1, 2.7 mL, 0.02 M) and added DDQ (25 mg, 0.109 mmol, 2 equiv.). The reaction was stirred at rt under N_2 for 5 h until TLC (pentane, EtOAc, 7:3) showed full conversion. The solution was quenched with $Na_2S_2O_3$ (sat. aq.) and diluted/extracted with EtOAc (x3). The combined organic phases were washed with $NaHCO_3$ (sat. aq.; x4) and brine (x1), dried over Na_2SO_4, filtrated and concentrated *in vacuo*. The crude was used without further purification. The residue was dissolved in pyridine (2 mL) and cooled to 0 °C and added Ac_2O (0.3 mL) and DMAP (catalytic amount) and stirred overnight at rt under N_2 until TLC (pentane/EtOAc, 7:3) showed full conversion. The mixture was dissolved in EtOAc, washed with 1 M HCl (x1), $NaHCO_3$ (sat. aq.; x1) and brine (x1), dried over Na_2SO_4 and concentrated *in vacuo*. Column chromatography (pentane/EtOAc, 8:2 → 6:4) yielded **35** in 57% (44 mg, 0.0312 mmol). **^{1}H NMR (400 MHz, $CDCl_3$)** δ 7.52 – 7.26 (m, 27H), 7.26 – 7.12 (m, 8H), 6.57 (d, *J* = 8.1 Hz, 1H), 5.28 – 5.14 (m, 4H), 5.09 – 5.03 (m, 1H), 4.94 (d, *J* = 11.5 Hz, 2H), 4.90 – 4.74 (m, 2H), 4.74 – 4.55 (m, 4H), 4.57 – 4.42 (m, 5H), 4.26 (t, *J* = 9.2 Hz, 1H), 4.18 – 4.08 (m, 1H), 4.04 – 3.88 (m, 3H), 3.88 – 3.72 (m, 2H), 3.72 – 3.63 (m, 2H), 3.57 (d, *J* = 2.7 Hz, 2H), 3.51 (dp, *J* = 7.5, 3.6 Hz, 1H), 3.46 – 3.13 (m, 4H), 2.02 (s, 3H), 1.72 – 1.47 (m, 6H), 1.41 – 1.21 (m, 5H), 1.16 (d, *J* = 6.6 Hz, 7H). **^{13}C NMR (101 MHz, $CDCl_3$)** δ 170.77, 167.78, 162.03, 138.52, 137.98, 137.74, 135.15, 129.32, 129.27, 128.90, 128.81, 128.77, 128.73, 128.68, 128.66, 128.59, 128.49, 128.42, 128.33, 128.25, 128.11,

128.07, 128.04, 127.94, 127.89, 127.83, 127.77, 127.70, 127.67, 127.37, 100.94, 98.74, 97.94, 78.50, 78.01, 75.68, 75.35, 74.75, 74.72, 73.15, 73.11, 71.95, 70.66, 69.68, 68.39, 67.76, 67.27, 65.30, 61.79, 56.99, 55.27, 50.65, 50.35, 47.19, 46.26, 29.82, 29.17, 23.92, 20.98, 17.13, 16.56.

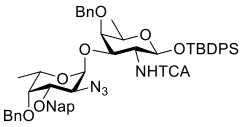
5-aminopentyl 2-acetamide-2-deoxy-β-D-fucopyranosyl-(1→4) 2-acetamide-2-deoxy-α-D-mannopyranosiduronsyl-(1→4)-2-*O*-acetyl-2-acetamide-2-deoxy-L-fucopyranoside (CP5-II)



35 (42 mg, 0.0302 mmol) was dissolved in THF (distilled, 3 mL) and added zinc powder (592 mg, 9.047 mmol, 300 equiv.), AcOH (1 mL) and Ac_2O (0.5 mL). The resulting mixture was stirred at 50 °C overnight until TLC (DCM/MeOH, 95:5) showed full conversion. The cooled mixture was filtered through Celite, evaporated *in vacuo* and co-evaporated with toluene (x3). The crude product was first purified by column chromatography (DCM/MeOH, 98:2 → 90:10) followed by HPLC given the acetamide intermediate in 21% yield (8 mg, 0.00605 mmol). The product (8 mg, 0.00605 mmol) was dissolved in *t*-BuOH (2.5 mL) and added AcOH (1 mL, 0.1 mL in 100 mL MilliQ). The solution was birched with argon for 20 min and then $Pd(OH)_2/C$ (catalytic amount) was added. The reaction was again birched with argon for 5 minutes before the atmosphere was changed for H_2. The mixture was stirred for 3 days under H_2 atmosphere, after which it was birched with argon for 20 min, filtered over a Whatman filter and lyophilized. Purification by a HW40 column with NH_4OAc followed by lyophilization gave **CP5-II** in 64% yield (3.1 mg, 0.00423 mmol). **^{1}H NMR (600 MHz, D_2O)** δ 4.94 (dd, *J* = 11.6, 3.0 Hz, 1H, H-3), 4.85 (d, *J* = 3.7 Hz, 1H, H-1), 4.71 (d, *J* = 1.4 Hz, 1H, H-1'), 4.64 (dd, *J* = 4.5, 1.4 Hz, 1H, H-2'), 4.39 (d, *J* = 8.4 Hz, 1H, H-1), 4.34 (dd, *J* = 11.6, 3.7 Hz, 1H, H-2), 4.18 – 4.12 (m, 2H, H-4, H-5), 3.86 (dd, *J* = 9.6, 4.4 Hz, 1H, H-3'), 3.85 – 3.77 (m, 4H, H-4'', H-2'', H-5'', H-4'), 3.75 (d, *J* = 3.8 Hz, 1H, H-5'), 3.72 – 3.66 (m, 2H, H-3'', CH_2-Linker), 3.58 (d, *J* = 9.5 Hz, 1H, H-5'), 3.50 (dt, *J* = 10.1, 6.2 Hz, 1H, CH_2-Linker), 3.00 (t, *J* = 7.7 Hz, 2H, CH_2-Linker), 2.13 (s, 3H, COCH_3), 2.05 (s, 3H, COCH_3), 2.04 (s, 3H, COCH_3), 1.99 (s, 3H, COCH_3), 1.71 – 1.60 (m, 4H, CH_2-Linker), 1.50 – 1.40 (m, 2H, CH_2-Linker), 1.28 (d, *J* = 6.4 Hz, 3H, H-6''), 1.24 (d, *J* = 6.5 Hz, 3H, H-6). **^{13}C NMR (151 MHz, D_2O)** δ 176.43 (C=O), 176.01 (C=O), 175.37 (C=O), 174.78 (C=O), 174.72 (C=O), 102.56 (C-1''), 100.99 (C-1'), 97.90 (C-1), 80.19 (C-4'), 78.32 (C-5'), 77.10 (C-4), 71.9 (C-5''/ C-3''), 71.92 (C-5''/ C-3''), 71.26 (C-5), 71.13 (C-3'), 70.95 (C-3), 68.87 (CH_2-Linker), 67.15 (C-5), 52.87 (C-2'/ C-2''), 48.17 (C-2), 40.28 (CH_2-Linker), 28.92 (CH_2-Linker), 27.38 (CH_2-Linker), 23.42 (COCH_3), 23.22 (CH_2-Linker), 22.91 (COCH_3), 22.72 (COCH_3), 21.22 (COCH_3), 16.33 (C-6''), 16.12 (C-6). **HRMS**: $[M+H]^+$ calculated for $C_{31}H_{52}N_4O_{16}H$: 737.34566; found 737.34526

