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Block synthesis of A tetrasaccharides (types 1, 3, and 4) related to the human ABO blood group system

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ABSTRACT

Blood group A tetrasaccharides of different types have the same terminal trisaccharide fragment that allows using a block scheme in their synthesis. 3-Aminopropyl glycosides of tetrasaccharides GalNAcα1-3(Fucα1-2)Galβ1-3GlcNAcβ (A type 1), GalNAcα1-3(Fucα1-2)Galβ1-3GalNAcα (A type 3), and GalNAcα1-3(Fucα1-2)Galβ1-3GalNAcβ (A type 4) were synthesised using acetylated Galα1-3(Fucα1-2)Gal trichloroacetimidate as a glycosyl donor at the key stage.

1. Introduction

ABO histo-blood group antigens are terminal oligosaccharides in glycosphingolipids and glycoproteins expressed on human erythrocytes and platelets that are widely distributed on vascular endothelia, epithelia, and primary sensory neurons; soluble forms of ABO antigens are found in plasma and body secretions, such as saliva and urine.[1–3] These antigens are major polymorphic alloantigens and potent immunogens, which cause harmful immune reactions in the cases of ABO incompatible transfusion, as well as in bone marrow and organ transplantation. The ABO antigens can be divided into six types in accordance with the structure of disaccharide core (Table 1).[4,5]

Each type of ABO antigens is expressed tissue selectively, so type 1 antigens are expressed mainly in tissues of endodermal origin, whereas type 2 antigens are expressed in various tissues of both ecto- and endodermal origin and are known as key structures of human erythrocytes. Expression of type 3 and type 4 antigens is less well characterized; biological significance of the structures was shown, for example, by Henry and co-workers.[6] The difference between two blood group A subgroups, A_1 and A_2, was first determined as a quantitative disparity of antigen presentation on the erythrocyte surface,[7,8] but later the difference in glycolipid structures was found. Thus A type 4 glycolipid was undetectable in the A_2 subgroup erythrocytes, but was present in the A_1 ones.[6] Aberrant expression of ABO antigens is often observed in the oncogenesis of various organs and in vascular inflammatory processes.[9–11]

The main aim of our investigation is to develop a synthetic approach that will allow us to obtain complete library of blood group ABO antigens. Such a library will provide a possibility for wide-range fundamental biological studies and practical use in the field of hematology and transplantology. To date, several chemical[5,12–19] and chemoenzymatic[20–22] syntheses of different types of histo-blood group antigens have been reported. The methodology of stepwise elongation of the carbohydrate chain starting from the reducing end is most often used in the synthesis of blood group tetrasaccharides.[5,12–18] In one paper the disaccharide glycosyl donor has been used for the synthesis of blood group tetrasaccharide A (type 3).[19] We have recently reported a block synthetic approach to obtaining blood group B tetrasaccharides (types 1, 3, and 4) that allows us to minimize the number of synthetic steps.[23] Similar strategy has been chosen here for the synthesis of blood group A tetrasaccharides **1–3**. Blood group tetrasaccharides A of various types contain the same structural trisaccharide fragment. This makes it possible to use the block scheme '3+1' in their synthesis, where '3' is a glycosyl donor, in particular, the acetylated glycosyl trichloroacetimidate **8** and '1' is the corresponding glucosamine **9** or galactosamine **12** or **13** glycosyl acceptors.

2. Results and discussion

The known aminopropyl glycoside **4**[24] was used as the precursor in trisaccharide glycosyl donor synthesis due to its availability

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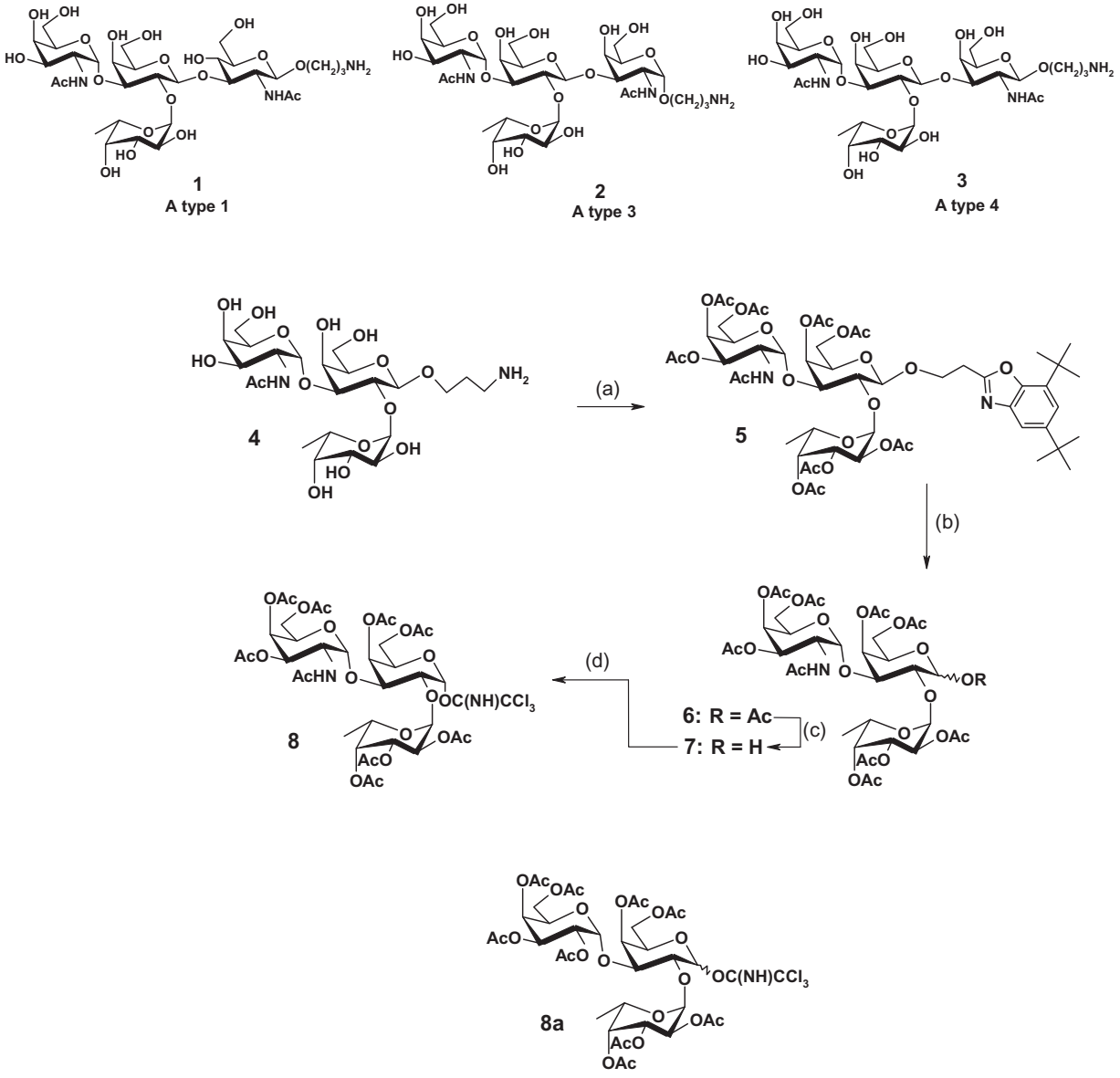
Table 1
Disaccharides cores of the ABO histo-blood group antigens

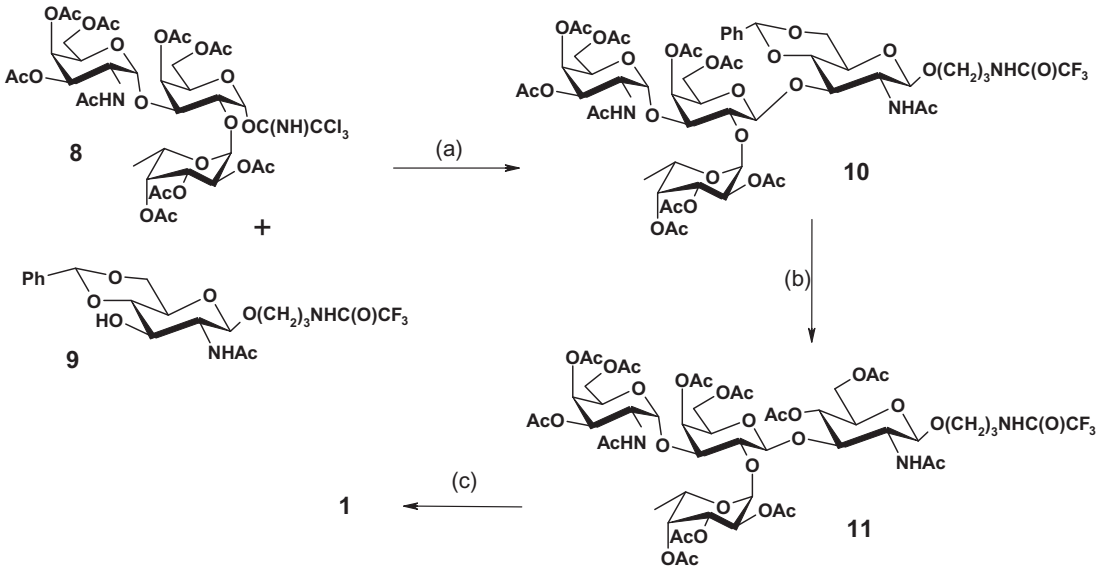
Type	Disaccharide core
Type 1	Gal[a]β1-3GlcNAcβ
Type 2	Galβ1-4GlcNAcβ
Type 3	Galβ1-3GalNAcα
Type 4	Galβ1-3GalNAcβ
Type 5	Galβ1-3Galβ
Type 6	Galβ1-4Glcβ

[a] All monosaccharide residues are in pyranose form.

in multi-gram quantity and its well-established scaled-upsynthesis. The method of aminopropyl spacer elimination previously published[25] and optimized[19] was used for obtaining of peracetylated trisaccharide **6**. Derivatization of aminopropyl glycoside **4** with 3,5-di-*tert*-butyl-1,2-benzoquinone, treatment of the resulting azomethine with oxalic acid dihydrate, and acetylation afforded the resulting benzoxazole derivative **5** in 92% yield (Scheme 1). The structure of compound **5** was confirmed by mass spectrometry (MS *m/z* 1123.4836 (M^++H)) and ^{1}H NMR spectroscopy data (two singlets at δ 1.37 (9H) and δ 1.49 (9H) related to *tert*-butyl groups and two doublets at δ 7.28 (1H) and δ 7.57 (1H) of aromatic protons). Base-catalyzed acetolysis/acetylation of benzoxazole **5** yielded the peracetylated trisaccharide **6** (96%). Then selective 1-O-deacetylation of derivative **6** with hydrazine acetate[26] and treating with trichloroacetonitrile in the presence of 1,8-diazobicyclo[5.4.0]undec-7-ene (DBU) gave glycosyl trichloroacetimidate **8**, which was isolated by flash chromatography on silica gel in 90% yield. We have to mention that only α-trichloroacetimidate **8** (δ 6.48, $J_{1,2}$ 3.7 Hz) of trisaccharide A was obtained. The synthesis of peracetylated B trisaccharide trichloroacetimidate **8a** reported earlier led to the mixture of α- and β-isomers under the same reaction conditions.[19] We cannot properly explain the differences in trichloroacetimidate formation, as the only disparity in the trisaccharide structures is the presence of an OAc-group (in the case of B trisaccharide) and an NHAc-group (in the case of A trisaccharide) in position 2 of the terminal galactose unit.



