Supporting Information

Development of a multifunctional aminoxy-based fluorescent linker for glycan immobilization and analysis

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Other *in solution* enzymatic modifications

Modifications of the type 2 LN disaccharide, such as fucosylation or α1,3-galactosylation was also possible by using *Ce* FUT-6 or commercial α1,3-galactosyltransferase (α1,3-GalT) (Figure S1, panel a). Thereby, a Lewis X type antigen and a Galili xenotransplantation epitope (the latter typical of many animals, but not Old world apes and humans) (Galili U. 2005) were easily prepared. In order to mimic O-Man glycosylation pathway, the linker displaying αMan*p* at the reducing end was also modified with a recombinant β1,2-*N*-acetylglucosaminyltransferase (POMGnT1) (Akasaka-Manya K. *et al.* 2011) and β1,4-GalT to produce a LacNAc structure as commonly also found on N-glycan antennae (Figure S1, panel b). Furthermore, starting with GalNAc-modified linker, the β-anomeric forms of T antigen and the STn antigen were respectively prepared using recombinant bacterial CgtB β1,3-galactosyltransferase (β1,3-GalT) (Persson K. *et al.* 2001) and α2,6-sialyltransferase (α2,6-SiaT; Pd2,6ST) (Yamamoto T. *et al.* 1998), respectively (Figure S1, panel c). A Gal-β1,3GalNAcβ motif, found on honeybee royal jelly glycoproteins (Kimura Y. *et al.* 2006), is a part of the *in vivo* lipooligosaccharide product of the CgtB galactosyltransferase (Persson K.*et al.* 2001) and is the terminal motif of the asialo-form of the human GM1 ganglioside (Svennerholm L. 1962); thus, our β-form of the T antigen is present in natural glycans, even if it does not correspond to the Gal-β1,3GalNAcα core of typical O-glycans. Due to the broad substrate specificity of Pd2,6ST, which sialylates both α- and β-linked GalNAc residues, the β-anomer of the STn antigen was made (Yu H. *et al.* 2006).

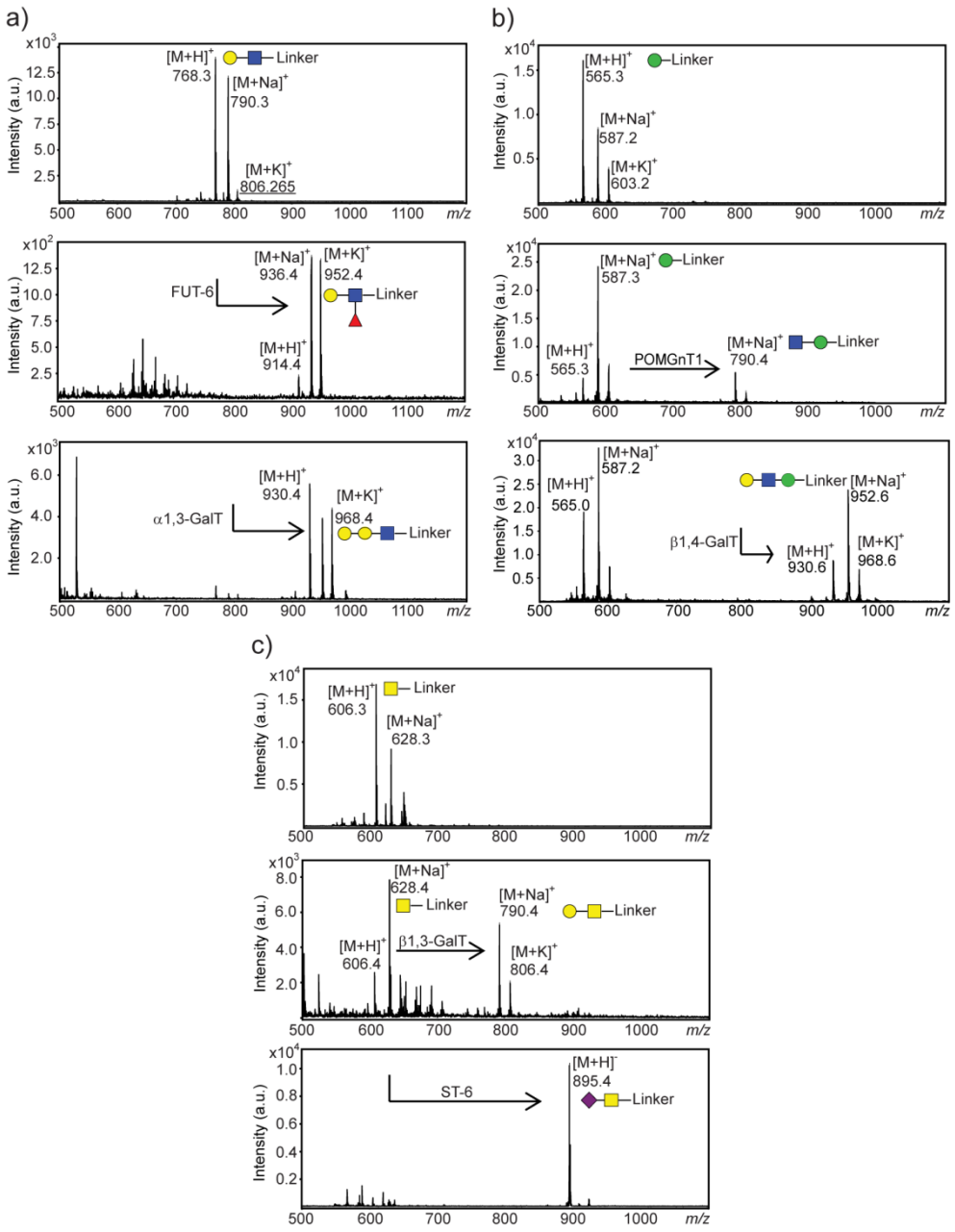


Fig. S1. ***In solution*** **enzymatic modification of chemically synthesized neoglycoconjugates.** a) Fucosylation or α1,3-galactosylation on the type 2 LN disaccharide using respectively *C. elegans* FUT-6 or α1,3-galactosyltransferase. b) Elongation of the Man-glycoconjugate with a GlcNAc and LacNAc unit using sequentially POMGnT1 and β1,4-galactosyltransferase. c) Extension of the GalNAc-modified glycoconjugate to create mimics of the T and STn antigens using respectively CgtB galactosyltransferase or Pd2,6ST α2,6-sialyltransferase. Aliquots of crude reaction products were analyzed by MALDI-TOF MS. In some cases, approximately 50% starting material was also observed in the MS spectra.