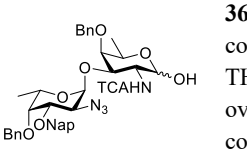
CP5-III: LF–DF–DM

***Tert*-butyldiphenylsilyl 2-azido-4-*O*-benzyl-2-deoxy-3-*O*-(2-naphthylmethyl)-α-L-fucopyranosyl-(1→3)-2-deoxy-2-*N*-trichloroacetamide-4-*O*-benzyl-β-D-fucopyranoside (36)**



Acceptor **9** (160 mg, 0.251 mmol, 1 2 equiv.) and donor **3** (222 mg, 0.376 mmol, 1.5 equiv.) were co-evaporated with toluene (x3), dissolved in dry DCM (2.5 mL, 0.1 M) and added activated 3Å molecular sieves and stirred for 30 min under argon at rt. TBSOTf (12 µL, 0.0501 mmol, 0.2 equiv.) was added at rt and the reaction was stirred for 30 min under argon until TLC (pentane/EtOAc, 8:2) showed full conversion. The reaction was quenched with Et_3N, diluted in EtOAc, washed $NaHCO_3$ (sat. aq.; x1) and brine (x1), dried over Na_2SO_4, filtered and concentrated *in vacuo*. Column chromatography (pentane/EtOAc, 95:5 → 80:20) yielded the α-1,3-linked disaccharide **36** in 89% yield (231 mg, 0.222 mmol). **^{1}H NMR (400 MHz, $CDCl_3$)** δ 7.90 – 7.79 (m, 4H, Ar-*H*), 7.79 – 7.74 (m, 2H, Ar-*H*), 7.71 – 7.64 (m, 3H, Ar-*H*), 7.55 – 7.46 (m, 4H, Ar-*H*), 7.44 – 7.27 (m, 20H, Ar-*H*), 4.98 (d, *J* = 3.7 Hz, 1H, H-1'), 4.95 (d, *J* = 7.6 Hz, 1H, H-1), 4.91 (d, *J* = 11.4 Hz, 1H, Ar-CH_2), 4.83 (s, 2H, Ar-CH_2), 4.75 (d, *J* = 8.2 Hz, 2H, Ar-CH_2), 4.71 (d, *J* = 4.2 Hz, 1H, Ar-CH_2), 4.60 (d, *J* = 11.5 Hz, 1H, Ar-CH_2), 4.20 – 4.12 (m, 1H, H-2), 4.06 (dd, *J* = 11.0, 2.8 Hz, 1H, H-3), 4.00 (dd, *J* = 10.6, 3.6 Hz, 1H, H-2'), 3.94 – 3.84 (m, 2H, H-5, H-3'), 3.60 (dd, *J* = 2.7, 1.2 Hz, 1H, H-4), 3.48 (d, *J* = 2.7 Hz, 1H, H-4'), 3.31 (q, *J* = 6.5 Hz, 1H, H-5'), 1.10 (s, 9H, (CH_3)$_3$), 1.08 (d, , *J* = 6.4 Hz, 3H, H-6), 1.06 (d, *J* = 6.4 Hz, 3H, H-6'). **^{13}C NMR (101 MHz, $CDCl_3$)** δ 161.82 (Ar-C_q), 138.86 (Ar-C_q), 138.09 (Ar-C_q), 136.33 (Ar-*C*), 136.18 (Ar-*C*), 136.10 (Ar-*C*), 135.95 (Ar-*C*), 135.08 (Ar-C_q), 133.68 (Ar-C_q), 133.46 (Ar-C_q), 133.38 (Ar-C_q), 133.13 (Ar-C_q), 129.68 (Ar-*C*), 129.58 (Ar-*C*), 128.65 (Ar-*C*), 128.45 (Ar-*C*), 128.42 (Ar-*C*), 128.38 (Ar-*C*), 128.07 (Ar-*C*), 127.94 (Ar-*C*), 127.82 (Ar-*C*), 127.63 (Ar-*C*), 127.51 (Ar-*C*), 127.29 (Ar-*C*), 127.28 (Ar-*C*), 126.48 (Ar-*C*), 126.34 (Ar-*C*), 126.17 (Ar-*C*), 125.66 (Ar-*C*), 99.52 (C-1'), 94.97 (C-1), 79.19 (C-4'), 78.48 (C-3), 78.44 (C-3'), 75.81 (C-4), 75.11 (Ar-*C*H_2), 75.01 (Ar-*C*H_2), 72.32 (Ar-*C*H_2), 70.60 (C-5'), 67.68 (C-5), 60.37 (C-2'), 57.45 (C-2), 27.12((*C*H_3)$_3$), 16.88(C-6), 16.76(C-6'). **HRMS**: $[M+Na]^+$ calculated for $C_{55}H_{59}Cl_3N_4O_8SiNa$: 1059.30654; found 1059.30600

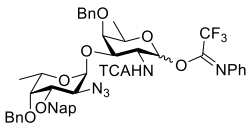
2-azido-4-*O*-benzyl-2-deoxy-3-*O*-(2-naphthylmethyl)-α-L-fucopyranosyl-(1→3)-2-deoxy-2-*N*-trichloroacetamide-4-*O*-benzyl-α/β-D-fucopyranose (37)



