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Investigation on the Synthesis of *Shigella flexneri* Specific Oligosaccharides Using Disaccharides as Potential Transglucosylase Acceptor Substrates

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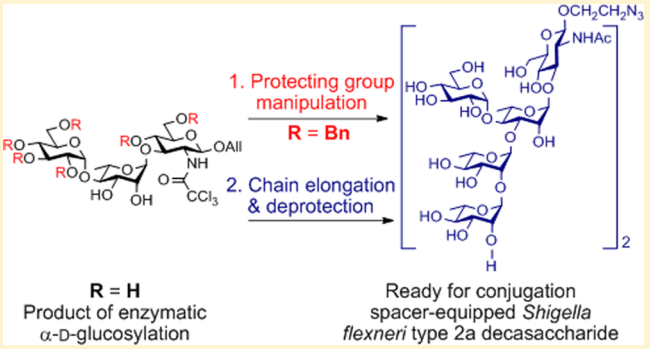
Supporting Information

ABSTRACT: Chemo-enzymatic strategies hold great potential for the development of stereo- and regioselective syntheses of structurally defined bioactive oligosaccharides. Herein, we illustrate the potential of the appropriate combination of a planned chemo-enzymatic pathway and an engineered biocatalyst for the multistep synthesis of an important decasaccharide for vaccine development. We report the stepwise investigation, which led to an efficient chemical conversion of allyl α-D-glucopyranosyl-(1→4)-α-L-rhamnopyranosyl-(1→3)-2-deoxy-2-trichloroacetamido-β-D-glucopyranoside, the product of site-specific enzymatic α-D-glucosylation of a lightly protected non-natural disaccharide acceptor, into a pentasaccharide building block suitable for chain elongation at both ends. Successful differentiation between hydroxyl groups features the selective acylation of primary alcohols and acetalation of a *cis*-vicinal diol, followed by a controlled per-*O*-benzylation step. Moreover, we describe the successful use of the pentasaccharide intermediate in the [5 + 5] synthesis of an aminoethyl aglycon-equipped decasaccharide, corresponding to a dimer of the basic repeating unit from the O-specific polysaccharide of *Shigella flexneri* 2a, a major cause of bacillary dysentery. Four analogues of the disaccharide acceptor were synthesized and evaluated to reach a larger repertoire of *O*-glucosylation patterns encountered among *S. flexneri* type-specific polysaccharides. New insights on the potential and limitations of planned chemo-enzymatic pathways in oligosaccharide synthesis are provided.



INTRODUCTION

The chemical assembly of complex oligosaccharides has progressed tremendously over the past decades.[1−5] Elegant chemical synthetic methods including modular solution-phase approaches[6] or automated stepwise solid-phase syntheses[7] of oligosaccharides encompassing more than 20 residues have been reported. Nevertheless, in the absence of general rules governing synthetic carbohydrate chemistry, the development of stereoselective syntheses of structurally defined oligosaccharides remains a challenging task. Complexity is increased owing to the impressive structural diversity of naturally occurring carbohydrates and, therefore, of synthetic targets. This is especially relevant in the case of glycans of microbial origin,[8] in which interest is growing. Efficient protecting group manipulation toward tailored-made building blocks on the one hand and their stereoselective chemical assembly on the other hand remain major issues, particularly with regard to the development of scalable synthetic processes or when multiple targets are involved. In some instances, enzymes have successfully emerged to circumvent these issues. Advantageously, synthesis using glycosyltransferases proceeds selectively in the absence of protecting groups,[9] while some glycosidases could be converted into efficient glycosynthases[10] or transglycosidases.[11] Evidently, access to naturally occurring carbohydrate-active enzymes is expanding, and in recent years, enzymatic syntheses and, as an extension, chemo-enzymatic pathways, have gained increasing attention owing to their powerful versatility in addition to high regio- and stereoselectivity.[3,4,12−14] Purposely tailored biocatalysts could help bypass specific limitations encountered with chemical glycosylation; we therefore chose to explore a chemo-enzymatic

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route in the context of *Shigella flexneri*, a family of Gram-negative enteroinvasive bacteria and one of the causes of shigellosis, otherwise known as bacillary dysentery, in humans.

With an estimated 122800 shigellosis-related deaths in 2010,[15] shigellosis is one of the diarrheal diseases causing the most burden worldwide, especially among children living in developing countries.[16] Rehydration is inadequate, and resistance to antibiotics is increasing.[17] Above all, the renewed awareness of the burden of shigellosis in the pediatric population has encouraged the search for novel vaccine strategies aimed at broad *Shigella* species and type coverage. As an alternative to important developments involving the detoxified lipopolysaccharide and/or analogues purified from biological sources,[18] we and others chose to investigate the impact of synthetic carbohydrates as active vaccine components.[19−21] Thus, synthetic oligosaccharides representing functional mimics of the O-specific polysaccharide moiety (O-SP) of the bacterial membrane-anchored lipopolysaccharides are meant to replace the natural antigens.[22] The efficient synthesis of tailored fragments of the *Shigella* O-SPs of interest is a prerequisite to the identification of oligosaccharides suitable for entering vaccine design and optimization.

S. flexneri encompasses a large diversity of types and subtypes which have been identified on the basis of the structure of their O-SPs. At least 15 of the identified O-SP repeating units share the same linear tetrasaccharide backbone, which is made of three L-rhamnose residues (A, B, C) and an *N*-acetyl-D-glucosamine residue (D), 1,2-*trans*-linked to one another. Type specificity is associated with the phage-encoded site-selective modification of the ABCD unit with α-D-glucopyranosyl residues (E), *O*-acetyl groups (Ac), and the more recently disclosed phosphoethanolamine residues (PEtN) (Figure 1).[23,24]

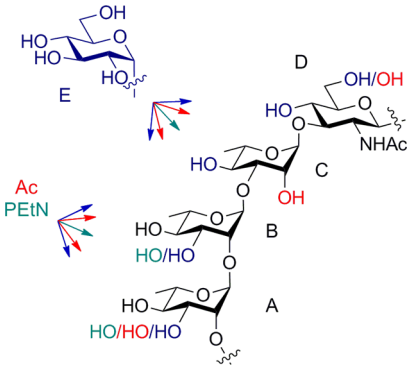

Figure 1. *S. flexneri* O-SP backbone (ABCD) showing type/group-specific substitution and sites of α-D-glucosyl (E) appendage (in blue).

α-D-Glucosylation can occur at any residue. It is always stoichiometric and is an essential feature of the most prevalent *S. flexneri* serotypes sharing the $\{ABCD\}_n$ backbone.[23,25] Concern for these important glucosyl side chains has been addressed repeatedly in the published chemical syntheses of *S. flexneri* oligosaccharides, most of which used readily available tetrabenzylglucosyl donors.[26−31] However, the 1,2-*cis* α-linkage has precluded stereospecific glucosylation, and α/β mixtures were isolated despite parameter optimization. Evolution from tetrabenzyl donors to those more diversely protected at remote positions such as analogues equipped with ester protecting groups at O-6[32] and conformationally constrained precursors[33−35] has been shown to enhance α/β ratios. Alternatively, prearranged donor/acceptor systems for intramolecular aglycon delivery,[36,37] mannose-to-glucose sequential oxidation−reduction,[38] and precursors designed for hydrogen bond mediated aglycon delivery[39] have been employed successfully. Furthermore, impressive progress in the area of stereocontrolled chemical 1,2-*cis* glucosylation has been achieved as exemplified by sophisticated donors enabling neighboring group participation by means of a C-2 (*S*)-auxiliary[40] or prepared in the form of precyclized 1,2-oxathiane precursors.[41−43] Nevertheless, despite remarkable realizations, concerns emerged due to increasing donor complexity and/or potential interferences with protecting group selection, branching patterns, subsequent chain elongation, substituent manipulation, or final deprotection. For systems such as *S. flexneri*, for which diversely substituted O-SPs and fragments thereof are identified as synthetic targets, these inherent drawbacks have to be evaluated on a case-to-case basis. Instead, we chose to investigate the input of enzymatic glucosylation.

Shigella type-specific O-SP glucosylation takes place on the periplasmic side of the cytoplasmic membrane on the growing membrane-anchored O-SP chain. Glucosylation involves serotype-specific glucosyltransferases featuring several transmembrane domains and a common membrane-anchored undecaprenyl-phosphate-β-glucose donor.[44] With this in mind, we chose to investigate novel chemo-enzymatic routes to diverse α-D-glucosylated oligosaccharides by use of sucrose-utilizing transglucosylases. Otherwise known as glucansucrases, the selected biocatalysts are α-retaining enzymes found in glycoside-hydrolase families 13 (GH13) and 70 (GH70) of the CAZy classification.[45] Owing to a remarkable plasticity, they have evolved into biocatalysts with novel substrate specificities.[46] Moreover, relying on computer-aided engineering, we have identified mutants of the amylosucrase from *Neisseria polysaccharea* (*Np*AS), which catalyze the regiospecific α-D-glucosylation of two non-natural monosaccharides into motifs of interest in the context of *S. flexneri*.[47,48] Featuring a related strategy, Armand et al. have shown that *Bacillus circulans* 251 cyclodextrin glucanotransferase (CGTase, EC 2.1.4.19) was able to accommodate non-natural rhamnoside acceptors and produce α-D-glucosylated products, identified as relevant building blocks for the synthesis of various *S. flexneri* O-SP fragments.[49]

As seen from our previous achievements,[47,49] the strategy that we have undertaken aims at developing target-oriented synthetic routes encompassing an early-stage enzymatic step. Therefore, acceptor substrates are fine-tuned in order to access a variety of glucosylation profiles compatible with subsequent chemical elongation. Having established the concept of programmed chemoenzymatic pathways to microbial carbohydrates by use of purposely tailored glycoenzymes in the case of monosaccharide acceptors,[47,49] our current effort aims at investigating the same approach in the case of non-natural disaccharide acceptors relevant to the design of a *S. flexneri* multivalent vaccine. The overall aim is to access oligosaccharides from several independent *S. flexneri* O-SPs starting from a single disaccharide, designed for use in acceptor reactions with properly engineered biocatalysts.

Obviously, the choice of the acceptor/transglucosylase system is a key issue governing the implementation of efficient tailored chemoenzymatic syntheses of complex glucosylated targets. Since the panel of known transglucosidases is highly diverse, acceptor selection controls the choice of the whole system. The acceptor has to fulfill several criteria, such as

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relevance to epidemiological data and potential for broad serotype coverage. In addition, compatibility with the chemical manipulation of the product of enzymatic glucosylation into *S. flexneri* type-specific oligosaccharides is an absolute must. Among the four possible disaccharides found on the backbone shared by most *S. flexneri* O-SPs (Figure 1), the L-rhamnosyl-(1→3)-2-*N*-acetyl-D-glucosamine moiety (CD) emerged as the one paving the way to the largest diversity of glucosylated products representative of any one of multiple *S. flexneri* serotypes.

To fulfill requirements for chemical chain elongation, the lightly protected CD analogue **1** was preferred to unprotected disaccharide CD as an acceptor substrate (Scheme 1).[50] Allyl glycoside **1** was shown to act as an acceptor substrate for several glucansucrases from the GH70 family,[50] including GBD-CD2, an α-transglucosylase engineered from *Leuconostoc mesenteroides* NRRL B-1299 dextransucrase (DSR-E). In particular, as a rewarding illustration of this powerful concept, we have recently communicated[50] on the high-yielding GBD-CD2-mediated enzymatic glucosylation of the selected disaccharide **1** into trisaccharide **2**, which features the α-D-Glc*p*-(1→4)-α-L-Rha*p* glucosylation pattern characteristic for *S. flexneri* type II[23] (Scheme 1) and on the efficient conversion of the latter into a pentadecasaccharide hapten entering in the composition of a synthetic carbohydrate-based vaccine candidate against *S. flexneri* 2a (SF2a) infection.[21]

Herein, we report on the stepwise investigation which led to an efficient conversion of the glucosylation product **2** into a pentasaccharide building block **4**, suitable for chain elongation at both ends (Scheme 2), as demonstrated in the synthesis of an aminoethyl aglycon-equipped dimer (**5**)[51] of the basic repeating unit from SF2a O-SP.[23]

Moreover, we also report on the synthesis of analogues of the GBD-CD2 acceptor substrate **1**, featuring a masked 4_C-OH (**6** and **7**) or modified at the reducing end (**8** and **9**) (Figure 2) and on their evaluation as substrates of available glucansucrases differing in terms of substrate and regiospecificity.

Scheme 1. Enzymatic Glucosylation of Acceptor Substrate 1 into Trisaccharide 2 Catalyzed by the GH70 Glucansucrase Called GBD-CD2[50]

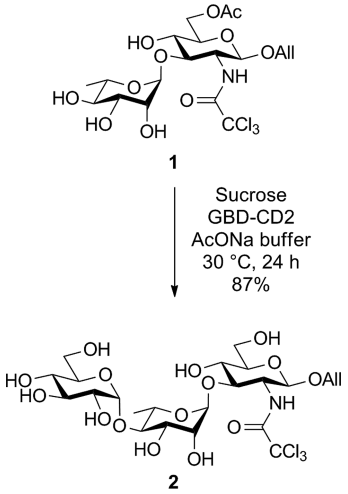
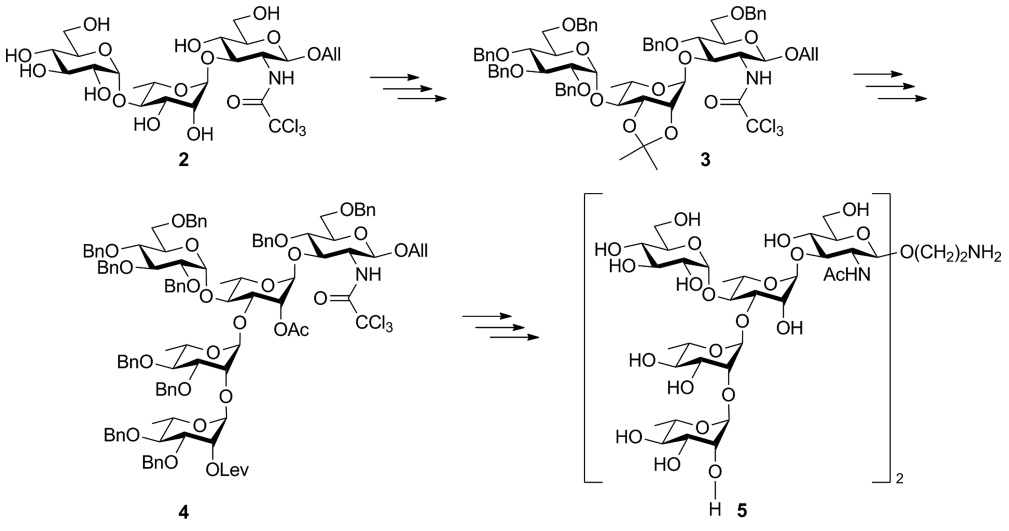


Figure 2. Chemical structures of the CD disaccharide acceptor analogues modified at position 4_C (**6**, **7**) or 1_D (**8**, **9**).

Scheme 2. Highlights of the Synthesis of Decasaccharide 5 from the Product of Enzymatic Glucosylation 2, Showing the Key Protected Intermediates 3 and 4[a]



[a]The development of efficient [2 → 3], [3 → 4], and [4 → 5] transformations, respectively, corresponds to issues addressed independently in the manuscript.

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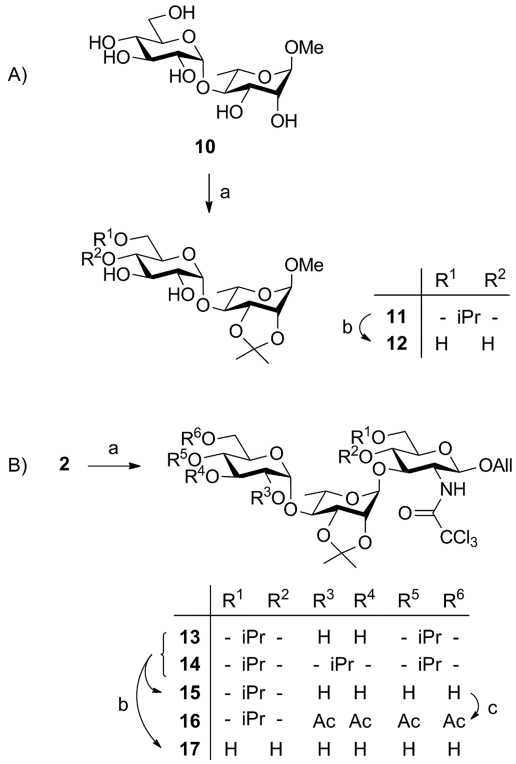
RESULTS AND DISCUSSION

Having produced trisaccharide **2** in high yield from the easily accessible GBD-CD2 acceptor substrate **1**,[50] our first objective was to demonstrate that the former could be converted into either a donor or an acceptor for use in glycosylation reactions. Since the CD disaccharide **1** had been designed toward this aim, the ECD trisaccharide **2** fulfills primary requirements at its D residue. It features a temporary allyl protecting group at the reducing end and a 2_D-*N*-trichloroacetyl moiety to ensure efficient anchimeric assistance in the D−A glycosylation step along with recovery of the 2_D-acetamido moiety upon final deprotection. However, trisaccharide **2** also has eight free hydroxyl groups, of which two are primary alcohols. Therefore, careful protection/deprotection sequences were required to allow elongation at the reducing end and/or at OH-3 of the C residue of precursor **2** so as to reach more complex SF2a oligosaccharides.

Owing to the *cis* configuration of the 2_C,3_C-diol moiety, the acetonide-protected **17** was seen as an ideal intermediate to the fully protected **3**. The higher stability of 1,3-*cis*-dioxolanes over 1,3-dioxanes toward hydrolysis is a well-acknowledged phenomenon, which has been thoroughly exemplified.[52−54] Following optimization of the reaction conditions, the expected chemoselectivity was obtained providing monoisopropylidene **12** in yields up to 89% using aqueous acetic acid at 0 °C for the selective hydrolysis of the 4,6-acetal in the EC disaccharide **11**, itself derived from the known methyl glycoside **10**[55] (Scheme 3A). Hence, trisaccharide **2** was submitted to a similar two-step process (Scheme 3B). Treatment of octaol **2** with excess 2-methoxypropene in acetone under acidic catalysis ensured its conversion into a mixture of the expected triisopropylidene **13** and the fully protected **14**, isolated in 49% and 39% yields, respectively. Acetalation of the 2_E,3_E-diol under these kinetic conditions has been reported previously,[56] but it was not perceived to be problematic due to the high lability of the 1,2-*trans* configuration. However, the selective deprotection of the isolated mixture of compounds **13** and **14** proved troublesome. Under mild hydrolysis conditions, ranging from conventional protic-mediated hydrolysis,[57,58] which was efficient on the EC model **11**, to less common protocols such as the use of molecular iodine in acetonitrile[59] or that of silica gel supported phosphomolybdic acid,[60] the deprotection pattern was similar despite different kinetics. As expected, the 2_E,3_E-*O*-isopropylidene was rapidly cleaved to give the sole intermediate **13**. Furthermore, the 6-membered 4_E,6_E-acetal was rather labile, providing access to tetraol **15** as unambiguously ascertained following isolation and per-acetylation into the fully protected **16**. In contrast, the 1,3-dioxane masking the 4_D,6_D-diol was comparably stable to the 1,3-*cis*-dioxolane blocking positions 2_C and 3_C. Because of this unexpected observation, the reaction had to be monitored carefully in order to avoid complete deprotection. The I_2/MeCN conditions gave the best yield of monoacetonide **17** (76%) in only a few hours. However, in our hands, the reaction suffered from lack of reproducibility, with yields ranging from 37% to 76%, and a more robust pathway to the fully protected **3** was required.

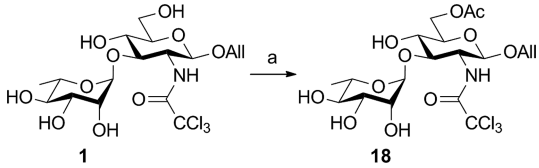
Toward this goal, preliminary protection of the primary alcohols before installing the 2_C,3_C-acetonide was investigated as an alternative. In order to extend the weight of the enzymatic input, regioselective *O*-acetylation by means of the well-known lipase B from *Candida antarctica* (CAL-B, also known as Novozym 435) in combination with vinyl acetate as donor was

Scheme 3. Synthesis of Monoacetonide 17: The Acetonide Route[a]



[a]Reagents and conditions: (A-a) 2,2-dimethoxypropane (DMP), cat. *p*-toluenesulfonic acid (*p*-TSA), DMF/acetone 1:1, 7.5 h, 86%, (A-b) 50% aq AcOH, 0 °C, 4.5 h, 89%; (B-a) 2-methoxypropene, cat. camphorsulfonic acid (CSA), acetone, 2 h, 49% for **13**, 39% for **14**; (B-b) cat. I_2, H_2O, MeCN, 2.75 h, 76%; (B-c) Ac_2O, cat. DMAP, Py, 2 h, 87%.

Scheme 4. Enzymatic *O*-Acetylation of Disaccharide 1[a]



[a]Reagents and conditions: (a) vinyl acetate, CAL-B, THF/Py (4:1) 45 °C, 17 h, 88%.

first attempted.[61−63] Once more, while the selective 6_D-*O*-acetylation of the CD disaccharide **1** proceeded in high yield (88%) to give monoacetate **18** (Scheme 4), adaptation to trisaccharide **2** was not as efficient. At best, 8% of the target 6_D,6_E-diacetate, albeit contaminated, was obtained, along with a mixture of monoacetates and starting material (not shown). Since these attempts also clearly demonstrated that the two acetate esters did not sufficiently increase lipophilicity to allow proper isolation of the target trisaccharide, the enzymatic route was abandoned.

Instead, we turned our attention to the selective chemical installment of benzoate esters. Optimization of the reaction conditions enabled the development of a process that accommodated a good balance in the solubility of the starting material **2** and expected product **19** as well as a higher reactivity of primary hydroxyls over secondary ones. Thus, octaol **2** was treated with benzoyl chloride and a hindered nucleophilic

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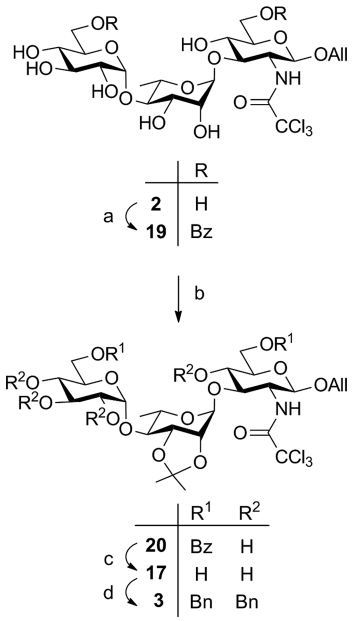
catalyst (*sym*-collidine) in a mixture of acetone/MeCN at −40 °C for 2 days to give dibenzoate **19** in a reproducible satisfactory 65% yield. The subsequent $2_C,3_C$-*O*-isopropylidenation of the latter into tetraol **20** (92%) and debenzoylation (94%) were uneventful and furnished monoacetonide **17** in high yield (Scheme 5). When masking the remaining hydroxyl groups of the latter as benzyl ethers while avoiding *N*-benzylation, the reaction was carried out at −10 °C using only a slight excess of benzyl bromide.[64] Indeed, available data suggest that the trichloroacetamide moiety of amino sugars is prone to *N*-benzylation or even cleavage when treated according to various *O*-benzylation protocols.[64] Moreover, after the sodium hydride in excess was quenched by addition of MeOH, the resulting sodium methoxide was neutralized by addition of acetic acid to protect the base-labile trichloroacetamide during evaporation of the volatiles.[64] Under these conditions, the fully protected trisaccharide **3** was obtained in 78% yield.

Scheme 5. Synthesis of Monoacetonide 17 and Subsequent Benzylation: The Ester Route[a]

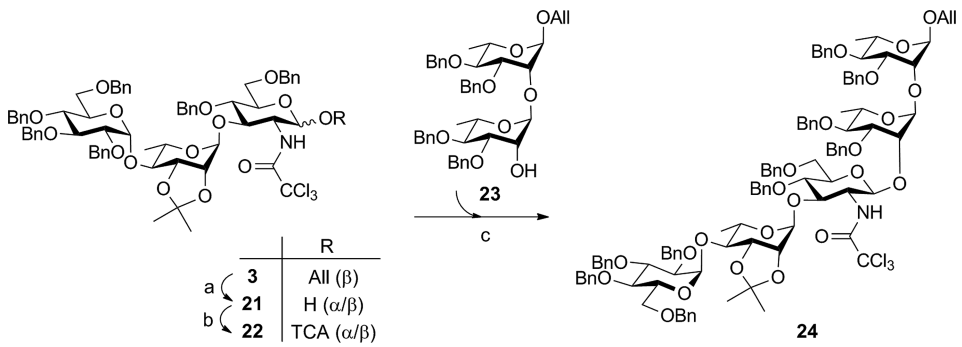


[a]Reagents and conditions: (a) BzCl, *sym*-collidine, MeCN/acetone (1:1), −40 °C, 46 h, 65%; (b) DMP, CSA, acetone, 2 h, 92%; (c) MeONa, MeOH, 3.3 h, 94%; (d) NaH, BnBr, DMF, −10 °C, 4 h, 78%.