Scheme 1. Reagents and conditions: (a) (i) 3,5-di-*tert*-butyl-1,2-benzoquinone, MeOH; (ii) $(COOH)_2{\cdot}2H_2O$ (pH 4); (iii) Ac_2O/Py, 92% (total for three stages); (b) NaOAc, 1:1 Ac_2O–AcOH, 100 °C, 10 days, 96%; (c) $N_2H_4{\cdot}HOAc$, DMF, 50 °C, 95%; (d) Cl_3CCN, DBU, CH_2Cl_2, 90%.



Scheme 2. Reagents and conditions: (a) TMSOTf, 3 Å MS, 1:1 CH_3CN–CH_2Cl_2; (b) (i) AcOH (80%), 80 °C; (ii) Ac_2O/Py, 56% (total for three stages); (c) (i) MeONa/MeOH; (ii) H_2O, 97%.

The key stage of the tetrasaccharides synthesis was the glycosylation of the known glycosyl acceptors **9**,[27] **12**,[28] and **13**[28] with trisaccharide trichloroacetimidate **8**.

2.1. Synthesis of 3-aminopropyl glycoside of the A (type 1) tetrasaccharide

The glycosylation of acceptor **9** with trichloroacetimidate **8** (Scheme 2) was carried out in acetonitrile–dichloromethane mixture at room temperature using a twofold excess of the glycosyl acceptor and trimethylsilyl triflate as the reaction promoter to yield tetrasaccharide **10**.

To facilitate purification, the product **10** was treated with AcOH to remove the benzylidene acetal, acetylated (Ac_2O/Py) followed by column chromatography on silica gel afforded the peracetylated derivative **11** in 56% yield (total for three stages). The stereochemistry of the glycosidic bond thus formed, was confirmed by NMR spectroscopy; **11** showed a signal of an anomeric proton at δ 4.37 as a doublet with J = 7.5 Hz, which is consistent with the β-configuration of glycosidic linkage. The corresponding α-isomer was not isolated. Complete deacetylation and removal of the trifluoroacetyl group in derivative **11** by aqueous alkali followed by cation-exchange chromatography afforded the 3-aminopropyglycoside of the A (type 1) tetrasaccharide **1**, in a yield of 97%.

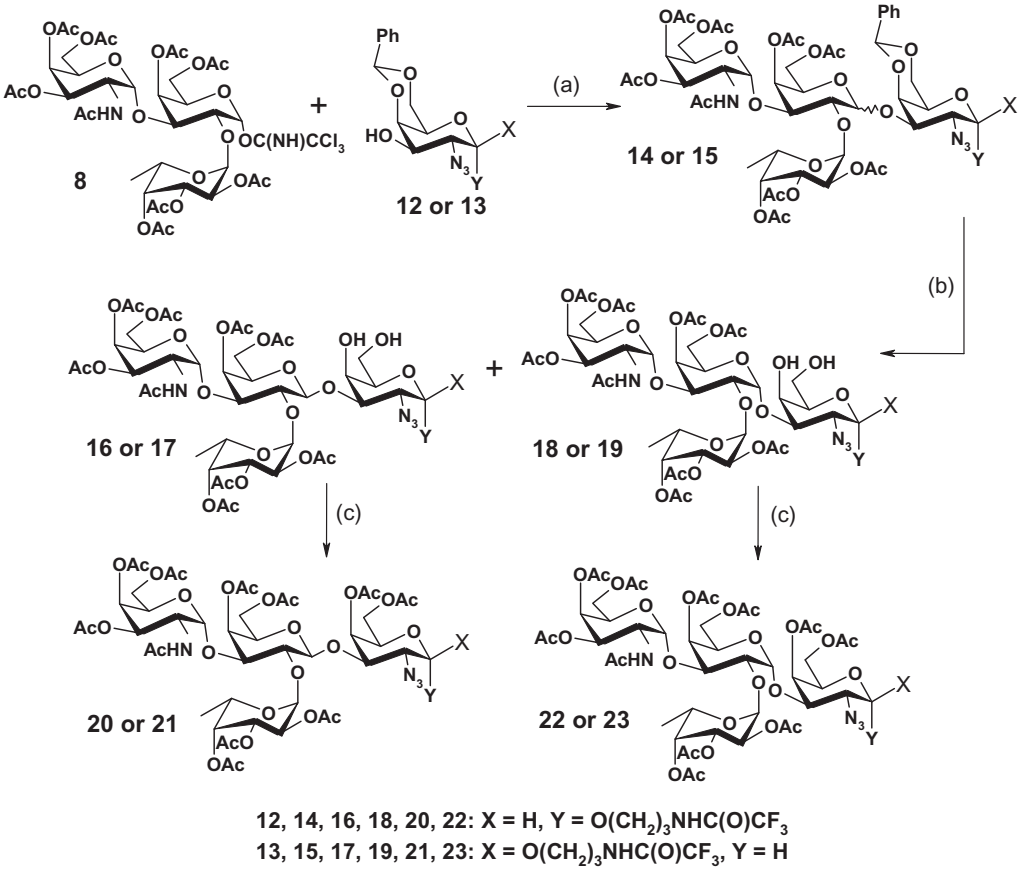
2.2. Synthesis of 3-aminopropyl glycosides of the A (type 3) and A (type 4) tetrasaccharides

In the synthesis of the A tetrasaccharides (type 3 and type 4), coupling of the trichloroacetimidate **8** with glycosyl acceptors **12** and **13** (Scheme 3) in acetonitrile in the presence of TMSOTf at room temperature gave corresponding mixtures **14** and **15** of the anomeric tetrasaccharides. To facilitate the separation of the type 3 tetrasaccharide, mixture **14** was treated with AcOH to remove the benzylidene acetal; subsequent column chromatography on silica gel afforded the required A (type 3) tetrasaccharide **16** (42%) and its α anomer **18** (18%). Both compounds were acetylated, and the structures of resulting derivatives **20** and **22** were confirmed by ^{1}H, 2D-COSY, and ^{13}C NMR and MS data. At this stage, the stereochemistry of the newly formed glycosylic bond was confirmed: ^{1}H NMR of **20** showed a signal of an anomeric proton at δ 4.69 as a doublet with J = 4.1 Hz, which is consistent with the β-configuration of the glycosidic linkage. ^{1}H NMR of **22** showed a signal of an anomeric proton at 5.28 as a doublet with a coupling constant of 4.1 Hz, consistent with the α-configuration of the glycosidic bond. For the separation of the type 4 tetrasaccharides, the anomeric tetrasaccharide mixture **15** was treated with AcOH and acetylated; subsequent column chromatography on silica gel afforded the required A (type 4) tetrasaccharide **21** (45%) and its α-anomer **23** (22%). ^{1}H NMR data confirmed the configuration of the formed glycosidic bond: an anomeric proton appeared as a doublet at δ 4.74 with J = 7.3 Hz (β-configuration) for tetrasaccharide **21**, and as a doublet at δ 5.45 with a coupling constant of 3.4 Hz (α-configuration) for tetrasaccharide **23**.

Though the β anomer is the major product of glycosylation under the reaction conditions described above, the α anomer is also generated, with the β/α ratio about 2.3:1 in the case of the A (type 3) and 2:1 in the case of the A (type 4) tetrasaccharides.

To check the possibility of improving the β-stereoselectivity, the coupling was performed at reduced temperature. The reaction was carried out in acetonitrile in the presence of TMSOTf.

It is proposed that if low temperatures are employed in the generation of the glycosyl cation, this may be attacked by acetonitrile along the kinetically favored axial direction to form an α-nitrillium ion.[29] As a result, β glycosides can predominate in the reaction products. In our case, performing the reaction at −20 °C did not result in the formation of any glycosylation product during 24 h. Only traces of trichloroacetimidate-derived by-products were registered. After the reaction temperature was increased to 4 °C, the glycosylation was completed in 1.5 h; the β/α-ratio was slightly improved and reached 3.3:1, as compared to that of 2.1:1, when the reaction proceeded at room temperature (Table 2). At the initial reaction temperature of −5 °C, only a trace amount of reaction products was registered after 3 h. The increase of the reaction temperature to 0 °C also had no effect on the glycosylation yield, resulting in accumulation of by-products. After the reaction temperature was raised to 4 °C, the glycosylation completed in 2 h, and the β/α-ratio was 3.0:1. The decrease of the yields of glycosylation products in the model experiments are likely caused by increased time without reaction progress at minus temperatures accompanied by the accumulation of by-products.



Scheme 3. Reagents and conditions: (a) TMSOTf, 3 Å MS, CH_3CN; (b) AcOH (80%), 80 °C; (c) Ac_2O/Py.

Table 2
Influence of temperature and solvent on the stereochemistry of the glycosylation reaction

Glycosyl acceptor	Solvent	Temperature (°C)	β/α[a]	Yield (%)
12	CH_3CN	rt	2.3:1	79
12	CH_3CN	−5→4	3.0:1	70
13	CH_3CN	rt	2.1:1	81
13	CH_3CN	−20→4	3.3:1	69
13	CH_2Cl_2	rt	1:1.6	77

[a] The values of β/α ratio were estimated from the ^1H NMR spectra of the anomeric mixtures.

The experiment using CH_2Cl_2 as a solvent was carried out for additional confirmation of the co-participation of acetonitrile in the coupling. In this case, the glycosylation proceeded with reverse stereoselectivity; the β/α-ratio was 1:1.6.

Thus, we can conclude that acetonitrile as a solvent and 4 °C are the most favorable conditions for preferential formation of β glycosides in glycosylation of galactosamine derivatives **12** and **13** with the trisaccharide trichloroacetimidate **8**.

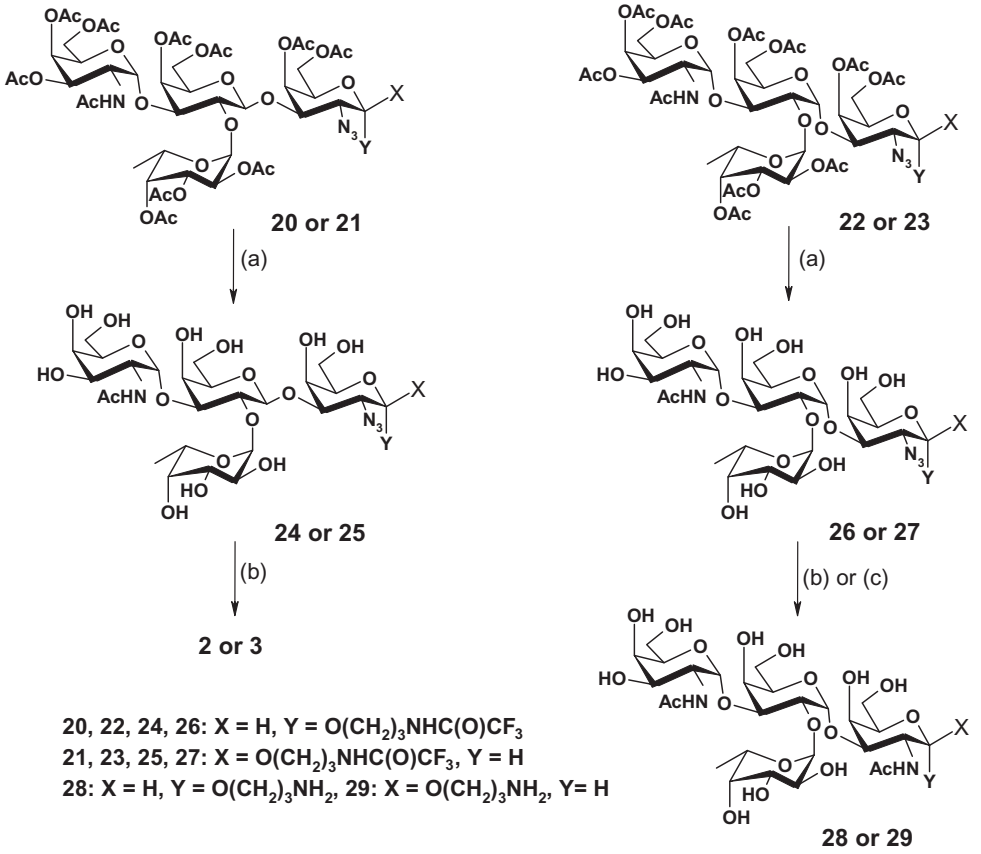
The target A tetrasaccharides **2** (type 3) and **3** (type 4) were obtained from the derivatives **20** and **21** by the following deprotection procedures: the Zemplén deacetylation, catalytic reduction of azide over 10% Pd/C followed by N-acetylation, and alkali treatment to remove the trifluoroacetic group (Scheme 4). Purification of the tetrasaccharides by cation-exchange chromatography completed the synthesis and afforded the tetrasaccharides **2** and **3** in 77% and 64% yields, respectively.