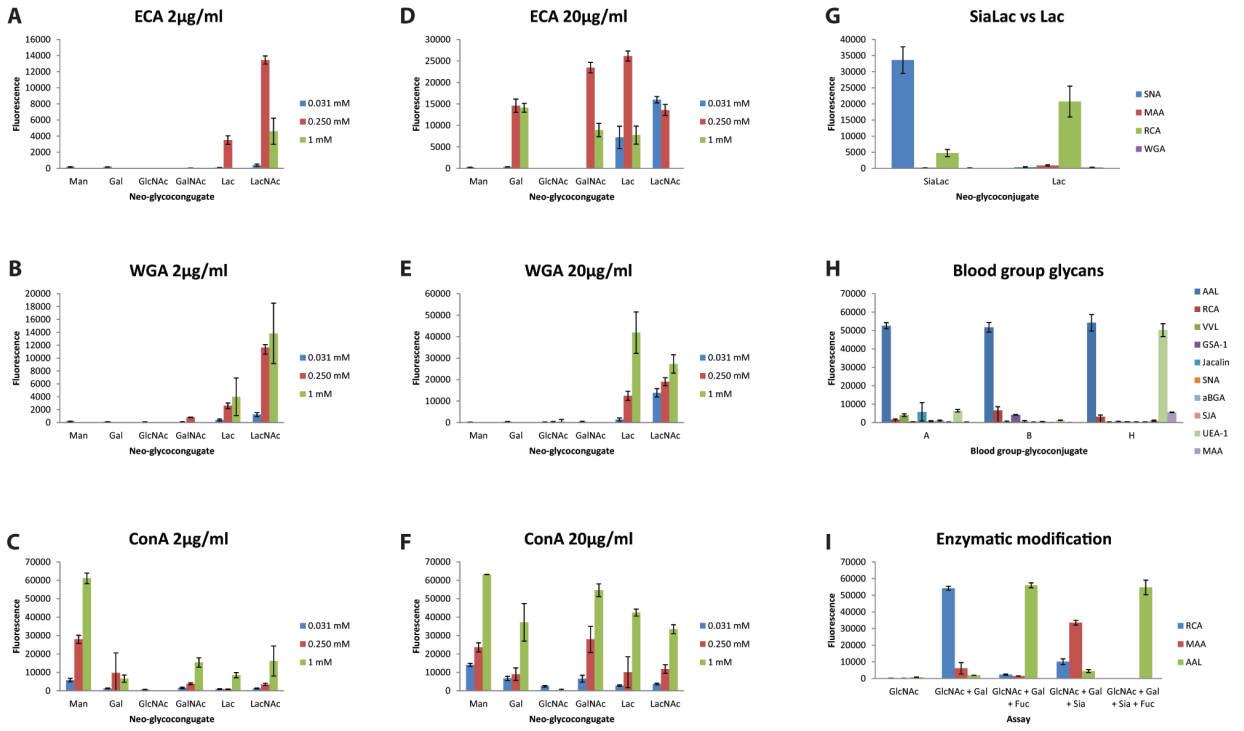


Fig. S2. Bar chart representations of lectin binding intensities. Fluorescence intensity data from Figure 3 (A-F; n=5) and Figure 4 (H-I; n=2) from the main text are presented as bar charts with error bars. AAL, *Aleuria* aurantia lectin; ConA, concanavalin A; ECA, *Erythrina cristagalli* lectin; GSA-I, *Grifonia* simplicifolia agglutinin I; MAA, *Maackia amurensis* agglutinin II; SNA, *Sambucus* nigra agglutinin; UEA, *Ulex europaeus* agglutinin I; VVA, *Vicia villosa* agglutinin; WGA, wheatgerm agglutinin. The blood group trisaccharides were also tested with jacalin, *Sophora japonica* agglutinin (SJA) and anti-blood group A (aBGA), but only insignificant binding was observed in these cases.

Materials

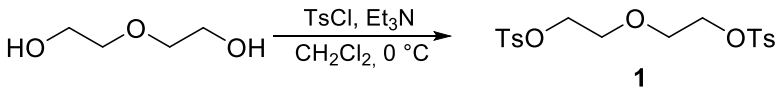
Chemicals were purchased from Sigma-Aldrich, Alfa Aesar, Polypeptide Laboratories AB, Fluorochem or TCI and used without further purification. Mono- and disaccharides were obtained from Carbosynth. Biotinylated lectins were purchased from Vector Laboratories and rabbit anti-horseradish peroxidase (anti-HRP) antibody was obtained from Sigma-Aldrich. Alexa Fluor® 647-conjugated streptavidin and Alexa Fluor® 647-conjugated goat anti-rabbit IgG were purchased from Invitrogen and abcam, respectively. Milli Q water was used for all aqueous preparations. HPLC solvents were purchased from Fisher Scientific. Glycosyltransferases (β1,4-GalT and α1,3-GalT) were purchased from Fluka and Sigma-Aldrich, respectively. Other glycosyltransferases (FUT-6, POMGnT1, ST-3, ST-6 and β1,3-GalT) were expressed and purified in the laboratory (see below for further details). Thin layer chromatography (TLC) was performed on pre-coated silica gel 60 F_{254} glass TLC plates and the substances were visualized by UV-irradiation (254 nm) or by dipping into anisaldehyde/H_2SO_4 reagent or ninhydrin reagent followed by heating to 250 °C.

Purification of compounds was performed by column chromatography using Merck silica gel (0.040-0.063 mm). For difficult separations, HPLC-column chromatography using pre-packed analytical column (YMC-Pack SIL-06, 0.005 mm, 250 x 10 mm) was employed. Integrity of neoglycoconjugates was evaluated either using a SeQuant ZIC-HILIC column (150 x 4.6 mm, 100 Å) on a Shimadzu LCMS-2020 equipped with an ESI-Q MS detector or a Tosoh Amide-80 column (Tosoh Bioscience, 4.6 x 250 mm, 100 Å) on a Shimadzu HPLC-30AD system equipped with a fluorescence detector (RF-20A XS). Neoglycoconjugates were quantified by integration of HPLC peaks or using Infinite® 200 PRO Tecan microplate reader. The fluorescence was measured at 280 nm (excitation) and 380 nm (emission), and the concentration was calculated by creating a standard curve using linker **10**.

Complex neoglycoconjugates were characterized by a Bruker Autoflex MALDI-TOF/TOF mass spectrometer using 6-aza-2-thio-thymine as matrix in either positive or negative ion reflectron modes. For microarrays Nexterion® Slides H were purchased from Schott, and the slides were scanned with an Agilent G2565CA Microarray scanner equipped with two lasers, a SHG-YAG laser (532 nm) and a helium-neon laser (633 nm). Integrated spot intensities were determined using Imagene software (Biodiscovery).

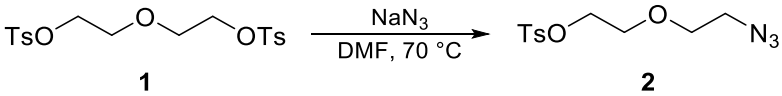
NMR spectra were acquired on a Bruker Avance 300 (300.13 MHz for ^{1}H, 75.46 MHz for ^{13}C) or Bruker Avance III 600 instrument (600.22 MHz for ^{1}H, 150.93 MHz for ^{13}C) using standard Bruker NMR software. ^{1}H spectra were referenced to tetramethylsilane (TMS, δ = 0) or by calibration with the residual solvent peak ($CDCl_3$ δ = 7.26, MeOD δ 3.31). ^{13}C spectra were referenced to 77.00 ($CDCl_3$) or 49.00 (MeOD), respectively. Assignments were based on COSY, HSQC and NOESY experiments.

Preparation of linker



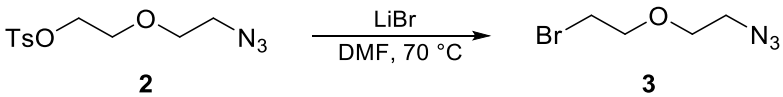
1

TsCl (76.3 g, 0.40 mol) was added in portions to an ice-cooled solution of diethylene glycol (19.0 mL, 0.20 mol) in CH_2Cl_2 (250 mL). After complete dissolution of TsCl, Et_3N (83.63 mL, 0.60 mol) was added and the reaction mixture was further stirred at 0 °C. After 1 h the mixture was allowed to warm up to rt. The solution was diluted with CH_2Cl_2 (50 mL) and consecutively washed with aq HCl (1 M, 2 x 100 mL), saturated aq $NaHCO_3$ solution (2 x 100 mL) and brine (100 mL). The organic phase was dried over $MgSO_4$, filtered and concentrated under reduced pressure giving the pure ditosylate compound **1** (74 g, 90%) as colorless crystals. NMR data are comparable to published data (Mohler D.L. *et al.* 2006).