In order to use the desired convergent [5 + 5] strategy to decasaccharide **5** (Scheme 1), conditions for a high-yielding glycosylation at the D−A linkage were required. Until recently, the investigation of convergent syntheses to SF2a oligosaccharides involving a disconnection at the D−A linkage was ruled out in our laboratory, essentially because of problems encountered by Bundle et al. in the synthesis of *S. flexneri* Y O-SP fragments.[65,66] Instead, we favored a disconnection at the C−D linkage.[51,67] However, the system described herein differs in many instances from the halide-equipped oligosaccharide donors bearing a 2_D-*N*-phthalimido participating group at their reducing end residue, which served in the *S. flexneri* Y studies.[65,66] Hence, the next focus was on using ECD as a donor in a [3 + 2] glycosylation to prepare the model ECDAB pentasaccharide **24** (Scheme 6). To this end, trisaccharide **3** was submitted to a selective two-step anomeric deallylation involving conventional allyl to propen-1-yl conversion by means of the $[Ir(COD)\{PCH_3(C_6H_5)_2\}]^+PF_6^-$ catalyst[68] and subsequent iodine-mediated hydrolysis,[69] giving rise to hemiacetal **21** (80%). The latter was in turn activated in the form of trichloroacetimidate **22** (70%, 93% corrected yield) by treatment with trichloroacetonitrile in the presence of a catalytic amount of DBU. Coupling of the known AB acceptor **23**[70] and donor **22** at low temperature in DCE containing 10 mol % of TMSOTf gave pentasaccharide **24** in a good 78% yield with complete β-selectivity.

Having demonstrated that the product of enzymatic glucosylation **2** could be converted into a potent donor, we turned our attention to the preparation of a fully protected AB(E)CD building block **4**. First, cleavage of the isopropylidene acetal in trisaccharide **3** by use of a biphasic aqueous TFA/CH_2Cl_2 system revealed the corresponding $2_C,3_C$-diol **25** (94%) (Scheme 7). Next, the 2_C-hydroxyl group was benzoylated through orthoester formation followed by regioselective acid-mediated opening,[28] yielding acceptor **26** (82%). The known rhamnosyl trichloroacetimidate **28**,[71] bearing a levulinyl ester orthogonal to the benzoyl group at position 2, was the selected donor. However, none of the [**26** + **28**] glycosylation attempts, differing in terms of solvent (Et_2O, CH_2Cl_2, toluene) and temperature (−15 °C, 25 °C, 70 °C), led to any tetrasaccharide formation (not described). Alcohol **26** was recovered unaffected while donor **28** eventually rearranged into trichloroacetamide **29**.[72] The total absence of glycosylated product was not anticipated, especially since a 2_C-benzoyl ester had been used in the synthesis of SF2a oligosaccharides, albeit on different substrates, in our group in the past.[51,67,73]

Scheme 6. Synthesis of Pentasaccharide 24 by Use of Donor 22[a]



[a]Reagents and conditions: (a) cat. $[Ir(COD)\{PCH_3(C_6H_5)_2\}]^+PF_6^-$ ([Ir]), H_2, THF, 2.5 h, then I_2, THF/H_2O, 5 h, 80%; (b) CCl_3CN, cat. DBU, DCE, −10 °C, 35 min, 70%; (c) TMSOTf (10 mol %), DCE, −35 to 0 °C, 30 min, 78%. TCA: trichloroacetimidoyl.

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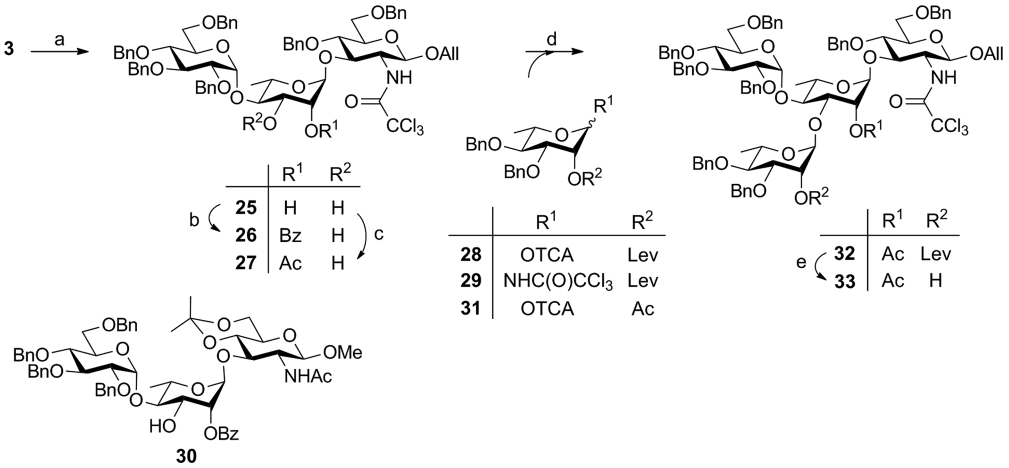
Scheme 7. Synthesis of the B(E)CD Acceptor 33[a]



[a]Reagents and conditions: (a) 50% aq TFA, CH_2Cl_2, 75 min, 94%; (b) $PhC(OMe)_3$, *p*-TSA, CH_2Cl_2, 20 min then 50% aq TFA, 45 min, 82%; (c) $MeC(OMe)_3$, *p*-TSA, MeCN, 35 min, then 80% aq AcOH, 0 °C, 25 min; (d) **28**, TMSOTf (5 mol %), Et_2O, −15 °C, 40 min, 81% from **25**, via **27**; (e) H_2NNH_2, Py/AcOH (3:2), 0 °C, 70 min, 92%.

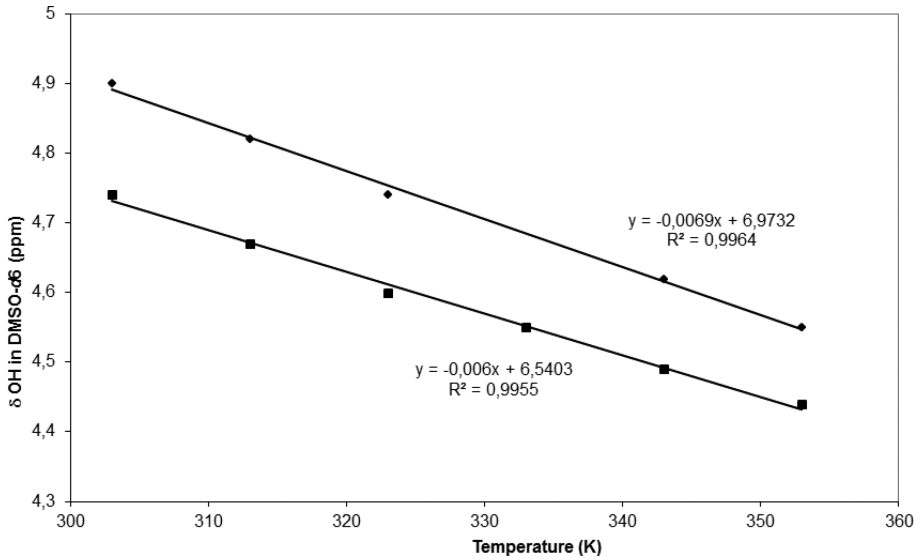


Figure 3. Temperature dependence of δ (3_C-OH) measured by ^{1}H NMR (400 MHz) for 15 mM solutions of **26** (♦) and **27** (■) in DMSO-d_6.

Interestingly, we have previously pointed to the somewhat poor reactivity of the 2_D-acetamido-4_D,6_D-*O*-isopropylidene-ECD acceptor **30**, which closely resembles trisaccharide **26**, when treated with the known 2-*O*-acetyl rhamnosyl donor **31**,[74] analogous to **28**. In spite of this observation and in contrast to the present finding, the target glycosylation product had been in this case obtained with moderate to good yield.[75] Whereas it was tempting to correlate the low reactivity of acceptor **30** with the known remote effect of the 2_D-acetamido moiety,[76] we had no explanation for the present outcome involving acceptor **26**. Changing the 2_C-benzoate in the latter for a 2_C-acetate provided analogue **27**, also obtained as a single product from diol **25** upon regioselective opening of an intermediate orthoester.[50] Satisfactorily, the [**27** + **28**] glycosylation proceeded smoothly to give tetrasaccharide **32** in a good 81% yield (from **25**), thus confirming that ECD could also serve as an acceptor. Delevulinylation of the fully protected **32** by means of hydrazine acetate then led to the B(E)CD acceptor **33** (92%).[50] The striking difference in reactivity between the 2_C-*O*-benzoyl acceptor and its 2_C-*O*-acetyl counterpart, **26** and **27**, respectively, was puzzling. We postulated that it could either be a consequence of a steric clash between the donor and acceptor **26**, or of an intramolecular hydrogen bond established between the 3_C-OH and the ester carbonyl moiety that would only occur in the case of the benzoate from acceptor **26**. It was hypothesized that if present, the latter phenomenon would either lower the electron density on the reactive hydroxyl group or constrain the molecule in a conformation less favorable for glycosylation. This hypothesis was addressed first by proper NMR analysis of the two hydroxy-esters in an aprotic solvent, which enables the detection of both intra- and intermolecular hydrogen bonds involving hydroxyl groups.[77] Measuring the ^{1}H NMR spectrum for each of the two samples in DMSO-d_6 solution at 10° intervals from 303 to 353 K clearly showed linear upfield shifts of the 3_C-OH, which were almost similar. The shift ranged between 0.002 and 0.008 ppm/°C, indicating that both hydroxyl groups were actually engaged in intramolecular hydrogen bonding (Figure 3).[78]

In contrast, ab initio quantum chemical calculations of the two disaccharides shed light on their different conformational

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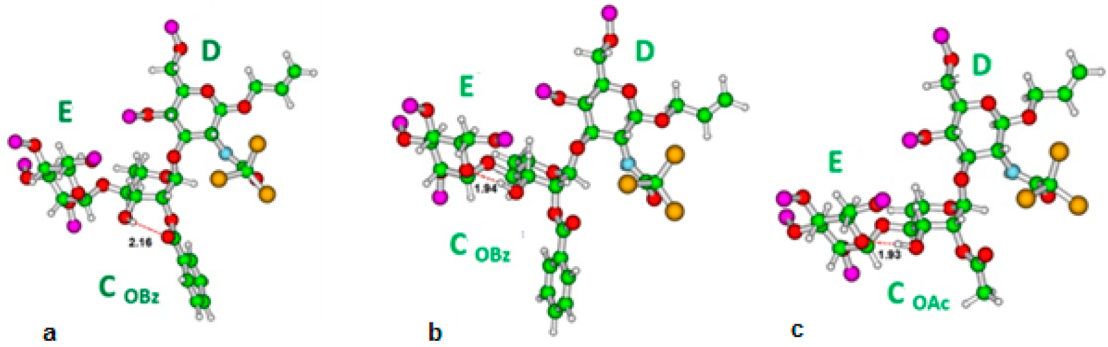
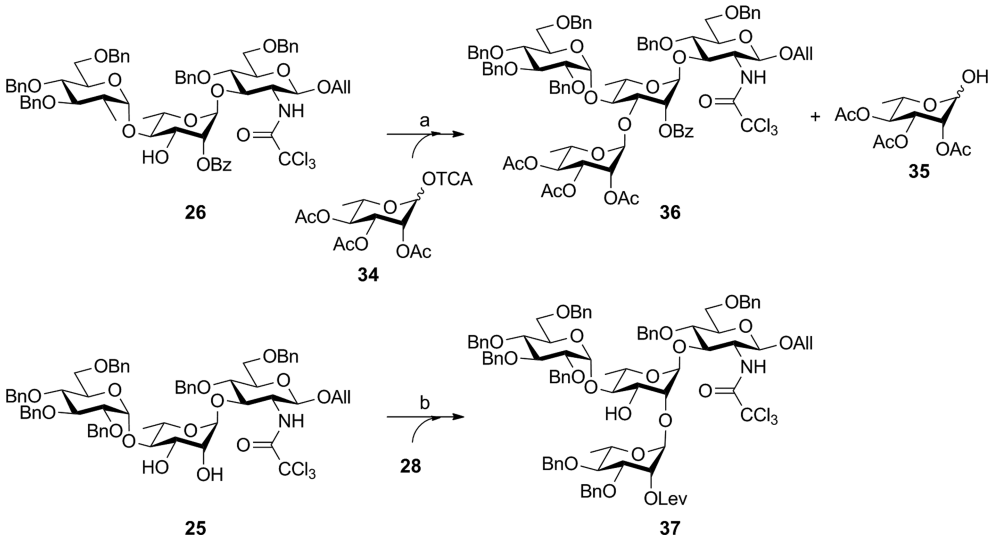


Figure 4. Local energy minima conformations adopted by the 2_C-*O*-benzoyl acceptor **26** (a and b) and by the 2_C-*O*-acetyl acceptor **27** (c). For clarity, benzyl ethers are truncated and represented as magenta circles. Distances between atoms are given in angstroms.

Scheme 8. Synthesis of Tetrasaccharides B(E)CD 36 and 37[a]



[a]Reagents and conditions: (a) 34, TMSOTf (9 mol %), toluene, 70 °C, 60 min, 20%, contaminated; (b) TMSOTf (5 mol %), toluene, −15 °C, 10 min, 80%.

behaviors. In the case of the 2_C-*O*-benzoyl trisaccharide **26**, two local energy minima were found upon optimization using the DFT/B3LYP/6-31G(3df,3pd) level (Figure 4a,b and Tables S1 and S2, Supporting Information), corresponding to geometries that mainly differ in the orientation of the 3_C-OH. In the first situation, the hydroxyl group is involved in a hydrogen-bonding interaction with the carbonyl of the vicinal ester, as hypothesized (Figure 4a). In the other minimal energy conformation, which is also more stable by ΔE = 4.8 kcal/mol, the 3_C-OH interacts with the endocyclic oxygen from the branched glucosyl residue E (Figure 4b). In contrast, the 2_C-*O*-acetyl trisaccharide **27** adopts a single energy minimum conformation, which closely resembles the more stable conformation in trisaccharide **26**. Analogously, it is stabilized by a hydrogen bond between the 3_C-OH group and the endocyclic oxygen from residue E (Figure 4c and Table S3, Supporting Information). All attempts to localize an energy minimum in which the 3_C-OH was involved in a hydrogen bond with the acetate carbonyl moiety in trisaccharide **27** were fruitless as calculations repeatedly converged toward one single energy well. The calculated Mulliken charges on the 3_C-oxygen atom for the observed energy minima conformations are 0.76 and 0.85 (Figure 4a,b, respectively) for the benzoylated **26** and 0.86 for the acetylated **27**, respectively. Since the 3_C-OH of the most stable conformations of the two hydroxy esters have similar charges, it could be assumed that the hydrogen bonding had no crucial influence on the electron density of the 3_C-oxygen involved in the B−C linkage. However, the most stable conformation of the benzoylated **26** clearly showed steric hindrance at the 3_C-OH brought by the aromatic ring of the vicinal benzoyl group, which could be held accountable for its lower reactivity.

With these new data in hand, the "steric hindrance" hypothesis was investigated more deeply by coupling acceptor **26** with the less hindered tri-*O*-acetyl rhamnosyl donor[79] **34** (Scheme 8). Despite the harsh conditions, a substantial amount of unreacted acceptor was recovered (70%), while most of the donor was hydrolyzed into the known hemiacetal[80] **35**. Nevertheless, a new product was isolated, even though impure, which was tentatively identified as the coupling product **36**, based on mass spectrometry analysis (HRMS (ESI^+) calcd for $C_{84}H_{92}Cl_3NO_{23}Na$ $[M + Na]^+$, 1610.5023, found *m*/*z* 1610.5002). It is noteworthy that from this point forward some NMR signals from the A or B rhamnoses in the B(E)CD tetrasaccharides and larger oligosaccharides would either be broadened or absent. As another example, the coupling of donor **28** and the less hindered diol **25** unambiguously furnished the 2_C-*O*-rhamnosyl tetrasaccharide **37** (Scheme 8).

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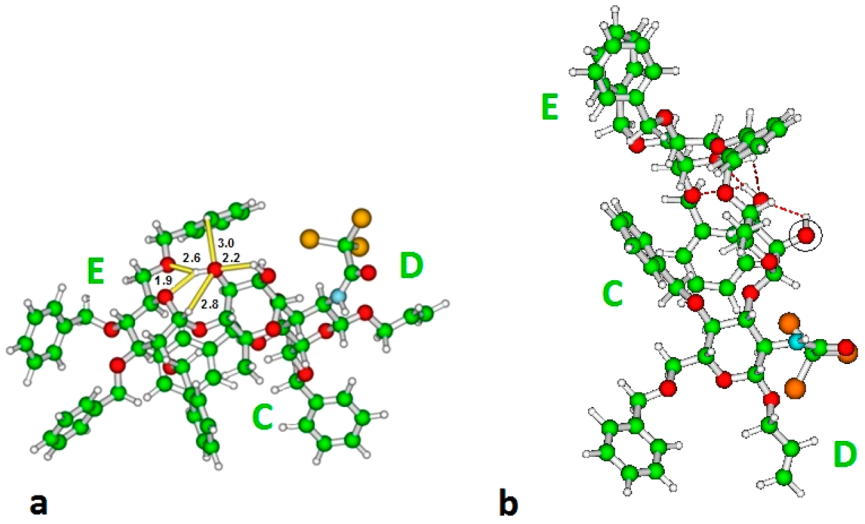

Figure 5. Conformation of the local energy minimum of diol **25** highlighting steric hindrance around the 3_C-OH (view a) and accessibility of the 2_C-OH (view b). Distances between atoms are given in angstroms. The 2_C-OH group is encircled.

This outcome is in total agreement with data from Kong and Du, who found that glycosylation at the 3-OH of 2,3,4-triol rhamnoside acceptors is strongly favored, whereas in the case of 2,3-diol rhamnosides, glycosylation occurs preferentially at the axial 2-OH.[81]

Molecular modeling of diol **25** using the DFT/B3LYP/6-31G(3df,3pd) level indicated two energy minima corresponding to geometries that mainly differ in the orientation of the 3_C-OH. In the first minimal energy conformation (Table S4, Supporting Information), the hydroxyl group is involved in a hydrogen-bonding interaction with the 2_C-oxygen. In the other major conformation (Figure 5 and Table S5, Supporting Information), which is also more stable by 7 kcal/mol, the 3_C-OH interacts with the endocyclic oxygen of residue E as also observed for trisaccharides **26** and **27**. The energy difference between these two conformers is such that only the lower local minimum is occupied. In the corresponding conformer, the charges on the oxygen atoms at positions 2_C and 3_C are similar, indicating that charge effects do not account for the observed regioselectivity. Rather, the latter is thought to arise from steric hindrance at the equatorial 3_C oxygen atom (Figure 5a). Indeed, the 3_C-OH takes part in three hydrogen bonds. It is in close proximity of H-1_E and also interacts with the phenyl ring of the 6_E-benzyl ether. In comparison to this highly constrained situation, the 2_C-OH group is easily accessible (Figure 5b, black circle).

We then moved on to the preparation of the $[AB(E)CD]_2$ decasaccharide (Scheme 9). To this end, acceptor **33** was coupled with donor **28** allowing the formation of pentasaccharide **38** (86%). Deallylation of the latter gave hemiacetal **39** (93%), which was in turn converted to the key trichloroacetimidate **40** (97%). In order to equip the acceptor for site-selective conjugation, donor **40** was glycosylated with bromoethanol and the bromine atom was substituted with sodium azide to reach the fully protected azidoethyl glycoside **43** (78%). A delevulinylation step led to acceptor **44** (90%), which was then glycosylated with the same key donor **40**. Although the reaction was run at 40 °C, very little condensation was detected. Instead, the donor rapidly evolved into the trichloro-oxazoline **45**. Satisfactorily, subsequent addition of TMSOTf to reach 15 mol % allowed the full consumption of the acceptor. Yet, despite several attempts at purifying the fully protected **46** by column chromatography using different elution systems, the latter could not be isolated as pure material. Indeed, a MALDI analysis clearly indicated that the decasaccharide was slightly contaminated with trichloroacetamide **42** (HRMS (ESI^+) calculated for $C_{111}H_{120}Cl_6N_2O_{26}Na$ $[M + Na]^+$ 2129.62, found m/z 2129.37), a rearranged form of donor **40**. Attempts at purifying the decasaccharide at the next step were disappointing (not shown). For that reason, trichloroacetimidate **40** was changed for the corresponding *N*-phenyltrifluoroacetimidoyl (PTFA) donor **41**,[50] obtained in 83% yield from hemiacetal **39**. Gratifyingly, the glycosylation between the new donor **41** and acceptor **44** performed at −10 °C gave a good 82% yield of the desired decasaccharide **46**. Methanolysis of the latter into triol **47** (72%) required unoptimized harsh conditions as previously experienced for a closely related decasaccharide.[51] Treatment of triol **47** with $Pd(OH)_2/C$ under a hydrogen atmosphere enabled the concomitant benzyl ether hydrogenolysis, trichloroacetamide hydrodechlorination, and azide reduction to give, in 58% yield following RP-HPLC purification, the $[AB(E)CD]_2$ target **5**, which corresponds to a two-unit segment of the *S. flexneri* 2a O-SP equipped with a linker enabling site-selective conjugation.

Capitalizing on the excellent outcome of the enzymatic regioselective α-D-glucosylation of the CD disaccharide **1** and the successful conversion of the obtained ECD **2** into relevant synthetic fragments of the O-SP from *S. flexneri* 2a as exemplified by decasaccharide **5**, a functional mimic of the homologous O-SP,[22] we questioned the potential of acceptor **1** as a precursor to α-D-glucosylation patterns characteristic of O-SPs from other *S. flexneri* serotypes (Figure 6). In particular, the α-D-glucosylation at O-4_D and O-6_D would open the way to oligosaccharides representative of O-SPs from *S. flexneri* type I and type IV, respectively.[23] In the following part of this article, we report on preliminary investigations toward this aim. To probe this hypothesis, minor modifications of the original acceptor **1** were considered, so as to identify any novel regioselectivity by use of GBD-CD2 and *Np*AS, a glucansucrase from GH13 family,[82] in combination with sucrose as donor. The former naturally forms α-(1→2) glucosidic bonds, whereas the latter catalyzes the α-(1→4) linkage synthesis.

The most straightforward approach to a modified CD acceptor was to mask the 4_C-OH in the starting allyl glycoside

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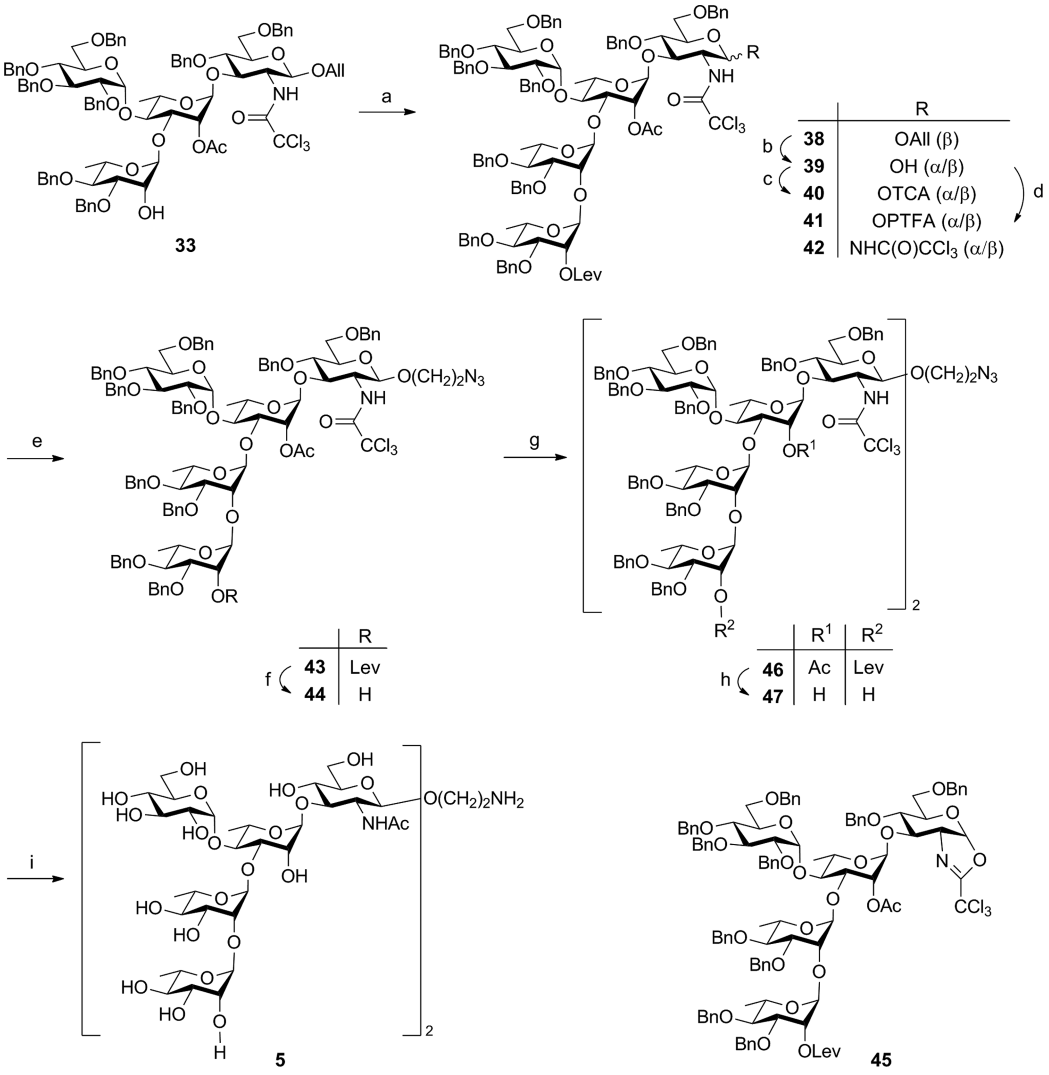
Scheme 9. Synthesis of the Target Decasaccharide 5[a]



[a]Reagents and conditions: (a) **28**, TMSOTf (7 mol %), Et_2O, −15 °C, 86%; (b) [Ir], H_2, THF, 2.5 h, then I_2, THF/H_2O (3:1), 4.5 h, 93%; (c) CCl_3CN, cat. DBU, DCE, −10 °C, 40 min, 97%; (d) $CF_3C(NPh)Cl$, Cs_2CO_3, acetone, rt, 2 h, 83%; (e) **40**, bromoethanol, TMSOTf (10 mol %), DCE, 0 °C, 25 min, then NaI, NaN_3, DMF, 80 °C, 2 h, 78%; (f) H_2NNH_2, AcOH/Py (2:3), 0 °C, 90%; (g) **41**, TMSOTf (20 mol %), toluene, −10 °C, 2 h, 82%; (h) MeONa, MeOH, 21 h, 71%; (i) H_2, $Pd(OH)_2$/C *t*-BuOH/CH_2Cl_2/H_2O, 3 d, 58%.