36 (196 mg, 0.189 mmol) was dissolved in THF (1.9 mL, 0.1 M) and cooled to 0 °C. AcOH (16 µL, mmol, 1.5 equiv.) and TBAF (1 M in THF; 0.3 mL, mmol, 1.5 equiv.) were added and the reaction was stirred overnight at rt under N_2 until TLC (pentane/EtOAc, 7:3) showed full conversion. The reaction was quenched with NH_4Cl (sat. aq.), diluted in EtOAc, washed with H_2O (x3) and brine (x1), dried over Na_2SO_4, filtered and concentrated *in vacuo*. Column chromatography (pentane/EtOAc, 80:20 → 50:50) furnished hemiacetal **37** in 96% (145 mg, 0.181 mmol). **^{1}H NMR (400 MHz, $CDCl_3$)** δ 7.35 – 7.28 (m, 8H, Ar-*H*), 7.24 – 7.19 (m, 2H, Ar-*H*), 7.15 – 7.11 (m, 2H, Ar-*H*), 6.86 – 6.77 (m, 2H, Ar-*H*), 5.47 (t, *J* = 5.3 Hz, 1H, H-1), 5.07 (d, *J* = 12.2 Hz, 1H, Ar-CH_2), 4.96 (d, *J* = 12.2 Hz, 1H, Ar-CH_2), 4.58 – 4.49 (m, 5H, H-5, Ar-CH_2), 4.13 (dd, *J* = 6.2, 5.2 Hz, 1H, H-4), 3.92 (dd, *J* = 6.2, 3.1 Hz, 1H, H-3), 3.80 (s, 3H, Ar-CH_3), 3.73 (dd, *J* = 5.6, 3.1 Hz, 1H, H-2). **^{13}C NMR (101 MHz, $CDCl_3$)** δ

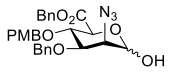
169.33 (*C*=O), 159.51 (Ar-C_q), 137.33 (Ar-C_q), 135.16 (Ar-C_q), 129.76 (Ar-*C*), 129.49 (Ar-C_q), 128.76 (Ar-*C*), 128.72 (Ar-*C*), 128.67 (Ar-*C*), 128.59 (Ar-*C*), 128.55 (Ar-*C*), 128.08 (Ar-*C*), 128.06 (Ar-*C*), 127.94 (Ar-*C*), 113.96 (Ar-*C*), 92.18 (C-1), 77.26 (C-3), 74.48 (C-4), 72.90 (Ar-*C*H_2), 72.73 (C-5), 67.38 (Ar-*C*H_2), 60.97 (C-2), 55.39 (PMB-*C*H_3). **HRMS**: $[M+Na]^+$ calculated for $C_{39}H_{41}Cl_3N_4O_8Na$: 821.18877; found 821.18822

2-azido-4-*O*-benzyl-2-deoxy-3-*O*-(2-naphthylmethyl)-α-L-fucopyranosyl-(1→3)-2-deoxy-2-*N*-trichloroacetamide-4-*O*-benzyl-1-*O*-(*N*-phenyl-2,2,2-trifluoroacetimidoyl)-α/β-D-fucopyranose (38)



37 (183 mg, 0.228 mmol) was co-evaporated with toluene (x3) and dissolved in dry acetone (1.2 mL, 0.2 M). K_2CO_3 (63 mg, 0.456 mmol, 2 equiv.) and ClC(=NPh)CF_3 (0.075 mL, 0.456 mmol, 2 equiv.) were added and the reaction was stirred overnight under N_2 at rt until TLC (pentane/EtOAc, 7:3) showed full conversion. The mixture was filtered and concentrated *in vacuo*. Column chromatography (pentane/EtOAc, 90:10 → 70:30) furnished imidate donor **38** in 89% (186 mg, 0.203 mmol). **^{1}H NMR (400 MHz, CD_3CN)** δ 7.94 – 7.67 (m, 7H), 7.64 – 7.40 (m, 6H), 7.42 – 7.19 (m, 16H), 7.12 (t, *J* = 7.5 Hz, 1H), 6.81 (d, *J* = 7.6 Hz, 2H), 6.56 – 6.17 (m, 1H), 5.23 (s, 1H), 4.98 – 4.85 (m, 3H), 4.85 – 4.78 (m, 1H), 4.74 (d, *J* = 11.6 Hz, 1H), 4.68 (d, *J* = 2.8 Hz, 1H), 4.64 (d, *J* = 11.1 Hz, 2H), 4.52 (d, *J* = 8.7 Hz, 1H), 4.27 (t, *J* = 7.3 Hz, 1H), 4.17 – 4.00 (m, 3H), 4.00 – 3.88 (m, 4H), 1.18 (s, 17H). **^{13}C NMR (101 MHz, CD_3CN)** δ 163.97, 139.76, 139.69, 136.61, 129.84, 129.33, 129.24, 129.18, 129.12, 129.05, 129.01, 128.97, 128.93, 128.87, 128.71, 128.68, 128.62, 128.56, 127.58, 127.33, 127.20, 127.16, 127.02, 126.89, 100.41, 98.47, 79.22, 78.53, 78.20, 77.39, 77.05, 76.37, 76.29, 75.89, 75.57, 72.27, 72.10, 71.66, 70.77, 69.28, 68.22, 66.88, 61.26, 55.13, 52.77, 29.26, 17.14, 17.06, 16.85. **HRMS**: $[M+Na]^+$ calculated for $C_{47}H_{45}Cl_3F_3N_3O_8Na$: 992.21835; found 992.21808

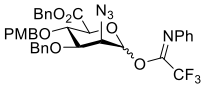
Benzyl (2-azido-2-deoxy-3-*O*-benzyl-4-*O*-*p*-methoxybenzyl)-α/β-D-mannopyranosiduronate (39)