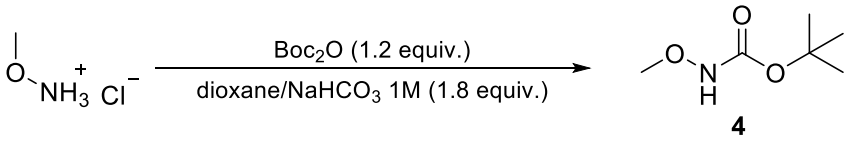
1 **2**

A solution of sodium azide (0.78 g, 12.1 mmol) in dry DMF (135 mL) was added dropwise to a solution of compound **1** (5.00 g, 12.1 mmol) in dry DMF (15 mL). The reaction was stirred at 70 °C under Ar. After 3 h, the mixture was cooled to rt, diluted with EtOAc (150 mL) and washed with water (3 x 100 mL). The aqueous phase was reextracted with EtOAc (2 x 75 mL). The combined organic phases were dried over $MgSO_4$, filtered and concentrated. The resulting residue was purified by flash chromatography (hexane/EtOAc 5:1) to give compound **2** (4.23 g, 62%) as colorless oil. NMR data are comparable to published data (Gill H.S. *et al.* 2009).



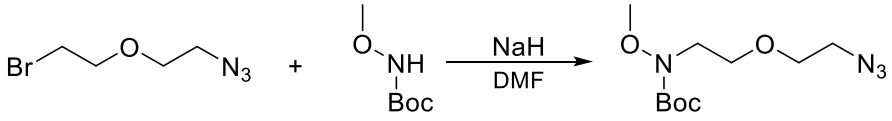
2 **3**

To a solution of compound **2** (4.2 g, 14.8 mmol) in dry DMF (95 mL), lithium bromide (12.8 g, 148 mmol) was added and the reaction mixture was heated to 70 °C. Upon complete conversion (1.5 h) the reaction mixture was cooled to rt and worked up by dilution with a saturated aq $NaHCO_3$ solution (475 mL). The aqueous phase was separated and extracted with hexane/EtOAc (2:1, 3 x 200 mL). The combined organic phases were washed three times with water (3 x 200 mL), once with brine (200 mL), dried over $MgSO_4$ and filtered. The solvent was removed by rotary evaporation to give compound **3** (2.35 g, 82%) as yellowish liquid and used without further purification. R_f 0.55 (hexane/EtOAc 2:1); ^{1}H NMR (300 MHz, $CDCl_3$): δ 3.83 (t, *J* = 6.2 Hz, 2H, $BrCH_2CH_2O$), 3.70 (t, *J* = 5.0 Hz, 2H, $OCH_2CH_2N_3$), 3.48 (t, *J* = 6.2 Hz, 2H, $BrCH_2$), 3.41 (t, *J* = 5.0 Hz, 2H, CH_2N_3); ^{13}C NMR (75 MHz, $CDCl_3$): δ 71.32 (CH_2, $BrCH_2CH_2O$), 70.09 (CH_2, $OCH_2CH_2N_3$), 50.85 (CH_2, CH_2N_3), 30.11 (CH_2, $BrCH_2$).



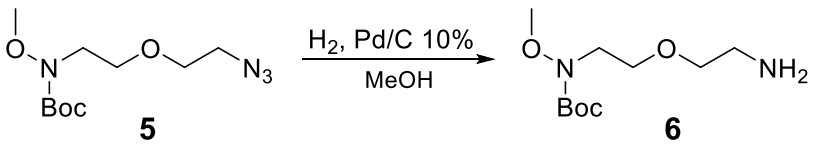
4

A solution of methoxyamine hydrochloride (16.7 g, 0.20 mol) in 1 M aq $NaHCO_3$ (170 mL, 0.17 mol) was added dropwise at rt to a stirred solution of di-*tert*-butyl dicarbonate (52.38 g, 0.24 mol) in 1,4-dioxane (240 mL). The resulting solution was stirred overnight at rt. Dioxane was removed under reduced pressure and the aqueous solution was acidified to pH 4 with solid citric acid. The mixture was extracted with CH_2Cl_2 (3 x 60 mL). The combined organic layers were washed with water (2 x 90 mL), aq $NaHCO_3$ solution (2 x 90 mL) and brine (90 mL), dried over $MgSO_4$, filtered and concentrated to give a crude product which was purified by vacuum distillation (b.p. 88-89 °C / 20 mbar) to yield compound **4** (21.7 g, 74%) as a colorless liquid. R_f 0.42 (hexane/EtOAc 2:1); ^{1}H NMR (300 MHz, $CDCl_3$): δ 3.72 (s, 3H, OC*H*$_3$), 1.49 [s, 9H, C(C*H*$_3$)$_3$]; ^{13}C NMR (75 MHz, $CDCl_3$): δ 156.94 (C_q, *C*=O), 81.86 [C_q, O*C*(CH$_3$)$_3$], 64.57 (CH_3, O*C*H$_3$), 28.33 (CH_3, *C*H$_3$C).



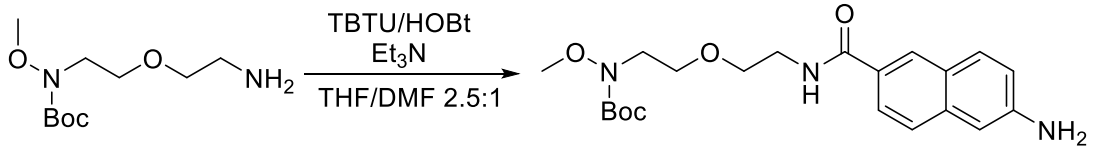
3 **4** **5**

Compound **4** (1.6 g, 10.91 mmol) was dissolved in dry DMF (100 mL), and NaH (60% suspension in mineral oil, 0.48 g, 12.0 mmol) was added portionwise. The reaction mixture was allowed to stir under Ar for 30 min at rt. Compound **3** (2.96 g, 15.3 mmol) dissolved in dry DMF (60 mL) was added in one portion to the stirred solution. The reaction mixture was stirred under Ar until complete conversion (2.5 h). Aq NH_4Cl (200 mL) was added followed by extraction with EtOAc (3 x 120 mL). The combined organic layers were washed with aq $NaHCO_3$ (2 x 120mL), water (120 mL) and brine (120 mL), dried over Na_2SO_4, filtered and concentrated. The crude residue was further purified by column chromatography (hexane/EtOAc 4:1) to give compound **5** (2.59 g, 91%) as yellowish liquid. R_f 0.22 (hexane/EtOAc 4:1); ^{1}H NMR (600 MHz, $CDCl_3$): δ 3.70 (s, 3H, OC*H*$_3$), 3.67-3.64 (m, 6H, CH$_3$ONC*H*$_2$C*H*$_2$OC*H*$_2$), 3.38 (t, *J* = 5.1 Hz, 2H, C*H*$_2$N$_3$), 1.50 [s, 9H, C(C*H*$_3$)$_3$]; ^{13}C NMR (151 MHz, $CDCl_3$): δ 156.50 (C_q, *C*=O), 81.55 (C_q, O*C*(CH$_3$)$_3$), 69.77 (CH_2, O*C*H$_2$CH$_2$N$_3$), 67.68 (CH_2, NCH$_2$*C*H$_2$O), 62.47 (CH_3, O*C*H$_3$), 50.88 (CH_2, N*C*H$_2$), 48.90 (CH_2, *C*H$_2$N$_3$), 28.40 [CH_3, C(*C*H$_3$)$_3$]; ESI-TOF HR-MS: *m/z* = 261.1555; calcd. for $C_{10}H_{21}N_4O_4$: 261.1557.