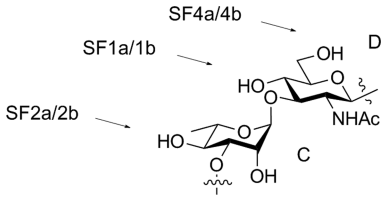


Figure 6. *S. flexneri* type-specific oligosaccharides in reach from CD following regioselective α-D-glucosylation at the positions indicated by the arrows.[23]

1. To this end, two different targets were envisaged. The 4_C-OH was blocked in the form of a methyl ether (**6**), and alternatively, it was oxidized into a 4_C-oxo moiety, therefore also interfering with the $^1C_4$ ring conformation of L-rhamnose C, to give allyl glycoside **7** bearing conformational properties different from that of native **1** (Scheme 10). Toward this aim, di-*O*-isopropylidenation of disaccharide **1** gave the common intermediate **48** (75%), which underwent *O*-methylation with methyl iodide in THF[83] to give the fully protected **49** (89%). It is noteworthy that performing the reaction in THF and using sodium hydride as the base was essential. Indeed, methylation attempts in DMF containing silver oxide or sodium hydride, or in THF containing silver oxide, resulted in partial *N*-methylation of the trichloroacetamide moiety (not shown), as already observed by Scharf and Jütten in their synthesis of evernitrose.[84] This phenomenon is attributed to the strong inductive effect of the trichloroacetyl group, which enhances the acidity of the amide proton. The fully protected **49** was then submitted to acid-mediated acetal hydrolysis, leading to the expected 4_C-OMe disaccharide **6** (82%). Alternatively, alcohol **48** was oxidized using Swern conditions into ketone **50** (84%), which was then deacetalated into the 4_C-oxo derivative **7** (69%).

We attempted to force glucosylation of disaccharide CD at positions different from 4_C-OH. To this end, acceptor reaction with disaccharides **6** and **7**, both lacking a free 4_C-OH, were performed with GBD-CD2. The LC−MS follow-up of the enzymatic conversion showed that no glucosylation product

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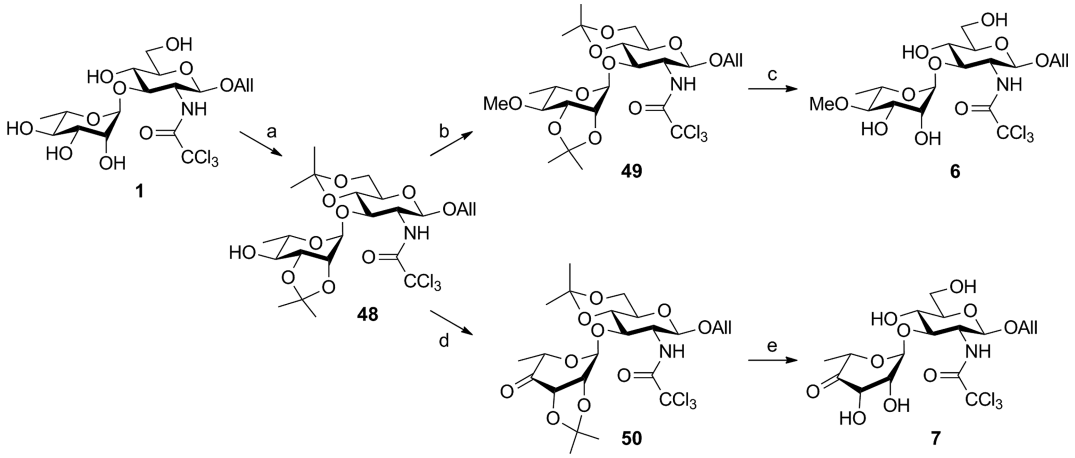
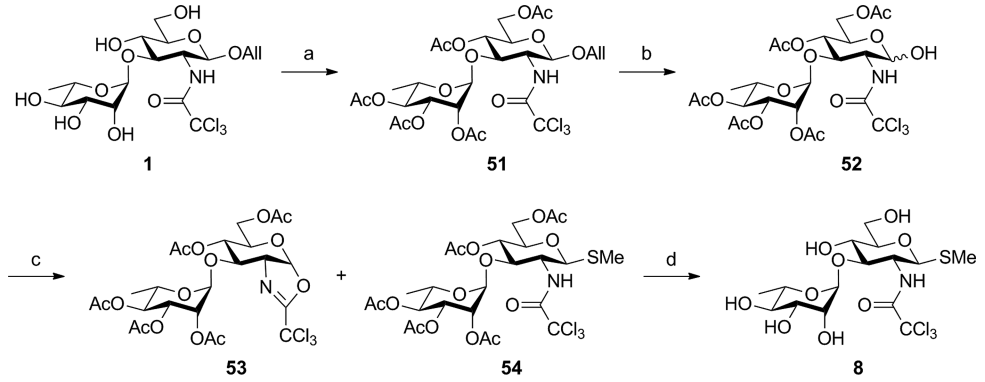
Scheme 10. Synthesis of the CD Disaccharides 6 and 7, Encompassing a Masked 4_C-OH[a]



[a]Reagents and conditions: (a) 2-methoxypropene, DMP, cat. CSA, DMF/acetone 1:1, 36 h, 75%; (b) NaH, MeI, THF, 3 h, 89%; (c) 50% aq TFA, CH_2Cl_2, 35 min, 82%; (d) $(COCl)_2$, DMSO, Et_3N, CH_2Cl_2, −78 °C→ rt, 75 min, 84%; (e) 80% aq AcOH, 80 °C, 15 h, 69%.

Scheme 11. Synthesis of the Methyl Thioglycoside 8[a]



[a]Reagents and conditions: (a) Ac_2O, cat. DMAP, Py, 14 h, 94%; (b) [Ir], H_2, THF, 4 h, then I_2, THF/H_2O (3:1), 16 h, 85%; (c) CCl_3CN, Cs_2CO_3, CH_2Cl_2, 60 min, then TMS-SMe, TMSOTf (5 mol %), 16 h, 50% for **53**, 31% for **54**; (d) MeONa, MeOH, 90 min, 91%.

was formed (not shown), indicating that blocking 4_C-OH does not facilitate any alternative glucosylation at another position.

A novel set of CD acceptor disaccharides, in this case differing from substrate **1** at their reducing end, was also envisioned to eliminate the possible steric hindrance of the allyl group with some amino acid residues constituting the active site that could prevent glucosylation by *Np*AS (docking results not shown). For that reason, the allyl aglycon in disaccharide **1** was replaced while keeping in mind that any group introduced at the anomeric position of the D residue should provide an access to a CD donor. On the one hand, introduction of a smaller methylthio group gave disaccharide **8**, and on the other hand, the allyl moiety in the key precursor **1** was simply cleaved to give the reducing analogue **9**, the opened form of which could lead to a complete rearrangement of the substrate inside the enzyme active site. Hence, per-acetylation of disaccharide **1** gave fully protected **51** (94%), and subsequent deallylation and reaction with methyl trimethylsilyl sulfide gave rise to mainly oxazoline **53** (50%) but also to the expected methyl thioglycoside **54**, albeit in an unoptimized 31% yield (Scheme 11). Finally, methanolysis gave the deprotected disaccharide **8** (91%).

For the preparation of analogue **9**, direct deallylation of disaccharide **1** was attempted under several conditions, including [Ir]/H_2/I_2 or $PdCl_2$/AcONa/AcOH, but none of them proved successful. Moreover, methanolysis of lactol **52** mediated by sodium methoxide or K_2CO_3 resulted in complete disaccharide degradation. Additionally, using the same conditions as for pentasaccharide **38**, deallylation of di-*O*-isopropylidene **48** followed by acidic hydrolysis gave the expected lactol **9**, albeit in poor yield (15% over two steps) (Scheme 12A). For those reasons, an alternative route was used (Scheme 12B). Thus, per-silylation of allyl glycoside **1** gave the fully protected intermediate **55** (86%). NMR data revealed a 3:2 equilibrium between two rotamers coexisting owing to the bulkiness of the five TBS groups. Deallylation of the persilylated **55** was uneventful (84%), and consecutive desilylation under mild conditions[85] provided the desired disaccharide **9** in an acceptable 70% yield. In contrast to its protected precursor, this novel potential acceptor substrate did not show any conformational constraints as inferred from NMR data.

Having disaccharides modified at their reducing end, **8** and **9**, in hand, we examined their potential as acceptor substrates for *Np*AS under standard conditions in the presence of excess sucrose. The LC−MS profiles of the enzymatic reaction mixtures revealed that *Np*AS could not glucosylate these novel CD analogues (not shown), indicating that the reduction

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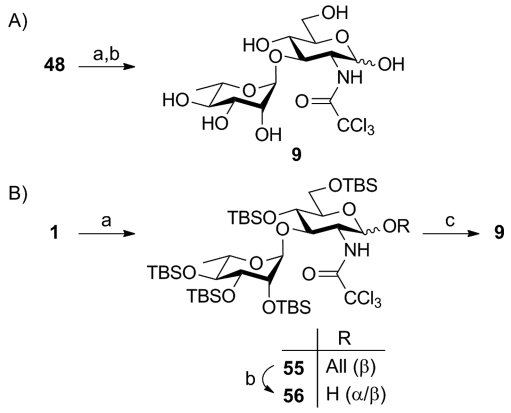
Scheme 12. Synthesis of the Reducing Disaccharide 9[a]



[a]A: from **48**. Reagents and conditions: (a) [Ir], H_2, THF, 75 min, then I_2, THF/H_2O (3:1), 3 h; (b) AcOH/H_2O 1:1, 80 °C, 3 h, 15% (over two steps). B: from **1**. Reagents and conditions: (a) TBSOTf, Py 60 °C, 15 h, 86%; (b) [Ir], H_2, THF, 2.5 h then I_2, THF/H_2O (2:1), 60 min, 84%; (c) Et_3N·3HF, THF, 50 °C, 26 h, 70%.

of the substituent size at C-1 did not influence the outcome of the enzymatic process.

It can be inferred from these results that non-natural acceptor substrate recognition by available biocatalysts is not a clear-cut process and that finding native glucansucrases able to glucosylate acceptor **1** at 4_D-OH as in *S. flexneri* type I or at 6_D-OH as in *S. flexneri* type IV is not straightforward. In such particular cases, enzyme engineering technologies combined with computer-aided design remain the best option. The potential of these strategies has recently been illustrated to reshape the active site of *Np*AS and generate catalysts to effect glucosylation of non-natural monosaccharide acceptor substrates[47,48] or a lightly protected DA disaccharide acceptor[86] with the requested stereo- and regiospecificities to achieve the chemo-enzymatic synthesis of other serotype-specific *S. flexneri* oligosaccharides.

CONCLUSION

This study is part of a program aimed at developing a synthetic carbohydrate-based vaccine against shigellosis. Toward this aim, the development of highly convergent synthetic strategies to several oligosaccharides identified as powerful functional mimics of a selected set of *S. flexneri* type specific O-SPs was undertaken. In addition to the study of purely chemical synthetic routes to the target oligosaccharides, we are investigating original chemo-enzymatic strategies involving an early enzymatic α-D-glucosylation step by use of tailored glucansucrases acting on non-natural lightly protected acceptors. Herein, we have reported the four-step chemical conversion of trisaccharide **2**, obtained from the GBD-CD2-mediated α-D-glucosylation at OH-4_C of disaccharide **1**, into a fully orthogonally protected ECD intermediate **3**. Differentiation between hydroxyl groups in trisaccharide **3** resulted from the selective benzoylation of primary alcohols under controlled conditions on the one hand and from the efficient selective isopropylidenation of *cis*-vicinal diols on the other hand. Trisaccharide **3** evolved into trichloroacetimidate **22** and into an acceptor, paving the way for chain elongation. While the former was easily validated as a donor to give pentasaccharide **24**, the identification of a suitable ECD acceptor revealed important steric hindrance at OH-3_C. The latter issue was overcome by the use of a small acetyl protecting group at OH-2_C providing acceptor **27**. Efficient chain elongation at OH-3_C of acceptor **27** with residues B and A, respectively, provided the fully protected pentasaccharide **38**. Following conversion of this key intermediate into PTFA donor **44** and acceptor **42**, a [5 + 5] glycosylation step gave the fully protected decasaccharide **46** in high yield, which was next completely deprotected into the known aminoethyl-equipped glycoside **5**. This achievement demonstrates the feasibility of the programmed chemo-enzymatic synthesis of *S. flexneri* oligosaccharides by use of glucansucrases in combination with carefully designed lightly protected acceptors, a disaccharide in this case. Broadening the programmed chemo-enzymatic concept onto four analogues of acceptor **1** also showed the limits of exploiting enzyme-natural promiscuity and that enzyme engineering toward novel tailored biocatalysts is required to address other *S. flexneri* serotypes.

EXPERIMENTAL SECTION

Chemical Synthesis. Purchased reagents and solvents were used as received. Air- and moisture-sensitive reactions were performed in dried glassware under argon. Anhydrous toluene, Et_2O, DCE, CH_2Cl_2, THF, DMF, MeCN, MeOH, and Py were delivered and stored on molecular sieves (MS). NaH (60% dispersion in mineral oil) was washed with anhyd pentane under a stream of argon before use. 4 Å MS were activated before use by heating at 250 °C under vacuum. Analytical TLC was performed with silica gel 60 F254, 0.25 mm precoated TLC plates. Compounds were visualized using UV_{254} and/or charring with orcinol (1 mg·mL^{-1}) in 10% aq H_2SO_4. Flash column chromatography was carried out using silica gel (particle size 40−63 μm or 15−40 μm). NMR spectra were recorded at 303 K and at 400 MHz (^{1}H) and 100 MHz (^{13}C) equipped with a Broadband Observe probe. Signal assignments were based on ^{1}H, COSY, DEPT-135, HSQC, ^{13}C, ^{13}C gated decoupling, and HMBC experiments. Signals are reported as m (multiplet), s (singlet), d (doublet), t (triplet), pt (pseudo triplet), dd (doublet of doublet), dq (doublet of quadruplet), br s (broad singlet), br d (broad doublet), and br t (broad triplet), and coupling constants are reported in hertz (Hz). Spectra were recorded in $CDCl_3$, CD_3OD, DMSO-d_6, and D_2O. Chemical shifts are reported in ppm (δ) relative to residual solvent peak, $CHCl_3$ in the case of $CDCl_3$, MeOH in the case of CD_3OD, HOD and 4,4-dimethyl-4-silapentane-1-sulfonic acid (DSS) in the case of D_2O, and DMSO in the case of DMSO-d_6, at 7.26/77.16, 3.31/49.0, 4.79/0.0, and 2.50/39.5 ppm for the ^{1}H and ^{13}C spectra, respectively. Of the two magnetically nonequivalent geminal protons at C-6, the one resonating at lower field is denoted H-6a, and the one at higher field is denoted H-6b. Sugar residues are serially lettered according to the lettering of the repeating unit of the *S. flexneri* 2a O-SP and identified by a subscript in the listing of signal assignments. HRMS were recorded in the positive-ion electrospray ionization (ESI+) mode. Solutions were prepared using 1:1 MeCN/H_2O containing 0.1% formic acid or MeOH/water containing 10 mM ammonium acetate in the case of sensitive compounds. HR-MALDI-TOF-MS were recorded in the positive-ion reflector mode using 2,5-dihydroxybenzoic acid as the matrix. Solutions were prepared in MeCN/0.1% aq TFA. Optical rotations were obtained using the sodium D line at ambient temperature.

Methyl (4,6-O-Isopropylidene-α-D-glucopyranosyl)-(1→4)-2,3-O-isopropylidene-α-L-rhamnopyranoside (11). To a solution of disaccharide **10**[55] (1.10 g, 3.23 mmol) in a 1:1 mixture of acetone and DMF (9 mL) were added DMP (2.4 mL, 19.5 mmol, 6.0 equiv) and *p*-TSA (37 mg, 0.19 mmol, 0.06 equiv). The reaction mixture was stirred at rt for 7.5 h. Et_3N was added, and volatiles were evaporated and coevaporated repeatedly with water, MeOH, and CH_2Cl_2 under vacuum. Column chromatography of the residue (CH_2Cl_2/MeOH, 100:0 → 95:5) gave the di-*O*-isopropylidene **11** (1.17 g, 2.78 mmol, 86%) as a white foam: R_f = 0.32 (CH_2Cl_2/MeOH 9.5:0.5); $[\alpha]^{23}_D$ = 66.4 (*c* 1.0; $CHCl_3$); ^{1}H NMR ($CDCl_3$) δ 4.98 (d, $J_{1,2}$ = 4.1 Hz, 1H, H-1_E), 4.84 (s, 1H, H-1_C), 4.15−4.10 (m, 2H, H-2_C, H-3_C), 3.90−3.84

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(m, 2H, H-5$_E$, H-6a$_E$), 3.78−3.68 (m, 3H, H-3$_E$, H-5$_C$, H-6b$_E$), 3.59 (dt, $J_{2,3}$ = 9.4 Hz, 1H, H-2$_E$), 3.52 (pt, $J_{3,4}$ = $J_{4,5}$ = 9.2 Hz, 1H, H-4$_E$), 3.40 (dd, $J_{4,5}$ = 9.8 Hz, $J_{3,4}$ = 6.8 Hz, 1H, H-4$_C$), 3.37 (s, 3H, OCH$_3$), 2.72 (br s, 1H, OH$_{3E}$), 2.24 (d, $J_{2,OH}$ = 9.4 Hz, OH$_{2E}$), 1.51 (s, 3H, H$_{iPr}$), 1.50 (s, 3H, H$_{iPr}$), 1.44 (s, 3H, H$_{iPr}$), 1.35 (s, 3H, H$_{iPr}$), 1.33 (d, $J_{5,6}$ = 6.3 Hz, 1H, H-6$_C$); ^{13}C NMR ($CDCl_3$) δ 109.4 (C$_{iPr\text{-}C}$), 99.8 (C$_{iPr\text{-}E}$), 99.2 (C-1$_E$), 98.1 (C-1$_C$), 80.7 (C-4$_C$), 76.7 (C-2$_C$), 76.0 (C-3$_C$), 73.4 (2C, C-2$_E$, C-4$_E$), 72.4 (C-3$_E$), 64.8 (C-5$_C$), 63.5 (C-5$_E$), 62.4 (C-6$_E$), 55.0 (OCH$_3$), 29.2, 28.2, 26.5, 19.3 (4C, C$_{iPr}$), 17.8 (C-6$_C$); HRMS (ESI$^+$) m/z calcd for $C_{19}H_{32}O_{10}Na$ [M + Na]$^+$ 443.1893, found 443.1861.

*Methyl α-D-Glucopyranosyl-(1→4)-2,3-O-isopropylidene-α-L-rhamnopyranoside (**12**).* A solution of di-*O*-isopropylidene derivative **11** (246 mg, 583 μmol) in 50% aq AcOH (2.0 mL) was stirred at 0 °C for 3.5 h and then at rt for 1 h. Toluene was added, and volatiles were evaporated. The residue was purified by column chromatography (CH_2Cl_2/MeOH, 100:0 → 90:10) to give the mono-*O*-isopropylidene **12** (199 mg, 523 μmol, 89%) as a white foam: R_f = 0.26 (CH_2Cl_2/MeOH 9:1); $[\alpha]^{23}_D$ = 65.0 (*c* 1.0; $CHCl_3$); ^{1}H NMR (DMSO-d_6) δ 4.79−4.77 (m, 3H, H-1$_C$, H-1$_E$, OH$_{4E}$), 4.73−4.71 (m, 2H, OH$_{2E}$, OH$_{3E}$), 4.17−4.14 (m, 1H, OH$_{6E}$), 4.08 (d, $J_{2,3}$ = 5.7 Hz, 1H, H-2$_C$), 3.95 (br t, 1H, H-3$_C$), 3.63−3.48 (m, 4H, H-5$_C$, H-5$_E$, H-6a$_E$, H-6b$_E$), 3.39 (dt, $J_{2,3}$ = 9.4 Hz, $J_{1,2}$ = 4.7 Hz, 1H, H-2$_E$), 3.26−3.14 (m, 3H, H-4$_C$, H-3$_E$, H-4$_E$), 1.42 (s, 3H, H$_{iPr}$), 1.26 (d, $J_{5,6}$ = 6.3 Hz, 1H, H-6$_C$), 1.25 (s, 3H, H$_{iPr}$); ^{13}C NMR (DMSO-d_6) δ 108.3 (C$_{iPr}$), 99.7 (C-1$_E$), 97.1 (C-1$_C$), 79.9 (C-4$_C$), 76.6 (C-3$_C$), 75.3 (C-2$_C$), 73.1 (C-2$_E$), 72.1 (2C, C-3$_E$, C-5$_E$), 69.5 (C-4$_E$), 64.7 (C-5$_C$), 60.1 (C-6$_E$), 54.1 (OCH$_3$), 27.9, 26.2 (2C, C$_{iPr}$), 17.3 (C-6$_C$); HRMS (ESI$^+$) m/z calcd for $C_{16}H_{28}O_{10}Na$ [M + Na]$^+$ 403.1580, found 403.1625; m/z calcd for $C_{32}H_{56}O_{20}Na$ [2 M + Na]$^+$ 783.3263, found 783.3432.

*Allyl (4,6-O-Isopropylidene-α-D-glucopyranosyl)-(1→4)-(2,3-O-isopropylidene-α-L-rhamnopyranosyl)-(1→3)-2-deoxy-4,6-O-isopropylidene-2-trichloroacetamido-β-D-glucopyranoside (**13**) and Allyl (2,3:4,6-Di-O-isopropylidene-α-D-glucopyranosyl)-(1→4)-(2,3-O-isopropylidene-α-L-rhamnopyranosyl)-(1→3)-2-deoxy-4,6-O-isopropylidene-2-trichloroacetamido-β-D-glucopyranoside (**14**).* To a solution of trisaccharide **2** (299 mg, 0.44 mmol) in acetone (10 mL), stirred under an argon atmosphere, were added CSA (20 mg, 0.09 mmol, 0.2 equiv) and 2-methoxypropene (253 μL, 2.64 mmol, 6.0 equiv). After the mixture was stirred for 2 h at rt, Et_3N (13 μL, 0.09 mmol, 0.2 equiv) was added and the reaction mixture was concentrated. The residue was purified by column chromatography (cHex/EtOAc, 70:30 → 50:50 to elute the first compound then 30:70 → 0:100 to elute the second compound) to give the fully protected trisaccharide **14** (143 mg, 0.17 mmol, 39%), followed by diol **13** (171 mg, 0.22 mmol, 49%). The two compounds were isolated as white amorphous solids. Diol **13**: R_f = 0.20 (cHex/EtOAc 3:7); $[\alpha]^{23}_D$ = +26 (*c* 1.0; $CHCl_3$); ^{1}H NMR ($CDCl_3$) δ 6.70 (d, $J_{NH,2}$ = 8.6 Hz, 1H, NH), 5.88−5.78 (m, 1H, C*H*=CH$_2$), 5.29−5.24 (m, J_{trans} = 17.3 Hz, 1H, CH=C*H*$_2$), 5.21−5.18 (m, J_{cis} = 10.4 Hz, 1H, CH=C*H*$_2$), 5.06 (s, 1H, H-1$_C$), 4.97 (d, $J_{1,2}$ = 4.2 Hz, 1H, H-1$_E$), 4.75 (d, $J_{1,2}$ = 8.4 Hz, 1H, H-1$_D$), 4.35−4.30 (m, 1H, −OCH$_{2All}$), 4.11−4.04 (m, 4H, −OCH$_{2All}$, H-2$_C$, H-3$_C$, H-3$_D$), 4.00−3.94 (m, 2H, H-6a$_D$, H-5$_C$), 3.89−3.78 (m, 3H, H-6b$_D$, H-6a$_E$, H-5$_E$), 3.77−3.64 (m, 4H, H-4$_D$, H-6b$_E$, H-2$_D$, H-3$_E$), 3.62−3.56 (m, 1H, H-2$_E$), 3.51 (pt, $J_{3,4}$ = $J_{4,5}$ = 9.2 Hz, 1H, H-4$_E$), 3.38−3.31 (m, 2H, H-4$_C$, H-5$_D$), 2.58 (bs, 1H, OH), 2.05 (bs, 1H, OH), 1.51 (s, 6H, H$_{iPr}$), 1.45 (s, 3H, H$_{iPr}$), 1.44 (s, 3H, H$_{iPr}$), 1.41 (s, 3H, H$_{iPr}$), 1.27 (bs, 6H, H-6$_C$, H$_{iPr}$); ^{13}C NMR ($CDCl_3$) δ 162.2 (NHCO), 133.4 (*C*H=CH$_2$), 118.4 (CH=*C*H$_2$), 109.3, 99.9, 99.8 (3C, C$_{iPr}$), 99.4 (C-1$_D$), 99.2 (C-1$_E$), 98.1 (C-1$_C$), 92.6 (CCl_3), 80.8 (C-4$_C$), 76.5, 76.4, 76.0 (3C, C-2$_C$, C-3$_C$, C-3$_D$), 73.5 (C-4$_E$), 73.4 (C-2$_E$), 73.0 (C-4$_D$), 72.3 (C-3$_E$), 70.5 (OCH$_{2All}$), 67.6 (C-5$_D$), 65.3 (C-5$_C$), 63.5 (C-5$_E$), 62.3, 62.2 (2C, C-6$_D$, C-6$_E$), 59.0 (C-2$_D$), 29.3, 29.2, 28.2, 26.5, 19.4, 19.3 (6C, C$_{iPr}$), 17.9 (C-6$_C$); HRMS (ESI$^+$) m/z calcd for $C_{32}H_{48}Cl_3NO_{15}Na$ [M + Na]$^+$ 814.1987, found 814.1950.