10 (249 mg, 0.407 mmol) was co-evaporated with toluene (x3) and dissolved in dry DCM (4 mL, 0.1 M) and cooled to 0 °C. NIS (137 mg, 0.610 mmol, 1.5 equiv.) and TFA (0.03 mL, 0.407 mmol, 1 equiv.) were added and the reaction stirred at 0 °C under N_2 for 1 h until TLC (pentane/EtOAc, 75:25) showed full conversion. The reaction was quenched with Et_3N and added $NaHCO_3$ (sat. aq.) and stirred vigorously. The mixture was diluted in EtOAc and washed with $Na_2S_2O_3$ (sat. aq.; x1), $NaHCO_3$ (sat. aq.; x1) and brine (x1), dried over Na_2SO_4, filtered and concentrated *in vacuo*. Column chromatography (pentane/EtOAc, 80:20 → 65:35) furnished hemiacetal **39** in 91% (192 mg, 03369 mmol). **^{1}H NMR (400 MHz, $CDCl_3$)** δ 7.35 – 7.28 (m, 8H, Ar-*H*), 7.24 – 7.19 (m, 2H, Ar-*H*), 7.15 – 7.11 (m, 2H, Ar-*H*), 6.86 – 6.77 (m, 2H, Ar-*H*), 5.47 (t, *J* = 5.3 Hz, 1H, H-1), 5.07 (d, *J* = 12.2 Hz, 1H, Ar-CH_2), 4.96 (d, *J* = 12.2 Hz, 1H, Ar-CH_2), 4.58 – 4.49 (m, 5H, Ar-CH_2, H-5), 4.13 (dd, *J* = 6.2, 5.2 Hz, 1H, H-4), 3.92 (dd, *J* = 6.2, 3.1 Hz, 1H, H-3), 3.80 (m, 3H, CH_3-PMB), 3.73 (dd, *J* = 5.6, 3.1 Hz, 1H, H-2). **^{13}C NMR (101 MHz, $CDCl_3$)** δ 169.33 (*C*=O), 159.51 (Ar-C_q), 137.33 (Ar-C_q), 135.16 (Ar-C_q), 129.76 (Ar-*C*), 129.49 (Ar-C_q), 128.76 (Ar-*C*), 128.72 (Ar-*C*), 128.67 (Ar-*C*), 128.59 (Ar-*C*), 128.55 (Ar-*C*), 128.08 (Ar-*C*), 128.06 (Ar-*C*), 127.94 (Ar-*C*), 113.96 (Ar-*C*), 92.18 (C-1), 77.26 (C-3), 74.48 (C-4), 73.19 (Ar-*C*H_2), 72.90

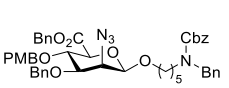
(Ar-*C*H$_2$), 72.73 (C-5), 67.38 (Ar-*C*H$_2$), 60.97 (C-2), 55.39 (*C*H$_3$-PMB). **HRMS**: [M+Na]$^+$ calculated for $C_{28}H_{29}N_3O_7Na$: 542.19032; found 543.18942

Benzyl (2-azido-2-deoxy-3-*O*-benzyl-4-*O*-*p*-methoxybenzyl-1-*O*-(*N*-phenyl-2,2,2-trifluoroacetimidoyl))-α/β-D-mannopyranosiduronate (40)



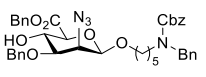
39 (291 mg, 0.559 mmol) was co-evaporated with toluene (x3) and dissolved in dry acetone (2.8 mL, 0.2 M). K_2CO_3 (155 mg, 1.12 mmol, 2 equiv.) and ClC(=NPh)CF_3 (0.18 mL, 1.12 mmol, 2 equiv.) were added to the reaction and it was stirred overnight under N_2 at rt until TLC (pentane/EtOAc, 7:3) showed full conversion. The mixture was filtered and concentrated *in vacuo*. Column chromatography (pentane/EtOAc, 90:10 → 70:30) furnished imidate donor **40** in 87% (334 mg, 0.484 mmol**). ^{1}H NMR (400 MHz, CD$_3$CN)** δ 7.40 – 7.25 (m, 18H), 7.21 – 7.11 (m, 4H), 6.88 – 6.80 (m, 5H), 5.19 – 5.09 (m, 1H), 5.09 – 5.00 (m, 2H), 4.69 – 4.59 (m, 4H), 4.50 (d, *J* = 10.9 Hz, 1H), 4.35 (m, 2H), 4.14 – 4.03 (m, 3H), 3.78 (s, 3H), 3.76 (s, 1H). **^{13}C NMR (101 MHz, CD$_3$CN)** δ 130.71, 129.84, 129.53, 129.45, 129.37, 129.33, 129.29, 129.11, 128.95, 114.60, 78.09, 74.84, 74.78, 73.35, 68.02, 59.99, 55.82, 29.65. **HRMS**: [M+Na]$^+$ calculated for $C_{36}H_{33}F_3N_4O_7Na$: 713.21990; found 713.21935

5-(Benzyl(benzyloxycarbonyl)amino)pentyl benzyl(2-azido-2-deoxy-4-*O*-*p*-methoxybenzyl-3-*O*-benzyl)-β-D-mannopyranosiduronate (41)