Deprotection procedures for the α anomer of the A (type 4) tetrasaccharide **23** were the same as those described for tetrasaccharide **21**. Unfortunately, the low yield at the step of catalytic reduction of the azido group resulted in only 25% total yield of tetrasaccharide **29**. Therefore, dithiothreitol (DTT) was used for the reduction of the azido group in the case of the α anomer of the A (type 3) tetrasaccharide. Application of DTT for reduction of the sugar azides in organic solvents, dichloromethane,[30] and acetonitrile[31] is known. The use of DTT aqueous media for reduction of azido nucleosides has also been reported.[32] Using DTT in 0.2 M aq $NaHCO_3$ (pH 8.2) for reduction of the azide in **26** allowed us to get tetrasaccharide **28** in 75% total yield.

3. Conclusions

Glycosylation of monosaccharide acceptors (**9**, **12**, and **13**) with A trisaccharide trichloroacetimidate **8** preferentially leads to the formation of β glycosides in spite of the absence of 2-*O*-acyl participation. We suppose that the glycosyl donor structure is the main reason for β-stereoselectivity: the bulky substituent at C-2, acetylated fucose, and conformational rigidity of the molecule due to the presence of two monosaccharide residues in adjacent positions [Fuc at C-2 and GalNAc at C-3] hinder the nucleophilic attack from the α-side, thus resulting in the formation of β-glycosides. Additionally, the use of acetonitrile as a solvent upon glycosylation also assists β-stereoselectivity.[29]

Synthesized here the A tetrasaccharides together with B tetrasaccharides described earlier[19] allowed us to perform two investigations of carbohydrate-binding proteins. First, with the help of Glyc-PAA-fluorescein probes [where Glyc are the blood group tetrasaccharides A and B; PAA is a poly(acrylamide) matrix] we have found that tandem type galectins (-4, -8, and -9) are anchored on the cell surface with the N-domain, whereas the C-carbohydrate binding domain is exposed for external binding and displays high



Scheme 4. Reagents: (a) MeONa/MeOH; (b) (i) H_2, Pd/C, MeOH/Ac_2O; (ii) NaOH; (c) (i) DTT, aq $NaHCO_3$ (pH 8.2), Ac_2O; (ii) NaOH.

affinity for the blood group A and B tetrasaccharides.[33] Second, using the corresponding PAA-probes, and a printed glycan array with the tetrasaccharides and Sepharose affinity adsorbents, we have demonstrated that blood of the A blood group donors contains formally autologous antibodies against the A antigen. These antibodies incapable of binding A (type 2) tetrasaccharide are specific to the small GalNAcα1-3Gal motif, and thus are not autoantibodies.[34]

4. Experimental

The reactions were performed with the use of commercial reagents (Acros, Aldrich, and Fluka); anhydrous solvents were purified according to the standard procedures. Column chromatography was performed on Silica Gel 60 0.040–0.063 mm (E. Merck), gel filtration was carried out on Sephadex LH-20 (Pharmacia) columns (elution with 1:1 $CHCl_3$–MeOH, unless otherwise specified). Solvents were removed in vacuum at 30–40 °C. Thin-layer chromatography (TLC) was performed on Silica Gel 60 F_{254} aluminium-backed plates (E. Merck). Spots of compounds were visualized by dipping a TLC plate into an aqueous solution of H_3PO_4 (8%) and subsequent heating (>150 °C). Deacetylation was carried out in absolute MeOH by the addition of catalytic amount of 2 M MeONa in MeOH (according to Zemplén). Na^+ ions were then removed by Dowex 50X4-400 (Acros) (H^+) cation exchanger, and the solution was evaporated. Hydrogenolysis was carried out on 10% Pd/C (E. Merck) in the atmosphere of H_2.

The values of optical rotations were measured on a Perkin–Elmer 341 polarimeter at 21 ± 2 °C. ^{1}H NMR spectra were recorded on a Bruker BioSpin GmbH (700 MHz) spectrometer at 30 °C; chemical shifts (δ-units) were referred to the peak of internal D_2O (δ 4.750), $CDCl_3$ (δ 7.270), or CD_3OD (δ 3.500); coupling constants (J) were measured in Hertz. Signals of ^{1}H NMR spectra were assigned to the corresponding protons using 2D spectroscopy (COSY). ^{13}C NMR spectra were recorded at 150 MHz. Symbols of monosaccharide residues in NMR spectra for trisaccharides: I—α/β-Gal (reducing end), II—α-Fuc, III—α-GalNAc; for tetrasaccharides: I—α/β-GalNAc or β-GlcNAc (reducing end), II—α/β-Gal, III—α-Fuc, IV—α-GalNAc. ESIMS spectra were recorded on an Exactive Orbitrap (Thermo Fisher Scientific, Germany) spectrometer; HRESIMS spectra were recorded on an Agilent 6224 TOF LC/MS instrument (USA).

4.1. 3-Aminopropyl 2-acetamido-2-deoxy-α-D-galactopyranosyl-(1→3)-[α-L-fucopyranosyl-(1→2)]-β-D-galactopyranosyl-(1→3)-2-acetamido-2-deoxy-β-D-glucopyranoside (1)

Tetrasaccharide **11** (85 mg, 0.065 mmol) was deacetylated using Zemplén procedures. Water (2 mL) was then added to the solution, MeOH was evaporated, and the reaction mixture was kept for 3 h. Ion-exchange chromatography on Dowex 50X4-400 (H^+) (elution with 5% aq ammonia) afforded 50 mg (97%) of product **1** as a white foam: $[\alpha]_D$ +18 (*c* 0.50, 1:1 H_2O–CH_3CN), R_f 0.10 (100:10:10:10:2 EtOH–BuOH–Py–H_2O–AcOH), ^{1}H NMR (D_2O): (characteristic signals) δ 1.25 (d, 3H, $J_{5,6}$ 6.6, H-6^{III}), 1.90–1.99 (m, 2H, CCH_2C), 2.04–2.09 (2s, 6H, 2 C(O)CH_3), 3.05–3.09 (m, 2H, NHCH_2), 4.34 (q, 1H, $J_{5,6}$ 6.6, H-5^{III}), 4.43 (d, 1H, $J_{1,2}$ 8.5, H-1^{I}), 4.71 (d, 1H, $J_{1,2}$ 7.5, H-1^{II}), 5.19 (d, 1H, $J_{1,2}$ 3.8, H-1^{IV}), 5.27 (d, 1H, $J_{1,2}$ 4.2, H-1^{III}); ^{13}C NMR (D_2O): δ 15.2 (C-6^{III}), 22.0 (CH_3C(O)), 22.3 (CH_3C(O)), 26.8 (CCH_2C), 37.5 (CCH_2N), 49.7 (C-2^{IV}), 54.8 (C-2^{I}), 60.7 (CCH_2O), 61.2, 61.4, 63.0 (C-6^{I}, C-6^{II}, C-6^{IV}), 66.7, 67.69, 67.70, 67.9, 68.6, 68.8, 69.8, 71.0, 71.9, 73.8, 74.9, 75.49, 75.53, 77.3 (C-2^{II}, C-2^{III}, C-3^{I}, C-3^{II}, C-3^{IV}, C-3^{III}, C-4^{I}, C-4^{II}, C-4^{IV}, C-4^{III}, C-5^{I}, C-5^{II}, C-5^{III}, C-5^{IV}), 91.3 (C-1^{IV}), 99.1, 100.0, 101.9 (C-1^{I}, C-1^{II},

C-1^{III}), 174.0 (CH_3C(O)), 174.9 (CH_3C(O)); HRESIMS, *m/z*: Calcd $[C_{31}H_{55}N_3O_{20}]H^+$: *m/z* 790.3452. Found: *m/z* 790.3453.

4.2. 3-Aminopropyl 2-acetamido-2-deoxy-α-D-galactopyranosyl-(1→3)-[α-L-fucopyranosyl-(1→2)]-β-D-galactopyranosyl-(1→3)-2-acetamido-2-deoxy-α-D-galactopyranoside (2)

Tetrasaccharide **20** (62 mg, 0.048 mmol) was deacetylated under Zemplén conditions, purified by gel filtration (1:1 CH_3CN–H_2O), and subjected to hydrogenolysis (3 h) in a mixture of MeOH (5 mL) and Ac_2O (200 μL). The reaction mixture was then filtered and concentrated. The residue was dissolved in H_2O (3 mL), an aq solution of NaOH (2 M, 5 μL) was added, and the reaction mixture was kept for 3 h at rt. Ion-exchange chromatography on Dowex 50X4-400 (H^+) (elution with 5% aq ammonia) gave 29 mg (77%) of product **2** as white foam: $[\alpha]_D$ +99 (*c* 0.4, 1:1 H_2O–CH_3CN), R_f 0.17 (100:10:10:10:2 EtOH–BuOH–Py–H_2O–AcOH), ^{1}H NMR (D_2O): (characteristic signals) δ 1.21 (d, 3H, $J_{5,6}$ 6.6, H-6^{III}), 1.95–2.00 (m, 2H, CCH_2C), 2.03–2.08 (2s, 6H, 2 C(O)CH_3), 3.09–3.13 (m, 2H, NHCH_2), 4.30 (q, 1H, $J_{5,6}$ 6.6, H-5^{III}), 4.66 (d, 1H, $J_{1,2}$ 7.5, H-1^{II}), 4.88 (d, 1H, $J_{1,2}$ 3.5, H-1^{I}), 5.16 (d, 1H, $J_{1,2}$ 3.8, H-1^{IV}), 5.27 (d, 1H, $J_{1,2}$ 4.2, H-1^{III}); ^{13}C NMR (D_2O): δ 15.3 (C-6^{III}), 21.9 (*C*H_3C(O)), 22.0 (*C*H_3C(O)), 26.8 (CCH_2C), 37.1 (CCH_2N), 49.4, 49.6 (C-2^{I}, C-2^{IV}), 61.1, 61.3, 61.5 (C-6^{I}, C-6^{II}, C-6^{IV}), 64.9 (CCH_2O), 63.1, 66.9, 67.7, 67.8, 68.6, 69.2, 70.0, 70.7, 71.0, 71.8, 72.8, 74.4, 74.9, 75.5 (C-2^{II}, C-2^{III}, C-3^{I}, C-3^{II}, C-3^{III}, C-3^{IV}, C-4^{I}, C-4^{II}, C-4^{III}, C-4^{IV}, C-5^{I}, C-5^{II}, C-5^{III}, C-5^{IV}), 91.3 (C-1^{IV}), 96.7 (C-1^{I}), 98.8 (C-1^{III}), 102.2 (C-1^{II}), 173.6 (CH_3C(O)), 174.9 (CH_3C(O)); HRESIMS, Calcd $[C_{31}H_{55}N_3O_{20}]H^+$: *m/z* 790.3452. Found: *m/z* 790.3451.