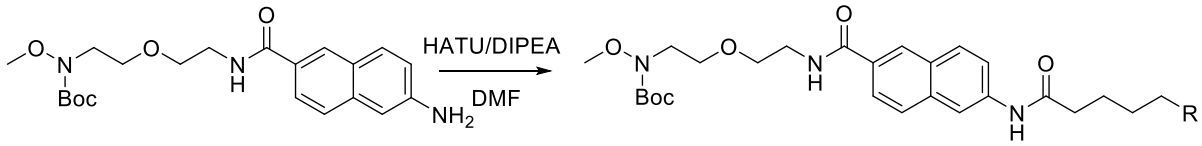
5 **6**

Compound **5** (1.27 g, 4.9 mmol) was dissolved in dry MeOH (635 mL) and the atmosphere was exchanged three times by evacuation and flushing with argon. Then, Pd-C (10 wt.%, 127 mg) was added to the flask and the atmosphere was exchanged by evacuation and flushing three times with hydrogen. The suspension was stirred for 1.5 h at rt, diluted with MeOH (200 mL), passed through a 0.45 µm syringe filter and the filtrate was concentrated under reduced pressure. The resulting residue was further purified by column chromatography ($CHCl_3$/MeOH 3:1 + Et_3N 1%) to give compound **6** (840 mg, 74%) as colorless liquid. R_f 0.58 ($CHCl_3$/MeOH/Et_3N 1:10:1); ^{1}H NMR (600 MHz, $CDCl_3$): δ 3.69 (s, 3H, OC*H*$_3$), 3.64 (m, 4H, ONC*H*$_2$C*H*$_2$O), 3.51 (t, *J* = 4.6 Hz, 2H, OC*H*$_2$CH$_2$NH$_2$), 2.87 (t, *J* = 4.4 Hz, 2H, C*H*$_2$NH$_2$), 1.50 [s, 9H, C(C*H*$_3$)]; ^{13}C NMR (151 MHz, $CDCl_3$): δ 156.71 (C_q, *C*=O), 81.48 [C_q, *C*(CH$_3$)$_3$], 72.94 (CH$_2$, O*C*H$_2$CH$_2$NH$_2$), 67.27 (CH$_2$, ONCH$_2$*C*H$_2$), 62.39 (CH$_3$, O*C*H$_3$), 48.86 (CH$_2$, ON*C*H$_2$), 41.83 (CH$_2$, *C*H$_2$NH$_2$), 28.40 [CH$_3$, C(*C*H$_3$)$_3$]; ESI-TOF HR-MS: *m/z* = 235.1654; calcd. for $C_{10}H_{23}N_2O_4$: 235.1652.



6 **7**

TBTU (1.23 g, 3.83 mmol) and HOBt (516 mg, 3.82 mmol) were added to a solution of 6-amino-2-naphthoic acid (714 mg, 3.81 mmol) in dry THF/DMF 2.5:1 (140 mL). Then, Et_3N (2.7 mL, 19.2 mmol) was added and the solution was purged with a stream of argon for 30 min. This mixture was added dropwise to a solution of compound **6** (895 mg, 3.82 mmol) in dry THF (20 mL). The reaction mixture was stirred at rt under protected atmosphere overnight. Upon completion, THF was removed under reduced pressure, and the mixture was diluted with EtOAc (40 mL), washed with water (2 x 40 mL), aq NH_4Cl (40 mL), aq $NaHCO_3$ (40 mL), brine (40 mL) and dried over Na_2SO_4. Concentration of the organic phase afforded the crude product which was purified by column chromatography ($CHCl_3$/MeOH 9:1) to yield compound **7** (1.2 g, 78%) as brown syrup. R_f 0.60 ($CHCl_3$/MeOH 9:1); ^{1}H NMR (600 MHz, MeOD): δ 8.20 (d, *J* = 1.8 Hz, 1H, Ar*H*), 7.73 (dd, *J* = 8.6, 1.9 Hz, 1H, Ar*H*), 7.71 (d, *J* = 8.8 Hz, 1H, Ar*H*), 7.57 (d, *J* = 8.6 Hz, 1H, Ar*H*), 7.05 (dd, *J* = 8.7, 2.3 Hz, 1H, Ar*H*), 6.98 (d, *J* = 2.3 Hz, 1H, Ar*H*), 3.68-3.66 (m, 6H, ONC*H*$_2$C*H*$_2$OC*H*$_2$), 3.64 (s, 3H, OC*H*$_3$), 3.59 (t, *J* = 5.6 Hz, 2H, C*H*$_2$NHCO), 1.44 [s, 9H, C(C*H*$_3$)]; ^{13}C NMR (151 MHz, MeOD): δ 170.70 (C_q, HN*C*=O), 158.43 (C_q, *C*=O), 149.20 (C_q, Ar*C*), 138.47 (C_q, Ar*C*), 131.19 (CH, Ar*C*), 128.94 (CH, Ar*C*), 128.22 (C_q, Ar*C*), 127.74 (C_q, Ar*C*), 126.69 (CH, Ar*C*H), 125.09 (CH, Ar*C*H), 120.31 (CH, Ar*C*H), 108.37 (CH, Ar*C*H), 82.78 [C_q, *C*(CH$_3$)$_3$], 70.52 (CH$_2$, O*C*H$_2$CH$_2$NHCO), 68.10 (CH$_2$, ONCH$_2$*C*H$_2$O), 62.57 (CH$_3$, O*C*H$_3$), 49.71 (CH$_2$, ON*C*H$_2$), 40.94 (CH$_2$, *C*H$_2$NHCO), 28.53 [CH$_3$, C(*C*H$_3$)$_3$]; ESI-TOF HR-MS: *m/z* = 404.2180; calcd. for $C_{21}H_{30}N_3O_5$: 404.2180.