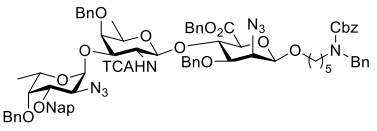
Donor **40** (244 mg, 0.353 mmol, 1 equiv.) and acceptor **12** (150 mg, 0.459 mmol, 1.3 equiv.) were co-evaporated with toluene (x3), dissolved in dry DCM (2.4 mL, 0.1 M), added activated 3Å molecular sieves and stirred for 30 min under argon at rt. The reaction mixture was cooled to -78 °C, followed by addition of TBSOTf (16 µL, 0.0706 mmol, 0.2 equiv.). The mixture was allowed to warm to -30 °C and stirred for 1 h until TLC (pentane/EtOAc, 8:2) showed full conversion. The reaction was quenched with Et_3N and diluted in EtOAc, washed with NaHCO3 (sat. aq.; x1) and brine (x1), dried over Na_2SO_4, filtered and concentrated *in vacuo*. Column chromatography (pentane/EtOAc, 85:15 → 70:30) yielded **41** in 69% (α: 42 mg, 0.0511 mmol; β: 157 mg, 0.192 mmol) in a α/β = 21:79. **^{1}H NMR (400 MHz, CDCl$_3$)** δ 7.41 – 7.27 (m, 20H), 7.06 – 7.02 (m, 2H, Ar-*H*), 6.81 – 6.76 (m, 2H, Ar-*H*), 5.28 – 5.14 (m, 4H, Ar-C*H*$_2$), 4.77 – 4.64 (m, 3H, Ar-C*H*$_2$), 4.50 (d, *J* = 6.9 Hz, 3H, Ar-C*H*$_2$, H-1), 4.40 (d, *J* = 10.2 Hz, 1H, Ar-C*H*$_2$), 4.05 (t, *J* = 9.3 Hz, 1H, H-4), 3.95 – 3.86 (m, 2H, H-3, H-2), 3.86 – 3.81 (m, 1H, C*H*$_2$-Linker), 3.78 (s, 4H, C*H*$_3$-PMB), 3.65 – 3.59 (m, 1H, H-5), 3.45 – 3.15 (m, 3H, C*H*$_2$-Linker), 1.59 – 1.49 (m, 3H, C*H*$_2$-Linker), 1.37 – 1.27 (m, 3H, C*H*$_2$-Linker). **^{13}C NMR (101 MHz, CDCl$_3$)** δ 167.93 (C=O), 159.39 (C=O), 138.02 (Ar-*C*$_q$), 137.49 (Ar-*C*$_q$), 135.27 (Ar-*C*$_q$), 129.78 (Ar-*C*), 128.73 (Ar-*C*), 128.69 (Ar-*C*), 128.65 (Ar-*C*), 128.58 (Ar-*C*), 128.55 (Ar-*C*), 128.21 (Ar-*C*), 128.02 (Ar-*C*), 127.96 (Ar-*C*), 127.94 (Ar-*C*), 127.37 (Ar-*C*), 113.83 (Ar-*C*), 100.31 (C-1), 79.99 (C-5), 75.37 (C-4), 74.70 (Ar-*C*H$_2$), 72.37 (Ar-*C*H$_2$), 70.06 (*C*H$_2$-Linker), 67.41 (Ar-*C*H$_2$), 61.64 (C-3), 55.38 (C-2), 50.60 (*C*H$_2$-Linker), 29.81 (*C*H$_2$-Linker), 29.18 (*C*H$_2$-Linker), 23.17 (*C*H$_2$-Linker). **HRMS**: [M+Na]$^+$ calculated for $C_{48}H_{52}N_4O_9Na$: 851.36320; found 851.36265

5-(Benzyl(benzyloxycarbonyl)amino)pentyl benzyl(2-azido-2-deoxy-3-*O*-benzyl)-β-D-mannopyranosiduronate (42)



41 (121 mg, 0.148 mmol) was dissolved in DCM/H_2O (20:1, 1.5 mL, 0.1 M) and added DDQ (67 mg, 0.296 mmol, 2 equiv.) and stirred at rt under N_2 for 2 h until TLC (pentane/EtOAc, 7:3) showed full conversion. The solution was quenched with $Na_2S_2O_3$ (sat. aq.) and diluted/extracted with EtOAc (x3). The combined organic phases were washed with $NaHCO_3$ (sat. aq.; x4) and brine (x1), dried over Na_2SO_4, filtrated and concentrated *in vacuo*. Column chromatography (pentane/EtOAc, 80:20 → 50:50) yielded **42** in 92% (96 mg, 0.136 mmol). **^{1}H NMR (400 MHz, $CDCl_3$)** δ 7.42 – 7.27 (m, 17H, Ar-*H*), 7.20 (m, 2H, Ar-*H*), 5.23 (d, *J* = 8.1 Hz, 2H, Ar-CH_2), 5.17 (d, *J* = 11.2 Hz, 2H, Ar-CH_2), 4.76 (d, *J* = 2.0 Hz, 2H, Ar-CH_2), 4.51 – 4.41 (m, 3H, H-1, Ar-CH_2), 4.14 (td, *J* = 9.4, 2.6 Hz, 1H, H-4), 3.95 – 3.79 (m, 2H, H-2, CH_2-Linker), 3.79 – 3.69 (m, 1H, H-5), 3.53 – 3.32 (m, 2H, H-3, CH_2-Linker), 3.30 – 3.11 (m, 2H, CH_2-Linker), 2.92 (dt, *J* = 2.6, 1.1 Hz, 1H, OH), 1.64 – 1.44 (m, 3H, CH_2-Linker), 1.40 – 1.19 (m, 3H, CH_2-Linker). **^{13}C NMR (101 MHz, $CDCl_3$)** δ 138.02 (Ar-C_q), 137.50 (Ar-C_q), 128.80 (Ar-*C*), 128.75 (Ar-*C*), 128.67 (Ar-*C*), 128.41 (Ar-*C*), 128.30 (Ar-*C*), 128.04 (Ar-*C*), 127.95 (Ar-*C*), 127.39 (Ar-*C*), 101.82 (C-1), 79.82 (C-3), 75.60 (C-5), 72.15 (Ar-*C*H_2), 70.12 (*C*H_2-Linker), 68.36 (C-4), 67.54 (Ar-*C*H_2), 67.28 (Ar-*C*H_2), 61.56 (C-2), 50.70 (*C*H_2-Linker), 29.22 (*C*H_2-Linker), 23.20 (*C*H_2-Linker). **HRMS**: $[M+Na]^+$ calculated for $C_{40}H_{44}N_4O_8Na$: 731.30568; found 731.30514

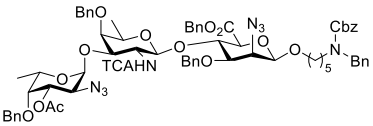
5-(Benzyl(benzyloxycarbonyl)amino)pentyl 2-azido-4-*O*-benzyl-2-deoxy-3-*O*-(2-naphthylmethyl)-α-L-fucopyranosyl-(1→3)-2-deoxy-2-*N*-trichloroacetamide-4-*O*-benzyl-β-D-fucopyranosyl-(1→4) benzyl(2-azido-2-deoxy-3-*O*-benzyl)-β-D-mannopyranosiduronate (43)