4.3. 3-Aminopropyl 2-acetamido-2-deoxy-α-D-galactopyranosyl-(1→3)-[α-L-fucopyranosyl-(1→2)]-β-D-galactopyranosyl-(1→3)-2-acetamido-2-deoxy-β-D-galactopyranoside (3)

Tetrasaccharide **21** (102 mg, 0.079 mmol) was deacetylated under Zemplén conditions. The product obtained was purified by gel filtration (1:1 CH_3CN–H_2O) and subjected to hydrogenolysis (5 h) in a mixture of MeOH (5 mL) and Ac_2O (300 μL). Reaction mixture was then filtered and concentrated. The residue was dissolved in H_2O (4 mL), an aq solution of NaOH (2 M, 5 μL) was added, and the reaction mixture was kept for 2 h. Cation-exchange chromatography on Dowex 50X4-400 (H^+) (elution with 5% aq ammonia) gave 40 mg (64%) of product **3** as white foam: $[\alpha]_D$ +3 (*c* 0.4, 1:1 H_2O–CH_3CN), R_f 0.14 (100:10:10:10:2 EtOH–BuOH–Py–H_2O–AcOH), ^{1}H NMR (D_2O): (characteristic signals) δ 1.21 (d, 3H, $J_{5,6}$ 6.6, H-6^{III}), 1.88–1.98 (m, 2H, CCH_2C), 2.03–2.07 (2s, 6H, 2C(O)CH_3), 3.06–3.09 (m, 2H, NHCH_2), 4.33 (d, 1H, $J_{1,2}$ 8.2, H-1^{I}), 4.66 (d, 1H, $J_{1,2}$ 7.8, H-1^{II}), 5.17 (d, 1H, $J_{1,2}$ 3.7, H-1^{IV}), 5.28 (d, 1H, $J_{1,2}$ 4.0, H-1^{III}); ^{13}C NMR (D_2O): δ 15.3 (C-6^{III}), 22.0 (*C*H_3C(O)), 22.3 (*C*H_3C(O)), 26.7 (CCH_2C), 37.7 (CCH_2N), 49.6, 51.3 (C-2^{I}, C-2^{IV}), 61.0, 61.1, 61.4 (C-6^{I}, C-6^{II}, C-6^{IV}), 63.1 (CCH_2O), 66.9, 67.7, 67.8, 68.0, 68.54, 68.56, 68.57, 69.9, 71.0, 71.8, 72.8, 74.9, 75.6, 77.1 (C-2^{II}, C-2^{III}, C-3^{I}, C-3^{II}, C-3^{III}, C-3^{IV}, C-4^{I}, C-4^{II}, C-4^{III}, C-4^{IV}, C-5^{I}, C-5^{II}, C-5^{III}, C-5^{IV}), 91.3 (C-1^{IV}), 98.8 (C-1^{III}), 102.2 (C-1^{II}), 102.7 (C-1^{I}) 174.0 (CH_3C(O)), 174.9 (CH_3C(O)), HRESIMS, Calcd $[C_{31}H_{55}N_3O_{20}]H^+$: *m/z* 790.3452. Found: *m/z* 790.3453.

4.4. 2-(4,6-Di-*tert*-butylbenzoxazol-2-yl)ethyl 2-acetamido-3,4,6-tri-*O*-acetyl-2-deoxy-α-D-galactopyranosyl-(1→3)-[2,3,4–tri-*O*-acetyl-α-L-fucopyranosyl-(1→2)]-4,6-di-*O*-acetyl-β-D-galactopyranoside (5)

3,5-Di-*tert*-butyl-*o*-benzoquinone (448 mg, 2.00 mmol) was added with stirring to a solution of trisaccharide **4** (590 mg, 1.01 mmol) in 30 mL of MeOH. The reaction mixture changed its color from dark brown to green in 5 min. The solution was stirred for 3 h, then treated with $(COOH)_2{\cdot}2H_2O$ (to pH 4), and the reaction mixture was left overnight at −20 °C. The solution was concentrated in vacuo; the residue was washed with 2:1 EtOAc–C_6H_6 (100 mL) and acetylated (Ac_2O/Py, 48 h). The reaction mixture was poured into ice and extracted with $CHCl_3$ (3 × 70 mL). The organic fraction was washed with H_2O (3 × 50 mL), the aq fraction was then extracted with $CHCl_3$ (2 × 20 mL). The combined $CHCl_3$ fractions were dried by filtration through cotton wool and concentrated. Column chromatography on silica gel (6:3:1 *n*-C_6H_{14}–$CHCl_3$–2-PrOH) yielded benzoxazole **5** (1.04 g, 92%) as white foam, $[\alpha]_D$ −2 (*c* 1, $CHCl_3$), R_f 0.50 (4:3:1 *n*-C_6H_{14}–$CHCl_3$–2-PrOH),^{1}H NMR ($CDCl_3$): δ 1.08 (d, 3H, $J_{5,6}$ 6.5, H-6^{II}), 1.37 (s, 9H, C(CH_3)$_3$), 1.49 (s, 9H, –C(CH_3)$_3$), 1.96–2.17 (9s, 27 H, 8 OC(O)CH_3, NHC(O)CH_3), 3.31–3.35 (m, 2H, CCH_2), 3.79–3.83 (m, 2H, H-2^{I}, OC*H*H), 3.88 (dd, 1H, $J_{2,3}$ 9.3, $J_{3,4}$ 3.5, H-3^{I}), 4.02–4.16 (m, 5H, H-6a^{I}, H-6b^{I}, H-6a^{III}, H-6b^{III}, OCH*H*), 4.27 (dd, 1H, $J_{5,6a}$ 6.1, $J_{5,6b}$ 6.1, H-5^{III}), 4.41–4.51 (m, 3H, H-5^{I}, H-2^{III}, H-5^{II}), 4.52 (d, 1H, $J_{1,2}$ 7.4, H-1^{I}), 5.02 (dd, 1H, $J_{2,3}$ 11.4, $J_{3,4}$ 3.2, H-3^{III}), 5.23–5.26 (m, 3H, H-1^{III}, H-3^{II}, H-4^{II}), 5.32 (dd, 1H, $J_{1,2}$ 3.7, $J_{2,3}$ 11.2, H-2^{II}), 5.38 (d, 1H, $J_{3,4}$ 3.2, H-4^{III}), 5.47 (d, 1H, $J_{3,4}$ 3.5, H-4^{I}), 5.51 (d, 1H, $J_{1,2}$ 3.7, H-1^{II}), 6.13 (d, 1H, $J_{2,NH}$ 8.4, N*H*Ac^{III}), 7.28 (d, 1H, *J* 1.7, Ar*H*), 7.57 (d, 1H, *J* 1.7, Ar*H*); ^{13}C NMR ($CDCl_3$): δ 15.9 (C-6^{II}), 20.6–20.6 (8OC(O)CH_3), 23.2 (NC(O)CH_3), 29.3 (CCH_2), 29.9 (C(CH_3)$_3$), 31.8 (C(CH_3)$_3$), 34.5 (*C*(CH$_3$)$_3$), 35.2 (*C*(CH$_3$)$_3$), 47.9 (C-2^{III}), 61.1 (C-6^{I}), 62.8 (C-6^{III}), 65.2 (C-5^{II}), 66.5 (C-4^{III}), 66.8 (OCH$_2$), 67.2 (C-4^{I}), 67.39 (C-2^{II}), 67.43 (C-3^{II}), 67.7 (C-3^{III}), 68.1 (C-5^{III}), 70.3 (C-5^{I}), 71.2 (C-4^{II}), 75.5 (C-2^{I}), 77.8 (C-3^{I}), 96.7 (C-1^{II}), 97.1 (C-1^{III}), 102.6 (C-1^{I}), 113.3, 119.9, 134.0, 139.1, 146.8, 148.4, 163.5 (C-Ar), 169.8–171.0 (9*C*(O)CH$_3$); HRESIMS: Calcd $[C_{53}H_{74}N_2O_{24}]H^+$: *m/z* 1123.4704. Found: *m/z* 1123.4836.

4.5. 2-Acetamido-3,4,6-tri-*O*-acetyl-2-deoxy-α-D-galactopyranosyl-(1→3)-[2,3,4–tri-*O*-acetyl-α-L-fucopyranosyl-(1→2)]-4,6-di-*O*-acetyl-β-D-galactopyranoside trichloroacetimidate (8)

A solution of benzoxazole **5** (1.04 g, 0.930 mmol) and NaOAc (2.5 g, 31 mmol) in a mixture of 1:1 AcOH–Ac_2O (37 mL) was sealed in glass ampoules and kept at 100 °C for 10 days. The ampoules were then opened, and reaction mixture was poured onto ice and extracted with $CHCl_3$ (3 × 75 mL).The organic fraction was washed with cold satd aq $NaHCO_3$ (2 × 150 mL) and H_2O (2 × 150 mL), and the aqueous fraction was extracted with $CHCl_3$ (2 × 100 mL). The combined $CHCl_3$ fractions were dried by filtration through cotton wool and concentrated. Column chromatography on silica gel (6:3:1 *n*-C_6H_{14}–$CHCl_3$–2-PrOH) yielded the mixture of anomeric acetates **6** (810 mg, 96%) as white foam, R_f 0.42 (4:3:1 *n*-C_6H_{14}–$CHCl_3$–2-PrOH), ESIMS: Calcd $[C_{38}H_{53}NO_{24}]H^+$ *m/z* 908.30. Found: *m/z* 908.34.

A solution of anomeric acetates **6** (300 mg, 0.330 mmol) in 3 mL of DMF was heated to 50 °C with stirring, and $N_2H_4{\cdot}$HOAc (43 mg, 0.46 mmol) was then added. The reaction was stirred for 20 min, diluted with $CHCl_3$ (10 mL), and washed with H_2O (3 × 20 mL). The aq fraction was extracted with $CHCl_3$ (2 × 5 mL). The combined $CHCl_3$ fractions were dried by filtration through cotton wool and concentrated. Column chromatography on silica gel (5:3:1 *n*-C_6H_{14}–$CHCl_3$–2-PrOH) afforded 270 mg (95%) of hemiacetal **7** as white foam, R_f 0.26 (4:3:1 *n*-C_6H_{14}–$CHCl_3$–2-PrOH).