7

8 R = NHBoc
9 R = N_3

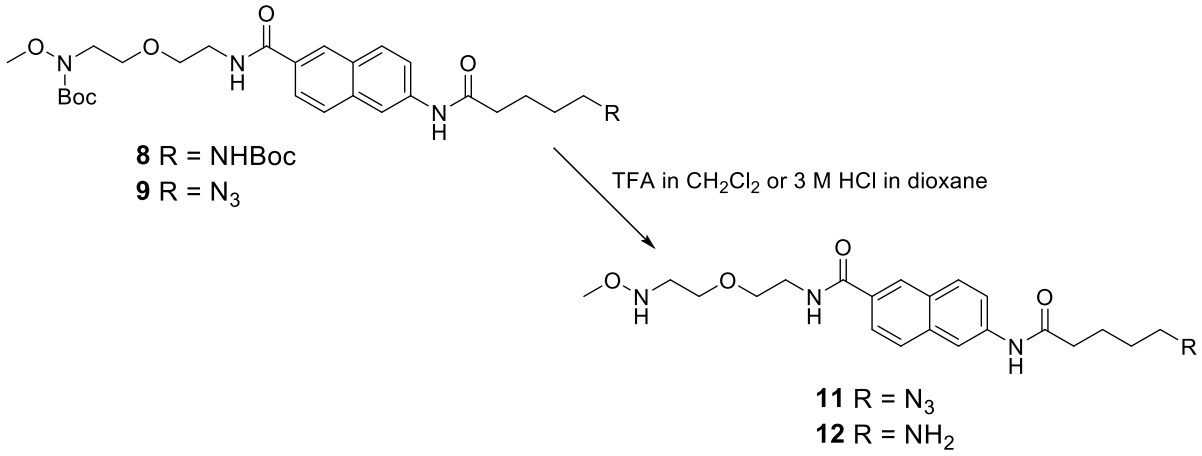
For the synthesis of compound **8**, HATU (0.43 g, 1.1 mmol), and DIPEA (0.6 mL, 3.4 mmol) were first added to a solution of Boc-5-aminopentanoic acid (0.25 g, 1.2 mmol) in dry DMF (21 mL) and allowed to react for 30 min under protected atmosphere. The solution was then added to a solution of compound **7** (500 mg, 1.24 mmol) in dry DMF (45 mL). The reaction mixture was stirred at rt under protected atmosphere for 18 h. The mixture was diluted with Et_2O (60 mL), washed with water (3 x 40 mL) and then acidified with aq HCl (250 mM, 2 x 20 mL). The organic layer was neutralized with aq $NaHCO_3$ (25 mL, caution!), washed with brine (50 mL), dried over $MgSO_4$, filtered and concentrated to yield compound **8** (642 mg, 92%) which was used without further purification. R_f 0.28 (hexane/EtOAc 1:10); ^{1}H NMR (600 MHz, $CDCl_3$): δ 8.38 (s, 1H, Ar*H*), 8.28 (s, 1H, Ar*H*), 7.93 (dd, *J* = 8.6, 1.7 Hz, 1H, Ar*H*), 7.88 (d, *J* = 8.9 Hz, 1H, Ar*H*), 7.80 (d, *J* = 8.6 Hz, 1H, Ar*H*), 7.55 (d, *J* = 8.7 Hz, 1H, Ar*H*), 3.70-3.66 (m, 8H, C*H*$_2$C*H*$_2$OC*H*$_2$C*H*$_2$), 3.63 (s, 3H, OC*H*$_3$), 3.19 [s, 2H, C*H*$_2$NH(Boc)], 2.47 (t, *J* = 7.5 Hz, 2H, NHCOC*H*$_2$), 1.83-1.78 [m, 2H, C*H*$_2$CH$_2$CH$_2$NH(Boc)], 1.61-1.55 [m, 2H,C*H*$_2$CH$_2$NH(Boc)], 1.46 (s, 9H, C*H*$_3$C), 1.45 [s, 9H, C(C*H*$_3$)$_3$]; ^{13}C NMR (151 MHz, $CDCl_3$): 167.67 (C_q, ArHN*C*=O), 157.47 (C_q, *C*=O), 137.33 (C_q, Ar*C*), 135.54 (C_q, Ar*C*), 130.71 (C_q, Ar*C*), 129.98 (CH, Ar*C*H), 129.77 (C_q, Ar*C*), 127.94 (CH, Ar*C*H), 127.65 (CH, Ar*C*H), 124.70 (CH, Ar*C*H), 120.61 (CH, Ar*C*H), 116.06 (CH, Ar*C*H), 81.72 [C_q, $(CH_3)_3C$], 70.15 (CH_2, O*C*H$_2$CH$_2$NHCO), 67.08 (CH_2, ONCH$_2$*C*H$_2$O), 62.26 (CH_3, O*C*H$_3$), 48.81 (CH_2, ON*C*H$_2$), 39.91 (CH_2, *C*H$_2$NHCO), 39.48 [CH_2, *C*H$_2$NH(Boc)], 37.02 (CH_2, NHCO*C*H$_2$), 29.68 [CH_2, *C*H$_2$CH$_2$NH(Boc)], 28.57 [CH_3, C(*C*H$_3$)$_3$], 28.38 [CH_3, C(*C*H$_3$)$_3$], 22.67 (CH_2, NHCOCH$_2$*C*H$_2$); ESI-TOF HR-MS: *m/z* = 603.3391; calcd. for $C_{31}H_{47}N_4O_8$: 603.3388.

Excess of starting material **7** was recovered by neutralization of the acidic aq phase with aq NaOH (250 mM, 40 mL) followed by extraction with Et_2O (2 x 50 mL). The organic phase containing compound **7** was washed with aq $NaHCO_3$ (50 mL), brine (50 mL), dried over $MgSO_4$, filtered and concentrated to give compound **7** (58 mg, 12%) as brown syrup.

For the synthesis of compound **9**, HATU (0.30 g, 0.79 mmol), and DIPEA (137.3 µL, 0.81 mmol) were first added to a solution of 5-azido-pentanoic acid (103.5 µL, 0.72 mmol) in dry DMF (30 mL) and allowed to react for 1 h under protected atmosphere. The solution was then added to a solution of compound **7** (350 mg, 0.87 mmol) in dry DMF (30 mL). The reaction mixture was stirred at rt under protected atmosphere for 21 h. The mixture was diluted with water (120 mL) and the product was extracted with Et_2O (3 x 60 mL). The combined organic layers were washed with water (2 x 90 mL) and then acidified with aq HCl (0.5 M, 90 mL), neutralized with aq $NaHCO_3$ (2 x 90 mL, caution!), washed with brine (90 mL) and dried

over $MgSO_4$, filtered and concentrated to yield compound **9** (319 mg, 77%) which was used without further purification. R_f 0.40 (CH_2Cl_2/MeOH 20:1); ^{1}H NMR (600 MHz, $CDCl_3$): δ 8.38 (s, 1H, Ar*H*), 8.26 (s, 1H, Ar*H*), 7.93 (d, *J* = 8.6 Hz, 1H, Ar*H*), 7.86 (d, *J* = 8.8 Hz, 1H, Ar*H*), 7.79 (d, *J* = 8.6 Hz, 1H, Ar*H*), 7.49 (dd, *J* = 8.9, 1.3 Hz, 1H, Ar*H*), 3.69-3.66 (m, 8H, CH_2CH_2OCH_2CH_2), 3.62 (s, 3H, OCH_3), 3.34 (t, *J* = 6.7 Hz, 2H, C$H_2$$N_3$), 2.47 (t, *J*= 7.3 Hz, 2H, NHCOCH_2), 1.88-1.83 (m, 2H, C$H_2$$CH_2CH_2N_3$), 1.73-1.68 (m, 2H, C$H_2$$CH_2N_3$), 1.46 [s, 9H, C(C$H_3$)$_3$]; ^{13}C NMR (151 MHz, $CDCl_3$): δ 171.11 (C_q, Ar*C*=O). 167.74 (C_q, ArHN*C*=O), 157.50 (C_q, *C*=O), 137.06 (C_q, Ar*C*), 135.52 (C_q, Ar*C*), 130.77 (C_q, Ar*C*), 129.85 (C_q, Ar*C*), 130.09 (CH, Ar*C*H), 127.98 (CH, Ar*C*H), 127.71 (CH, Ar*C*H), 124.77 (CH, Ar*C*H), 120.54 (CH, Ar*C*H), 116.21 (CH, Ar*C*H), 81.73 [C_q, *C*(CH$_3$)$_3$], 70.12 (CH_2, O*C*$H_2$$CH_2$NHCO), 67.09 ($CH_2$, ONCH$_2$*C*$H_2$O), 62.25 ($CH_3$, O*C*$H_3$), 51.35 ($CH_2$, *C*$H_2$$N_3$), 48.83 ($CH_2$, ON*C*$H_2$), 39.97 ($CH_2$, *C*$H_2$NHCO), 37.05 ($CH_2$, NHCO*C*$H_2$), 28.49 ($CH_2$, *C*$H_2$$CH_2N_3$), 28.39 [$CH_3$, C(*C*H$_3$)$_3$], 22.80 ($CH_2$, NHCOCH$_2$*C*$H_2$); ESI-TOF HR-MS: *m/z* = 529.2775; calcd. for $C_{26}H_{37}N_6O_6$: 529.2769.

Excess of starting material **7** (95 mg, 27%) was recovered as described above.