Fully protected **14**: R_f = 0.25 (cHex/EtOAc 7:3); $[\alpha]^{23}_D$ = +16 (*c* 1.0; $CHCl_3$); ^{1}H NMR ($CDCl_3$) δ 6.68 (d, $J_{NH,2}$ = 8.6 Hz, 1H, NH), 5.88−5.78 (m, 1H, C*H*=CH$_2$), 5.29−5.24 (m, J_{trans} = 17.3 Hz, 1H, CH=C*H*$_2$), 5.22 (d, $J_{1,2}$ = 3.1 Hz, 1H, H-1$_E$), 5.21−5.18 (m, J_{cis} = 10.5 Hz, 1H, CH=C*H*$_2$), 5.07 (s, 1H, H-1$_C$), 4.75 (d, $J_{1,2}$ = 8.4 Hz, 1H, H-1$_D$), 4.35−4.30 (m, 1H, −OCH$_{2All}$), 4.13−4.03 (m, 4H, H-2$_C$, −OCH$_{2All}$, H-3$_C$, H-3$_D$), 4.00−3.94 (m, 3H, H-3$_E$, H-5$_C$, H-6a$_D$), 3.90−3.72 (m, 6H, H-2$_D$, H-4$_E$, H-5$_E$, H-6b$_D$, H-6a$_E$, H-6b$_E$), 3.66 (pt, *J* = 9.3 Hz, 1H, H-4$_D$), 3.48 (dd, $J_{2,3}$ = 9.1 Hz, 1H, H-2$_E$), 3.38−3.30 (m, 2H, H-4$_C$, H-5$_D$), 1.54 (s, 3H, H$_{iPr}$), 1.50 (s, 3H, H$_{iPr}$), 1.44 (bs, 9H, H$_{iPr}$), 1.41 (s, 3H, H$_{iPr}$), 1.40 (s, 3H, H$_{iPr}$), 1.28 (s, 3H, H$_{iPr}$), 1.24 (d, $J_{5,6}$ = 6.3 Hz, 3H, H-6$_C$); ^{13}C NMR ($CDCl_3$) δ 162.1 (NHCO), 133.4 (*C*H=CH$_2$), 118.4 (CH=*C*H$_2$), 111.5, 109.3 (2C, C$_{iPr\text{-}C}$), 99.9, 99.8 (2C, C$_{iPr\text{-}D}$), 99.4 (C-1$_D$), 98.7 (C-1$_E$), 97.9 (C-1$_C$), 92.6 (CCl_3), 82.2 (C-4$_C$), 77.3 (C-2$_E$), 76.8, 76.0, 75.5 (3C, C-2$_C$, C-3$_C$, C-3$_D$), 74.1 (C-3$_E$), 73.9 (C-4$_E$), 73.0 (C-4$_D$), 70.6 (OCH$_{2All}$), 67.7 (C-5$_D$), 65.3 (C-5$_C$), 65.2 (C-5$_E$), 62.6 (C-6$_D$), 62.3 (C-6$_E$), 59.2 (C-2$_D$), 29.2 (2C, C$_{iPr}$), 28.2, 27.1, 26.5, 26.4, 19.4, 19.3 (6C, C$_{iPr}$), 17.5 (C-6$_C$); HRMS (ESI$^+$) m/z calcd for $C_{35}H_{52}Cl_3NO_{15}Na$ [M + Na]$^+$ 854.2300, found 854.2335.

*Allyl α-D-Glucopyranosyl-(1→4)-(2,3-O-isopropylidene-α-L-rhamnopyranosyl)-(1→3)-2-deoxy-4,6-O-isopropylidene-2-trichloroacetamido-β-D-glucopyranoside (**15**).* A suspension of fully protected **14** (342 mg, 411 μmol) in AcOH/H_2O (1:1 v/v, 6 mL) was stirred at rt for 15 h. The reaction mixture was concentrated and then repeatedly coevaporated with cyclohexane and toluene. The residue was purified by column chromatography (CH_2Cl_2/MeOH, 95:5 → 80:20) to elute first tetraol **15** (63 mg, 85 μmol, 20%) as a white amorphous solid and then hexaol **17** (191 mg, 268 μmol, 65%) as a white amorphous solid: R_f = 0.66 (CH_2Cl_2/MeOH 8.5:1.5); $[\alpha]^{23}_D$ = +29 (*c* 1.0; $CHCl_3$); ^{1}H NMR (MeOD) δ 5.92−5.83 (m, 1H, C*H*=CH$_2$), 5.31−5.25 (m, J_{trans} = 17.2 Hz, 1H, CH=C*H*$_2$), 5.17−5.13 (m, J_{cis} = 10.5 Hz, 1H, CH=C*H*$_2$), 5.07 (br s, 1H, H-1$_C$), 4.92 (d, $J_{1,2}$ = 3.8 Hz, 1H, H-1$_E$), 4.69−4.67 (m, 1H, H-1$_D$), 4.33−4.27 (m, 1H, −OCH$_{2All}$), 4.12−4.05 (m, 3H, H-2$_C$, H-3$_C$, −OCH$_{2All}$,), 4.04−3.97 (m, 1H, H-5$_C$), 3.96−3.90 (m, 3H, H-2$_C$, H-3$_C$, H-6a$_D$), 3.87−3.84 (m, 2H, H-5$_E$, H-6b$_D$), 3.80−3.76 (m, 2H, H-6a$_E$, H-6b$_E$), 3.71−3.62 (m, 2H, H-4$_D$, H-3$_E$), 3.47−3.40 (m, 2H, H-2$_E$, H-4$_E$), 3.37−3.29 (m, 2H, H-4$_C$, H-5$_D$), 1.56 (s, 3H, H$_{iPr}$), 1.49 (s, 3H, H$_{iPr}$), 1.43 (s, 3H, H$_{iPr}$), 1.31 (d, $J_{5,6}$ = 6.2 Hz, 3H, H-6$_C$), 1.26 (s, 3H, H$_{iPr}$); ^{13}C NMR (MeOD) δ 164.4 (NHCO), 135.1 (*C*H=CH$_2$), 117.5 (CH=*C*H$_2$), 110.1 (C$_{iPr\text{-}C}$), 101.6 (C-1$_D$), 101.5 (C-1$_E$), 101.0 (C$_{iPr\text{-}D}$), 99.4 (C-1$_C$), 99.4 (CCl_3), 82.5 (C-4$_C$), 78.7 (C-3$_D$), 78.2 (C-3$_C$), 77.3 (C-2$_C$), 74.8 (C-3$_D$), 74.1 (C-4$_D$), 73.7 (C-2$_E$), 73.2 (C-5$_E$), 71.3 (2C, C-4$_E$, OCH$_{2All}$), 68.7 (C-5$_D$), 66.6 (C-5$_C$), 63.1 (C-6$_D$), 62.1 (C-6$_E$), 59.4 (C-2$_D$), 29.6, 28.5, 26.6, 19.3 (4C, C$_{iPr}$), 18.3 (C-6$_C$); HRMS (ESI$^+$) m/z calcd for $C_{29}H_{44}Cl_3NO_{15}Na$ [M + Na]$^+$ 774.1674, found 774.1688.

*Allyl (2,3,4,6-Tetra-O-acetyl-α-D-glucopyranosyl)-(1→4)-(2,3-O-isopropylidene-α-L-rhamnopyranosyl)-(1→3)-2-deoxy-4,6-O-isopropylidene-2-trichloroacetamido-β-D-glucopyranoside (**16**).* To a solution of tetraol **15** (267 mg, 354 μmol) in anhyd Py (10 mL), stirred under an argon atmosphere, was added DMAP (4 mg, 33 μmol, 0.1 equiv). The reaction mixture was cooled to 0 °C, acetic anhydride (1.0 mL, 10.6 mmol, 30 equiv) was added, and the reaction mixture was allowed to warm to rt. After being stirred for 2 h, the reaction mixture was cooled to 0 °C, and the reaction was quenched by slow addition of MeOH (10 mL). Volatiles were evaporated, and the remaining Py was removed by repeated coevaporation with toluene. The residue was purified by column chromatography (cHex/EtOAc, 75:25 → 60:40) to give tetra-acetate **16** (283 mg, 307 μmol, 87%) as a white amorphous solid: R_f = 0.46 (cHex/EtOAc 1:1); $[\alpha]^{23}_D$ = +49 (*c* 1.0; $CHCl_3$); ^{1}H NMR ($CDCl_3$) δ 6.75 (d, $J_{NH,2}$ = 8.7 Hz, 1H, NH), 5.87−5.78 (m, 1H, C*H*=CH$_2$), 5.47 (pt, *J* = 9.6 Hz, 1H, H-3$_E$), 5.29−5.23 (m, J_{trans} = 17.2 Hz, 1H, CH=C*H*$_2$), 5.21−5.17 (m, J_{cis} = 10.4 Hz, 1H, CH=C*H*$_2$), 5.14 (pt, *J* = 9.6 Hz, 1H, H-4$_E$), 5.08 (d, $J_{1,2}$ = 3.8 Hz, 1H, H-1$_E$), 5.05 (s, 1H, H-1$_C$), 4.92 (dd, $J_{2,3}$ = 10.2 Hz, 1H, H-2$_E$), 4.73 (d, $J_{1,2}$ = 8.4 Hz, 1H, H-1$_D$), 4.36−4.27 (m, 3H, H-5$_E$, H-6a$_E$, −OCH$_{2All}$), 4.10−4.03 (m, 5H, H-2$_C$, H-3$_C$, H-3$_D$, H-6b$_E$, −OCH$_{2All}$), 3.97−3.89 (m, 2H, H-6a$_D$, H-5$_C$), 3.84−3.73 (m, 2H, H-2$_D$, H-6b$_D$), 3.66 (pt, J = 9.3 Hz, 1H, H-4$_D$), 3.33 (dt, $J_{5,6}$ = 5.3 Hz, 1H, H-5$_D$), 3.25 (dd, $J_{4,5}$ = 10.3 Hz, $J_{3,4}$ = 7.0 Hz, 1H, H-4$_C$), 2.08, 2.02, 2.01, 2.00 (4s, 12H, H$_{Ac}$), 1.54, 1.46, 1.42, 1.39 (4s, 12H, H$_{iPr}$), 1.12 (d, $J_{5,6}$ = 6.1 Hz, 3H, H-6$_C$); ^{13}C NMR ($CDCl_3$) δ 171.0, 170.4, 169.9, 169.7 (4C, C$_{Ac}$), 162.1 (NHCO), 133.3 (*C*H=CH$_2$), 118.4 (CH=*C*H$_2$), 109.4 (C$_{iPr}$), 99.8 (C$_{iPr}$), 99.4 (C-1$_D$), 97.8 (C-1$_C$), 96.9 (C-1$_E$), 92.5 (CCl_3), 87.8

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(C-4$_C$), 77.4, 76.6, 76.0 (3C, C-2$_C$, C-3$_C$, C-3$_D$), 72.9 (C-4$_D$), 70.9 (C-2$_E$), 70.5 (OCH$_{2\ All}$), 70.4 (C-3$_E$), 68.3 (C-4$_E$), 67.6 (C-5$_D$), 67.4 (C-5$_E$), 65.1 (C-5$_C$), 62.2 (C-6$_D$), 61.4 (C-6$_E$), 59.0 (C-2$_D$), 29.2, 28.3, 26.4 (3C, C$_{iPr}$), 20.9, 20.8, 20.7, 20.6 (4C, C$_{Ac}$), 19.4 (C$_{iPr}$), 17.3 (C-6$_C$); HRMS (ESI$^+$) m/z calcd for $C_{37}H_{52}Cl_3NO_{19}Na$ [M + Na]$^+$ 942.2097, found 942.2035.

*Allyl α-D-Glucopyranosyl-(1→4)-(2,3-O-isopropylidene-α-L-rhamnopyranosyl)-(1→3)-2-deoxy-2-trichloroacetamido-β-D-glucopyranoside (**17**). Route 1.* To a solution of diol **13** (100 mg, 126 μmol) in MeCN (3 mL) were added I_2 (8 mg, 32 μmol, 0.25 equiv) and H_2O (20 μL). After the mixture was stirred for 2 h 45 at rt, Et_3N (100 μL) was added, and the volatiles were removed under vacuum. The residue was purified by column chromatography (CH_2Cl_2/MeOH, 85:15) to give monoisopropylidene **17** (69 mg, 95 μmol, 76%) as a white amorphous solid.

Route 2. MeONa (25 wt %, 230 μL, 1.01 mmol, 0.6 equiv) was added to a solution of tetraol **20** (1.54 g, 1.68 mmol) in anhyd MeOH (20 mL) stirred under an argon atmosphere. The reaction mixture was stirred for 3 h 40, and the reaction was quenched by addition of Dowex 50Wx8-200. The suspension was filtered over a pad of Celite, and volatiles were removed under vacuum. The residue was purified by column chromatography (CH_2Cl_2/MeOH, 90:10 → 80:20) to give hexaol **17** (1.12 g, 1.57 mmol, 94%) as a white amorphous solid: R_f = 0.36 (CH_2Cl_2/MeOH 4:1); $[\alpha]^{24}_D$ = +29.4 (*c* 1.0, MeOH); ^{1}H NMR (D_2O) δ 5.98−5.89 (m, 1H, C*H*═CH$_2$), 5.37−5.32 (m, J_{trans} = 17.3 Hz, 1H, CH═C*H*$_2$), 5.30−5.26 (m, J_{cis} = 10.5 Hz, 1H, CH═C*H*$_2$), 5.13 (br s, 1H, H-1$_C$), 5.07 (d, $J_{1,2}$ = 3.8 Hz, 1H, H-1$_E$), 4.79 (overlapped with D_2O, 1H, H-1$_D$), 4.41−4.36 (m, 1H, −OCH$_{2All}$), 4.28 (dd, $J_{3,4}$ = 7.5 Hz, $J_{2,3}$ = 5.4 Hz, 1H, H-3$_C$), 4.25−4.19 (m, 1H, −OCH$_{2All}$), 4.16 (d, 1H, H-2$_C$), 4.08 (dq, $J_{4,5}$ = 10.2 Hz, 1H, H-5$_C$), 3.99−3.77 (m, 7H, H-6a$_D$, H-3$_D$, H-6a$_E$, H-4$_D$, H-6b$_E$, H-2$_D$, H-6b$_D$), 3.71 (pt, *J* = 9.5 Hz, 1H, H-3$_E$), 3.60−3.50 (m, 4H, H-5$_D$, H-2$_E$, H-4$_E$, H-5$_E$), 3.45 (dd, 1H, H-4$_C$), 1.56 (s, 3H, H$_{iPr}$), 1.36 (s, 3H, H$_{iPr}$), 1.31 (d, $J_{5,6}$ = 6.3 Hz, 3H, H-6$_C$); ^{13}C NMR (D_2O) δ 167.5 (NHCO), 135.6 (*C*H═CH$_2$), 121.3 (CH═*C*H$_2$), 112.8 (C$_{iPr}$), 102.1 (C-1$_E$), 101.5 (C-1$_D$), 100.7 (C-1$_C$), 94.2 (CCl$_3$), 84.4 (C-3$_D$), 82.7 (C-4$_C$), 79.0 (C-3$_C$), 78.7 (C-5$_D$), 78.3 (C-2$_C$), 75.3 (C-3$_E$), 74.1 (2C, C-5$_E$, C-2$_E$), 73.4 (OCH$_{2All}$), 71.7 (C-4$_E$), 71.1 (C-4$_D$), 68.6 (C-5$_C$), 63.3 (C-6$_D$), 62.4 (C-6$_E$), 59.9 (C-2$_D$), 29.7, 28.1 (2C, C$_{iPr}$), 18.8 (C-6$_C$); HRMS (ESI$^+$) m/z calcd for $C_{26}H_{40}Cl_3NO_{15}Na$ [M + Na]$^+$ 734.1361, found 734.1402.

*Allyl α-L-Rhamnopyranosyl-(1→3)-6-O-acetyl-2-deoxy-2-trichloroacetamido-β-D-glucopyranoside (**18**).* Vinyl acetate (1.0 mL, 10.8. mmol, 54 equiv) and novozyme 435 (50 mg) were added to a solution of disaccharide **1** (102 mg, 0.20 mmol) in THF/Py (4:1 v/v, 5 mL) under an argon atmosphere. The reaction mixture was heated to 45 °C, stirred for 17 h, and then filtered on a pad of Celite, and the filtrate was concentrated. The residue was purified by column chromatography (CH_2Cl_2/MeOH 90:10) to give acetate **18** (97 mg, 175 μmol, 88%) as a white amorphous solid: R_f = 0.30 (CH_2Cl_2/MeOH 9:1); $[\alpha]^{23}_D$ = −45 (*c* 1.0; MeOH); ^{1}H NMR (D_2O) δ 5.97−5.87 (m, 1H, C*H*═CH$_2$), 5.36−5.31 (m, J_{trans} = 17.3 Hz, 1H, CH═C*H*$_2$), 5.29−5.26 (m, J_{cis} = 10.4 Hz, 1H, CH═C*H*$_2$), 4.91 (d, $J_{1,2}$ = 1.6 Hz, 1H, H-1$_C$), 4.80 (overlapped with D_2O, 1H, H-1$_D$), 4.47 (dd, $J_{6a,6b}$ = 12.2 Hz, $J_{5,6a}$ = 2.0 Hz, 1H, H-6a$_D$), 4.37−4.32 (m, 2H, H-6b$_D$, −OCH$_{2All}$), 4.22−4.16 (m, 1H, −OCH$_{2All}$), 4.00 (dq, $J_{4,5}$ = 9.7 Hz, 1H, H-5$_C$), 3.93−3.79 (m, 3H, H-2$_D$, H-2$_C$, H-3$_D$), 3.76−3.65 (m, 3H, H-3$_C$, H-4$_D$, H-5$_D$), 3.45 (pt, $J_{3,4}$ = 9.7 Hz, 1H, H-4$_C$), 2.17 (s, 3H, H$_{Ac}$), 1.25 (d, $J_{5,6}$ = 6.3 Hz, 3H, H-6$_C$); ^{13}C NMR (D_2O) δ 176.7 (C$_{Ac}$), 167.4 (NHCO), 135.7 (*C*H═CH$_2$), 121.5 (CH═*C*H$_2$), 103.9 (C-1$_C$), 101.7 (C-1$_D$), 94.2 (CCl$_3$), 83.1 (C-3$_D$), 76.0 (C-5$_D$), 74.5 (C-4$_C$), 73.6 (−OCH$_{2All}$), 73.2 (C-2$_C$), 73.0 (C-3$_C$), 71.6 (C-5$_C$), 71.1 (C-4$_D$), 65.8 (C-6$_D$), 59.9 (C-2$_D$), 22.8 (C$_{Ac}$), 19.1 (C-6$_C$); HRMS (ESI$^+$) m/z calcd for $C_{19}H_{28}Cl_3NO_{11}Na$ [M + Na]$^+$ 574.0626, found 574.0602.

*Allyl (6-O-Benzoyl-α-D-glucopyranosyl)-(1→4)-α-L-rhamnopyranosyl-(1→3)-6-O-benzoyl-2-deoxy-2-trichloroacetamido-β-D-glucopyranoside (**19**).* Trisaccharide **2** (1.82 g, 2.70 mmol) was suspended under an argon atmosphere in MeCN/acetone (1:1 v/v, 54 mL), and collidine (3.5 mL, 26.5 mmol, 9.8 equiv) was added. The reaction mixture was cooled to −40 °C, and benzoyl chloride was added (2.2 mL, 19.0 mmol, 7.0 equiv). After 46 h of stirring at this temperature, MeOH (20 mL) was added. After another 1 h, the reaction mixture was concentrated and repeatedly coevaporated with toluene, and the residue was purified by column chromatography (CH_2Cl_2/MeOH, 100:0 → 90:10). Fractions containing the expected dibenzoate were evaporated then taken up in CH_2Cl_2 (150 mL), washed with a 5% citric acid aq solution (2 × 20 mL) and brine (1 × 20 mL), and the aqueous layer was extracted twice with CH_2Cl_2 (2 × 10 mL). The combined organics were dried (Na_2SO_4), filtered, and concentrated to give dibenzoate **19** (1.55 g, 1.76 mmol, 65%) as a white amorphous solid: R_f = 0.36 (CH_2Cl_2/MeOH 9:1); $[\alpha]^{24}_D$ = +2.9 (*c* 1.0, MeOH); ^{1}H NMR (MeOD) δ 8.07−8.05 (m, 4H, Ph), 7.64−7.59 (m, 2H, Ph), 7.51−7.47 (m, 4H, Ph), 5.90−5.80 (m, 1H, C*H*═CH$_2$), 5.25−5.19 (m, J_{trans} = 17.3 Hz, 1H, CH═C*H*$_2$), 5.11−5.07 (m, J_{cis} = 10.6 Hz, 1H, CH═C*H*$_2$), 4.96 (d, $J_{1,2}$ = 3.9 Hz, 1H, H-1$_E$), 4.90 (d, $J_{1,2}$ = 1.5 Hz, 1H, H-1$_C$), 4.69−4.64 (m, 3H, H-6a$_E$, H-1$_D$, H-6a$_D$), 4.53 (dd, $J_{6a,6b}$ = 11.9 Hz, $J_{5,6}$ = 5.5 Hz, 1H, H-6b$_D$), 4.43 (dd, $J_{6a,6b}$ = 12.0 Hz, $J_{5,6}$ = 4.9 Hz, 1H, H-6b$_E$), 4.31−4.24 (m, 2H, H-5$_E$, −OCH$_{2All}$), 4.19 (dq, $J_{4,5}$ = 9.7 Hz, 1H, H-5$_C$), 4.09−4.04 (m, 1H, −OCH$_{2All}$), 3.88−3.80 (m, 4H, H-2$_C$, H-3$_C$, H-3$_D$, H-2$_D$), 3.68−3.64 (m, 2H, H-5$_D$, H-3$_E$), 3.60−3.55 (m, 1H, H-4$_D$), 3.51−3.43 (m, 3H, H-4$_E$, H-2$_E$, H-4$_C$), 1.34 (d, $J_{5,6}$ = 6.2 Hz, 3H, H-6$_C$); ^{13}C NMR (MeOD) δ 168.2, 167.9 (2C, C$_{Bz}$), 164.4 (NHCO), 135.2 (*C*H═CH$_2$), 134.4, 134.3 (2C, Ph), 131.4, 131.3 (2C$_{quat}$, Ph), 130.7, 130.6, 129.7, 129.6 (8C, Ph), 117.6 (CH═*C*H$_2$), 102.7 (C-1$_C$), 101.2 (C-1$_E$), 101.0 (C-1$_D$), 94.2 (CCl$_3$), 83.6 (C-4$_C$), 82.6 (C-3$_D$), 75.5 (C-5$_D$), 74.7 (C-3$_E$), 73.6 (C-2$_E$), 72.6 (C-2$_C$), 71.8 (C-4$_E$), 71.7 (C-5$_E$), 71.2 (OCH$_{2All}$), 71.1 (C-4$_D$), 70.8 (C-3$_C$), 69.0 (C-5$_C$), 65.3 (C-6$_E$), 65.0 (C-6$_D$), 58.8 (C-2$_D$), 18.1 (C-6$_C$); HRMS (ESI$^+$) m/z calcd for $C_{37}H_{44}Cl_3NO_{17}Na$ [M + Na]$^+$ 902.1572, found 902.1567.