A stirred mixture of derivative **7** (270 mg, 0.312 mmol), Cl_3CCN (334 μL, 3.37 mmol), and CH_2Cl_2 (5 mL) was cooled to −20 °C at stirring, and then DBU (5 μL, 0.03 mmol) was added. The reaction mixture was stirred for 3.5 h and concentrated. Column chromatography on silica gel (1:3 $PhCH_3$–EtOAc, 1% Et_3N) gave trichloroacetimidate **8** (284 mg, 90%) as white foam: R_f 0.25 (1:1 *n*-C_6H_{14}–

acetone, 1% Et_3N), ^1H NMR ($CDCl_3$): δ 1.09 (d, 3H, $J_{5,6}$ 6.5, H-6^{II}), 1.94–2.22 (9s, 27 H, 8 OC(O)CH_3, NHC(O)CH_3), 4.04–4.09 (m, 3H, H-6a^{I}, H-6a^{III}, H-6b^{III}), 4.11 (dd, 1H, $J_{1,2}$ 3.7, $J_{2,3}$ 9.8, H-2^{I}), 4.18–4.23 (m, 2H, H-6b^{I}, H-5^{II}), 4.28 (dd, 1H, $J_{5,6a}$ 6.6, $J_{5,6b}$ 6.6, H-5^{III}), 4.30 (dd, 1H, $J_{2,3}$ 9.8, $J_{3,4}$ 3.7, H-3^{I}), 4.37 (dd, 1H, $J_{5,6a}$ 6.6, $J_{5,6b}$ 6.6, H-5^{I}), 4.55 (ddd, 1H, $J_{1,2}$ 3.6, $J_{2,3}$ 11.3, $J_{2,NH}$ 8.8, H-2^{III}), 5.01 (dd, 1H, $J_{2,3}$ 11.3, $J_{3,4}$ 3.1,H-3^{III}), 5.22–5.24 (m, 2H, H-3^{II}, H-4^{II}), 5.31 (d, 1H, $J_{1,2}$ 3.6, H-1^{III}), 5.32–5.35 (m, 1H, H-2^{II}), 5.39 (d, 1H, $J_{1,2}$ 3.6, H-1^{II}), 5.44 (d, 1H, $J_{3,4}$ 3.1, H-4^{III}), 5.54 (d, 1H, $J_{3,4}$ 3.7, H-4^{I}), 5.90 (d, 1H, $J_{2,NH}$ 8.8, NHAc^{III}), 6.48 (d, 1H, $J_{1,2}$ 3.7, H-1^{I}), 8.62 (s, 1H, C(NH)CCl$_3$); ^{13}C NMR ($CDCl_3$): δ 16.0 (C-6^{II}), 20.5–20.8 (8 OC(O)CH_3), 23.2 (NC(O)CH_3), 47.9 (C-2^{III}), 61.4 (C-6^{I}), 62.1 (C-6^{III}), 65.8 (C-5^{II}), 66.9 (C-2^{II}), 67.1 (C-4^{I}), 67.28 (C-4^{III}), 67.34 (C-3^{II}), 67.76 (C-5^{III}), 68.8 (C-5^{I}), 71.1 (C-4^{II}), 73.5 (C-3^{I}), 74.6 (C-2^{I}), 90.7 (C(NH)CCl$_3$), 94.7 (C-1^{I}), 96.8 (C-1^{III}), 98.3 (C-1^{II}), 160.8 (C(NH)CCl$_3$), 169.8–170.6 (9C(O)CH$_3$), ESIMS: Calcd [$C_{38}H_{51}N_2O_{23}Cl_3$]H$^+$: *m/z* 1011.20. Found: *m/z* 1011.24.

4.6. 3-Trifluoroacetamidopropyl 2-acetamido-3,4,6-tri-*O*-acetyl-2-deoxy-α-D-galactopyranosyl-(1→3)-[2,3,4-tri-*O*-acetyl-α-L-fucopyranosyl-(1→2)]-4,6-di-*O*-acetyl-β-D-galactopyranosyl-(1→3)-2-acetamido-4,6-di-*O*-acetyl-2-deoxy-β-D-glucopyranoside (11)

A mixture of trichloroacetimidate **8** (269 mg, 0.266 mmol), glycosyl acceptor **9** (246 mg, 0.532 mmol), anhyd CH_3CN (7 mL), and anhyd CH_2Cl_2 (7 mL) was stirred with 3 Å molecular sieves (500 mg) for 30 min under an atmosphere of N_2. A solution of TMSOTf (16 μL, 0.090 mmol) in anhyd CH_3CN (0.3 mL) was then added, and the reaction mixture was stirred for 5 h. The mixture was neutralized with Et_3N (50 μL) and filtered. The filtrate was concentrated and subjected to gel filtration followed by column chromatography on silica gel (1:1→1:2 *n*-C_6H_{14}–acetone) to afford compound **10** (282 mg) as a colorless oil containing about 10% of impurities, which was immediately used for the next step. The colorless oil was dissolved in AcOH (80%), kept for 1 h at 80 °C, concentrated, and co-evaporated with $PhCH_3$ (2 × 15 mL). The residue was acetylated (Ac_2O/Py, 10 h), concentrated, co-evaporated with $PhCH_3$ (4 × 10 mL), and subjected to column chromatography on silica gel (2:3 *n*-C_6H_{14}–acetone) to give tetrasaccharide **11** (194 mg, 56%) as white foam: $[\alpha]_D$ +13 (*c* 1, $CHCl_3$), R_f 0.22 (2:3 *n*-C_6H_{14}–acetone), ^1H NMR ($CDCl_3$): δ 1.21 (d, 3H, $J_{5,6}$ 6.5, H-6^{III}), 1.85–1.94 (m, 2H, CCH_2C), 1.98–2.18 (12s, 36 H, 10 OC(O)CH_3, 2 NHC(O)CH_3), 2.91–2.98 (m, 1H, H-2^{I}), 3.48–3.54 (m, 2H, NCH_2), 3.68–3.78 (m, 4H, H-5^{I}, H-3^{II}, H-5^{II}, OC*H*H), 3.81 (dd, 1H, $J_{1,2}$ 7.5, $J_{2,3}$ 9.6, H-2^{II}), 3.90–3.95 (m, 1H, OCH*H*), 3.98–4.04 (m, 1H, H-6a^{IV}), 4.05–4.10 (m, 3H, H-6a^{I}, H-6b^{I}, H-6b^{IV}), 4.11–4.15 (m, 1H, H-5^{IV}), 4.18 (dd, 1H, $J_{5,6a}$ 2.4, $J_{6a,6b}$ 12.3, H-6a^{II}), 4.24 (dd, 1H, $J_{5,6b}$ 4.9, $J_{6a,6b}$ 12.3, H-6b^{II}), 4.37 (d, 1H, $J_{1,2}$ 7.5, H-1^{II}), 4.48 (ddd, 1H, $J_{1,2}$ 3.5, $J_{2,3}$ 11.5, $J_{2,NH}$ 9.1, H-2^{IV}), 4.68–4.75 (m, 2H, H-3^{I}, H-5^{III}), 4.84 (dd, 1H, $J_{3,4}$ 9.4, $J_{4,5}$ 9.6, H-4^{I}), 4.98 (dd, 1H, $J_{2,3}$ 11.5, $J_{3,4}$ 3.1, H-3^{IV}), 5.02 (d, 1H, $J_{2,1}$ 8.0, H-1^{I}), 5.23 (d, 1H, $J_{1,2}$ 3.5, H-1^{IV}), 5.27 (br s, 1H, H-4^{III}), 5.31–5.35 (m, 2H, H-2^{III}, H-3^{III}), 5.36 (d, 1H, $J_{3,4}$, H-4^{II}), 5.41 (br s, 1H, H-4^{IV}), 5.63 (d, 1H, $J_{2,1}$ 2.6, H-1^{III}), 6.30–6.45 (m, 1H, NHAc^{IV}), 6.92 (d, 1H, $J_{2,NH}$ 6.1, NHAc^{I}), 7.14–7.21 (m, 1H, NHC(O)CF$_3$); ^{13}C NMR ($CDCl_3$): δ 15.4 (C-6^{III}), 20.6–20.8 (10 OC(O)CH$_3$), 23.0 (NC(O)CH$_3$), 23.1 (NC(O)CH$_3$), 28.3 (CCH$_2$C), 37.4 (CCH$_2$N), 48.1 (C-2^{IV}), 58.8 (C-2^{I}), 60.6 (C-6^{I}), 62.2 (C-6^{II}), 62.8 (H-6^{IV}), 65.5 (H-5^{III}), 66.9 (C-4^{II}), 67.2 (C-3^{IV}), 67.29 (C-2^{III}), 67.35 (C-4^{IV}), 67.7 (CCH$_2$O), 68.1 (C-3^{III}), 68.4 (C-5^{IV}), 68.9 (C-4^{I}), 70.5 (C-5^{II}), 71.4 (C-4^{III}), 72.0 (C-5^{I}), 72.6 (C-2^{II}), 75.3 (C-3^{I}), 79.6 (C-3^{II}), 95.7 (C-1^{III}), 98.3 (C-1^{IV}), 99.3 (C-1^{I}), 101.5 (C-1^{II}), 116.0 (C(O)CF$_3$), 157.3 (C(O)CF$_3$), 169.3–172.3 (12C(O)CH$_3$); HRESIMS: Calcd [$C_{53}H_{74}N_3O_{31}F_3$]H$^+$: *m/z* 1306.4331. Found: *m/z* 1306.4353.

4.7. 3-Trifluoroacetamidopropyl 2-acetamido-3,4,6-tri-*O*-acetyl-2-deoxy-α-D-galactopyranosyl-(1→3)-[2,3,4-tri-*O*-acetyl-α-L-fucopyranosyl-(1→2)]-4,6-di-*O*-acetyl-β-D-galactopyranosyl-(1→3)-4,6-di-*O*-acetyl-2-azido-2-deoxy-α-D-galactopyranoside (20) and 3-trifluoroacetamidopropyl 2-acetamido-3,4,6-tri-*O*-acetyl-2-deoxy-α-D-galactopyranosyl-(1→3)-[2,3,4-tri-*O*-acetyl-α-L-fucopyranosyl-(1→2)]-4,6-di-*O*-acetyl-α-D-galactopyranosyl-(1→3)-4,6-di-*O*-acetyl-2-azido-2-deoxy-α-D-galactopyranoside (22)

A solution of trichloroacetimidate **8** (411 mg, 0.407 mmol) and glycosyl acceptor **12** (366 mg, 0.821 mmol) in anhyd CH_3CN (18 mL) was stirred with 3 Å molecular sieves (700 mg) for 30 min at rt under an atmosphere of N_2. A solution of TMSOTf (7 μL, 0.04 mmol) in anhyd CH_3CN (100 μL) was added, the mixture was stirred for 3 h, and it was then neutralized by Et_3N (10 μL), filtered, and concentrated. Gel filtration followed by column chromatography on silica gel (6:3:1→4:3:1 *n*-C_6H_{14}–$CHCl_3$–2-PrOH) yielded a mixture of anomers **14** (416 mg, 79%) with a β/α ratio of 2.5:1.

A mixture of anomeric glycosides **14** (416 mg, 0.322) was dissolved in 20 mL of AcOH (80%), kept for 1 h at 80 °C, concentrated, and co-evaporated with $PhCH_3$ (2 × 15 mL). Column chromatography on silica gel (3:3:1 *n*-C_6H_{14}–$CHCl_3$–2-PrOH) resulted in a fraction (212 mg) containing tetrasaccharide **16**, R_f 0.12 (3:3:1 *n*-C_6H_{14}–$CHCl_3$–2-PrOH), and pure tetrasaccharide **18** (91 mg, 18%) as white foam, $[\alpha]_D$ +90 (*c* 1, $CHCl_3$), R_f 0.25 (3:3:1 *n*-C_6H_{14}–$CHCl_3$–2-PrOH).