Acceptor **38** (88 mg, 0.125 mmol, 1 equiv.) and donor **42** (186 mg, 0.203 mmol, 1.6 equiv.) were co-evaporated with toluene (x3), dissolved in dry DCM (1.2 mL, 0.1 M), added activated 3Å molecular sieves and stirred for 30 min under argon at rt. The mixture was cooled to -78 °C, after which TBSOTf (6 µL, 0.0244 mmol, 0.2 equiv.) was added. The reaction mixture was stirred for 1 h until TLC (pentane/EtOAc, 7:3) showed full conversion. The reaction mixture was quenched with Et_3N, diluted in EtOAc, washed with $NaHCO_3$ (sat. aq.; x1) and brine (x1), dried over Na_2SO_4, filtered and concentrated *in vacuo*. Column chromatography (pentane/EtOAc, 75:25 → 65:35) furnished **43** in 81% yield (153 mg, 0.101 mmol). **^{1}H NMR (400 MHz, $CDCl_3$)** δ 7.88 – 7.71 (m, 5H, Ar-*H*), 7.53 – 7.43 (m, 4H, Ar-*H*), 7.43 – 7.26 (m, 31H, Ar-*H*), 7.25 – 7.12 (m, 6H, Ar-*H*), 6.93 (d, *J* = 8.5 Hz, 1H, N*H*), 5.18 (d, *J* = 10.7 Hz, 2H, Ar-CH_2), 4.96 (d, *J* = 3.7 Hz, 1H, H-1''), 4.95 – 4.89 (m, 2H, Ar-CH_2), 4.88 – 4.76 (m, 5H, Ar-CH_2), 4.72 – 4.63 (m, 3H, Ar-CH_2), 4.60 (dd, *J* = 11.5, 4.6 Hz, 2H, Ar-CH_2), 4.49 (d, *J* = 8.4 Hz, 4H, CH_2-Linker, H-1', H-1), 4.28 (t, *J* = 8.2 Hz, 1H, H-4), 4.20 (dt, *J* = 11.0, 8.4 Hz, 1H, H-2'), 4.04 (dd, *J* = 10.6, 3.6 Hz, 1H, H-2''), 3.93 – 3.90 (m, 1H, H-5''), 3.89 (d, *J* = 2.1 Hz, 1H, H-3''), 3.85 (d, *J* = 8.3 Hz, 1H, H-5), 3.82 – 3.71 (m, 3H, H-2), 3.69 – 3.57 (m, 3H, H-3', H-4'', H-3), 3.50 – 3.39 (m, 2H, H-4', CH_2-Linker), 3.34 (d, *J* = 7.2 Hz, 1H, H-5'), 3.31 – 3.14 (m, 3H, CH_2-Linker), 1.62 – 1.46 (m, 2H, CH_2-Linker), 1.39 – 1.21 (m, 6H, CH_2-Linker), 1.17 (d, *J* = 6.3 Hz, 3H, H-6''), 1.10 (d, *J* = 6.5 Hz, 3H, H-6'). **^{13}C NMR (101 MHz, $CDCl_3$)** δ 168.82 (C=O), 161.94 (C=O), 138.67 (Ar-C_q), 138.28 (Ar-C_q), 138.14 (Ar-C_q), 138.03 (Ar-C_q),

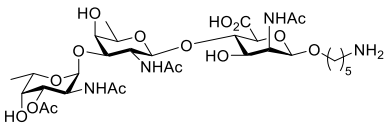
135.35 (Ar-C_q), 135.10 (Ar-C_q), 133.37 (Ar-C_q), 133.13 (Ar-C_q), 128.96 (Ar-*C*), 128.77 (Ar-*C*), 128.73 (Ar-*C*), 128.68 (Ar-*C*), 128.63 (Ar-*C*), 128.49 (Ar-*C*), 128.43 (Ar-*C*), 128.39 (Ar-*C*), 128.31 (Ar-*C*), 128.29 (Ar-*C*), 128.09 (Ar-*C*), 128.02 (Ar-*C*), 127.95 (Ar-*C*), 127.93 (Ar-*C*), 127.81 (Ar-*C*), 127.77 (Ar-*C*), 127.72 (Ar-*C*), 127.63 (Ar-*C*), 127.53 (Ar-*C*), 127.34 (Ar-*C*), 126.57 (Ar-*C*), 126.45 (Ar-*C*), 126.32 (Ar-*C*), 126.16 (Ar-*C*), 125.73 (Ar-*C*), 125.57 (Ar-*C*), 99.84 (C-1', C-1), 99.69 (C-1''), 79.60 (C-3'), 78.66 (C-5''), 78.42 (C-4'), 75.98 (C-3), 75.69 (C-4''), 75.08 (Ar-*C*H_2), 74.45 (C-4, C-5), 73.43 (Ar-*C*H_2), 72.50 (Ar-*C*H_2), 70.85 (C-5'), 70.04 (C-3''), 67.86 (Ar-*C*H_2), 67.50 (Ar-*C*H_2), 67.24 (Ar-*C*H_2), 6-.96 (C-2), 60.32 (C-2''), 54.45 (C-2'), 50.59 (CH_2-Linker), 50.27 (CH_2-Linker), 46.95 (CH_2-Linker), 29.82 (CH_2-Linker), 29.15 (CH_2-Linker), 23.19 (CH_2-Linker), 16.99 (C-6', C-6''), 16.91 (C-6', C-6''). **HRMS**: $[M+Na]^+$ calculated for $C_{79}H_{83}Cl_3N_8O_{15}Na$: 1511.49412; found 1511.49357

5-(Benzyl(benzyloxycarbonyl)amino)pentyl 3-*O*-Acetyl-2-azido-4-*O*-benzyl-2-deoxy-α-L-fucopyranosyl-(1→3)-2-deoxy-2-*N*-trichloroacetamide-4-*O*-benzyl-β-D-fucopyranosyl-(1→4) benzyl(2-azido-2-deoxy-3-*O*-benzyl)-β-D-mannopyranosiduronate (44)