Compound **9** (470 mg, 0.89 mmol) was dissolved in CH_2Cl_2 (14 mL), and TFA (2.9 mL, 37.64 mmol) was added gradually over 15 min. The reaction mixture was stirred under protected atmosphere at rt for 1.5 h. Upon complete deprotection, the reaction mixture was diluted with CH_2Cl_2 (280 mL), neutralized with aq $NaHCO_3$ (2 x 150 mL) and washed with brine (2 x 100 mL). Concentration of the organic phase afforded the desired compound **11** (352 mg, 92%) as light orange syrup, which was used without further purification. R_f 0.35 ($CHCl_3$/MeOH 20:1); ^{1}H NMR (600 MHz, $CDCl_3$): δ 8.25 (s, 1H, Ar*H*), 8.23 (s, 1H, Ar*H*), 7.80 (d, *J* = 8.8 Hz, 1H, Ar*H*), 7.79 (dd, *J* = 8.6,1.6 Hz, 1H, Ar*H*), 7.76 (d, *J* = 8.6 Hz, 1H, Ar*H*), 7.48 (dd, *J* = 8.8, 1.8 Hz, 1H, Ar*H*), 3.73-3.68 (m, 4H, OCH_2CH_2NHCO), 3.66 (t, *J* = 5.2 Hz, 2H, CH_3ONHCH$_2$CH_2O), 3.53 (s, 3H, OCH_3), 3.33 (t, *J* = 6.7 Hz, 2H, C$H_2$$N_3$), 3.13 (t, *J* = 5.2 Hz, 2H, CH_3ONHCH_2), 2.47 (t, *J* = 7.3 Hz, 2H, NHCOCH_2), 1.88-1.83 (m, 2H, C$H_2$$CH_2CH_2N_3$), 1.78-1.67 (m, 2H, C$H_2$$CH_2N_3$). ^{13}C NMR (151 MHz, $CDCl_3$): δ 171.16 (C_q, Ar*C*=O), 167.85 (C_q, ArHN*C*=O), 137.12 (C_q, Ar*C*), 135.50 (C_q, Ar*C*), 130.88 (C_q, Ar*C*), 129.93 (CH, Ar*C*H), 128.16 (CH, Ar*C*H), 127.42 (CH, Ar*C*H), 124.35

(CH, Ar*C*H), 120.75 (CH, Ar*C*H), 116.30 (CH, Ar*C*H), 69.83 (CH_2, O*C*$H_2$$CH_2$NHCO), 67.10 ($CH_2$, CH_3ONHCH_2*C*H_2O), 61.74 (CH_3, O*C*H_3), 51.33 (CH_2, *C*H_2N_3), 51.10 (CH_2, CH_3ONH*C*H_2), 39.97 (CH_2, *C*H_2NHCO), 37.04 (CH_2, NHCO*C*H_2), 28.48 (CH_2, *C*$H_2CH_2N_3$), 22.79 (NHCOCH_2*C*H_2); ESI-TOF HR-MS: *m/z* = 429.2260; calcd. for $C_{21}H_{29}N_6O_4$: 429.2245.

Compound **8** (210 mg, 0.35 mmol) was dissolved in a 3 M HCl solution in dioxane (11.4 mL) and the reaction mixture was stirred under protected atmosphere at rt for 1.5 h. Upon complete deprotection, the reaction mixture was diluted with Et_2O and the precipitate was separated from the supernatant by centrifugation. The precipitate was washed three times with Et_2O, and then dried under reduced pressure. The precipitate was then dissolved in MeOH (pH 2) and the solution was neutralized by addition of anion exchange resin (AG1-X8, HCO_3^--form). Filtration and concentration of the filtrate afforded compound **12** (132 mg, 94%) in sufficient purity. R_f 0.29 ($CHCl_3$/MeOH/NH_4OH 8:3.5:0.4); ^{1}H NMR (600 MHz, MeOD): δ 8.32 (d, *J* = 0.8 Hz, 1H, Ar*H*), 8.30 (d, *J* = 1.9 Hz, 1H, Ar*H*), 7.93 (d, *J* = 8.9 Hz, 1H, Ar*H*), 7.88 (dd, *J* = 8.6, 1.8 Hz, 1H, Ar*H*), 7.84 (d, *J* = 8.6 Hz, 1H, Ar*H*), 7.65 (dd, *J* = 8.9, 2.1 Hz, 1H, Ar*H*), 3.67 (t, *J* = 4.9 Hz, 2H, CH_2OC*H*$_2$), 3.65-3.63 (m, 4H, C*H*$_2$OCH_2C*H*$_2$), 3.48 (s, 3H, OC*H*$_3$) 3.05 (t, *J* = 5.3 Hz, 2H, CH_3ONC*H*$_2$), 2.93 (t, *J* = 7.4 Hz, 2H, C*H*$_2$$NH_2$), 2.52 (t, *J* = 7.1 Hz, 2H, NHCOC*H*$_2$$CH_2CH_2CH_2NH_2$), 1.84-1.79 (m, 2H, C*H*$_2$$CH_2CH_2NH_2$), 1.75-1.70 (m, 2H, C*H*$_2$$CH_2NH_2$); ^{13}C NMR (151 MHz, MeOD): δ 130.72 (CH, Ar*C*H), 128.84 (CH, Ar*C*H), 128.57 (CH, Ar*C*H), 125.51 (CH, Ar*C*H), 122.04 (CH, Ar*C*H), 117.20 (CH, Ar*C*H), 70.68 (CH_2, O*C*$H_2$$CH_2$NHCO), 68.29 ($CH_2$, ONC$H_2$*C*$H_2$O), 61.43 ($CH_3$, O*C*$H_3$), 51.81 ($CH_2$, ON*C*$H_2$), 41.07 ($CH_2$, *C*$H_2$NHCO), 40.74 ($CH_2$, *C*$H_2$$NH_2$), 37.14 ($CH_2$, NHCO*C*$H_2$), 29.07 ($CH_2$, *C*$H_2$$CH_2NH_2$), 23.47 ($CH_2$, NHCOC$H_2$*C*$H_2$); ESI-TOF HR-MS: *m/z* = 403.2360; calcd. for $C_{21}H_{30}N_4O_4$: 404.2340.

Hydrogenation of azido-functionalized compounds to obtain amino-functionalized compounds 10, 22-33

Linker (**9**) or neoglycoconjugates (**13**-**21**) were dissolved in MeOH and the atmosphere was exchanged three times by evacuation and flushing with argon. Afterwards, Pd/C (10 wt. %) was added to the reaction mixture and the atmosphere was exchanged three times to argon and hydrogen by evacuation and flushing. After 2 h, the reaction mixture was diluted with MeOH, passed through a 0.45 µm syringe filter and the filtrate was concentrated under reduced pressure to yield the corresponding amino-functionalized compounds as yellowish syrups in 99% yield for linker **10** and in 75% average yield for neoglycoconjugates (**22**-**33**). All amino-functionalized compounds were used without further purification.