The 'mix fraction' was acetylated (Ac_2O/Py, 12 h) and the solution was evaporated. Column chromatography on silica gel (6:3:1 *n*-C_6H_{14}–$CHCl_3$–2-PrOH) afforded tetrasaccharide **20** (220 mg, 42%) as white foam, $[\alpha]_D$ +64 (*c* 1, $CHCl_3$), R_f 0.50 (3:3:1 *n*-C_6H_{14}–$CHCl_3$–2-PrOH). ^1H NMR ($CDCl_3$): δ 1.11 (d, 3H, $J_{5,6}$ 6.6, H-6^{III}), 1.94–2.00 (m, 14 H, CCH_2C, 4 OC(O)CH_3), 2.06–2.20 (7s, 21 H, 6 OC(O)CH_3, NHC(O)CH_3), 3.52 (dd, 1H, $J_{1,2}$ 3.6, $J_{2,3}$ 10.9, H-2^{I}), 3.53–3.58 (m, 2H, NCH_2), 3.59–3.64 (m, 1H, OC*H*H), 3.78 (dd, 1H, $J_{1,2}$ 7.4, $J_{2,3}$ 9.3, H-2^{II}), 3.82–3.86 (m, 2H, H-5^{II}, OCH*H*), 3.87 (dd, 1H, $J_{2,3}$ 9.3, $J_{3,4}$ 3.4, H-3^{II}), 4.00–4.19 (m, 7H, H-5^{I}, H-6a^{I}, H-6b^{I}, H-6a^{II}, H-6b^{II}, H-6a^{IV}, H-6b^{IV}), 4.19–4.23 (m, 2H, H-3^{I}, H-5^{IV}), 4.47 (ddd, 1H, $J_{1,2}$ 3.4, $J_{2,3}$ 11.2, $J_{2,NH}$ 8.2, H-2^{IV}), 4.67 (q, 1H, $J_{5,6}$ 6.6, H-5^{III}), 4.69 (d, 1H, $J_{1,2}$ 7.4, H-1^{II}), 5.01 (dd, 1H, $J_{2,3}$ 11.2, $J_{3,4}$ 3.1, H-3^{IV}), 5.06 (d, 1H, $J_{1,2}$ 3.6, H-1^{I}), 5.26 (d, 1H, $J_{1,2}$ 3.4, H-1^{IV}), 5.28–5.35 (m, 3H, H-2^{III}, H-3^{III}, H-4^{III}), 5.38 (d, 1H, $J_{3,4}$ 3.4, H-4^{II}), 5.47 (br s, 1H, H-4^{IV}), 5.52–5.55 (m, 2H, H-4^{I}, H-1^{III}), 6.31 (d, 1H, $J_{2,NH}$ 8.2, NHAc^{IV}), 6.83–6.90 (m, 1H, NHC(O)CF$_3$); ^{13}C NMR ($CDCl_3$): δ 15.8 (C-6^{III}), 20.5–20.8 (10 OC(O)CH$_3$), 23.0 (NC(O)CH$_3$), 28.7 (CCH$_2$C), 37.7 (CCH$_2$N), 48.2 (C-2^{IV}), 58.6 (C-2^{I}), 61.0 (C-6^{I}), 62.4 (C-6^{II}), 62.8 (H-6^{IV}), 64.9 (H-5^{III}), 66.5 (CCH$_2$O), 66.8 (C-4^{II}), 67.2 (C-2^{III}), 67.4 (C-3^{IV}), 67.5 (C-4^{IV}), 67.6 (C-5^{IV}), 67.8 (C-3^{III}), 68.8 (C-5^{I}), 70.0 (C-4^{I}), 70.6 (C-5^{II}), 71.6 (C-4^{III}), 73.0 (C-3^{I}), 74.1 (C-2^{II}), 78.9 (C-3^{II}), 96.2 (C-1^{III}), 98.1 (C-1^{IV}), 98.8 (C-1^{I}), 102.5 (C-1^{II}), 115.9 (C(O)CF$_3$), 157.2 (C(O)CF$_3$), 169.3–171.2 (11C(O)CH$_3$); HRESIMS: Calcd [$C_{50}H_{70}N_5O_{30}F_3$]H$^+$: *m/z* 1290.4130. Found: *m/z* 1290.4047.

Tetrasaccharide **18** was acetylated (Ac_2O/Py, 12 h), and the solution was evaporated. Column chromatography on silica gel (*n*-C_6H_{14}–$CHCl_3$–2-PrOH, 6:3:1) gave tetrasaccharide **22** (90 mg, 92%) as white foam: $[\alpha]_D$ +68 (*c* 1, $CHCl_3$), R_f 0.53 (3:3:1 *n*-C_6H_{14}–$CHCl_3$–2-PrOH), ^1H NMR ($CDCl_3$): δ 1.17 (d, 3H, $J_{5,6}$ 6.5, H-6^{III}), 1.90–1.96 (m, 5H, CCH_2C, OC(O)CH_3), 1.97–2.21 (10s, 30 H, 9 OC(O)CH_3, NHC(O)CH_3), 3.33–3.41 (m, 1H, NC*H*H), 3.52–3.59 (m, 1H, OC*H*H), 3.63–3.70 (m, 1H, NCH*H*), 3.76–3.84 (m, 1H, H-2^{I}), 3.87–3.83 (m, 1H, OCH*H*), 3.97–4.06 (m, 4H, H-2^{II}, H-6a^{II}, H-6b^{II}, H-6a^{IV}), 4.10–4.18 (m, 5H, H-3^{I}, H-5^{I}, H-6a^{I}, H-6b^{I}, H-6b^{IV}), 4.18–4.22 (m, 1H, H-5^{IV}), 4.24 (dd, 1H, $J_{2,3}$ 8.5, $J_{3,4}$ 3.3, H-3^{II}), 4.25–4.29 (m, 2H, H-5^{II}, H-5^{III}), 4.51 (ddd, 1H, $J_{1,2}$ 3.5, $J_{2,3}$ 11.8, $J_{2,NH}$ 8.6, H-

2^{IV}), 5.00 (d, 1H, $J_{1,2}$ 3.6, H-1^{I}), 5.07–5.13 (m, 1H, H-3^{IV}), 5.16 (dd, 1H, $J_{2,3}$ 10.8, $J_{3,4}$ 3.6, H-3^{III}), 5.25 (d, 1H, $J_{1,2}$ 3.5, H-1^{IV}), 5.27 (d, 1H, $J_{3,4}$ 3.6, H-4^{III}), 5.28 (d, 1H, $J_{1,2}$ 4.1, H-1^{II}), 5.32 (dd, 1H, $J_{1,2}$ 2.4, $J_{2,3}$ 10.8, H-2^{III}), 5.37–5.40 (m, 1H, H-1^{III}), 5.42 (d, 1H, $J_{3,4}$ 2.7, H-4^{IV}), 5.47 (dd, 1H, $J_{3,4}$ 3.3, $J_{4,5}$ 3.1, H-4^{II}), 5.69 (d, 1H, $J_{3,4}$ 2.65, H-4^{I}), 6.26 (d, 1H, $J_{2,NH}$ 8.6, N*H*Ac^{IV}), 7.38–7.42 (m, 1H, N*H*C(O)CF_3); ^{13}C NMR ($CDCl_3$): δ 15.9 (C-6^{III}), 20.5–20.8 (10 OC(O)*C*H_3), 23.0 (NC(O)CH_3), 28.3 (*C*CH_2C), 38.1 (*C*CH_2N), 48.3 (C-2^{IV}), 59.8 (C-2^{I}), 60.7 (C-6^{II}), 62.2 (C-6^{I}), 62.5 (H-6^{IV}), 65.0 (C-5^{III}), 66.8 (C-3^{III}), 66.9 (C-4^{I}), 67.3 (*C*CH_2O), 67.4 (C-4^{IV}), 67.47 (C-3^{IV}), 67.53 (C-3^{I}), 67.6 (C-4^{II}), 68.21 (C-2^{III}), 68.23 (C-5^{IV}), 68.6 (C-5^{II}), 70.9 (C-4^{III}), 72.37 (C-5^{I}), 72.39 (C-2^{II}), 74.2 (C-3^{II}), 94.8 (C-1^{II}), 96.3 (C-1^{III}), 97.9 (C-1^{I}), 98.3 (C-1^{IV}), 116.0 (C(O)*C*F_3), 157.3 (*C*(O)CF_3), 169.8–170.7 (11*C*(O)CH_3); HRESIMS: Calcd $[C_{50}H_{70}N_5O_{30}F_3]H^+$: *m/z* 1290.4130. Found: *m/z* 1290.4024.

4.8. 3-Trifluoroacetamidopropyl 2-acetamido-3,4,6-tri-*O*-acetyl-2-deoxy-α-D-galactopyranosyl-(1→3)-[2,3,4-tri-*O*-acetyl-α-L-fucopyranosyl-(1→2)]-4,6-di-*O*-acetyl-β-D-galactopyranosyl-(1→3)-4,6-di-*O*-acetyl-2-azido-2-deoxy-β-D-galactopyranoside (21) and 3-trifluoroacetamidopropyl 2-acetamido-3,4,6-tri-*O*-acetyl-2-deoxy-α-D-galactopyranosyl-(1→3)-[2,3,4-tri-*O*-acetyl-α-L-fucopyranosyl-(1→2)]-4,6-di-*O*-acetyl-α-D-galactopyranosyl-(1→3)-4,6-di-*O*-acetyl-2-azido-2-deoxy-β-D-galactopyranoside (23)

A solution of trichloroacetimidate **8** (400 mg, 0.396 mmol) and glycosyl acceptor **13** (353 mg, 0.791 mmol) in anhyd CH_3CN (18 mL) was stirred with 3 Å molecular sieves (700 mg) for 30 min at rt under an atmosphere of N_2. A solution of TMSOTf (7 μL, 0.04 mmol) in anhyd CH_3CN (100 μL) was then added, and the reaction mixture was stirred for 3 h. The mixture was neutralized with Et_3N (10 μL), filtered, and concentrated. Column chromatography on silica gel (1:1→2:3 *n*-C_6H_{14}–acetone) afforded the mixture of anomeric tetrasaccharides **15** (413 mg, 81%) with a β/α ratio of 2:1.

Pure tetrasaccharide **15-β** was also isolated in analytical amounts as white foam: $[\alpha]_D$ −18 (*c* 1, $CHCl_3$), R_f 0.25 (4:3:1 *n*-C_6H_{14}–$CHCl_3$–2-PrOH), ^{1}H NMR ($CDCl_3$): δ 0.70 (d, 3H, $J_{5,6}$ 6.4, H-6^{III}), 1.95–2.06 (m, 17 H, CC*H*$_2$C, 5 OC(O)C*H*$_3$), 2.11–2.18 (4s, 12H, 3 OC(O)C*H*$_3$, NHC(O)C*H*$_3$), 3.44 (br s, 1H, H-6a^{I}), 3.51–3.58 (m, 1H, NC*H*H), 3.60 (dd, 1H, $J_{2,3}$ 10.7, $J_{3,4}$ 3.2, H-3^{I}), 3.61–3.65 (m, 1H, NCH*H*), 3.74–3.78 (m, 1H, OC*H*H), 3.80 (dd, 1H, $J_{5,6a}$ 6.6, $J_{5,6b}$ 6.6, H-5^{II}), 3.87 (dd, 1H, $J_{2,3}$ 9.4, $J_{3,4}$ 3.3, H-3^{II}), 3.92 (dd, 1H, $J_{1,2}$ 7.4, $J_{2,3}$ 9.4, H-2^{II}), 3.97–4.00 (m, 1H, H-6a^{IV}), 4.02 (dd, 1H, $J_{1,2}$ 7.9, $J_{2,3}$ 10.7, H-2^{I}), 4.06–4.14 (m, 5H, H-5^{I}, H-6a^{II}, H-5^{IV}, H-6b^{IV}, OCH*H*), 4.19 (dd, 1H, $J_{5,6b}$ 6.6, $J_{6,6b}$ 11.2, H-6b^{II}), 4.24 (dd, 1H, $J_{3,4}$ 3.2, H-4^{I}), 4.30 (d, 1H, $J_{5,6a}$ 11.8, H-6b^{I}) 4.45 (d, 1H, $J_{1,2}$ 7.9, H-1^{I}), 4.47 (ddd, 1H, $J_{1,2}$ 3.4, $J_{2,3}$ 11.4, $J_{2,NH}$ 8.4, H-2^{IV}), 4.53 (q, 1H, $J_{5,6}$ 6.4, H-5^{III}), 4.87 (d, 1H, $J_{1,2}$ 7.4, H-1^{II}), 4.98 (dd, 1H, $J_{2,3}$ 11.4, $J_{3,4}$ 3.0, H-3^{IV}), 5.19 (d, 1H, $J_{3,4}$ 3.1, H-4^{III}), 5.25 (d, 1H, $J_{1,2}$ 3.4, H-1^{IV}), 5.28 (dd, 1H, $J_{1,2}$ 3.4, $J_{2,3}$ 11.0, H-2^{III}), 5.31 (dd, $J_{2,3}$ 11.0, $J_{3,4}$ 3.1, H-3^{III}), 5.38 (d, 1H, $J_{3,4}$ 3.0 H-4^{IV}), 5.40 (d, 1H, $J_{3,4}$ 3.3, H-4^{II}), 5.56 (s, 1H, PhC*H*), 5.59 (d, 1H, $J_{1,2}$ 3.4, H-1^{III}), 6.39 (d, 1H, $J_{2,NH}$ 8.4, N*H*Ac^{IV}), 7.12–7.16 (m, 1H, N*H*C(O)CF_3), 7.32–7.38 (m, 3H, Ar*H*), 7.47–7.50 (m, 2H, Ar*H*), ESIMS: Calcd $[C_{54}H_{70}N_5O_{28}F_3]Na^+$: *m/z* 1316.41. Found: *m/z* 1316.46.