*Allyl (6-O-Benzoyl-α-D-glucopyranosyl)-(1→4)-(2,3-O-isopropylidene-α-L-rhamnopyranosyl)-(1→3)-6-O-benzoyl-2-deoxy-2-trichloroacetamido-β-D-glucopyranoside (**20**).* To a solution of dibenzoate **19** (806 mg, 0.91 mmol) in acetone (14 mL) stirred under an argon atmosphere were added CSA (85 mg, 0.37 mmol, 0.4 equiv) and DMP (450 μL, 3.66 mmol, 4.0 equiv). After being stirred for 2 h at rt, Et_3N (100 μL, 0.72 mmol, 0.8 equiv) was added, and the reaction mixture was concentrated. The residue was purified by column chromatography (CH_2Cl_2/MeOH, 96:4 → 95:5, then 90:10) to give tetraol **20** (779 mg, 0.85 mmol, 92%) as a white amorphous solid, followed by the starting dibenzoate **19** (20 mg, 0.02 mmol, corrected yield: 95%): R_f = 0.34 (CH_2Cl_2/MeOH 9.2:0.8); $[\alpha]^{24}_D$ = +29.1 (*c* 1.0, $CHCl_3$); ^{1}H NMR (MeOD) δ 8.09−8.03 (m, 4H, Ph), 7.65−7.60 (m, 2H, Ph), 7.53−7.47 (m, 4H, Ph), 5.91−5.81 (m, 1H, C*H*═CH$_2$), 5.26−5.20 (m, J_{trans} = 17.3 Hz, 1H, CH═C*H*$_2$), 5.12−5.08 (m, J_{cis} = 10.5 Hz, 1H, CH═C*H*$_2$), 5.10 (br s, 1H, H-1$_C$), 4.93 (d, $J_{1,2}$ = 3.9 Hz, 1H, H-1$_E$), 4.70−4.61 (m, 3H, H-6a$_E$, H-1$_D$, H-6a$_D$), 4.56−4.48 (m, 2H, H-6b$_E$, H-6b$_D$), 4.30−4.25 (m, 1H, −OCH$_{2All}$), 4.16−4.05 (m, 5H, −OCH$_{2All}$, H-5$_C$, H-2$_C$, H-3$_C$, H-5$_E$), 3.91−3.82 (m, 2H, H-3$_D$, H-2$_D$), 3.68−3.63 (m, 2H, H-5$_D$, H-3$_E$), 3.61−3.54 (m, 2H, H-4$_D$, H-4$_E$), 3.43 (dd, $J_{2,3}$ = 9.5 Hz, 1H, H-2$_E$), 3.37−3.32 (m, 1H, H-4$_C$), 1.50 (s, 3H, H$_{iPr}$), 1.29 (d, $J_{5,6}$ = 6.2 Hz, 3H, H-6$_C$), 1.21 (s, 3H, H$_{iPr}$); ^{13}C NMR (MeOD) δ 168.0, 167.8 (2C, C$_{Bz}$), 164.4 (NHCO), 135.1 (*C*H═CH$_2$), 134.4, 134.3 (2C, Ph), 131.4, 131.3 (2C$_{quat}$, Ph), 130.6, 130.5, 129.6 (8C, Ph), 117.5 (CH═*C*H$_2$), 110.1 (C$_{iPr}$), 101.2 (C-1$_E$), 100.9 (C-1$_D$), 99.9 (C-1$_C$), 94.1 (CCl$_3$), 82.5 (C-3$_D$), 82.3 (C-4$_C$), 78.3 (C-2$_C$), 77.4 (C-3$_C$), 75.5 (C-5$_D$), 74.9 (C-3$_E$), 73.7 (C-2$_E$), 71.5 (C-4$_D$), 71.2 (C-5$_E$), 71.1 (OCH$_{2All}$), 70.9 (C-4$_E$), 66.9 (C-5$_C$), 65.0 (C-6$_D$), 64.5 (C-6$_E$), 58.9 (C-2$_D$), 28.6, 26.8 (2C, C$_{iPr}$), 17.7 (C-6$_C$); HRMS (ESI$^+$) m/z calcd for $C_{40}H_{48}Cl_3NO_{17}Na$ [M + Na]$^+$ 942.1885, found 942.1855.

*Allyl (2,3,4,6-Tetra-O-benzyl-α-D-glucopyranosyl)-(1→4)-(2,3-O-isopropylidene-α-L-rhamnopyranosyl)-(1→3)-4,6-di-O-benzyl-2-deoxy-2-trichloroacetamido-β-D-glucopyranoside (**3**).* NaH (272 mg, 11.3 mmol, 14 equiv) was added portionwise to a solution of hexaol **17** (578 mg, 0.81 mmol) in anhyd DMF (16 mL), stirred at −10 °C under an argon atmosphere. After 20 min, benzyl bromide (0.7 mL, 5.89 mmol, 7.3 equiv) was added dropwise. After another 4 h, while the bath temperature was slowly raised to 0 °C, MeOH (20 mL) was slowly added. The reaction mixture was acidified by dropwise addition of AcOH until pH = 6 was reached, and then volatiles were

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evaporated under vacuum and coevaporated with toluene. The residue was taken up in EtOAc (100 mL) and washed with water (2 × 15 mL). The organic layer was dried with Na_2SO_4, filtered, and concentrated. The residue was purified by column chromatography (cHex/EtOAc, 90:10 → 80:20) to give the fully protected trisaccharide **3** (795 mg, 0.63 mmol, 78%) as a white foam: R_f = 0.42 (cHex/EtOAc 7.6:2.4); $[\alpha]^{24}_D$ = +12.7 (*c* 1.0, $CHCl_3$); ^{1}H NMR ($CDCl_3$) δ 7.41−7.25 (m, 29H, Ph, NH), 7.21−7.19 (m, 2H, Ph), 5.93−5.84 (m, 1H, C*H*═CH_2), 5.31−5.26 (m, J_{trans} = 17.2 Hz, 1H, CH═CH_2), 5.29 (s, 1H, H-1_C), 5.21−5.17 (m, J_{cis} = 10.5 Hz, 1H, CH═CH_2), 5.00 (d, $J_{1,2}$ = 3.5 Hz, 1H, H-1_E), 4.96−4.52 (m, 13H, $12H_{Bn}$, H-1_D), 4.39−4.34 (m, 1H, −OCH_{2All}), 4.13 (pt, *J* = 6.2 Hz, 1H, H-3_D), 4.10−4.01 (m, 5H, H-2_D, H-2_C, H-3_C, H-5_E, −OCH_{2All}), 3.98 (pt, *J* = 9.5 Hz, 1H, H-3_E), 3.93−3.89 (m, 1H, H-5_D), 3.86−3.80 (m, 5H, H-5_C, H-4_E, H-$6a_E$, H-$6a_D$, H-$6b_D$), 3.73 (pt, *J* = 5.9 Hz, 1H, H-4_D), 3.67 (dd, $J_{6a,6b}$ = 10.4 Hz, $J_{5,6b}$ = 1.9 Hz, 1H, H-$6b_E$), 3.62 (dd, $J_{2,3}$ = 9.8 Hz, 1H, H-2_E), 3.38−3.32 (m, 1H, H-4_C), 1.45 (s, 3H, H_{iPr}), 1.28 (s, 3H, H_{iPr}), 1.25 (d, $J_{5,6}$ = 6.3 Hz, 3H, H-6_C); ^{13}C NMR ($CDCl_3$) δ 161.7 (NHCO), 138.9, 138.5, 138.1 ($3C_{quat}$, Ph), 138.0 ($2C_{quat}$, Ph), 137.2 (C_{quat}, Ph), 133.7 (*C*H═CH_2), 128.7, 128.6, 128.5, 128.4, 128.3, 128.2, 128.1, 128.0, 127.9, 127.8, 127.7, 127.6 (30C, Ph), 117.5 (CH═CH_2), 109.1 (C_{iPr}), 98.8 (C-1_E), 98.4 (C-1_D), 96.2 (C-1_C), 92.5 (CCl_3), 82.3 (C-3_E), 81.2 (C-4_C), 80.1 (C-2_E), 78.0 (C-4_E), 76.8 (C-4_D), 76.1, 76.0 (2C, C-2_C, C-3_C), 75.6, 75.2 (2C, C_{Bn}), 74.3 (C-5_D), 74.2, 73.9, 73.7, 73.6 (4C, C_{Bn}), 73.5 (C-3_D), 70.6 (C-5_E), 69.8 (C-6_D), 69.7 (OCH_{2All}), 68.1 (C-6_E), 66.2 (C-5_C), 53.7 (C-2_D), 28.2, 26.5 (2C, C_{iPr}), 17.4 (C-6_C); HRMS (ESI$^+$) *m*/*z* calcd for $C_{68}H_{76}Cl_3NO_{15}Na$ $[M + Na]^+$ 1274.4178, found 1274.4186.

2,3,4,6-Tetra-O-benzyl-α-D-glucopyranosyl-(1→4)-(2,3-O-isopropylidene-α-L-rhamnopyranosyl)-(1→3)-4,6-di-O-benzyl-2-deoxy-2-trichloroacetamido-α/β-D-glucopyranose (**21**). 1,5-Cyclooctadiene−bis(methyldiphenylphosphine)iridium hexafluorophosphate (14 mg, 17 μmol, 0.03 equiv) was dissolved in THF (5 mL) under an argon atmosphere. Hydrogen was bubbled through the solution for 20 min, causing the color to change from red to yellow. The solution was degassed by complete evaporation of the solvent under vacuum. The activated iridium complex was dissolved in THF (2 mL) under an argon atmosphere, and a solution of allyl glycoside **3** (619 mg, 494 μmol) in THF (8 mL) was added. The reaction mixture was stirred for 2 h 30 at rt, and then a solution of iodine (238 mg, 938 μmol, 1.9 equiv) in THF/water (10 mL, 4:1 v/v) was added. After 5 h, the excess iodine was quenched by addition of 10% aq sodium bisulfite (10 mL). The reaction mixture was concentrated under reduced pressure to remove THF, water (5 mL) was added, and the aqueous phase was extracted with CH_2Cl_2 (2 × 40 mL). The combined organics were dried (Na_2SO_4), filtered, and concentrated to dryness. Column chromatography of the residue (toluene/EtOAc, 90:10 → 80:20) afforded lactol **21** (480 mg, 396 μmol, 80%) as a colorless oil (α/β, 9:1): R_f = 0.32 (α), 0.12 (β) (toluene/EtOAc 4:1). α: ^{1}H NMR ($CDCl_3$) δ 7.37−7.14 (m, 30H, Ph), 7.04 (d, $J_{NH,2}$ = 9.3 Hz, 1H, NH), 5.26−5.24 (m, 2H, H-1_D, H-1_C), 4.96 (d, $J_{1,2}$ = 3.5 Hz, 1H, H-1_E), 4.96−4.48 (m, 12H, H_{Bn}), 4.24 (pdt, $J_{2,3}$ = 9.8 Hz, $J_{1,2}$ = 3.5 Hz, 1H, H-2_D), 4.16−3.90 (m, 7H, H-3_D, H-2_C, H-3_C, H-5_D, H-5_E, H-3_E, H-5_C), 3.81−3.76 (m, 2H, H-$6a_E$, H-4_E), 3.72 (dd, $J_{6a,6b}$ = 10.6 Hz, $J_{5,6a}$ = 4.7 Hz, 1H, H-$6a_D$), 3.66−3.60 (m, 3H, H-$6b_E$, H-$6b_D$, H-4_D), 3.58 (dd, $J_{2,3}$ = 9.7 Hz, 1H, H-2_E), 3.37 (dd, $J_{4,5}$ = 10.2 Hz, $J_{3,4}$ = 7.2 Hz, 1H, H-4_C), 3.20 (bs, 1H, OH), 1.42 (s, 3H, H_{iPr}), 1.23 (s, 3H, H_{iPr}), 1.16 (d, $J_{5,6}$ = 6.2 Hz, 3H, H-6_C); ^{13}C NMR ($CDCl_3$) δ 161.9 (NHCO), 138.9, 138.5 ($2C_{quat}$, Ph), 138.1 ($2C_{quat}$, Ph), 137.5, 137.4 ($2C_{quat}$, Ph), 128.6, 128.5, 128.4, 128.3, 128.2, 128.0, 127.9, 127.8, 127.7, 127.6 (30C, Ph), 109.0 (C_{iPr}), 98.8 (C-1_E), 97.7 (C-1_C), 92.4 (CCl_3), 91.4 (C-1_D), 82.2 (C-3_E), 81.6 (C-4_C), 80.1 (C-2_E), 78.0 (C-4_E), 77.4 (C-4_D), 76.8 (C-2_C), 76.2 (C-3_C), 75.7 (C_{Bn}), 75.5 (C-3_D), 75.4, 75.2, 73.9, 73.6, 73.5 (5C, C_{Bn}), 70.9 (C-5_D), 70.6 (C-5_E), 68.7 (C-6_D), 68.1 (C-6_E), 66.2 (C-5_C), 55.8 (C-2_D), 28.2, 26.5 (2C, C_{iPr}), 17.2 (C-6_C); HRMS (ESI$^+$) *m*/*z* calcd for $C_{65}H_{72}Cl_3NO_{15}Na$ $[M + Na]^+$ 1234.3865, found 1234.3800.

2,3,4,6-Tetra-O-benzyl-α-D-glucopyranosyl-(1→4)-(2,3-O-isopropylidene-α-L-rhamnopyranosyl)-(1→3)-4,6-di-O-benzyl-2-deoxy-2-trichloroacetamido-α/β-D-glucopyranosyl Trichloroacetimidate (**22**). To a solution of lactol **21** (426 mg, 0.34 mmol) in anhyd DCE (4 mL) stirred at −10 °C and under argon atmosphere were added trichloroacetonitrile (170 μL, 1.70 mmol, 5.0 equiv) and DBU (15 μL, 0.10 mmol, 0.3 equiv). The reaction mixture was stirred for 35 min at −10 °C and volatiles were evaporated at rt under reduced pressure. The residue was purified by column chromatography (toluene/EtOAc, 90:10 + 1% Et_3N) to give imidate **22** (322 mg, 0.24 mmol, 70%) as a white amorphous solid (α/β ratio 9:1), followed by lactol **21** (118 mg, 0.09 mmol, corrected yield: 93%). R_f = 0.35 (toluene/EtOAc 9:1 + 1% Et_3N). α: ^{1}H NMR ($CDCl_3$) δ 8.78 (s, 1H, C═NH), 7.39−7.19 (m, 30H, Ph), 7.10 (d, $J_{NH,2}$ = 8.9 Hz, 1H, NH), 6.45 (d, $J_{1,2}$ = 3.6 Hz, 1H, H-1_D), 5.31 (bs, 1H, H-1_C), 4.99 (d, $J_{1,2}$ = 3.3 Hz, 1H, H-1_E), 4.99 (d, *J* = 10.9 Hz, 1H, H_{Bn}), 4.90−4.45 (m, 12H, $11H_{Bn}$, H-2_D), 4.21 (pt, *J* = 9.6 Hz, 1H, H-3_D), 4.17−4.11 (m, 2H, H-2_C, H-3_C), 4.06−3.92 (m, 5H, H-5_D, H-5_E, H-3_E, H-4_D, H-5_C), 3.90−3.79 (m, 3H, H-$6a_D$, H-$6a_E$, H-4_E), 3.73 (dd, $J_{6a,6b}$ = 10.9 Hz, $J_{5,6b}$ = 1.8 Hz, 1H, H-$6b_D$), 3.67 (dd, $J_{6a,6b}$ = 10.5 Hz, $J_{5,6b}$ = 1.7 Hz, 1H, H-$6b_E$), 3.62 (dd, $J_{2,3}$ = 9.8 Hz, 1H, H-2_E), 3.30 (dd, $J_{4,5}$ = 10.2 Hz, $J_{3,4}$ = 7.1 Hz, 1H, H-4_C), 1.45 (s, 3H, H_{iPr}), 1.27 (s, 3H, H_{iPr}), 1.24 (d, $J_{5,6}$ = 6.2 Hz, 3H, H-6_C). ^{13}C NMR ($CDCl_3$) δ 161.9 (NHCO), 160.3 (C═NH), 138.9, 138.6 ($2C_{quat}$, Ph), 138.1 ($2C_{quat}$, Ph), 137.8, 137.5 ($2C_{quat}$, Ph), 128.7, 128.6, 128.5, 128.4, 128.3, 128.2, 128.0, 127.9, 127.8, 127.7, 127.6 (30C, Ph), 109.3 (C_{iPr}), 98.9 (C-1_E), 97.6 (C-1_C), 94.9 (C-1_D), 92.2 (C(O)CCl_3), 90.9 (C(NH)CCl_3), 82.2 (C-3_E), 81.2 (C-4_C), 80.1 (C-2_E), 78.0 (C-4_E), 77.0 (C-3_C), 76.2 (2C, C-4_D, C-2_C), 75.7, 75.6, 75.2 (3C, C_{Bn}), 75.0 (C-3_D), 74.2 (C_{Bn}), 73.8 (C-5_D), 73.7, 73.6 (2C, C_{Bn}), 70.6 (C-5_E), 68.2 (C-6_E), 67.9 (C-6_D), 66.8 (C-5_C), 55.2 (C-2_D), 28.2, 26.4 (2C, C_{iPr}), 17.3 (C-6_C).

Allyl (2,3,4,6-Tetra-O-benzyl-α-D-glucopyranosyl)-(1→4)-(2,3-O-isopropylidene-α-L-rhamnopyranosyl)-(1→3)-(4,6-di-O-benzyl-2-deoxy-2-trichloroacetamido-β-D-glucopyranosyl)-(1→2)-(3,4-di-O-benzyl-α-L-rhamnopyranosyl)-(1→2)-3,4-di-O-benzyl-α-L-rhamnopyranoside (**24**). A suspension of donor **22** (100 mg, 73.6 μmol), acceptor **23** (57 mg, 80.1 μmol, 1.1 equiv), and freshly activated 4 Å MS (200 mg) in anhyd DCE (1 mL), was stirred for 15 min at rt under an argon atmosphere. The reaction mixture was cooled to −35 °C, and TMSOTf (2 μL, 7.7 μmol, 0.1 equiv) was added. After themixture was stirred for 30 min at this temperature, Et_3N (10 μL) was added, and then the reaction mixture was filtered on a pad of Celite. The filtrate was evaporated, and the residue was purified by column chromatography (toluene/EtOAc 95:5) to give pentasaccharide **24** (110 mg, 57.6 μmol, 78%) as a white amorphous solid: R_f = 0.28 (toluene/EtOAc 9:1); $[\alpha]^{24}_D$ = +5.9 (*c* 1.0; $CHCl_3$). ^{1}H NMR ($CDCl_3$) δ 7.41−7.18 (m, 50H, Ph), 7.06 (d, $J_{NH,2}$ = 8.0 Hz, 1H, NH), 5.93−5.84 (m, 1H, C*H*═CH_2), 5.29−5.24 (m, 2H, CH═CH_2, H-1_C), 5.21−5.18 (m, J_{cis} = 10.4 Hz, 1H, CH═CH_2), 5.04 (d, $J_{1,2}$ = 1.5 Hz, 1H, H-1_A), 5.00 (d, $J_{1,2}$ = 3.5 Hz, 1H, H-1_E), 4.94−4.39 (m, 22H, $20H_{Bn}$, H-1_D, H-1_B), 4.16−4.11 (m, 1H, −OCH_{2All}), 4.10−4.03 (m, 5H, H-2_D, H-5_E, H-2_A, H-3_C, H-2_C), 3.99−3.87 (m, 5H, H-2_B, H-3_A, H-3_D, H-3_E, −OCH_{2All}), 3.85−3.77 (m, 5H, H-$6a_E$, H-4_E, H-5_A, H-5_C, H-3_B), 3.72−3.56 (m, 6H, H-5_D, H-2_E, H-$6a_D$, H-5_B, H-4_D, H-$6b_E$), 3.52 (dd, $J_{6a,6b}$ = 9.6 Hz, $J_{5,6b}$ = 3.6 Hz, 1H, H-$6b_D$), 3.47 (pt, *J* = 9.4 Hz, 1H, H-4_A), 3.37 (pt, *J* = 9.4 Hz, 1H, H-4_B), 3.29−3.25 (m, 1H, H-4_C), 1.43 (s, 3H, H_{iPr}), 1.31 (d, $J_{5,6}$ = 6.2 Hz, 6H, H-6_A, H-6_B), 1.23 (s, 3H, H_{iPr}), 1.19 (d, $J_{5,6}$ = 6.2 Hz, 3H, H-6_C); ^{13}C NMR ($CDCl_3$) δ 161.7 (NHCO), 138.9, 138.7, 138.6, 138.5, 138.3, 138.2, 138.1, 138.0, 137.5 ($10C_{quat}$, Ph), 133.9 (*C*H═CH_2), 128.7, 128.6, 128.5, 128.4, 128.3, 128.2, 128.1, 128.0, 127.9, 127.8, 127.7, 127.6, 127.5 (50C, Ph), 117.2 (CH═CH_2), 109.0 (C_{iPr}), 101.5 (C-1_A), 101.2 (C-1_D, $^1J_{CH}$ = 162.3 Hz), 98.8 (C-1_E), 97.9 (C-1_B), 96.9 (C-1_C), 92.5 (CCl_3), 82.2 (C-3_E), 81.6 (C-4_C), 81.1 (C-4_A), 80.6 (C-4_B), 80.2 (C-2_E), 79.5 (C-3_A), 79.2 (C-3_B), 78.0 (C-4_E), 76.9, 76.7, 76.4, 76.2, 76.1, 76.0, 75.8 (7C, C-5_D, C-4_D, C-3_D, C-3_C, C-2_C, C-2_B, C-2_A), 75.6, 75.5, 75.4, 75.1, 74.1, 74.0, 73.6, 73.5, 73.4, 71.5 (10C, C_{Bn}), 70.6 (C-5_E), 69.3 (C-6_D), 68.5 (C-5_B), 68.2 (C-6_E), 67.8 (2C, C-5_A, OCH_{2All}), 66.0 (C-5_C), 56.4 (C-2_D), 28.3 (C_{iPr}), 26.6 (C_{iPr}), 18.1 (2C, C-6_A, C-6_B), 17.3 (C-6_C); HRMS (ESI$^+$) *m*/*z* calcd for $C_{108}H_{120}Cl_3NO_{23}Na$ $[M + Na]^+$ 1926.7214, found 1926.7100.

Allyl (2,3,4,6-Tetra-O-benzyl-α-D-glucopyranosyl)-(1→4)-(α-L-rhamnopyranosyl)-(1→3)-4,6-di-O-benzyl-2-deoxy-2-trichloroace-

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*tamido-β-D-glucopyranoside (**25**).* To a solution of fully protected trisaccharide **3** (657 mg, 0.52 mmol) in CH_2Cl_2 (7 mL) was added TFA (50% aq, 3 mL). The biphasic mixture was vigorously stirred for 75 min and then repeatedly coevaporated with toluene and cyclohexane. The residue was purified by column chromatography (cHex/EtOAc, 70:30 → 65:35) to give diol **25** (601 mg, 0.50 mmol, 94%) as a white amorphous solid: R_f = 0.26 (cHex/EtOAc 6.5:3.5); $[\alpha]^{24}_D$ = −4.1 (*c* 1.0; $CHCl_3$); ^{1}H NMR ($CDCl_3$) δ 7.40−7.22 (m, 29H, Ph, NH), 7.18−7.15 (m, 2H, Ph), 5.95−5.85 (m, 1H, C*H*=CH_2), 5.33−5.28 (m, J_{trans} = 17.2 Hz, 1H, CH=CH_2), 5.23−5.17 (m, J_{cis} = 10.5 Hz, 1H, CH=CH_2), 5.16 (bs, 1H, H-1_C), 4.95 (d, $J_{1,2}$ = 3.7 Hz, 1H, H-1_E), 4.92 (d, *J* = 11.0 Hz, 1H, H_{Bn}), 4.86 (d, *J* = 10.9 Hz, 1H, H_{Bn}), 4.78−4.49 (m, 11H, 10 H_{Bn}, H-1_D), 4.39−4.35 (m, 1H, −OCH_{2All}), 4.15 (pt, *J* = 7.1 Hz, 1H, H-3_D), 4.11−3.98 (m, 4H, H-2_C, H-2_D, H-3_E, −OCH_{2All}), 3.97−3.75 (m, 7H, H-4_D, H-3_C, H-$6a_D$, H-5_D, H-$6b_D$, H-5_E, H-5_C), 3.69−3.58 (m, 3H, H-2_E, H-$6a_E$, H-$6b_E$), 3.52 (pt, *J* = 9.5 Hz, 1H, H-4_E), 3.41 (pt, *J* = 9.1 Hz, 1H, H-4_C), 2.81 (bs, 1H, OH), 1.33 (d, $J_{5,6}$ = 6.1 Hz, H-6_C); ^{13}C NMR ($CDCl_3$) δ 161.8 (NHCO), 138.7, 138.1, 138.0, 137.9, 137.5, 137.4 (6C_{quat}, Ph), 133.7 (CH=CH_2), 128.6, 128.5, 128.2, 128.0, 127.9, 127.8, 127.7 (30C, Ph), 117.7 (CH=CH_2), 99.4 (C-1_E), 98.8 (C-1_C), 98.5 (C-1_D), 92.5 (CCl_3), 86.0 (C-4_C), 81.6 (C-3_E), 79.9 (C-2_E), 77.9 (C-4_E), 76.4 (C-4_D), 75.7, 75.2 (2C, C_{Bn}), 75.1 (C-3_D), 74.4 (C-5_D), 74.1, 73.7, 73.6, 73.5 (4C, C_{Bn}), 71.4 (C-5_E), 70.8, 70.0 (2C, C-2_C, C-3_C), 69.9 (OCH_{2All}), 69.3 (C-6_D), 68.7 (C-6_E), 67.1 (C-5_C), 55.2 (C-2_D), 17.8 (C-6_C); HRMS (ESI^+) *m*/*z* calcd for $C_{65}H_{72}Cl_3NO_{15}Na$ $[M + Na]^+$ 1234.3865, found 1234.3870.