Structural characterization of linker 10 and neoglycoconjugates (13-33)

***Tert*-butyl *N*-{2-[2-(6-(5-aminopentanamido)-2-naphthamido)ethoxy]ethyl}-*N*-methoxycarbamate (10)**

R_f 0.36 ($CHCl_3$/MeOH/NH_4OH 8:3.5:0.4); ^{1}H NMR (600 MHz, MeOD) δ 8.34 (d, *J* = 0.9 Hz, 1H, Ar*H*), 8.30 (d, *J* = 1.7 Hz, 1H, Ar*H*), 7.93 (d, *J* = 9.0 Hz, 1H, Ar*H*), 7.88 (dd, *J* = 8.6,1.8 Hz, 1H, Ar*H*), 7.84 (d, *J* = 8.8 Hz, 1H, Ar*H*), 7.64 (dd, *J* = 8.9, 2.1 Hz, 1H, Ar*H*), 3.69 (t, *J* = 5.6 Hz, 2H, OC$H_2$$CH_2$NHCO), 3.67 (dd, *J* = 3.9, 2.7 Hz, 4H, CH_3ONHCH_2CH_2O), 3.64 (s, 3H, OCH_3), 3.61 (t, *J* = 5.5 Hz, 2H, CH_2NHCO), 2.76 (t, *J* = 7.3 Hz, 2H, C$H_2$$NH_2$), 2.48 (t, *J* = 7.4 Hz, 2H, NHCOCH_2), 1.81-1.76 (m, 2H, C$H_2$$CH_2$$CH_2$$NH_2$), 1.65-1.60 (m, 2H, C$H_2$$CH_2$$NH_2$); ^{13}C NMR (151 MHz, MeOD): δ 174.57 (C_q, Ar*C*=O), 170.25 (C_q, ArHN*C*=O), 158.44 (C_q, *C*=O), 139.31 (C_q, Ar*C*), 136.86 (C_q, Ar*C*), 131.75 (C_q, Ar*C*), 130.97 (C_q, Ar*C*), 130.76 (CH, Ar*C*H), 128.83 (CH, Ar*C*H), 128.62 (CH, Ar*C*H), 125.53 (CH, Ar*C*H), 122.00 (CH, Ar*C*H), 117.17 (CH, Ar*C*H), 82.77 [C_q, *C*(CH_3)$_3$], 70.44 (CH_2, O*C*$H_2$$CH_2$NHCO), 68.10 ($CH_2$, ONC$H_2$*C*$H_2$O), 62.55 ($CH_3$, O*C*$H_3$), 49.71 ($CH_2$, ON*C*$H_2$), 41.78 ($CH_2$, *C*$H_2$$NH_2$), 41.03 ($CH_2$, *C*$H_2$NHCO), 37.57 ($CH_2$, NHCO*C*$H_2$) 32.11 ($CH_2$, *C*$H_2$$CH_2$$NH_2$), 28.52 [$CH_3$, C(*C*$H_3$)$_3$], 23.90 [$CH_2$, NHC(O)$CH_2$*C*$H_2$]; ESI-TOF HR-MS: *m/z* 537.2493; calcd. for $C_{26}H_{38}ClN_4O_6$: 537.2485.

6-(5-Azidopentanamido)-*N*-{2-[2-(*N*-2-acetamido-2-deoxy-β-D-glucopyranosyl-*N*-methoxy-amino)ethoxy]ethyl}-2-naphthamide (13)

R_f 0.19 (CH_2Cl_2/MeOH 5:1); ^{1}H-NMR (600 MHz, MeOD): δ 8.34 (d, *J* = 1.8 Hz, 1H, Ar*H*), 8.30 (d, *J* = 2.0 Hz, 1H, Ar*H*), 7.93 (d, *J* = 8.9 Hz, 1H, Ar*H*), 7.88 (dd, *J* = 8.6, 1.8 Hz, 1H, Ar*H*), 7.84 (d, *J* = 8.7 Hz, 1H, Ar*H*), 7.64 (dd, *J* = 8.8, 2.1 Hz, 1H, Ar*H*), 4.27 (d, $J_{1,2}$ = 9.8 Hz, 1H, H-1), 3.83 (dd, $J_{6a,6b}$ = 12.2 Hz, $J_{5,6a}$ = 2.4 Hz ,1H, H-6a), 3.82 (t, $J_{1,2}$ = 9.8 Hz, 1H, H-2), 3.71-3.64 (m, 7H, H-6b, CH_2OCH_2, CH_2NHCO), 3.49 (s, 3H, OCH_3), 3.42 (dd, $J_{2,3}$ = 9.9 Hz, $J_{3,4}$ = 8.9 Hz, 1H, H-3), 3.37 (t, *J* = 6.8 Hz, 2H, C$H_2$$N_3$), 3.28 (t, $J_{4,5}$ = 8.8 Hz, 1H, H-4), 3.24-3.19 (m, 2H, H-5, CH_3ONCH_2), 3.14 (dt, *J* = 13.6, 6.1 Hz, 1H, CH_3ONCH_2), 2.49 (t, *J* = 7.4 Hz, 2H, HNCOCH_2), 1.94 (s, 3H, CH_3CO), 1.85-1.80 (m, 2H, NHCOCH_2), 1.72-1.67 (m, 2H, C$H_2$$CH_2$$N_3$); ^{13}C-NMR (151 MHz, MeOD): δ 174.39 (C_q, Ar*C*=O), 173.46 (C_q, CH_3*C*=O), 170.30 (C_q, ArNH*C*=O), 139.31 (C_q, Ar*C*), 136.87 (C_q, Ar*C*), 131.77 (C_q, Ar*C*), 130.98 (C_q, Ar*C*), 130.87 (CH, Ar*C*H), 128.94 (CH, Ar*C*H) , 128.72 (CH, Ar*C*H), 125.51 (CH, Ar*C*H) , 122.03 (CH, Ar*C*H), 117.20 (CH, Ar*C*H), 93.06 (CH, C-1), 79.87 (CH, C-5), 77.77 (CH, C-3), 72.07 (CH, C-4), 70.47 (CH_2, CH_2O*C*H_2), 70.07 (CH_2, *C*H_2OCH_2), 62.97 (CH_2, C-6), 62.17 (CH_3, O*C*H_3), 54.18 (CH, C-2), 52.37 (CH_2, CH_3ON*C*H_2), 52.18 (CH_2, *C*$H_2$$N_3$), 41.20 ($CH_2$, *C*$H_2$NHCO), 37.41 ($CH_2$, HNCO*C*$H_2$), 29.50 ($CH_2$, *C*$H_2$$CH_2$$N_3$), 24.08 ($CH_2$, HNCOC$H_2$*C*$H_2$), 23.05 ($CH_3$, *C*$H_3$CO); ESI-TOF HR-MS: *m/z* 632.3058; calcd. for $C_{29}H_{42}N_7O_9$: 632.3039.

6-(5-Azidopentanamido)-*N*-{2-[2-(*N*-β-D-galactofuranosyl-*N*-methoxyamino)ethoxy]ethyl}-2-naphthamide (14a)