A mixture of anomeric tetrasaccharides **15** (413 mg, 0.319 mmol) was dissolved in 20 mL of AcOH (80%), kept for 3 h at 70 °C, concentrated, and co-evaporated with $PhCH_3$ (4 × 15 mL). Column chromatography on silica gel (3:3:1 *n*-C_6H_{14}–$CHCl_3$–2-PrOH) gave a fraction containing tetrasaccharide **17** (230 mg), R_f 0.11 (3:3:1 *n*-C_6H_{14}–$CHCl_3$–2-PrOH), and a fraction containing tetrasaccharide **19** (111 mg), R_f 0.22 (3:3:1 *n*-C_6H_{14}–$CHCl_3$–2-PrOH). Both fractions also contained some impurities. Both fractions were separately acetylated (Ac_2O/Py, 15 h) and subjected to column chromatography on silica gel (4:3:1 *n*-C_6H_{14}–$CHCl_3$–2-PrOH, for tetrasaccharide **17**, and 5:3:1 *n*-C_6H_{14}–$CHCl_3$–2-PrOH, for tetrasaccharide **19** correspondingly) to afford tetrasaccharide **21** (232 mg, 45%), and tetrasaccharide **23** (114 mg, 22%).

Compound **21**: white foam, $[\alpha]_D$ +4 (*c* 1, $CHCl_3$), R_f 0.25 (*n*-C_6H_{14}–$CHCl_3$–*i*-PrOH, 4:3:1), ^{1}H NMR ($CDCl_3$): δ 1.17 (d, 3H, $J_{5,6}$ 6.5, H-6^{III}), 1.94–2.00 (m, 11 H, CC*H*$_2$C, 3 OC(O)C*H*$_3$), 2.05–2.19 (8s, 24 H, 7 OC(O)C*H*$_3$, NHC(O)C*H*$_3$), 3.50–3.54 (m, 1H, NC*H*H), 3.55–3.60 (m, 1H, NCH*H*), 3.64–3.70 (m, 2H, H-2^{I}, H-3^{I}), 3.75 (dd, 1H, $J_{1,2}$ 7.3, $J_{2,3}$ 9.1, H-2^{II}), 3.76–3.81 (m, 2H, H-3^{II}, H-5^{II}), 3.83–3.87 (m, 2H, H-5^{I}, OC*H*H), 3.97–4.01 (m, 1H, OCH*H*), 4.05–4.19 (m, 6H, H-6a^{I}, H-6b^{I}, H-6a^{II}, H-6b^{II}, H-6a^{IV}, H6b^{IV}), 4.23 (dd, 1H, $J_{5,6a}$ 5.8, $J_{5,6b}$ 5.8, H-5^{IV}), 4.42 (d, 1H, $J_{1,2}$ 7.2, H-1^{I}), 4.49 (ddd, 1H, $J_{1,2}$ 3.3, $J_{2,3}$ 11.3, $J_{2,NH}$ 7.2, H-2^{IV}), 4.56 (q, 1H, $J_{5,6}$ 6.5, H-5^{III}), 4.74 (d, 1H, $J_{1,2}$ 7.3, H-1^{II}), 5.03 (dd, 1H, $J_{2,3}$ 11.3, $J_{3,4}$ 2.9, H-3^{IV}), 5.26 (d, 1H, $J_{1,2}$ 3.3, H-1^{IV}), 5.30 (dd, 1H, $J_{1,2}$ 3.4, $J_{2,3}$ 10.2, H-2^{III}), 5.31–5.34 (m, 2H, H-3^{III}, H-4^{III}), 5.37–5.39 (m, 2H, H-4^{I}, H-4^{II}), 5.47 (d, 1H, H-4^{IV}), 5.56 (d, 1H, $J_{1,2}$ 3.37, H-1^{III}), 6.21 (d, 1H, $J_{2,NH}$ 7.2, N*H*Ac^{IV}), 6.97–7.02 (m, 1H, N*H*C(O)CF_3); ^{13}C NMR ($CDCl_3$): δ 16.1 (C-6^{III}), 20.5–20.7 (10OC(O)*C*H_3), 23.0 (NC(O)*C*H_3), 28.5 (*C*CH_2C), 38.0 (*C*CH_2N), 48.1 (C-2^{IV}), 61.1 (C-6^{I}), 62.1 (C-6^{II}), 62.3 (C-2^{I}), 62.6 (C-6^{IV}), 65.3 (C-5^{III}), 66.3 (C-4^{II}), 67.4 (C-4^{IV}), 67.5 (C-3^{IV}), 67.67 (C-3^{III}), 67.72 (C-2^{III}), 68.4 (C-5^{IV}), 68.7 (C-4^{I}), 68.8 (*C*CH_2O), 70.6 (C-3^{I}), 71.4 (C-4^{III}), 71.8 (C-5^{I}), 74.8 (C-2^{II}), 75.4 (C-3^{II}), 77.6 (C-5^{II}), 96.2 (C-1^{III}), 97.2 (C-1^{IV}), 102.1 (C-1^{I}), 102.8 (C-1^{II}), 115.9 (C(O)*C*F_3), 157.2 (*C*(O)CF_3), 169.5–170.9 (11*C*(O)CH_3); HRESIMS: Calcd $[C_{50}H_{70}N_5O_{30}F_3]H^+$: *m/z* 1290.4130. Found: *m/z* 1290.4047.

Compound **23**: white foam, $[\alpha]_D$ +39 (*c* 1, $CHCl_3$), R_f 0.36 (4:3:1 *n*-C_6H_{14}–$CHCl_3$–2-PrOH), ^{1}H NMR ($CDCl_3$–CD_3OD, 1:1): δ 1.37 (d, 3H, $J_{5,6}$ 6.5, H-6^{III}), 2.08–2.12 (m, 2H, CC*H*$_2$C), 2.13–2.39 (11s, 33 H, 10 OC(O)C*H*$_3$, NHC(O)C*H*$_3$), 3.57–3.68 (m, 2H, NC*H*$_2$), 3.81 (dd, 1H, $J_{1,2}$ 7.8, $J_{2,3}$ 10.6, H-2^{I}), 3.86–3.90 (m, 1H, OC*H*H), 3.92 (dd, 1H, $J_{2,3}$ 10.6, $J_{3,4}$ 3.0, H-3^{I}), 4.05 (dd, 1H, $J_{5,6a}$ 6.3, $J_{5,6b}$ 6.3, H-5^{I}), 4.15–4.19 (m, 1H, OCH*H*), 4.22 (dd, 1H, $J_{5,6a}$ 6.9, $J_{6,6b}$ 11.1, H-6a^{I}), 4.24–4.30 (m, 3H, H-2^{II}, H-6a^{I}, H-6a^{II}), 4.30–4.36 (m, 3H, H-6b^{I}, H-6b^{II}, H6b^{IV}), 4.36–4.39 (m, 1H, H-5^{IV}), 4.40 (dd, $J_{2,3}$ 9.3, $J_{3,4}$ 2.9, H-3^{II}), 4.44–4.49 (m, 1H, H-5^{III}), 4.57 (d, 1H, $J_{1,2}$ 7.8, H-1^{I}), 4.58–4.60 (m, 1H, H-5^{II}), 4.62 (dd, 1H, $J_{1,2}$ 3.4, $J_{2,3}$ 11.6, H-2^{IV}), 5.27 (d, 1H, $J_{2,3}$ 11.6, H-3^{IV}), 5.41–5.44 (m, 2H, H-1^{IV}, H-2^{III}), 5.45 (d, 1H, $J_{1,2}$ 3.4, H-1^{II}), 5.47 (dd, 1H, $J_{2,3}$ 10.8, $J_{3,4}$ 3.3, H-3^{III}), 5.50 (d, 1H, $J_{3,4}$ 3.3, H-4^{III}), 5.61 (d, 1H, $J_{3,4}$ 2.5, H-4^{IV}), 5.62–5.64 (m, 1H, H-1^{III}), 5.66–5.70 (m, 2H, H-4^{I}, H-4^{II}); ^{13}C NMR ($CDCl_3$): δ 16.0 (C-6^{III}), 20.5–20.8 (10 OC(O)*C*H_3), 23.0 (NC(O)*C*H_3), 28.4 (*C*CH_2C), 38.2 (*C*CH_2N), 48.4 (C-2^{IV}), 61.2 (C-6^{I}), 61.8 (C-6^{II}), 62.4 (C-6^{IV}), 62.7 (C-2^{I}), 65.1 (C-5^{III}), 65.4 (C-4^{II}), 67.1 (C-2^{III}), 67.4 (C-4^{IV}), 67.5 (C-3^{IV}), 67.6 (C-4^{I}), 68.0 (C-3^{III}), 68.1 (C-5^{IV}), 68.9 (C-5^{II}), 69.1 (*C*CH_2O), 71.0 (C-4^{III}), 71.3 (C-3^{I}), 72.4 (C-5^{I}), 73.6 (C-3^{II}), 74.4 (C-2^{II}), 94.5 (C-1^{II}), 96.4 (C-1^{III}), 97.6 (C-1^{IV}), 102.5 (C-1^{I}), 116.0 (C(O)*C*F_3), 157.1 (*C*(O)CF_3), 169.8–170.7 (11*C*(O)CH_3); HRESIMS: Calcd $[C_{50}H_{70}N_5O_{30}F_3]H^+$: *m/z* 1290.4130. Found: *m/z* 1290.4046.