43 (143 mg, 0.095 mmol) was dissolved in DCM/H_2O (4:1, 2 mL, 0.05 M) and added DDQ (43 mg, 0.19 mmol, 2 equiv.). The reaction was stirred at rt under N_2 for 3 h until TLC (pentane, EtOAc, 7:3) showed full conversion. The solution was quenched with $Na_2S_2O_3$ (sat. aq.) and extracted with EtOAc (x3). The combined organic phases were washed with $NaHCO_3$ (sat. aq.; x4) and brine (x1), dried over Na_2SO_4, filtrated and concentrated *in vacuo*. The crude was used without further purification. The residue was dissolved in pyridine (2 mL) and cooled to 0 °C and added Ac_2O (0.3 mL) and DMAP (catalytic amount) and stirred at rt under N_2 overnight until TLC (pentane/EtOAc, 7:3) showed full conversion. The mixture was dissolved in EtOAc, washed with 1 M HCl (x1), $NaHCO_3$ (sat. aq.; x1) and brine (x1), dried over Na_2SO_4 and concentrated *in vacuo*. Column chromatography (pentane/EtOAc, 8:2 → 6:4) yielded **44** in 92% yield (123 mg, 0.087 mmol). **^{1}H NMR (400 MHz, $CDCl_3$)** δ 7.44 – 7.27 (m, 36H), 7.25 – 7.10 (m, 8H), 6.88 (d, *J* = 8.3 Hz, 1H), 5.25 (d, *J* = 12.2 Hz, 1H), 5.23 – 5.12 (m, 5H), 5.13 – 5.00 (m, 2H), 4.98 (d, *J* = 3.7 Hz, 1H), 4.90 – 4.83 (m, 3H), 4.71 – 4.63 (m, 4H), 4.62 – 4.55 (m, 2H), 4.56 – 4.53 (m, 2H), 4.52 – 4.45 (m, 4H), 4.31 – 4.23 (m, 1H), 4.16 – 4.04 (m, 2H), 3.98 – 3.88 (m, 3H), 3.88 – 3.80 (m, 3H), 3.79 – 3.70 (m, 3H), 3.62 (dd, *J* = 7.9, 3.6 Hz, 2H), 3.51 – 3.38 (m, 2H), 3.33 – 3.12 (m, 4H), 2.06 (d, *J* = 6.3 Hz, 4H), 1.39 – 1.19 (m, 10H), 1.19 (d, *J* = 6.3 Hz, 4H), 1.11 (d, *J* = 6.5 Hz, 5H). **^{13}C NMR (101 MHz, $CDCl_3$)** δ 170.27, 161.92, 138.39, 138.21, 138.03, 137.62, 135.38, 128.91, 128.76, 128.72, 128.67, 128.64, 128.61, 128.56, 128.51, 128.40, 128.34, 128.32, 128.24, 128.02, 127.94, 127.68, 127.58, 127.36, 99.85, 99.51, 78.84, 78.28, 75.82, 74.94, 74.37, 73.28, 72.28, 70.94, 70.05, 67.47, 67.32, 67.25, 58.18, 54.78, 50.52, 50.07, 46.82, 46.26, 29.15, 23.19, 20.98, 17.11, 16.59. **HRMS**: $[M+Na]^+$ calculated for $C_{70}H_{77}Cl_3N_8O_{16}Na$: 1413.44308; found 1413.48622

5-aminopentyl 3-*O*-Acetyl-2-acetamide-deoxy-α-L-fucopyranosyl-(1→3)-2-deoxy-2-acetamide-β-D-fucopyranosyl-(1→4) 2-acetamide-2-deoxy-β-D-mannopyranosiduronate (CP5-III)



44 (36 mg, mmol) was dissolved in THF (distilled, 3 mL) and added zinc powder (500 mg, 7.69 mmol, 300 equiv.), AcOH (1 mL) and Ac_2O (0.5 mL). The resulting mixture was stirred at 50 °C overnight until TLC (DCM/MeOH, 95:5) showed full conversion. The cooled mixture was filtered through Celite, evaporated *in vacuo* and co-evaporated with toluene (x3). The crude product was first purified by column chromatography (DCM/MeOH, 98:2 → 90:10) followed by HPLC given the acetamide product in 35 % yield (12 mg, 0.0091 mmol). SM (12 mg, 0.0091 mmol) was dissolved in *t*-BuOH (2.5 mL) and added AcOH (1 mL, 0.1 mL in 100 mL MilliQ). The solution was birched with argon for 20 min and added $Pd(OH)_2/C$ (catalytic amount). The reaction was again birched with argon for 5 minutes before the atmosphere was changed for H_2. The mixture was stirred for 3 days under H_2 atmosphere until NMR showed full conversion, after which it was burched with argon for 20 min, filtered over a Whatman filter and lyophilized. Purification by a HW40 column with NH_4OAc followed by lyophilization gave **CP5-III** in 57% yield (4.3 mg, 0.0058 mmol). However migration of the 3-*O*-acetyl on the L-Fuc residue to the C-4 of the L-Fuc was observed. NMR reported for the migrated product. **^{1}H NMR (600 MHz, D_2O)** δ 5.25 – 5.23 (m, 0H), 5.17 (dd, *J* = 11.4, 3.1 Hz, 0H), 5.02 (dd, *J* = 7.8, 3.8 Hz, 1H), 4.96 (d, *J* = 4.0 Hz, 0H), 4.75 (s, 2H), 4.49 (dd, *J* = 4.3, 1.4 Hz, 1H), 4.45 – 4.35 (m, 1H), 4.25 (q, *J* = 6.7 Hz, 1H), 4.20 – 4.13 (m, 1H), 4.13 – 4.07 (m, 1H), 4.00 – 3.90 (m, 2H), 3.88 – 3.73 (m, 7H), 3.69 – 3.62 (m, 2H), 2.99 – 2.95 (m, 2H), 2.22 (s, 2H), 2.09 (s, 1H), 2.06 (s, 3H), 2.03 (s, 2H), 2.02 – 1.97 (m, 4H), 1.90 (s, 1H), 1.69 – 1.56 (m, 4H), 1.44 – 1.35 (m, 2H), 1.29 – 1.24 (m, 3H), 1.21 (dd, *J* = 6.6, 4.5 Hz, 2H), 1.12 (d, *J* = 6.6 Hz, 2H). **^{13}C NMR (151 MHz, D_2O)** δ 176.12, 175.52, 175.45, 175.17, 175.12, 175.07, 174.84, 174.11, 102.03, 100.31, 99.99, 79.28, 79.19, 78.38, 78.31, 78.29, 77.86, 77.71, 77.54, 74.34, 71.94, 71.88, 71.67, 71.29, 71.23, 70.72, 69.70, 68.51, 68.05, 67.77, 66.98, 66.87, 54.61, 53.02, 52.12, 50.76, 50.40, 48.04, 40.30, 28.87, 27.22, 23.43, 23.39, 23.15, 23.09, 22.97, 22.95, 22.94, 21.12, 16.29, 16.22, 16.16, 16.13. **HRMS**: $[M+H]^+$ calculated for $C_{31}H_{52}N_4O_{16}H$: 737.34566; found 737.34511

Supporting information

SPR experiments

The SPR experiments were conducted using a Surface Plasmon Resonance Biacore X100 from GE Healthcare Biacore Life Science. CP5-biotin (CP5-biotin, lot EB23GIU16, M=351.6 µg/mL) was immobilized on a SA-chip using a 20 µg/mL solution. After the run 311.7 AU was immobilized on the chip. For the SPR-experiments a 20 nM rat anti-CP5 mAb 331 concentration was used together with the CP5-OS concentrations as summarized in Table S1. From the Biacore X100 control software the binding levels were collected and used for calculation of the inhibition percentage.