*Allyl (2,3,4,6-Tetra-O-benzyl-α-D-glucopyranosyl)-(1→4)-(2-O-benzoyl-α-L-rhamnopyranosyl)-(1→3)-4,6-di-O-benzyl-2-deoxy-2-trichloroacetamido-β-D-glucopyranoside (**26**).* To a solution of diol **25** (99 mg, 82 μmol) in anhyd CH_2Cl_2 (200 μL) and under an argon atmosphere were added trimethyl orthobenzoate (96 μL, 559 μmol, 6.8 equiv) and *p*-TSA (1 mg, 5 μmol, 0.06 equiv). The reaction mixture was stirred for 20 min at rt, and 50% aq TFA (1 mL) was added. The reaction mixture was stirred for another 45 min before being repeatedly coevaporated with toluene. The residue was purified by column chromatography (cHex/EtOAc, 85:15 → 70:30) to give alcohol **26** (89 mg, 68 μmol, 82%) as a white amorphous solid: R_f = 0.44 (cHex/EtOAc 7:3); $[\alpha]^{24}_D$ = +16.8 (*c* 1.0; $CHCl_3$); ^{1}H NMR ($CDCl_3$) δ 8.06−8.04 (m, 2H, H_{Bz}), 7.63−7.58 (m, 1H, H_{Bz}), 7.50−7.46 (m, 2H, H_{Bz}), 7.39−7.19 (m, 29H, Ph, NH), 7.17−7.13 (m, 2H, Ph), 5.96−5.86 (m, 1H, C*H*=CH_2), 5.44−5.43 (m, 1H, H-2_C), 5.33−5.27 (m, J_{trans} = 17.2 Hz, 1H, CH=CH_2), 5.33−5.19 (m, 2H, H-1_C, CH=CH_2), 4.96 (d, $J_{1,2}$ = 3.7 Hz, 1H, H-1_E), 4.92−4.43 (m, 13H, 12H_{Bn}, H-1_D), 4.40−4.34 (m, 1H, −OCH_{2All}), 4.28−4.22 (m, 1H, H-3_D), 4.11−3.97 (m, 5H, H-5_C, H-5_E, H-3_C, −OH, −OCH_{2All}), 3.92−3.74 (m, 6H, H-4_D, H-5_D, H-$6a_D$, H-$6b_D$, H-3_E, H-2_D), 3.65−3.55 (m, 4H, H-4_E, H-2_E, H-$6a_E$, H-$6b_E$), 3.46 (pt, *J* = 9.0 Hz, 1H, H-4_C), 1.37 (d, $J_{5,6}$ = 6.2 Hz, H-6_C); ^{13}C NMR ($CDCl_3$) δ 165.9 (C_{Bz}), 161.9 (NHCO), 138.8, 138.2, 138.1, 138.0, 137.7, 137.5 (6C_{quat}, Ph), 133.7 (CH=CH_2), 133.3 (C_{Bz}), 130.1 (C_{quat}, C_{Bz}), 130.0 (C_{Bz}), 128.7, 128.6, 128.5, 128.2, 128.1, 128.0, 127.9, 127.8, 127.7 (32C, Ph), 117.8 (CH=CH_2), 99.3 (C-1_E), 98.2 (C-1_D), 97.2 (C-1_C), 92.5 (CCl_3), 85.7 (C-4_C), 81.8 (C-3_E), 80.1 (C-2_E), 77.9 (C-4_E), 76.7 (C-4_D), 75.7 (C_{Bn}), 75.5 (C-3_D), 75.2 (C_{Bn}), 74.4 (C-5_D), 74.2, 73.9, 73.7, 73.6 (4C, C_{Bn}), 72.6 (C-2_C), 71.5 (C-5_E), 70.1 (OCH_{2All}), 69.3 (C-6_D), 68.6 (C-6_E), 68.5 (C-3_C), 67.7 (C-5_C), 55.9 (C-2_D), 18.1 (C-6_C); HRMS (ESI^+) *m*/*z* calcd for $C_{72}H_{76}Cl_3NO_{16}$ $[M + H]^+$ 1316.4308, found 1316.4205.

*Allyl (2,3,4-Tri-O-acetyl-α-L-rhamnopyranosyl)-(1→3)-[2,3,4,6-tetra-O-benzyl-α-D-glucopyranosyl)-(1→4)]-(2-O-benzoyl-α-L-rhamnopyranosyl)-(1→3)-4,6-di-O-benzyl-2-deoxy-2-trichloroacetamido-β-D-glucopyranoside (**36**).* A suspension of acceptor **26** (82 mg, 62.2 μmol), the known rhamnosyl donor[79] **34** (40 mg, 92.0 μmol, 1.5 equiv), and freshly activated 4 Å MS (100 mg) in anhyd toluene (2 mL) was heated to 70 °C under an argon atmosphere. TMSOTf (1.0 μL, 5.4 μmol, 0.09 equiv) was added to the reaction mixture, which was stirred at 70 °C for 60 min and then cooled to rt. Et_3N (5 μL) was added, and the mixture was filtered over a pad of Celite. The filtrate was concentrated to dryness, and the residue was purified by column chromatography (toluene/EtOAc, 95:5 → 85:15) to give tetrasaccharide **36** (20 mg, 12.6 μmol, 20%) as a white amorphous solid along with recovered acceptor **26** (57 mg, 43.5 μmol, 70%): R_f = 0.44 (toluene/EtOAc 4:1); $[\alpha]^{24}_D$ = −4.0 (*c* 1.0; $CHCl_3$); ^{1}H NMR ($CDCl_3$) δ 8.10−8.07 (m, 2H, H_{Bz}), 7.64−7.60 (m, 1H, H_{Bz}), 7.51−7.47 (m, 2H, H_{Bz}), 7.39−7.26 (m, 28H, Ph, NH), 7.20−7.16 (m, 3H, Ph), 5.93−5.86 (m, 1H, C*H*=CH_2), 5.48 (br s, 1H, H-2_C), 5.34−5.16 (m, 6H, CH=CH_2, H-1_C, H-2_B, H-3_B, H-1_E), 5.06 (br s, 1H, H-1_B), 4.97−4.90 (m, 2H, H_{Bn}, H-4_B), 4.86−4.50 (m, 11H, H_{Bn}), 4.41 (d, *J* = 12.1 Hz, 1H, H_{Bn}), 4.37−4.25 (m, 3H, −OCH_{2All}, H-3_C, H-3_D), 4.10−3.90 (m, 5H, −OCH_{2All}, H-2_D, H-3_E, H-5_C, H-5_E), 3.87−3.59 (m, 10H, H-2_E, H-4_C, H-4_D, H-4_E, H-5_B, H-5_D, H-$6a_D$, H-$6b_D$, H-$6a_E$, H-$6b_E$), 2.10 (s, 3H, H_{Ac}), 1.88, 1.86 (2s, 6H, H_{Ac}), 1.23 (d, $J_{5,6}$ = 5.9 Hz, 1H, H-6_C), 0.91 (br s, 3H, H-6_B); ^{13}C NMR (partial, $CDCl_3$) δ 170.0, 169.8 (3C, C_{Ac}), 165.8 (C_{Bz}), 161.8 (NHCO), 139.0, 138.8, 138.5, 138.3, 138.2, 137.5 (6C_{quat}, Ph), 133.8 (CH=CH_2), 133.5 (C_{Bz}), 130.0 (C_{Bz}), 129.8 (C_{quat}, C_{Bz}), 128.6, 128.5, 128.4, 128.3, 128.1, 128.0, 127.9, 127.8, 127.7, 127.6, 127.5 (32C, Ph), 117.8 (CH=CH_2), 98.2 (C-1_D), 96.1 (C-1_C), 92.6 (CCl_3), 81.8 (C-3_E), 81.0 (C-2_E), 78.0 (C-4_E), 76.3 (C-4_D), 75.6 (C_{Bn}), 75.0 (C_{Bn}), 74.5 (C-5_D), 74.1, 73.8, 73.6, 73.2 (4C, C_{Bn}), 73.0 (C-2_B), 71.6 (C-5_E), 71.1 (C-4_B), 70.1 (OCH_{2All}), 69.9 (C-2_C), 69.4 (C-6_E), 69.1 (C-3_B), 69.0 (C-6_D), 68.2 (C-5_C), 67.2 (C-5_B), 56.1 (C-2_D), 21.0, 20.8, 20.7 (3C, C_{Ac}), 18.9 (C-6_C), 17.2 (C-6_B); HRMS (ESI^+) *m*/*z* calcd for $C_{84}H_{92}Cl_3NO_{23}Na$ $[M + Na]^+$ 1610.5023, found 1610.5002.

*Allyl (3,4-Di-O-benzyl-2-O-levulinyl-α-L-rhamnopyranosyl)-(1→2)-[2,3,4,6-tetra-O-benzyl-α-D-glucopyranosyl)-(1→4)]-(α-L-rhamnopyranosyl)-(1→3)-4,6-di-O-benzyl-2-deoxy-2-trichloroacetamido-β-D-glucopyranoside (**37**).* A suspension of diol **25** (144 mg, 119 μmol), the known rhamnosyl donor **28**[71] (71 mg, 121 μmol, 1.0 equiv), and freshly activated 4 Å MS (160 mg) in anhyd toluene (2.4 mL) was stirred for 10 min under an argon atmosphere. The reaction mixture was cooled to −15 °C, and TMSOTf (1.5 μL, 5.8 μmol, 0.05 equiv) was added. After the mixture was stirred for 10 min at −15 °C, Et_3N (10 μL) was added, and the reaction mixture was filtered over a pad of Celite and concentrated to dryness. The residue was purified by column chromatography (toluene/EtOAc, 84:16) to give tetrasaccharide **37** (161 mg, 98.3 μmol, 80%) as a white amorphous solid contaminated with trichloroacetamide: R_f = 0.33 (toluene/EtOAc 4:1); ^{1}H NMR ($CDCl_3$) δ 7.49−7.11 (m, 41H, Ph, NH), 5.92−5.82 (m, 1H, C*H*=CH_2), 5.50 (dd, $J_{2,3}$ = 2.9 Hz, $J_{1,2}$ = 1.9 Hz, 1H, H-2_B), 5.29−5.24 (m, J_{trans} = 17.3 Hz, 1H, CH=CH_2), 5.19−5.15 (m, J_{cis} = 10.4 Hz, 1H, CH=CH_2), 5.11 (bs, 1H, H-1_C), 5.00 (br s, 1H, H-1_C), 4.97 (d, $J_{1,2}$ = 3.7 Hz, 1H, H-1_E), 4.91−4.86 (m, 2H, H_{Bn}), 4.81 (d, *J* = 10.8 Hz, 1H, H_{Bn}), 4.75−4.67 (m, 6H, H-1_D, H_{Bn}), 4.63−4.45 (m, 8H, H_{Bn}), 4.34−4.29 (m, 1H, −OCH_{2All}), 4.08−4.01 (m, 3H, H-3_D, H-5_E, −OCH_{2All}), 3.94−3.78 (m, 11H, H-2_C, H-2_D, H-3_B, H-3_E, H-5_B, H-5_C, H-5_D, H-$6a_D$, H-$6b_D$, H-$6a_E$, H-$6b_E$), 3.72 (pt, *J* = 5.9 Hz, 1H, H-4_D), 3.66−3.63 (m, 2H, H-3_C, OH), 3.60−3.54 (m, 2H, H-2_E, H-4_E), 3.39 (pt, *J* = 9.4 Hz, 1H, H-4_B), 3.29 (pt, *J* = 9.2 Hz, 1H, H-4_B), 2.75−2.67 (m, 4H, 2 × CH_{2Lev}), 2.16 (s, 3H, CH_{3Lev}), 1.32 (d, $J_{5,6}$ = 6.1 Hz, 3H, H-6_C), 1.28 (d, $J_{5,6}$ = 6.2 Hz, 3H, H-6_B); ^{13}C NMR ($CDCl_3$) δ 206.5 (C_{Lev}), 171.9 (C_{Lev}), 161.8 (NHCO), 138.8, 138.6, 138.3, 138.2, 138.1, 138.0, 137.8, 137.3 (8C_{quat}, Ph), 133.7 (CH=CH_2), 128.6, 128.5, 128.4, 128.2, 128.1, 128.0, 127.9, 127.8, 127.7 (40C, Ph), 117.8 (CH=CH_2), 99.8 (C-1_B), 99.0 (C-1_E), 98.3 (C-1_D), 98.0 (C-1_C), 92.4 (CCl_3), 85.4 (C-4_C), 81.8 (C-3_E), 80.1, 80.0 (2C, C-2_E,C-4_B), 77.9 (2C, C-2_C, C-3_B), 77.8 (C-4_E), 76.3 (C-4_D), 75.6, 75.5, 75.2 (3C, C_{Bn}), 74.4 (C-3_D), 74.1 (C-5_D), 73.8, 73.7, 73.6 (4C, C_{Bn}), 71.8 (C_{Bn}), 71.3 (C-5_E), 69.9 (OCH_{2All}), 69.8 (C-3_C), 69.6 (C-6_D), 69.1 (C-2_B), 68.6 (C-6_E), 68.5 (C-5_B), 67.8 (C-5_C), 53.7 (C-2_D), 38.2 (CH_{2Lev}), 30.0 (CH_{3Lev}), 28.3 (CH_{2Lev}), 18.1, 18.0 (2C, C-6_B, C-6_C); HRMS (ESI^+) *m*/*z* calcd for $C_{90}H_{100}Cl_3NO_{21}Na$ $[M + Na]^+$ 1658.5751, found 1658.5919.

*(3,4-Di-O-benzyl-2-levulinyl-α-L-rhamnopyranosyl)-(1→2)-(3,4-di-O-benzyl-α-L-rhamnopyranosyl)-(1→3)-[2,3,4,6-tetra-O-benzyl-α-D-glucopyranosyl)-(1→4)]-(2-O-acetyl-α-L-rhamnopyranosyl)-(1→3)-4,6-di-O-benzyl-2-deoxy-2-trichloroacetamido-α/β-D-glucopyranosyl trichloroacetimidate (**40**).* To a solution of hemiacetal **39** (2.24 g, 1.14 mmol) in anhyd DCE (6 mL) and under argon

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atmosphere was added DBU (51 μL, 0.34 mmol, 0.3 equiv). The reaction mixture was cooled to 0 °C, and then trichloroacetonitrile (570 μL, 5.68 mmol, 5.0 equiv) was added. After 40 min of stirring at 0 °C, the volatiles were partially evaporated at room temperature, and the residue was purified by column chromatography (toluene/EtOAc, 90:10 + 2% Et_3N) to give imidate **40** (2.36 g, 1.12 mmol, 97%) as a white foam (α/β ratio 9:1): R_f = 0.56 (toluene/EtOAc 8.6:1.4 + 2% Et_3N); ^{1}H NMR ($CDCl_3$) δ (α) 8.77 (s, 1H, C═NH), 7.37−7.13 (m, 50H, Ph), 6.88 (d, $J_{NH,2}$ = 9.0 Hz, NH), 6.43 (d, $J_{1,2}$ = 2.9 Hz, 1H, H-1_D), 5.56 (bs, 1H, H-2_A), 5.19 (d, $J_{1,2}$ = 3.2 Hz, 1H, H-1_C), 5.12 (bs, 1H, H-2_C), 5.05 (bs, 1H, H-1_A), 4.97−4.36 (m, 24H, 20H$_{Bn}$, H-1_B, H-1_E, H-2_B, H-2_D), 4.20−3.37 (m, 20H, H-2_E, H-3_A, H-3_B, H-3_C, H-3_D, H-3_E, H-4_A, H-4_B, H-4_C, H-4_D, H-4_E, H-5_A, H-5_B, H-5_C, H-5_D, H-5_E, H-$6a_D$, H-$6b_D$, H-$6a_E$, H-$6b_E$), 2.68−2.53 (m, 4H, 2 × CH_{2Lev}), 2.11, 2.10 (bs, 6H, CH_{3Lev}, H_{Ac}), 1.30−1.18 (m, 9H, H-6_A, H-6_B, H-6_C); ^{13}C NMR (partial, $CDCl_3$) δ 206.1 (C_{Lev}), 171.8 (C_{Lev}), 170.0 (C_{Ac}), 161.7 (NHCO), 160.4 (C═NH), 138.9, 138.8, 138.7, 138.4, 138.3, 138.2, 137.9, 137.6 (10C_{quat}, Ph), 128.6, 128.5, 128.4, 128.3, 128.1, 128.0, 127.9, 127.8, 127.7, 127.6, 127.5 (50C, Ph), 99.4 (C-1_A), 97.1 (C-1_C), 94.8 (C-1_D), 92.3 (C(O)CCl_3), 90.9 (C(NH)CCl_3), 54.9 (C-2_D), 38.2 (CH_{2Lev}), 29.9 (CH_{3Lev}), 28.2 (CH_{2Lev}), 21.1 (C_{Ac}), 18.8, 18.3, 18.1 (3C, C-6_A, C-6_B, C-6_C).

*2-Azidoethyl (3,4-Di-O-benzyl-2-O-levulinyl-α-L-rhamnopyranosyl)-(1→2)-(3,4-di-O-benzyl-α-L-rhamnopyranosyl)-(1→3)-[2,3,4,6-tetra-O-benzyl-α-D-glucopyranosyl)-(1→4)]-(2-O-acetyl-α-L-rhamnopyranosyl)-(1→3)-4,6-di-O-benzyl-2-deoxy-2-trichloroacetamido-β-D-glucopyranoside (**43**).* A solution of donor **40** (1.20 g, 0.57 mmol) and bromoethanol (120 μL, 1.69 mmol, 3.0 equiv) in anhyd DCE (11 mL) containing 4 Å MS (1.2 g) was stirred under argon for 5 min. After the mixture was cooled to 0 °C, TMSOTf (10 μL, 55 μmol, 0.1 equiv) was added. The reaction mixture was stirred for 25 min at this temperature, Et_3N (50 μL) was added, and the mixture was filtered over a pad of Celite and concentrated to dryness. The crude residue was taken up under argon atmosphere in anhyd DMF (11 mL), and then NaI (427 mg, 2.85 mmol, 5.0 equiv) followed by NaN_3 (183 mg, 2.81 mmol, 4.9 equiv) were added. The reaction mixture was heated to 80 °C, stirred for 2 h, and then concentrated. The residue was dissolved in EtOAc (100 mL), washed with water (20 mL) and then brine (20 mL), and the aqueous layer was back-extracted with CH_2Cl_2 (20 mL). The combined organics were dried (Na_2SO_4), filtered, and concentrated. The residue was purified by column chromatography (toluene/EtOAc, 100:0 → 80:20) to give azide **43** (909 mg, 0.45 mmol, 78%) as a white foam. Analytical data were as reported previously.[50]

*O-(3,4-Di-O-benzyl-2-O-levulinyl-α-L-rhamnopyranosyl)-(1→2)-(3,4-di-O-benzyl-α-L-rhamnopyranosyl)-(1→3)-[2,3,4,6-tetra-O-benzyl-α-D-glucopyranosyl)-(1→4)]-(2-O-acetyl-α-L-rhamnopyranosyl)-(1→3)-2-trichloromethyl-(4,6-di-O-benzyl-1,2-dideoxy-α-D-glucopyrano)-[2,1-d]-2-oxazoline (**45**).* A suspension of acceptor **42** (1.0 g, 517 μmol), donor **40** (1.43 g, 676 μmol, 1.3 equiv), and freshly activated 4 Å MS (350 mg) in anhyd DCE (5 mL) was stirred for 15 min under an argon atmosphere. The reaction mixture was heated to 40 °C, and then TMSOTf (5 μL, 27 μmol, 0.05 equiv) was added. After 20 min, TLC analysis showed full conversion of the donor into a slightly less polar compound. TMSOTf (5 μL, 27 μmol, 0.05 equiv) was added, the reaction mixture was stirred for 30 min, and then TMSOTf (5 μL, 27 μmol, 0.05 equiv) was added once more. After the mixture was stirred for another 1 h 30 at 40 °C, Et_3N (50 μL) was added, and the reaction mixture was filtered over a pad of Celite and concentrated to dryness. The residue was purified by column chromatography (toluene/EtOAc, 95:5 → 85:15) to elute first oxazoline **45** (178 mg, 91 μmol, 18%) as a white amorphous solid and then decasaccharide **46** (1.32 g) slightly contaminated with trichloroacetamide **43**. Oxazoline **45**: R_f = 0.55 (toluene/EtOAc, 8.4:1.6); ^{1}H NMR ($CDCl_3$) δ 7.28−7.04 (m, 51H, Ph, NH), 6.29 (d, $J_{1,2}$ = 7.2 Hz, H-1_D), 5.50 (dd, $J_{2,3}$ = 3.0 Hz, $J_{1,2}$ = 1.9 Hz, 1H, H-2_A), 5.02−4.98 (m, 3H, H-1_A, H-1_C, H-2_C), 4.88−4.83 (m, 5H, 3H$_{Bn}$, H-1_B, H-1_E), 4.76−4.27 (m, 20H, 17H$_{Bn}$,H-2_B, H-2_D, H-3_D), 3.92−3.30 (m, 19H, H-2_E, H-3_A, H-3_B, H-3_C, H-3_E, H-4_A, H-4_B, H-4_C, H-4_D, H-4_E, H-5_A, H-5_B, H-5_C, H-5_D, H-5_E, H-$6a_D$, H-$6b_D$, H-$6a_E$, H-$6b_E$), 2.59−2.43 (m, 4H, 2 × CH_{2Lev}), 2.05 (s, 3H, H_{Ac}), 2.00 (s, 3H, CH_{3Lev}), 1.28, 1.24, 1.13 (d, $J_{5,6}$ = 6.2 Hz, 9H, H-6_A, H-6_B, H-6_C); ^{13}C NMR (partial, $CDCl_3$) δ 206.0 (C_{Lev}), 171.8 (C_{Lev}), 170.4 (C_{Ac}), 162.9 (C═N), 138.7, 138.6, 138.5, 138.4, 138.3, 138.2, 137.9, 137.5 (10C_{quat}, Ph), 128.5, 128.4, 128.3, 128.2, 128.1, 128.0, 127.9, 127.8, 127.6, 127.5, 127.4, 127.3 (50C, Ph), 104.2 (C-1_D), 101.0 (C-1_B), 99.3 (C-1_A), 98.3 (C-1_E), 96.8 (C-1_C), 86.4 (CCl_3), 66.0 (C-2_D), 38.1 (CH_{2Lev}), 29.8 (CH_{3Lev}), 28.2 (CH_{2Lev}), 21.2 (C_{Ac}), 18.6, 18.5, 18.1 (3C, C-6_A, C-6_B, C-6_C); HRMS (ESI$^+$) m/z calcd for $C_{109}H_{118}Cl_3NO_{25}Na$ [M + Na]$^+$ 1968.6957, found 1968.7250.

*2-Azidoethyl (3,4-Di-O-benzyl-α-L-rhamnopyranosyl)-(1→2)-(3,4-di-O-benzyl-α-L-rhamnopyranosyl)-(1→3)-[2,3,4,6-tetra-O-benzyl-α-D-glucopyranosyl)-(1→4)]-(α-L-rhamnopyranosyl)-(1→3)-(4,6-di-O-benzyl-2-deoxy-2-trichloroacetamido-β-D-glucopyranosyl)-(1→2)-(3,4-di-O-benzyl-α-L-rhamnopyranosyl)-(1→2)-(3,4-di-O-benzyl-α-L-rhamnopyranosyl)-(1→3)-[2,3,4,6-tetra-O-benzyl-α-D-glucopyranosyl)-(1→4)]-(α-L-rhamnopyranosyl)-(1→3)-(4,6-di-O-benzyl-2-deoxy-2-trichloroacetamido-β-D-glucopyranoside (**47**).* To a solution of fully protected decasaccharide **46** (244 mg, 62.8 μmol) in DCM/MeOH (1:3, 4.0 mL) was added methanolic MeONa (25 wt %, 72 μL, 0.32 mmol, 5.1 equiv). The reaction mixture was heated to 50 °C and stirred for 15 h, and then more methanolic MeONa (25 wt %, 43 μL, 0.19 mmol, 3.0 equiv) was added, and the reaction mixture was heated to 60 °C. After being stirred for 6 h, the reaction mixture was neutralized by addition of Dowex 50Wx8-200 (H^+), filtered over a pad of Celite, and concentrated. Column chromatography of the residue (toluene/EtOAc, 90:10 → 84:16) afforded triol **47** (166 mg, 44.8 μmol, 71%) as a white amorphous solid. Decasaccharide **47**: R_f = 0.32 (toluene/EtOAc, 4:1); ^{1}H NMR ($CDCl_3$) δ 7.38−6.94 (m, 102H, Ph, NH),5.18−4.12 (m, 54H, 40H$_{Bn}$, H-1_A, H-$1_A'$, H-1_B, H-$1_B'$, H-1_C, H-$1_C'$, H-1_D, H-$1_D'$, H-1_E, H-$1_E'$, H-2_C, H-$2_C'$, H-3_D, H-$3_D'$), 4.01−3.28 (m, 46H, H-2_B, H-$2_B'$, H-2_D, H-$2_D'$, H-2_E, H-$2_E'$, H-3_A, H-$3_A'$, H-3_B, H-$3_B'$, H-3_C, H-$3_C'$, H-3_E, H-$3_E'$, H-4_A, H-$4_A'$, H-4_B, H-$4_B'$, H-4_C, H-$4_C'$, H-4_D, H-$4_D'$, H-4_E, H-$4_E'$, H-5_A, H-$5_A'$, H-5_B, H-$5_B'$, H-5_C, H-$5_C'$, H-5_D, H-$5_D'$, H-5_E, H-$5_E'$, H-$6a_D$, H-$6a_D'$, H-$6b_D$, H-$6b_D'$, H-$6a_E$, H-$6a_E'$, H-$6b_E$, H-$6b_E'$, −$OCH_2CH_2N_3$), 1.32−1.07 (m, 18H, H-6_A, H-$6_A'$, H-6_B, H-$6_B'$, H-6_C, H-$6_C'$); ^{13}C NMR (partial, $CDCl_3$) δ 162.0 (NHCO), 161.9 (NHCO), 139.0, 138.8, 138.7, 138.6, 138.5, 138.4, 138.2, 138.0, 137.8, 137.5 (20C_{quat}, Ph), 128.9, 128.6, 128.5, 128.4, 128.3, 128.1, 128.0, 127.9, 127.8, 127.7, 127.6, 127.5, 127.4, 127.3 (100C, Ph), 92.6 (CCl_3), 92.5 (CCl_3), 50.9 (CH_2N_3), 18.6, 18.5, 18.4, 18.3, 18.2 (6C, C-6_A, C-$6_A'$, C-6_B, C-$6_B'$, C-6_C, C-$6_C'$); HRMS (ESI$^+$) m/z calcd for $C_{206}H_{229}Cl_6N_6O_{45}$ [M + NH_4]$^+$ 3717.4026, found 3716.8455; m/z calcd for $C_{206}H_{233}Cl_6N_7O_{45}$ [M + 2NH_4]$^{2+}$ 1867.2145, found 1867.2684.