4.9. 3-Aminopropyl 2-acetamido-2-deoxy-α-D-galactopyranosyl-(1→3)-[α-L-fucopyranosyl-(1→2)]-α-D-galactopyranosyl-(1→3)-2-acetamido-2-deoxy-α-D-galactopyranoside (28)

Tetrasaccharide **22** (22 mg, 0.017 mmol) was deacetylated under Zemplén conditions, and product **26** that was obtained was purified by gel filtration (1:1 CH_3CN–H_2O). A mixture of tetrasaccharide **26** (15 mg, 0.017 mmol), DTT (8 mg. 0.05 mmol), and aq $NaHCO_3$ (1.5 mL, 50 mM, pH 8.2) was stirred for 1.5 h under an atmosphere of Ar, Ac_2O (50 μL) was then added and the reaction was stirred for 45 min. The mixture was subjected to gel filtration (1:1 CH_3CN–H_2O) followed by column chromatography (elution with 6:5:1 CH_2Cl_2–EtOH–H_2O) and concentrated. The residue was

dissolved in H_2O (1 mL), aq NaOH (2 M, 2 μL) was added, and the reaction mixture was kept for 3 h. Cation-exchange chromatography on Dowex 50X4-400 (H^+) (elution with 5% aq ammonia) gave 10 mg (75%) of product **28** as white foam: $[\alpha]_D$ +129 (*c* 0.9, H_2O–CH_3CN, 1:1), R_f 0.19 (100:10:10:10:2 EtOH–BuOH–Py–H_2O–AcOH), ^1H NMR (D_2O): (characteristic signals) δ 1.27 (d, 3H, $J_{5,6}$ 6.6, H-6^{III}), 1.98–2.05 (m, 2H, CCH_2C), 2.05–2.08 (2s, 6H, 2 C(O)CH_3), 3.11–3.16 (m, 2H, NHCH_2), 4.93 (d, 1H, $J_{1,2}$ 3.8, H-1^I), 5.18 (d, 1H, $J_{1,2}$ 3.7, H-1^{IV}), 5.19 (d, 1H, $J_{1,2}$ 3.8, H-1^{II}), 5.28 (d, 1H, $J_{1,2}$ 4.0, H-1^{III}); ^{13}C NMR (D_2O): δ 15.4 (C-6^{III}), 22.0 (*C*H_3C(O)), 22.1 (*C*H_3C(O)), 26.8 (CCH_2C), 37.1 (CCH_2N), 48.0, 49.6 (C-2^I, C-2^{IV}), 61.11, 61.14, 61.2 (C-6^I, C-6^{II}, C-6^{IV}), 65.0 (CCH_2O), 63.8, 65.2, 67.4, 67.8, 67.9, 68.5, 69.4, 69.6, 70.7, 70.9, 71.3, 71.75, 71.76, 72.8 (C-2^{II}, C-2^{III}, C-3^I, C-3^{II}, C-3^{III}, C-3^{IV}, C-4^I, C-4^{II}, C-4^{III}, C-4^{IV}, C-5^I, C-5^{II}, C-5^{III}, C-5^{IV}), 91.3 (C-1^{IV}), 94.6 (C-1^{II}), 97.2 (C-1^I), 99.2 (C-1^{III}), 174.2 (CH_3*C*(O)), 174.8 (CH_3*C*(O)), HRESIMS, Calcd $[C_{31}H_{55}N_3O_{20}]H^+$: *m/z* 790.3452. Found: *m/z* 790.3439.

4.10. 3-Aminopropyl 2-acetamido-2-deoxy-α-D-galactopyranosyl-(1→3)-[α-L-fucopyranosyl-(1→2)]-α-D-galactopyranosyl-(1→3)-2-acetamido-2-deoxy-β-D-galactopyranoside (29)

Tetrasaccharide **23** (59 mg, 0.046 mmol) was deacetylated under Zemplén conditions. The product obtained was purified by gel filtration (1:1 CH_3CN–H_2O) and subjected to hydrogenolysis (7 h) in a mixture of MeOH (4 mL) and Ac_2O (200 μL). The reaction mixture was then filtered and concentrated. The residue was dissolved in H_2O (3 mL), an aq solution of NaOH (2 M, 3 μL) was added, and the reaction mixture was kept for 4 h. Cation-exchange chromatography on Dowex 50X4-400 (H^+) (elution with 5% aq ammonia) gave 9 mg (25%) of product **29** as white foam: $[\alpha]_D$ +91 (*c* 0.9, H_2O–CH_3CN, 1:1), R_f 0.18 (100:10:10:10:2 EtOH–BuOH–Py–H_2O–AcOH), ^1H NMR (D_2O): (characteristic signals) δ 1.27 (d, 3H, $J_{5,6}$ 6.6, H-6^{III}), 1.94–2.01 (m, 2H, CCH_2C), 2.05–2.07 (2s, 6H, 2 C(O)CH_3), 3.10–3.13 (m, 2H, NHCH_2), 4.53 (d, 1H, $J_{1,2}$ 8.5, H-1^I), 5.18 (m, 2H, H-1^{II}, H-1^{IV}), 5.28 (d, 1H, $J_{1,2}$ 4.0, H-1^{III}); ^{13}C NMR (D_2O): δ 15.4 (C-6^{III}), 22.0 (*C*H_3C(O)), 22.3 (*C*H_3C(O)), 26.7 (CCH_2C), 37.7 (CCH_2N), 49.6, 50.7 (C-2^I, C-2^{IV}), 60.9, 61.0, 61.2 (C-6^I, C-6^{II}, C-6^{IV}), 63.7 (CCH_2O), 64.5, 67.4, 67.87, 67.90, 68.0, 68.5, 69.3, 69.6, 70.9, 71.2, 71.7, 71.8, 74.7, 75.5 (C-2^{II}, C-2^{III}, C-3^I, C-3^{II}, C-3^{III}, C-3^{IV}, C-4^I, C-4^{II}, C-4^{III}, C-4^{IV}, C-5^I, C-5^{II}, C-5^{III}, C-5^{IV}), 91.3 (C-1^{IV}), 94.9 (C-1^{II}), 99.2 (C-1^{III}), 101.6 (C-1^I) 174.6 (CH_3*C*(O)), 174.9 (CH_3*C*(O)); HRESIMS: Calcd $[C_{31}H_{55}N_3O_{20}]H^+$: *m/z* 790.3452. Found: *m/z* 790.3459.

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Supplementary data

Supplementary data associated with this article can be found, in the online version, at doi:10.1016/j.carres.2011.12.013.

References

1. Okamura, Y. *Histochemistry* **1990**, *94*, 489–496.
2. Eastlund, T. *Transfusion* **1998**, *38*, 975–988.
3. Milland, J.; Sandrin, M. S. *Tissue Antigens* **2006**, *68*, 459–466.
4. Mollicone, R.; Gibaud, A.; François, A.; Ratcliffe, M.; Oriol, R. *Eur. J. Biochem.* **1990**, *191*, 169–176.
5. Meloncelli, P. J.; Lowary, T. L. *Carbohydr. Res.* **2010**, *345*, 2305–2322.
6. Svensson, L.; Rydberg, L.; de Mattos, L. C.; Henry, S. M. *Vox Sang.* **2009**, *96*, 56–61.
7. Economidou, J.; Hughes-Jones, N. C.; Gardner, B. *Vox Sang.* **1967**, *12*, 321–328.
8. Makela, O.; Ruoslahti, E.; Ehnholm, C. *J. Immunol.* **1969**, *102*, 763–771.
9. Gao, S.; Bennet, E. P.; Reibel, J.; Chen, X.; Christensen, M. E.; Krogdahl, A.; Dabelsteen, E. *APMIS* **2004**, *112*, 11–20.
10. Paré, G.; Chasman, D. I.; Kellog, M.; Zee, R. Y. L.; Rifai, N.; Badola, S.; Miletich, J. P.; Ridker, P. M. *PLoS Genet.* **2008**, *4*, 1–8.
11. Nosaka, M.; Ishida, Y.; Tanaka, A.; Hayashi, T.; Miyashita, T.; Kaminaka, C.; Eisenmenger, W.; Furukawa, F.; Kimura, A. *J. Histochem. Cytochem.* **2008**, *56*, 223–231.
12. Milat, M.-L.; Sinaÿ, P. *Carbohydr. Res.* **1981**, *92*, 183–189.
13. Bovin, N. V.; Zurabyan, S. E.; Khorlin, A. Y. *Russ. Chem. Bull.* **1982**, *31*, 1023–1030.
14. Zimmermann, P.; Greilich, U.; Schmidt, R. R. *Tetrahedron Lett.* **1990**, *13*, 1849–1852.
15. Udodong, U. E.; Rao, C. S.; Fraser-Reid, B. *Tetrahedron* **1992**, *48*, 4713–4724.
16. Macindoe, W. M.; Ijima, H.; Nakahara, Y.; Ogawa, T. *Carbohydr. Res.* **1995**, *269*, 227–257.
17. Pazynina, G. V.; Tyrtysh, T. V.; Bovin, N. V. *Mendeleev Commun.* **2002**, 143–145.
18. Meloncelli, P. J.; Lowary, T. L. *Carbohydr. Res.* **2011**, *346*, 1406–1426.
19. Macindoe, W. M.; Ijima, H.; Nakahara, Y.; Ogawa, T. *Tetrahedron Lett.* **1994**, *35*, 1735–1738.
20. Blixt, O.; Razi, N. *Methods Enzymol.* **2006**, *415*, 137–153.
21. Yi, W.; Shao, J.; Zhu, L.; Li, M.; Singh, M.; Lu, Y.; Lin, S.; Li, H.; Ryu, K.; Shen, J.; Guo, H.; Yao, O.; Bush, C. A.; Wang, P. G. *J. Am. Chem. Soc.* **2005**, *127*, 2040–2041.
22. Compston, C. A.; Condon, C.; Hanna, C. R.; Mazid, M. A. *Carbohydr. Res.* **1993**, *239*, 167–176.
23. Korchagina, E. Y.; Ryzhov, I. M.; Byrgazov, K. A.; Popova, I. S.; Pokrovsky, S. N.; Bovin, N. V. *Mendeleev Commun.* **2009**, *19*, 152–154.
24. Korchagina, E. Y.; Bovin, N. V. *Bioorg. Khim.* **1992**, *18*, 283–298 (*Russ. J. Bioorg. Chem.* **1992**, *18*, 153–165).
25. Shipova, E. V.; Bovin, N. V. *Mendeleev Commun.* **2000**, 63–65.
26. Excoffier, G.; Cagnaire, D. *Carbohydr. Res.* **1975**, *39*, 368–373.
27. Bovin, N. V.; Zemlyanikina, T. V.; Chagiashvilli, C. N.; Khorlin, A. Y. *Khim. Prir. Soedin.* **1988**, *6*, 777–785 [*Chem. Nat. Compd. (Engl. Transl.)* **1988**, *6*, 659–665].
28. Ovchinnikova, T. V.; Ter-Grigoryan, A. G.; Pazynina, G. V.; Bovin, N. V. *Bioorg. Khim.* **1997**, *23*, 61–74 (*Russ. J. Bioorg. Chem.* **1997**, *23*, 55–68).
29. Schmidt, R. R.; Kinzy, W. *Adv. Carbohydr. Chem. Biochem.* **1994**, *50*, 21–121.
30. Meijohannes, E.; Meldal, M.; Jensen, T.; Werdelin, O.; Galli-Stampino, L.; Mouritsen, S.; Bock, K. *J. Chem. Soc., Perkin Trans. 1* **1997**, 871–884.
31. Komarova, B. S.; Tsvetkov, Y. E.; Pier, G. B.; Nifantiev, N. E. *J. Org. Chem.* **2008**, *73*, 8411–8421.
32. Handlon, A. L.; Oppenheimer, N. J. *Pharm. Res.* **1988**, *5*, 297–299.
33. Vokhmyanina, O. A.; Rapoport, E. M.; Ryzhov, I. M.; Korchagina, E. Yu.; Pazynina, G. V.; Severov, V. V.; Kaltner, H.; André, S.; Gabius, H.-J.; Bovin, N. V. *Biochemistry (Moscow)* **2011**, *76*, 1185–1192.
34. Obukhova, P.; Korchagina, E.; Henry, S.; Bovin, N. *Transfusion* **2011**. doi: 10.1111/j1537-2995.2011.03381.x.