Competitor concentrations (µg/mL)	CP5-I	CP5-II	Hexasaccharide	Nonasaccharide	CP5-PS
	1000	50	20	5	0.0781
	500	25	10	2.5	0.0391
	250	12.5	5	1.25	0.0195
	125	6.25	2.5	0.625	0.00977
	62.5	3.125	1.25	0.313	0.00488
	31.25	1.563	0.625	0.156	0.00244
	15.63	0.781	0.313	0.0781	0.00122
	7.81	0.391	0.156	0.0391	0.000610
	3.91	0.195	0.0781	0.0195	0.000305
	1.95	0.0977	0.0391	0.00977	0.000153
	0.977	0.0488	0.0195	0.00488	7.629E-05
	0.488	0.0244	0.00977	0.00244	3.815E-05
	0	0	0	0	0

Table S1: CP5-OS concentrations for the SPR-experiments

SPR IC50 values

The calculation of IC50 values were performed with GraphPad Prism software using Kruskal-Wallis with Dunn's multiple comparisons; "***" denotes the significant result within $p < 0.001$, "ns" means not significant.

Structural studies

Acknowledgements: Luca Unione from CIC BioGune is acknowledged for his contribution to the conformational analysis.

To gain insight into the structure of the trisaccharide that showed best binding, **CP5-II** was investigated using a combination of NMR-methodologies (*J*-couplings and NOE-interactions) and computational protocols (MM). As depicted in Figure 3 and as analyzed by the intra-residual NOE and *J*-couplings, the three pyranosides (C: D-ManNAcA, B: L-FucNAc and A: D-FucNAc) were found to adopt the common chair conformations ($^4C_1$ for the D-ManNAcA and D-FucNAc and $^1C_4$ for L-FucNAc), in line with the structural findings in Chapter 3 for the **CP5-I** trisaccharide. For the conformation around the C-A bond, the strong NOE between the H1(A)-H4(C)/ H6(C) proton pair and the H2(A)-H6(C) proton pair, suggested, together with the predicted MM calculations, a conformational equilibrium between the exo-*syn*-ϕ/*syn*(+)-ψ and the exo-*syn*-ϕ/*syn*(-)-ψ conformations with the exo-*syn*-ϕ/*syn*(+)-ψ being the major populated on. For the C-B glycosidic linkage the strong NOE between the H1(B)-H4(C) proton pair and the

weaker NOE between the H1(B)-H3(C) and H1(B)- H5(C) proton pairs suggested an exo-*syn*-ϕ/*syn*(-)-ψ conformation.

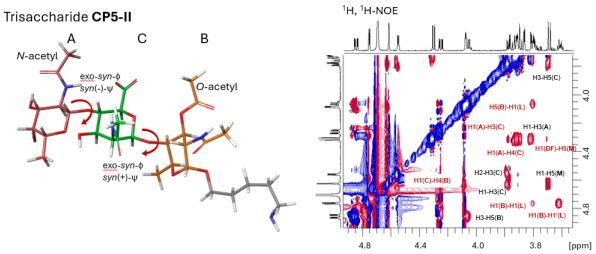


Figure 3: Conformational analysis of **CP5-II** as established by NMR and MM calculations. Zoom area of 2D NOESY spectrum of **CP5-II** and its main conformation as defined by NOE analysis and MM calculations. Monosaccharidic residues are labeled with a letter code. The main conformation at each glycosidic linkage and the spatial orientation of the acetyl are reported.

Structure and conformational studies

NMR methods. NMR experiments were performed in a Bruker Avance III 800 MHz spectrometer equipped with a TCI cryoprobe. Samples were dissolved in D_2O at 1.0 mM concentration. Experiments were acquired at the temperature of 298 K.

^{1}H and ^{13}C NMR resonances of the molecules **CP5-II** were assigned through standard 2D-TOCSY, 2D-ROESY, 2D-NOESY, 2D ^{1}H-^{13}C-HSQC. 2D-TOCSY experiments were acquired with 30 ms mixing time, 1.0 s of relaxation delay, 4 scans, and 4096x256 (F2xF1) points with a spectral width of 6556.0 Hz. 2D-ROESY experiment was acquired with mixing time of 200 ms, 1.0 s of relaxation delay, 48 scans, and 4096x256 (F2xF1) points with a spectral width of 6880.7 Hz. 2D-NOESY experiment was acquired with mixing time of 200 ms, 1.5 s of relaxation delay, 32 scans, and 4096x256 (F2xF1) points with a spectral width of 6242.2 Hz. 2D ^{1}H,^{13}C-HSQC experiments were acquired with 1.0 s of relaxation delay, 48 scans, and 4096x220 (F2xF1) points with a spectral width of 6250.0 Hz (F2) and 24144.6 Hz (F1). The data were processed with Topspin 4.2 (Bruker Biospin) using a 90° shifted qsine window function to a total of 16K × 2K data points (F2 × F1), followed by automated baseline- and phase correction.

Molecular Mechanics Calculations. The geometry optimization was performed by using the Jaguar/Schroedinger package (version 13.5) and the AMBER* force field, with the GB/SA continuum solvent model for water. The glycosidic torsion angles were defined as ϕ (H1'-C1'-Ox-Cx) and ψ (C1'-Ox-Cx-Hx). Extended nonbonded cut-off distances (van der Waals cut-off of 8.0 Å and electrostatic cut-off of 20.0 Å) were used. The conformers for **CP5-II** were generated employing geometric restrictions to respect the *exo*-anomeric effect. The possible staggered rotamers around ψ were selected and minimized. The coordinates of the obtained local minima were employed to measure the key inter-proton distances that were then compared to those obtained experimentally by the ROESY and NOESY NMR experiments through integration of the observed NOEs cross peaks using the ISPA approximation.

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