*2-Aminoethyl α-L-Rhamnopyranosyl-(1→2)-α-L-rhamnopyranosyl)-(1→3)-[α-D-glucopyranosyl)-(1→4)]-α-L-rhamnopyranosyl-(1→3)-2-acetamido-2-deoxy-β-D-glucopyranosyl-(1→2)-α-L-rhamnopyranosyl-(1→2)-α-L-rhamnopyranosyl-(1→3)-[α-D-glucopyranosyl)-(1→4)]-α-L-rhamnopyranosyl-(1→3)-2-acetamido-2-deoxy-β-D-glucopyranoside (**5**).* Triol **47** (24 mg, 6.5 μmol) in solution in *t*-BuOH/CH_2Cl_2/H_2O (4:7:1, 4.0 mL) was treated with Pd(OH)$_2$/C (24 mg), and the suspension was stirred an hydrogen atmosphere for 3 days. More Pd(OH)$_2$ (5.0 mg), K_2HPO_4 (3.8 mg), and H_2O (1.0 mL) were added, and stirring went on for an additional 24 h. Once more, Pd(OH)$_2$/C (3.0 mg) and H_2O (1.0 mL) were added. After 24 h, an HPLC control indicated reaction completion. The suspension was filtered through a pad of Celite, and the filtrate was concentrated. RP-HPLC purification of the residue (5 μm C18, 100 Å, 10 × 250 mm column) eluting with CH_3CN/0.08% aq TFA (0 → 15% over 20 min at a flow rate of 5.5 mL.min^{-1}) gave the target decasaccharide **5** (6.3 mg, 58%) as a white powder following repeated freeze−drying. Analytical RP-HPLC (5 μm, C18 100 Å, 4.6 × 150 mm, 215 nm) for decasaccharide **5** eluting with CH_3CN/0.08% aq TFA (0 → 15% over 20 min at a flow rate of 1.0 mL.min^{-1}) indicated t_R = 4.76 min: ^{1}H NMR (D_2O, partial) δ 5.26 (br s, 2H, H-1_E, H-$1_E'$), 5.20, 5.13, 5.09, 5.04, 4.90, 4.88 (6s, 6H, H-1_A, H-$1_A'$, H-1_B, H-$1_B'$, H-1_C, H-$1_C'$), 4.78 (overlapped with D_2O, 1H, H-$1_D'$), 4.65 (d, $J_{1,2}$ = 8.5 Hz, 1H, H-1_D), 4.26−3.37 (m, 50H), 3.34−3.24 (m, 2H, CH_2NH_2), 2.13 (s, 6H, H_{Ac}), 1.42−1.35 (m, 18H, H-6_A, H-$6_A'$, H-6_B, H-$6_B'$, H-6_C, H-$6_C'$); ^{13}C NMR (partial, D_2O) δ 177.1 (NHCO), 177.0 (NHCO), 104.9 (C-$1_D'$), 103.1 (C-1_D), 100.1 (2C, C-1_E, C-$1_E'$), 68.3 (OCH_2), 63.3 (C-

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6_D, C-$6_D'$, C-6_E, C-$6_E'$), 58.3, 57.7 (2C, C-2_D, C-$2_D'$), 42.1 (CH_2NH_2), 25.0, 24.9 (2C, NHAc), 20.5, 19.5, 19.3, 19.2 (6C, C-6_A, C-$6_A'$, C-6_B, C-$6_B'$, C-6_C, C-$6_C'$); HRMS (ESI$^+$) m/z calcd for $C_{66}H_{113}N_3O_{45}$ [M + H]$^+$ 1668.6725, found 1668.6951.Other analytical data were as described.[51]

*Allyl (2,3-O-Isopropylidene-α-L-rhamnopyranosyl)-(1→3)-2-deoxy-4,6-O-isopropylidene-2-trichloroacetamido-β-D-glucopyranoside (**48**).* To a solution of disaccharide **1** (512 mg, 1.0 mmol) in anhyd DMF/acetone (1:1 v/v, 10 mL) stirred at 0 °C under an argon atmosphere were added CSA (48 mg, 0.21 mmol, 0.2 equiv) and 2-methoxypropene (144 μL, 1.50 mmol, 1.5 equiv). After 1 h at this temperature, DMP (184 μL, 1.50 mmol, 1.5 equiv) was added, and the reaction mixture was allowed to reach rt. After 15 h, more CSA (47 mg, 0.21 mmol, 0.2 equiv) and DMP (184 μL, 1.50 mmol, 1.5 equiv) were added. The reaction mixture was stirred for 20 h, then Et_3N (150 μL, 1.10 mmol, 1.1 equiv) was added. Volatiles were removed under vacuum, and the residue was purified by column chromatography (cHex/EtOAc 70:30 → 40:60) to give di-*O*-isopropylidene **48** (441 mg, 0.75 mmol, 75%) as a white amorphous solid: R_f = 0.21 (cHex/EtOAc 3:2); $[\alpha]^{24}_D$ = −40 (*c* 1.0; MeOH); ^{1}H NMR (MeOD) δ 5.91−5.81 (m, 1H, C*H*═CH$_2$), 5.29−5.23 (m, J_{trans} = 17.2 Hz, 1H, CH═C*H*$_2$), 5.15−5.12 (m, J_{cis} = 10.5 Hz, 1H, CH═C*H*$_2$), 5.07 (s, 1H, H-1_C), 4.65 (bd, $J_{1,2}$ = 8.3 Hz, 1H, H-1_D), 4.30−4.26 (m, 1H, −OCH$_{2All}$), 4.09−4.03 (m, 2H, −OCH$_{2All}$, H-2_C), 3.92−3.80 (m, 6H, H-2_D, H-3_D, H-$6a_D$, H-$6b_D$, H-3_C, H-5_C), 3.67 (m, $J_{4,5}$ = 9.3 Hz, 1H, H-4_D), 3.35−3.29 (m, 1H, H-5_D), 3.20 (dd, $J_{4,5}$ = 10.2 Hz, $J_{3,4}$ = 7.7 Hz, 1H, H-4_C), 1.52 (s, 3H, H_{iPr}), 1.44 (s, 3H, H_{iPr}), 1.40 (s, 3H, H_{iPr}), 1.26 (s, 3H, H_{iPr}), 1.18 (d, $J_{5,6}$ = 6.2 Hz, 3H, H-6_C); ^{13}C NMR (MeOD) δ 164.4 (NHCO), 135.0 (*C*H═CH$_2$), 117.5 (CH═*C*H$_2$), 110.2 ($C_{iPr\text{-}C}$), 101.6 (C-1_D), 100.9 ($C_{iPr\text{-}D}$), 99.3 (C-1_C), 94.0 (CCl_3), 79.6 (C-3_C), 78.2 (C-3_D), 77.2 (C-2_C), 75.6 (C-4_C), 74.2 (C-4_D), 71.3 (−*C*H$_{2All}$), 68.7 (C-5_D), 67.1 (C-5_C), 63.2 (C-6_D), 59.5 (C-2_D), 29.7, 28.4, 26.6, 19.4 (4C, C_{iPr}), 18.0 (C-6_C); HRMS (ESI$^+$) m/z calcd for $C_{23}H_{34}Cl_3NO_{10}Na$ [M + Na]$^+$ 612.1146, found 612.1105.

*Allyl (2,3-O-Isopropylidene-4-O-methyl-α-L-rhamnopyranosyl)-(1→3)-2-deoxy-4,6-O-isopropylidene-2-trichloroacetamido-β-D-glucopyranoside (**49**).* To a solution of alcohol **48** (299 mg, 0.51 mmol) in anhyd THF (8 mL) stirred at 0 °C under an argon atmosphere was added NaH (60% in mineral oil, 60 mg, 1.50 mmol, 3.0 equiv). After 15 min, MeI (95 μL, 1.53 mmol, 3.0 equiv) was added, and the reaction mixture was allowed to warm to rt. After 3 h of stirring, the solution was cooled to 0 °C, MeOH (8 mL) was slowly added, and the volatiles were evaporated. The residue was purified by column chromatography (cHex/EtOAc, 75:25 → 50:50) to give the methyl ether **49** (275 mg, 0.45 mmol, 89%) as a white amorphous solid: R_f = 0.28 (cHex/EtOAc 7:3); $[\alpha]^{24}_D$ = −45 (*c* 1.0; $CHCl_3$); ^{1}H NMR ($CDCl_3$) δ 6.66 (d, $J_{NH,2}$ = 8.7 Hz, 1H, NH), 5.88−5.78 (m, 1H, C*H*═CH$_2$), 5.29−5.24 (m, J_{trans} = 17.3 Hz, 1H, CH═C*H*$_2$), 5.21−5.17 (m, J_{cis} = 10.5 Hz, 1H, CH═C*H*$_2$), 5.04 (bs, 1H, H-1_C), 4.73 (d, $J_{1,2}$ = 8.5 Hz, 1H, H-1_D), 4.35−4.29 (m, 1H, −OCH$_{2All}$), 4.10−4.02 (m, 4H, −OCH$_{2All}$, H-2_C, H-3_C, H-3_D), 3.94 (dd, $J_{6a,6b}$ = 10.8 Hz, $J_{5,6a}$ = 5.4 Hz, 1H, H-$6a_D$), 3.86−3.73 (m, 3H, H-2_D, H-$6b_D$, H-5_C), 3.63 (pt, *J* = 9.2 Hz, 1H, H-4_D), 3.52 (s, 3H, −OCH$_3$), 3.37−3.31 (m, 1H, H-5_D), 2.92 (dd, $J_{4,5}$ = 10.1 Hz, $J_{3,4}$ = 7.0 Hz, 1H, H-4_C), 1.49 (s, 3H, H_{iPr}), 1.48 (s, 3H, H_{iPr}), 1.41 (s, 3H, H_{iPr}), 1.28 (s, 3H, H_{iPr}), 1.22 (d, $J_{5,6}$ = 6.2 Hz, 3H, H-6_C); ^{13}C NMR ($CDCl_3$) δ 162.1 (NHCO), 133.3 (CH═CH$_2$), 118.4 (CH═CH$_2$), 109.1 ($C_{iPr\text{-}C}$), 99.9 ($C_{iPr\text{-}D}$), 99.3 (C-1_D), 98.2 (C-1_C), 92.5 (CCl_3), 83.7 (C-4_C), 78.2 (C-2_C), 76.1, 76.0 (C-3_C, C-3_D), 72.9 (C-4_D), 70.5 (−OCH$_{2All}$), 67.7 (C-5_D), 65.2 (C-5_C), 62.2 (C-6_D), 59.8 (−OCH$_3$), 59.1 (C-2_D), 29.2, 28.2, 26.4, 19.2 (4C, C_{iPr}), 17.9 (C-6_C); HRMS (ESI$^+$) m/z calcd for $C_{24}H_{36}Cl_3NO_{10}Na$ [M + Na]$^+$ 626.1302, found 626.1290.

*Allyl (4-O-Methyl-α-L-rhamnopyranosyl)-(1→3)-2-deoxy-2-trichloroacetamido-β-D-glucopyranoside (**6**).* To a solution of fully protected disaccharide **49** (106 mg, 175 μmol) in CH_2Cl_2 (2 mL) was added TFA (50% aq, 2 mL). The biphasic mixture was vigorously stirred for 35 min and then repeatedly coevaporated with toluene and cyclohexane. The residue was purified by column chromatography (EtOAc) to give tetraol **6** (75 mg, 143 μmol, 82%) as a white amorphous solid: R_f = 0.17 (EtOAc); $[\alpha]^{24}_D$ = −54 (*c* 1.0; MeOH); ^{1}H NMR (D_2O) δ 6.03−5.93 (m, 1H, C*H*═CH$_2$), 5.42−5.36 (m, J_{trans} = 17.3 Hz, 1H, CH═C*H*$_2$), 5.35−5.31 (m, J_{cis} = 10.4 Hz, 1H, CH═C*H*$_2$), 4.94 (d, $J_{1,2}$ = 1.7 Hz, 1H, H-1_C), 4.82 (d, $J_{1,2}$ = 8.5 Hz, 1H, H-1_D), 4.45−4.40 (m, 1H, −OCH$_{2All}$), 4.28−4.23 (m, 1H, −OCH$_{2All}$), 4.04 (dq, $J_{4,5}$ = 9.8 Hz, 1H, H-5_C), 4.01 (dd, $J_{6a,6b}$ = 12.4 Hz, $J_{5,6a}$ = 2.2 Hz, 1H, H-$6a_D$), 3.95−3.81 (m, 5H, H-$6b_D$, H-2_D, H-3_D, H-2_C, H-3_C), 3.61 (s, 3H, −OCH$_3$), 3.66−3.54 (m, 2H, H-5_D, H-4_D), 3.25 (pt, $J_{3,4}$ = 9.7 Hz, 1H, H-4_C), 1.33 (d, $J_{5,6}$ = 6.2 Hz, 3H, H-6_C); ^{13}C NMR (D_2O) δ 164.8 (NHCO), 133.1 (*C*H═CH$_2$), 118.8 (CH═*C*H$_2$), 101.1 (C-1_C), 99.1 (C-1_D), 91.6 (CCl_3), 82.2 (C-4_C), 80.6 (C-3_D), 76.1 (C-5_D), 70.9 (−OCH$_{2All}$), 70.8 (C-2_C), 70.1 (C-3_C), 68.7 (C-4_D), 68.0 (C-5_C), 60.8 (C-6_D), 60.0 (−OCH$_3$), 57.4 (C-2_D), 16.7 (C-6_C); HRMS (ESI$^+$) m/z calcd for $C_{18}H_{28}Cl_3NO_{10}Na$ [M + Na]$^+$ 546.0676, found 546.0681.

*Allyl (2,3-O-Isopropylidene-α-L-rhamnopyranos-4-ulosyl)-(1→3)-2-deoxy-4,6-O-isopropylidene-2-trichloroacetamido-β-D-glucopyranoside (**50**).* A solution of DMSO (301 μL, 4.23 mmol, 5.0 equiv) in CH_2Cl_2 (1.0 mL) was added to a stirred solution of oxalyl chloride (179 μL, 2.12 mmol, 2.5 equiv) in CH_2Cl_2 (10 mL) at −78 °C under an argon atmosphere. After 30 min, a solution of alcohol **48** (500 mg, 0.85 mmol) in CH_2Cl_2 (2.0 mL) was added at −78 °C, and the reaction was stirred at this temperature for a further 45 min. After this time, Et_3N (850 μL, 6.09 mmol, 7.2 equiv) was added, and the reaction was warmed to room temperature and diluted with CH_2Cl_2 (50 mL). The combined organics were successively washed with a saturated aqueous solution of $NaHCO_3$ (20 mL) and brine (20 mL) before being dried (Na_2SO_4) and concentrated to a pale yellow solid, which was purified by column chromatography (CH_2Cl_2/Me_2CO 9:1 → 8:2) to give ketone **50** (416 mg, 0.71 mmol, 84%) as a white amorphous solid: R_f = 0.38 (CH_2Cl_2/Me_2CO 4:1); ^{1}H NMR (DMSO-d_6) δ 9.18 (d, $J_{NH,2}$ = 9.2 Hz, 1H, NH), 5.88−5.75 (m, 1H, C*H*═CH$_2$), 5.28−5.20 (m, J_{trans} = 17.3 Hz, 1H, CH═C*H*$_2$), 5.15−5.09 (m, J_{cis} = 10.7 Hz, 1H, CH═C*H*$_2$), 5.07 (bs, 1H, H-1_C), 4.70 (q, $J_{5,6}$ = 6.5 Hz, 1H, H-5_C), 4.64 (d, $J_{1,2}$ = 8.2 Hz, 1H, H-1_D), 4.62 (d, $J_{2,3}$ = 5.5 Hz, 1H, H-3_C), 4.25−4.15 (m, 2H, −OCH$_{2All}$, H-2_C), 4.06−3.98 (m, 1H, −OCH$_{2All}$), 3.99 (pt, *J* = 9.5 Hz, H-3_D), 3.92−3.82 (m, 2H, H-2_D, H-$6a_D$), 3.77 (pt, *J* = 10.4 Hz, 1H, H-$6b_D$) 3.70 (pt, *J* = 9.3 Hz, 1H, H-4_D), 3.31−3.22 (m, 1H, H-5_D), 1.46 (s, 3H, H_{iPr}), 1.31 (s, 3H, H_{iPr}), 1.27 (s, 3H, H_{iPr}), 1.20 (s, 3H, H_{iPr}), 1.13 (d, $J_{5,6}$ = 6.5 Hz, 3H, H-6_C) ppm; ^{13}C NMR (DMSO-d_6) δ 204.7 (C-4_C), 161.8 (NHCO), 134.1 (CH═CH$_2$), 116.6 (CH═CH$_2$), 109.7 ($C_{iPr\text{-}C}$), 100.1 (C-1_D), 99.2 ($C_{iPr\text{-}D}$), 97.1 (C-1_C), 92.9 (CCl_3), 78.9 (C-2_C), 77.1 (C-3_D), 75.6 (C-3_C), 72.1 (C-4_D), 69.5 (−OCH$_{2All}$), 68.3 (C-5_C), 65.9 (C-5_D), 61.4 (C-6_D), 57.5 (C-2_D), 28.9, 26.7, 25.5, 19.0 (4C, C_{iPr}), 14.6 (C-6_C); HRMS (ESI$^+$) m/z calcd for $C_{23}H_{32}Cl_3NO_{10}Na$ [M + Na]$^+$ 702.0735, found 702.0714.

*Allyl (α-L-Rhamnopyranos-4-ulosyl)-(1→3)-2-deoxy-2-trichloroacetamido-β-D-glucopyranoside (**7**).* A solution of fully protected disaccharide **50** (1.0 g, 1.70 mmol) in AcOH (80% aq, 34 mL) was stirred at 80 °C overnight and then repeatedly coevaporated with a 2:1 mixture of cyclohexane and toluene. The residue was purified by column chromatography (CH_2Cl_2/MeOH 9:1 → 8:2) to give tetraol **7** (600 mg, 1.18 mmol, 69%) as a white amorphous solid: R_f = 0.16 (CH_2Cl_2/MeOH 4:1); ^{1}H NMR (DMSO-d_6 + D_2O) δ 5.87−5.75 (m, 1H, C*H*═CH$_2$), 5.25−5.17 (m, J_{trans} = 17.3 Hz, 1H, CH═C*H*$_2$), 5.12−5.07 (m, J_{cis} = 10.5 Hz, 1H, CH═C*H*$_2$), 4.91 (d, $J_{1,2}$ = 2.1 Hz, 1H, H-1_C), 4.73 (qd, $J_{5,6}$ = 6.5 Hz, $J_{3,5}$ = 0.6 Hz, 1H, H-5_C), 4.54 (d, $J_{1,2}$ = 8.4 Hz, 1H, H-1_D), 4.41 (dd, $J_{2,3}$ = 3.5 Hz, $J_{3,5}$ = 0.6 Hz, 1H, H-3_C), 4.26−4.19 (m, 1H, −OCH$_{2All}$), 4.04−3.97 (m, 1H, −OCH$_{2All}$), 3.94 (dd, $J_{2,3}$ = 3.5 Hz, $J_{1,2}$ = 2.1 Hz, 1H, H-2_C), 3.86 (dd, $J_{2,3}$ = 10.3 Hz, $J_{3,4}$ = 8.8 Hz, 1H, H-3_D), 3.73−3.61 (m, 2H, H-$6a_D$, H-2_D), 3.52 (dd, $J_{6a,6b}$ = 12.0 Hz, $J_{5,6b}$ = 5.6 Hz, 1H, H-$6b_D$), 3.30 (dd, $J_{4,5}$ = 9.9 Hz, $J_{3,4}$ 8.8 Hz, 1H, H-4_D), 3.22 (ddd, $J_{4,5}$ = 9.9 Hz, $J_{5,6b}$ = 5.6 Hz, $J_{5,6a}$ = 1.8 Hz, 1H, H-5_D), 1.06 (d, $J_{5,6}$ = 6.5 Hz, 3H, H-6_C) ppm; ^{13}C NMR (DMSO-d_6) δ 207.9 (C-4_C), 161.6 (NHCO), 134.4 (*C*H═CH$_2$), 116.3 (CH═*C*H$_2$), 99.9 (C-1_C), 99.4 (C-1_D), 93.0 (CCl_3), 79.3 (C-3_D), 76.8 (C-5_D), 75.0 (C-2_C), 73.3 (C-3_C), 70.3 (C-5_C), 69.0 (−OCH$_{2All}$), 68.9 (C-4_D), 60.7 (C-6_D), 57.4 (C-2_D), 13.8 (C-6_C); HRMS (ESI$^+$) m/z calcd for $C_{17}H_{24}Cl_3NO_{10}Na$ [M + Na]$^+$ 530.0364, found 530.0280.

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*Allyl (2,3,4-Tri-O-acetyl-α-L-rhamnopyranosyl)-(1→3)-4,6-di-O-acetyl-2-deoxy-2-trichloroacetamido-β-D-glucopyranoside (**51**).* To a solution of disaccharide **1** (1.01 g, 1.97 mmol) in anhyd Py (30 mL) under an argon atmosphere was added DMAP (25 mg, 0.20 mmol, 0.1 equiv). The reaction mixture was cooled to 0 °C acetic anhydride (20 mL, 212 mmol, 107 equiv) was added, and the reaction mixture was allowed to warm to rt. After 14 h of stirring, the reaction mixture was cooled to 0 °C. The reaction was quenched by the slow addition of MeOH (20 mL), and the mixture was concentrated then coevaporated twice with toluene. The residue was purified by column chromatography (cHex/EtOAc, 50:50) to give the fully protected disaccharide **51** (1.34 g, 1.86 mmol, 94%) as a white amorphous solid: R_f = 0.45 (cHex/EtOAc 1:1); $[\alpha]^{24}_D$ = −2 (*c* 1.0; $CHCl_3$); ^{1}H NMR ($CDCl_3$) δ 7.16 (d, $J_{NH,2}$ = 6.9 Hz, 1H, NH), 5.91−5.81 (m, 1H, C*H*═CH_2), 5.29−5.24 (m, J_{trans} = 17.3 Hz, 1H, CH═CH_2), 5.22−5.17 (m, 3H, CH═CH_2, H-2$_C$, H-3$_C$), 5.09 (d, $J_{1,2}$ = 8.1 Hz, 1H, H-1$_D$), 5.11−4.98 (m, 2H, H-4$_C$, H-4$_D$), 4.83 (d, $J_{1,2}$ = 1.6 Hz, 1H, H-1$_C$), 4.53 (dd, $J_{2,3}$ = 10.0 Hz, $J_{3,4}$ = 8.6 Hz, 1H, H-3$_D$), 4.36−4.31 (m, 1H, −OCH_{2All}), 4.23 (dd, $J_{6a,6b}$ = 12.3 Hz, $J_{5,6a}$ = 5.1 Hz, 1H, H-6a$_D$), 4.13−4.06 (m, $J_{5,6b}$ = 2.7 Hz, 2H, −OCH_{2All}, H-6b$_D$), 3.86 (dq, $J_{4,5}$ = 9.8 Hz, 1H, H-5$_C$), 3.64 (ddd, $J_{4,5}$ = 7.9 Hz, 1H, H-5$_D$), 3.42−3.35 (m, 1H, H-2$_D$), 2.10 (s, 6H, H$_{Ac}$), 2.09, 2.03, 1.94 (3s, 9H, H$_{Ac}$), 1.16 (d, $J_{5,6}$ = 6.2 Hz, 1H, H-6$_C$); ^{13}C NMR ($CDCl_3$) δ 170.8, 170.1, 170.0, 169.9, 169.8 (5C, C$_{Ac}$), 162.5 (NHCO), 133.4 (CH═CH_2), 118.6 (CH═CH_2), 99.7 (C-1$_C$), 97.4 (C-1$_D$), 92.1 (CCl_3), 78.6 (C-3$_D$), 72.0 (C-5$_D$), 70.9 (−OCH_{2All}), 70.7 (C-4$_C$), 70.4 (C-4$_D$), 70.0 (C-2$_C$), 68.9 (C-3$_C$), 67.9 (C-5$_C$), 62.4 (C-6$_D$), 59.1 (C-2$_D$), 21.3 (C$_{Ac}$), 21.0 (C$_{Ac}$), 20.9 (2C, C$_{Ac}$), 20.8 (C$_{Ac}$), 17.4 (C-6$_C$); HRMS (ESI$^+$) *m*/*z* calcd for $C_{27}H_{36}Cl_3NO_{15}Na$ [M + Na]$^+$ 742.1048, found 742.0972.

*2,3,4-Tri-O-acetyl-α-L-rhamnopyranosyl-(1→3)-4,6-di-O-acetyl-2-deoxy-2-trichloroacetamido-α/β-D-glucopyranose (**52**).* 1,5-Cyclooctadiene−bis(methyldiphenylphosphine)iridium hexafluorophosphate (63 mg, 74 μmol, 0.03 equiv) was dissolved in anhyd THF (20 mL) under an argon atmosphere. Hydrogen was bubbled through the solution for 15 min, causing the color to change from red to yellow. The solution was degassed by complete evaporation of the solvent under vacuum. The activated iridium complex was dissolved under an argon atmosphere in anhyd THF (10 mL) and added to a solution of **51** (1.72 g, 2.39 mmol) in anhyd THF (30 mL). The reaction mixture was stirred for 4 h at rt, and then a solution of iodine (1.21 g, 4.77 mmol, 2.0 equiv) in THF/water (40 mL, 3:1 v/v) was added. After 16 h, the excess iodine was quenched by addition of a 10% aq sodium bisulfite solution (30 mL). The reaction mixture was concentrated under reduced pressure to remove THF, and the aqueous phase was extracted with CH_2Cl_2 (2 × 60 mL). The combined organics were washed with brine (1 × 20 mL), dried (Na_2SO_4), filtered, and concentrated to dryness. Column chromatography of the residue (cHex/EtOAc, 60:40 → 30:70) afforded hemiacetal **52** (1.39 g, 2.04 mmol, 85%) as a pale yellow amorphous solid (α/β, 9:1). α: R_f = 0.28 (cHex/EtOAc 1:1); ^{1}H NMR ($CDCl_3$) δ 6.92 (d, $J_{NH,2}$ = 9.1 Hz, 1H, NH), 5.35 (pt, *J* = 3.7 Hz, 1H, H-1$_D$), 5.23 (dd, $J_{3,4}$ = 10.0 Hz, $J_{2,3}$ = 3.4 Hz, 1H, H-3$_C$), 5.15−5.08 (m, 2H, H-4$_D$, H-2$_C$), 4.98 (pt, *J* = 9.9 Hz, 1H, H-4$_C$), 4.88 (d, $J_{1,2}$ = 1.8 Hz, 1H, H-1$_C$), 4.30−4.24 (m, 1H, H-2$_D$), 4.19−4.05 (m, 4H, H-3$_D$, H-5$_D$, H-6a$_D$, H-6b$_D$), 3.90 (dq, $J_{4,5}$ = 9.9 Hz, 1H, H-5$_C$), 2.12, 2.11, 2.09, 2.01, 1.93 (5s, 15H, H$_{Ac}$), 1.16 (d, $J_{5,6}$ = 6.3 Hz, 1H, H-6$_C$); ^{13}C NMR ($CDCl_3$) δ 171.1, 170.5, 170.2, 169.7, 169.4 (5C, C$_{Ac}$), 162.1 (NHCO), 99.4 (C-1$_C$), 92.4 (CCl_3), 91.2 (C-1$_D$), 78.5 (C-3$_D$), 71.2 (C-4$_C$), 70.7 (C-2$_C$), 69.9 (C-4$_D$), 68.4 (C-3$_C$), 68.0 (C-5$_D$), 67.7 (C-5$_C$), 62.2 (C-6$_D$), 54.7 (C-2$_D$), 21.2 (C$_{Ac}$), 21.0 (C$_{Ac}$), 20.9 (2C, C$_{Ac}$), 20.7 (C$_{Ac}$), 17.2 (C-6$_C$); HRMS (ESI$^+$) *m*/*z* calcd for $C_{24}H_{32}Cl_3NO_{15}Na$ [M + Na]$^+$ 702.0735, found 702.0714.

*Methyl (2,3,4-Tri-O-acetyl-α-L-rhamnopyranosyl)-(1→3)-4,6-di-O-acetyl-2-deoxy-1-thio-2-trichloroacetamido-β-D-glucopyranoside (**54**) and O-(2,3,4-Tri-O-acetyl-α-L-rhamnopyranosyl)-(1→3)-2-trichloromethyl-(4,6-di-O-acetyl-1,2-dideoxy-α-D-glucopyrano)-[2,1-d]-2-oxazoline (**53**).* To a solution of hemiacetal **52** (668 mg, 0.98 mmol) in anhyd CH_2Cl_2 (10 mL) under an argon atmosphere were added Cs_2CO_3 (64 mg, 0.20 mmol, 0.2 equiv) and trichloroacetonitrile (490 μL, 4.89 mmol, 5.0 equiv). The reaction mixture was stirred for 60 min, filtered on Celite, and concentrated. The residue was dissolved in CH_2Cl_2 (10 mL) under an argon atmosphere, and freshly activated MS were added (530 mg). The reaction mixture was cooled to −15 °C, and then methyl trimethylsilyl sulfide (710 μL, 5.00 mmol, 5.0 equiv) followed by TMSOTf (13 μL, 0.05 mmol, 0.05 equiv) was added. The reaction mixture was stirred overnight while being allowed to slowly reach rt, and then Et_3N (20 μL) was added. The mixture was filtered on Celite and concentrated, and then column chromatography of the residue (cHex/EtOAc, 90:10 → 50:50) afforded first oxazoline **53** (324 mg, 0.49 mmol, 50%) and then the expected thioglycoside **54** (221 mg, 0.31 mmol, 31%), both as white amorphous solids. Oxazoline **53**: R_f = 0.51 (cHex/EtOAc 1:1); $[\alpha]^{24}_D$ = −30 (*c* 1.0; $CHCl_3$); ^{1}H NMR ($CDCl_3$) δ 6.37 (d, $J_{1,2}$ = 7.4 Hz, 1H, H-1$_D$), 5.25 (dd, $J_{3,4}$ = 10.1 Hz, $J_{2,3}$ = 3.4 Hz, 1H, H-3$_C$), 5.20 (dd, $J_{1,2}$ = 1.7 Hz, 1H, H-2$_C$), 5.09 (pt, $J_{4,5}$ = 9.9 Hz, 1H, H-4$_C$), 5.04−5.02 (m, $J_{4,5}$ = 8.3 Hz, 1H, H-4$_D$), 4.99 (d, 1H, H-1$_C$), 4.47−4.44 (m, $J_{2,3}$ = 2.7 Hz, 1H, H-2$_D$), 4.35−4.34 (m, 1H, H-3$_D$), 4.28 (dd, $J_{6a,6b}$ = 12.2 Hz, $J_{5,6a}$ = 3.1 Hz, 1H, H-6a$_D$), 4.16 (dd, $J_{5,6b}$ = 6.0 Hz, 1H, H-6b$_D$), 4.09−4.02 (dq, 1H, H-5$_C$), 3.79 (ddd, 1H, H-5$_D$), 2.16, 2.11, 2.08, 2.05, 1.99 (5s, 15H, H$_{Ac}$), 1.24 (d, $J_{5,6}$ = 6.3 Hz, 1H, H-6$_C$); ^{13}C NMR ($CDCl_3$) δ 170.7, 170.3, 170.0, 169.9, 169.7 (5C, C$_{Ac}$), 163.4 (C═N), 103.5 (C-1$_D$), 96.5 (C-1$_C$), 86.2 (CCl_3), 72.3 (C-3$_D$), 70.8 (C-4$_C$), 69.9 (C-2$_C$), 68.8 (C-3$_C$), 68.6, 68.5 (2C, C-4$_C$, C-5$_D$), 67.6 (C-5$_C$), 64.2 (C-2$_D$), 63.6 (C-6$_D$), 21.0, 20.9, 20.8 (3C, C$_{Ac}$), 20.7 (2C, C$_{Ac}$), 17.4 (C-6$_C$); HRMS (ESI$^+$) *m*/*z* calcd for $C_{24}H_{30}Cl_3NO_{14}Na$ [M + Na]$^+$ 684.0630, found 684.0669.

Thioglycoside **54**: R_f = 0.38 (cHex/EtOAc 1:1); $[\alpha]^{24}_D$ = 0 (*c* 1.0; $CHCl_3$); ^{1}H NMR ($CDCl_3$) δ 7.14 (d, $J_{NH,2}$ = 7.6 Hz, 1H, NH), 5.20−5.16 (m, 2H, H-2$_C$, H-3$_C$), 5.07−4.97 (m, 3H, H-4$_C$, H-1$_D$, H-4$_D$), 4.85 (d, $J_{1,2}$ = 1.7 Hz, 1H, H-1$_C$), 4.37 (t, *J* = 9.6 Hz, 1H, H-3$_D$), 4.20 (dd, $J_{6a,6b}$ = 12.4 Hz, $J_{5,6a}$ = 5.2 Hz, 1H, H-6a$_D$), 4.10 (dd, $J_{5,6b}$ = 2.6 Hz, 1H, H-6b$_D$), 3.89−3.82 (dq, $J_{4,5}$ = 9.9 Hz, 1H, H-5$_C$), 3.67−3.57 (m, 2H, H-2$_D$, H-5$_D$), 2.21 (s, 3H, SCH_3), 2.10, 2.09, 2.08, 2.01, 1.94 (5s, 15H, H$_{Ac}$), 1.15 (d, $J_{5,6}$ = 6.2 Hz, 1H, H-6$_C$); ^{13}C NMR ($CDCl_3$) δ 170.3, 170.1, 170.0, 169.9, 169.8 (5C, C$_{Ac}$), 162.2 (NHCO), 99.6 (C-1$_C$), 92.1 (CCl_3), 82.2 (C-1$_D$), 80.1 (C-3$_D$), 76.2 (C-5$_D$), 70.8 (3C, C-2$_C$, C-4$_C$, C-4$_D$), 68.8 (C-3$_C$), 67.9 (C-5$_C$), 62.6 (C-6$_D$), 57.5 (C-2$_D$), 21.2, 21.0, 20.9, 20.8, 20.7 (5C, C$_{Ac}$), 12.5 (S-CH_3); HRMS (ESI$^+$) *m*/*z* calcd for $C_{25}H_{34}Cl_3NO_{14}SNa$ [M + Na]$^+$ 732.0663, found 732.0718.

*Methyl α-L-Rhamnopyranosyl-(1→3)-2-deoxy-1-thio-2-trichloroacetamido-β-D-glucopyranoside (**8**).* To a solution of fully protected thioglycoside **54** (269 mg, 0.38 mmol) in MeOH (5 mL) was added methanolic MeONa (25 wt %, 87 μL, 0.38 mmol, 1.0 equiv). The reaction mixture was stirred for 90 min, neutralized by addition of Dowex 50Wx8−200, filtered over a pad of Celite, and concentrated. Column chromatography of the residue (CH_2Cl_2/MeOH, 90:10 → 80:20) afforded pentaol **8** (174 mg, 0.35 mmol, 91%) as a white amorphous solid: R_f = 0.30 (CH_2Cl_2/MeOH 8.5:1.5); $[\alpha]^{24}_D$ = −61 (*c* 1.0; MeOH); ^{1}H NMR (D_2O) δ 4.98 (d, $J_{1,2}$ = 1.7 Hz, 1H, H-1$_C$), 4.78 (overlapped with D_2O, 1H, H-1$_D$), 4.11−3.99 (m, 3H, H-5$_C$, H-2$_D$, H-6a$_D$), 3.91−3.78 (m, 4H, H-2$_C$, H-3$_C$, H-3$_D$, H-6b$_D$), 3.66 (dd, $J_{3,4}$ = 9.9 Hz, $J_{4,5}$ = 8.4 Hz, 1H, H-4$_D$), 3.62−3.59 (m, 1H, H-5$_D$), 3.51 (pt, *J* = 9.7 Hz, 1H, H-4$_C$), 2.28 (s, 3H, SCH_3), 1.31 (d, $J_{5,6}$ = 6.2 Hz, 1H, H-6$_C$); ^{13}C NMR (D_2O) δ 167.2 (NHCO), 104.0 (C-1$_C$), 94.1 (CCl_3), 85.8 (C-1$_D$), 84.8 (C-3$_D$), 82.9 (C-5$_D$), 74.5 (C-4$_C$), 73.3(C-2$_C$), 73.0 (C-3$_C$), 71.6 (C-5$_C$), 71.2 (C-4$_D$), 63.5 (C-6$_D$), 58.5 (C-2$_D$), 19.1 (C-6$_C$), 14.2 (S-CH_3); HRMS (ESI$^+$) *m*/*z* calcd for $C_{15}H_{24}Cl_3NO_9SNa$ [M + Na]$^+$ 522.0135, found 522.0093.

*Allyl (2,3,4-Tris-O-(tert-butyldimethylsilyl)-α-L-rhamnopyranosyl)-(1→3)-4,6-bis-O-(tert-butyldimethylsilyl)-2-deoxy-2-trichloroacetamido-β-D-glucopyranoside (**55**).* To a solution of disaccharide **1** (511 mg, 1.00 mmol) in dry Py (10 mL), under an argon atmosphere at 0 °C, was added *tert*-butyldimethylsilyl trifluoromethanesulfonate (2.3 mL, 10.0 mmol, 10.0 equiv) dropwise. The reaction mixture was heated to 60 °C for 15 h and then cooled to 0 °C. The reaction was quenched by addition of MeOH (10 mL). Volatiles were concentrated and coevaporated twice with toluene. Column chromatography of the residue (cHex/EtOAc, 100:0 → 98:2) afforded the fully protected disaccharide **55** (930 mg, 0.86 mmol, 86%) as a colorless oil (rotameric mixture 3:2): R_f = 0.78 (cHex/EtOAc 90:10); $[\alpha]^{24}_D$ =

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−33 (*c* 1.0; $CHCl_3$); ^{1}H NMR ($CDCl_3$) δ 7.76 (d, $J_{NH,2}$ = 8.7 Hz, 0.6H, NH), 7.40 (d, $J_{NH,2}$ = 8.8 Hz, 0.4H, NH), 5.89−5.83 (m, 1H, CH=CH_2), 5.29−5.14 (m, 2H, CH=CH_2), 4.78−4.75 (m, 1H, H-1_C), 4.72 (br s, 1H, H-1_D), 4.28−4.24 (m, 1H, −OCH_{2All}), 4.10−3.95 (m, 4H, −OCH_{2All}, H-2_D, H-4_D, H-$6a_D$), 3.88−3.54 (m, 6.6H, H-2_C, H-3_C, H-4_C, H-5_C, H-3_D, H-5_D, H-$6b_D$), 3.41 (br d, *J* = 5.0 Hz, 0.4H, H-4_C), 1.22 (br s, 3H, H-6_C), 0.93−0.88 (m, 45H, CH_{3tBuSi}), 0.17−0.06 (m, 30H, $SiCH_3$); ^{13}C NMR ($CDCl_3$) δ 161.5 (NHCO major), 161.2 (NHCO minor), 134.1 (CH=CH_2), 117.7 (CH=CH_2 major), 117.4 (CH=CH_2 minor), 102.3 (C-1_C minor), 99.6 (C-1_C major), 98.0 (C-1_D minor), 97.4 (C-1_D major), 92.8 (CCl_3 minor), 92.6 (CCl_3 major), 79.9 (C-3_D major), 79.6 (C-3_D minor), 78.5 (C-4_C minor), 77.19 (C-4_C major), 76.7 (C-3_C minor), 74.1 (C-5_D major), 73.9 (C-5_D minor), 73.4, 73.2 (C-2_C major, C-3_C major), 71.9 (C-5_C major), 71.4, 71.3 (C-2_C minor, C-5_C minor), 69.6 (−OCH_{2All} major), 69.3 (−OCH_{2All} minor), 69.1 (C-4_D minor), 68.4 (C-4_D major), 64.0 (C-6_D major), 63.8 (C-6_D minor), 53.9 (C-2_D minor), 49.7 (C-2_D major), 27.1, 26.9, 26.5, 26.3, 26.2, 26.0, 25.9 (15C, CH_{3tBuSi}), 20.1 (C-6_C minor), 18.8 (C-6_C major), 18.6, 18.4, 18.3, 18.0 (5C, Si−C_{tBu}), −2.3, −3.3, −3.6, −3.7, −3.9, −4.2, −4.3, −4.4, −4.6, 4.8, −4.9, 5.1 (10C, Si-CH_3); HRMS (ESI$^+$) *m*/*z* calcd for $C_{47}H_{96}Cl_3NO_{10}Si_5Na$ $[M + Na]^+$ 1102.4844, found 1102.4854.

*(2,3,4-Tris-(O-tert-butyldimethylsilyl)-α-L-rhamnopyranosyl)-(1→3)-4,6-bis-O-(tert-butyldimethylsilyl)-2-deoxy-2-trichloroacetamido-α/β-D-glucopyranose (**56**).* 1,5-Cyclooctadiene−bis(methyldiphenylphosphine)iridium hexafluorophosphate (19 mg, 22 μmol, 0.03 equiv) was dissolved in anhyd THF (5 mL) under an argon atmosphere. Hydrogen was bubbled through the solution for 15 min, causing the color to change from red to yellow. The solution was degassed by complete evaporation of the solvent under vacuum. The activated iridium complex was dissolved under an argon atmosphere in anhyd THF (10 mL) and added to a solution of **55** (810 mg, 0.75 mmol) in anhyd THF (10 mL). The reaction mixture was stirred for 2 h 30 at rt, and then a solution of iodine (383 mg, 1.51 mmol, 2.0 equiv) in THF/water (10 mL, 2:1 v/v) was added. After 60 min, the excess iodine was consumed by addition of 10% aq sodium bisulfite (8 mL). The reaction mixture was concentrated under reduced pressure to remove THF, and the aqueous phase was extracted with CH_2Cl_2 (3 × 30 mL). The combined organics were dried (Na_2SO_4), filtered, and concentrated to dryness. Column chromatography of the residue (cHex/EtOAc, 97:3 → 90:10) afforded hemiacetal **56** (653 mg, 0.63 mmol, 84%) as a white amorphous solid (*α*/*β* ratio 2:1, rotameric mixture 7:3): R_f = 0.30 (cHex/EtOAc 9:1). *α*: ^{1}H NMR ($CDCl_3$) δ 7.72 (d, $J_{NH,2}$ = 8.8 Hz, 0.7H, NH), 7.63 (d, $J_{NH,2}$ = 8.4 Hz, 0.3H, NH), 5.22 (dd, $J_{1,OH}$ = 7.1 Hz, $J_{1,2}$ = 2.5 Hz, 0.7H, H-1_D), 4.82−4.81 (m, 1H, H-1_C), 4.23−3.56 (m, 9H, H-2_C, H-2_D, H-3_C, H-3_D, H-4_D, H-5_C, H-5_D, H-$6a_D$, H-$6b_D$), 3.46−3.42 (m, 1H, H-4_C), 3.17 (d, 0.7H, OH), 1.27−1.21 (br s, 3H, H-6_C), 0.93−0.89 (m, 45H, CH_{3tBuSi}), 0.18−0.07 (m, 30H, $SiCH_3$). ^{13}C NMR (partial, $CDCl_3$) δ 162.7 (NHCO major), 162.5 (NHCO minor), 102.1 (C-1_C major), 102.0 (C-1_C minor), 93.5 (CCl_3 minor), 92.7 (CCl_3 major), 89.0 (C-1_D major), 88.7 (C-1_D minor), 62.3 (C-6_D minor), 62.1 (C-6_D major), 53.1 (C-2_D major), 52.9 (C-2_D minor), 26.8, 26.5, 26.2, 26.1, 26.0, 25.9, 25.8 (15C, CH_{3tBuSi}), 20.1 (C-6_C), 18.9, 18.5, 18.3, 18.0 (5C, Si−C_{tBu}), −3.8, −3.9, −4.2, −4.3, −4.4, −4.5, −5.0, −5.2 (10C, Si-CH_3); HRMS (ESI$^+$) *m*/*z* calcd for $C_{44}H_{92}Cl_3NO_{10}Si_5Na$ $[M + Na]^+$ 1062.4531, found 1062.4498.

*α-L-Rhamnopyranosyl-(1→3)-2-deoxy-2-trichloroacetamido-α/β-D-glucopyranose (**9**). Route 1.* 1,5-Cyclooctadiene−bis(methyldiphenylphosphine)iridium hexafluorophosphate (21 mg, 24 μmol, 0.03 equiv) was dissolved in THF (10 mL) under an argon atmosphere. Hydrogen was bubbled through the solution for 20 min, causing the color to change from red to yellow. The solution was degassed by complete evaporation of the solvent under vacuum. The activated iridium complex was dissolved in THF (10 mL) under an argon atmosphere, and a solution of allyl glycoside **48** (499 mg, 845 μmol) in THF (15 mL) was added. The reaction mixture was stirred for 75 min at rt, and then a solution of iodine (430 mg, 1.69 mmol, 2.0 equiv) in THF/water (15 mL, 4:1 v/v) was added. After 3 h, the excess iodine was quenched by addition of 10% aq sodium bisulfite (10 mL). The reaction mixture was concentrated under reduced pressure to remove THF, water (5 mL) was added, and the aqueous phase was extracted with CH_2Cl_2 (2 × 40 mL). The aqueous phase was concentrated and then suspended in EtOH. The residue was dissolved in AcOH/H_2O (1:1 v/v, 10 mL), and the reaction mixture was heated at 80 °C for 3 h. The volatiles were removed under vacuum and then coevaporated repeatedly with cyclohexane and toluene. Reversed-phase column chromatography of the residue (H_2O/MeOH 80:20) afforded lactol **9** (59 mg, 126 μmol, 15%) as a white amorphous solid (*α*/*β*, 8:2).

Route 2. To a solution of hemiacetal **56** (202 mg, 194 μmol) in anhyd THF (5 mL) under an argon atmosphere was added $Et_3N{\cdot}3HF$ (175 μL, 975 μmol, 5.0 equiv). The reaction mixture was stirred for 19 h, more $Et_3N{\cdot}3HF$ (70 μL, 390 μmol, 2.0 equiv) was added, and the reaction mixture was heated to 50 °C. After 6 h, $Et_3N{\cdot}3HF$ (105 μL, 390 μmol, 2.0 equiv) was added once more, and the reaction mixture was stirred at 50 °C for 20 h before being neutralized by addition of Et_3N (1 mL). Volatiles were removed under vacuum. Column chromatography of the residue (CH_2Cl_2/MeOH, 95:5 → 75:25) followed by several freeze−dry cycles afforded the fully deprotected disaccharide **9** (65 mg, 138 μmol, 70%) as a white amorphous solid (*α*/*β*, 7:3): R_f = 0.39 (*α*), 0.24 (*β*) (CH_2Cl_2/MeOH 4:1); ^{1}H NMR (D_2O) δ 5.33 (d, $J_{1,2}$ = 3.5 Hz, 0.4H, H-$1_D\alpha$), 5.02 (d, $J_{1,2}$ = 1.5 Hz, 0.4H, H-$1_C\alpha$), 4.98−4.96 (m, 1.2H, H-$1_C\beta$, H-$1_D\beta$), 4.18 (dd, $J_{2,3}$ = 10.4 Hz, 0.4H, H-$2_D\alpha$), 4.11−3.78 (m, 7H, H-2_C, H-3_C, H-5_C, H-$2_D\beta$, H-$5_D\alpha$, H-$6a_D$, H-$6b_D$), 3.69−3.62 (m, 1H, H-4_D), 3.60−3.55 (m, 0.6H, H-$5_D\beta$), 3.50 (pt, *J* = 9.8 Hz, H-4_C), 1.31 (d, $J_{5,6}$ = 6.3 Hz, 1.2H, H-$6_C\alpha$), (d, $J_{5,6}$ = 6.3 Hz, 1.8H, H-$6_C\beta$); ^{13}C NMR (D_2O) δ 167.4 (NHCO *β*), 167.1 (NHCO *α*), 103.9 (C-$1_C\beta$), 103.7 (C-$1_C\alpha$), 96.5 (C-$1_D\beta$), 94.3 ($CCl_3\beta$), 94.1 ($CCl_3\alpha$), 93.2 (C-$1_D\alpha$), 83.6 (C-$3_D\beta$), 81.5 (C-$3_D\alpha$), 78.8 (C-$5_D\beta$), 74.5 (C-4_C), 74.4 (C-$5_D\alpha$), 73.3 (C-2_C), 73.0 (C-3_C), 71.6 (C-5_C), 71.3 (C-4_D), 63.4 (C-$6_D\beta$), 63.3 (C-$6_D\alpha$), 61.1 (C-$2_D\beta$), 58.4 (C-$2_D\alpha$), 19.1 (C-6_C); HRMS (ESI$^+$) *m*/*z* calcd for $C_{14}H_{22}Cl_3NO_{10}Na$ $[M + Na]^+$ 492.0207, found 492.0186.

Computational Procedures. All calculations were performed using the hybrid density functional methods[87] integrated in the Gaussian 09 set of programs. The Lee, Yang, and Parr correlation functional method (B3LYP) was chosen.[88,89] The 6-31G(3df,3pd) basis set was employed for each atom. Structural parameters and associated energies result from full geometry optimization in the gas phase, with no imposed constraints. The convergence criteria were set up so that the maximum atomic force is negligible.

Enzymatic Assays. Procedures were as described previously.[50]

■ ASSOCIATED CONTENT

Supporting Information

The Supporting Information is available free of charge on the ACS Publications website at DOI: 10.1021/acs.joc.5b01407.

^{1}H and ^{13}C NMR spectra for the target decasaccharide **5** and all novel compounds described in the Experimental Section (**3**, **6**−**9**, **11**−**22**, **24**−**26**, **36**, **37**, **40**, **45**, **47**−**56**); HPLC profile for decasaccharide **5**; tables of atom coordinates and absolute energies for compounds **25**−**27** (PDF)

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Notes

The authors declare no competing financial interest.

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