R_f 0.41 (CH_2Cl_2/MeOH 5:1); ^{1}H NMR (600 MHz, MeOD): δ 8.32 (d, *J* = 0.8 Hz, 1H, Ar*H*), 8.29 (d, *J* = 1.9 Hz, 1H, Ar*H*), 7.93 (d, *J* = 9.0 Hz, 1H, Ar*H*), 7.87 (dd, *J* = 8.6, 1.7 Hz, 1H, Ar*H*), 7.84 (d, *J* = 8.7 Hz, 1H, Ar*H*), 7.64 (dd, *J* = 8.9, 2.1 Hz, 1H, Ar*H*), 4.49 (d, $J_{1,2}$ = 5.6 Hz, 1H, H-1), 4.20 (dd, $J_{2,3}$ = 6.8 Hz, 1H, H-2), 4.08 (dd, $J_{3,4}$ = 8.3 Hz, 1H, H-3), 3.85 (dd, $J_{4,5}$ = 2.7 Hz, 1H, H-4), 3.73 (t, *J* = 6.0 Hz, 2H, CH_2OCH$_2$), 3.70 (t, *J* = 5.8 Hz, 2H, CH$_2$OCH_2), 3.66-3.63 (m, 3H, CH_2NHCO, H-5), 3.60 (s, 2H, H-6a, H-6b), 3.59 (s, 3H, OCH_3), 3.37 (t, *J* = 6.8 Hz, 2H, CH_2N$_3$), 3.13 (dt, *J* = 13.6, 5.6 Hz, 1H, CH$_3$ONCH_2), 3.04 (dt, *J* = 13.7, 5.8 Hz, 1H, CH$_3$ONCH_2), 2.49 (t, *J* = 7.4 Hz, 2H, HNCOCH_2), 1.85-1.80 (m, 2H, HNCOCH$_2$CH_2), 1.72-1.67 (m, 2H, CH_2CH$_2$N$_3$). ^{13}C NMR (151 MHz, MeOD): δ 130.44 (CH, Ar*C*H), 128.56 (CH, Ar*C*H), 128.22 (CH, Ar*C*H), 125.03 (CH, Ar*C*H), 121.76 (CH, Ar*C*H), 116.85 (CH, Ar*C*H), 99.57 (CH, C-1), 82.97 (CH, C-4), 78.33 (CH, C-2), 77.30 (CH, C-3), 71.84 (CH, C-5), 70.05 (CH$_2$, CH$_2$O*C*H$_2$), 69.38 (CH$_2$, *C*H$_2$OCH$_2$), 64.55 (CH$_2$, C-6), 62.70 (CH$_3$, O*C*H$_3$), 53.59 (CH$_2$, CH$_3$ON*C*H$_2$), 52.02 (CH$_2$, *C*H$_2$N$_3$), 40.65 (CH$_2$, *C*H$_2$NHCO), 37.01 (CH$_2$, HNCO*C*H$_2$), 29.27 (CH$_2$, *C*H$_2$CH$_2$N$_3$), 23.55 (CH$_2$, HNCOCH$_2$*C*H$_2$); ESI-TOF HR-MS: *m/z* 591.2772; calcd. for $C_{27}H_{39}N_6O_9$: 591.2773.

6-(5-Azidopentanamido)-*N*-{2-[2-(*N*-β-D-galactopyranosyl-*N*-methoxy-amino)ethoxy]ethyl}-2-naphthamide (14b)

R_f 0.28 (CH_2Cl_2/MeOH 5:1); ^{1}H NMR (600 MHz, MeOD): δ 8.33 (d, *J* = 1.4 Hz, 1H, Ar*H*), 8.29 (d, *J* = 1.7 Hz, 1H, Ar*H*), 7.94 (d, *J* = 8.9 Hz, 1H, Ar*H*), 7.88 (dd, *J* = 8.6, 1.8 Hz, 1H, Ar*H*), 7.84 (d, *J* = 8.7 Hz, 1H, Ar*H*), 7.64 (dd, *J* = 8.9, 2.1 Hz, 1H, Ar*H*), 4.06 (d, $J_{1,2}$ = 9.1 Hz, 1H, H-1), 3.79 (dd, $J_{3,4}$ = 3.4 Hz, $J_{4,5}$ = 0.8 Hz, 1H, H-4), 3.75-3.72 (m, 6H, H-6a, H-6b, CH_2OCH_2), 3.68 (t, $J_{1,2}$ = $J_{2,3}$ = 9.3 Hz, 1H, H-2), 3.67-3.64 (m, 2H, CH_2NHCO), 3.57 (s, 3H, OCH_3), 3.47 (dd, $J_{2,3}$ = 9.3 Hz, $J_{3,4}$ = 3.4 Hz, 1H, H-3), 3.45 (ddd, $J_{4,5}$ = 1.0 Hz, $J_{5,6}$ = 5.3 Hz, 1H, H-5), 3.37 (t, *J* = 6.8 Hz, 2H, CH_2N$_3$), 3.24 (dt, *J* = 14.0, 5.8 Hz, 1H, CH$_3$ONCH_2), 3.12 (dt, *J* = 14.0, 5.4 Hz, 1H, CH$_3$ONCH_2), 2.49 (t, *J* = 7.4 Hz, 2H, NHCOCH_2), 1.84-1.80 (m, 2H, NHCOCH$_2$CH_2), 1.72-1.67 (m, 2H, CH_2CH$_2$N$_3$); ^{13}C NMR (151 MHz, MeOD): δ 174.42 (C$_q$, Ar*C*=O), 170.33 (C$_q$, ArHN*C*=O), 139.30 (C$_q$, Ar*C*), 136.86 (C$_q$, Ar*C*), 131.76 (C$_q$, Ar*C*), 130.97 (C$_q$, Ar*C*), 130.79 (CH, Ar*C*H), 128.90 (CH, Ar*C*H), 128.58 (CH, Ar*C*H), 125.46 (CH, Ar*C*H), 122.04 (CH, Ar*C*H), 117.19 (CH, Ar*C*H), 95.39 (CH, C-1), 78.40 (CH, C-5), 76.23 (CH, C-3), 70.52 (CH, C-4), 70.49 (CH$_2$, CH$_2$O*C*H$_2$), 69.72 (CH$_2$, *C*H$_2$OCH$_2$), 69.00 (CH, C-2), 62.66 (CH$_2$, C-6), 62.42 (CH$_3$, O*C*H$_3$), 52.56 (CH$_2$, CH$_3$ON*C*H$_2$), 52.20 (CH$_2$, *C*H$_2$N$_3$), 40.98 (CH$_2$, *C*H$_2$NHCO), 37.38 (CH$_2$, HNCO*C*H$_2$), 29.48 (CH$_2$, *C*H$_2$CH$_2$N$_3$), 23.99 (CH$_2$, HNCOCH$_2$*C*H$_2$); ESI-TOF HR-MS: *m/z* 591.2774; calcd. for $C_{27}H_{39}N_6O_9$: 591.2773.

6-(5-Azidopentanamido)-*N*-{2-[2-(*N*-β-D-glucopyranosyl-*N*-methoxyamino)ethoxy]ethyl}-2-naphthamide (15)

R_f 0.25 (CH_2Cl_2/MeOH 5:1); ^{1}H NMR (600 MHz, MeOD): δ 8.33 (d, *J* = 0.8 Hz, 1H, Ar*H*), 8.29 (d, *J* = 1.7

Hz, 1H, Ar*H*), 7.94 (d, *J* = 8.9 Hz, 1H, Ar*H*), 7.87 (dd, *J* = 8.6, 1.8 Hz, 1H, Ar*H*)), 7.84 (d, *J* = 8.8 Hz, 1H, Ar*H*), 7.64 (dd, *J* = 8.8, 2.0 Hz, 1H, Ar*H*), 4.09 (d, *J* = 8.8 Hz, 1H, H-1), 3.82 (dd, $J_{5,6a}$ = 2.3 Hz, $J_{6a,6b}$ = 12.0 Hz, 1H, H-6a), 3.73 (dd, $J_{5,6b}$ = 5.8 Hz, $J_{6a,6b}$ = 10.6 Hz, 1H, H-6b), 3.72 (dd, *J* = 6.5, 1.4 Hz, 4 H, C*H*$_2$OC*H*$_2$), 3.65 (t, *J* = 5.5 Hz, 2H, C*H*$_2$NHCO), 3.59 (s, 3H, OC*H*$_3$), 3.42 (t, $J_{2,3}$ = 8.9 Hz, 1H, H-2), 3.37 (t, $J_{3,4}$ = 8.9 Hz, 1H, H-3), 3.37 (t, *J* = 6.7 Hz, 2H, C*H*$_2$N$_3$), 3.27 (dd, $J_{4,5}$ = 9.7 Hz, 1H, H-4), 3.25-3.20 (m, 2H, H-5, CH$_3$ONC*H*$_2$), 3.13 (td, *J* = 13.9, 5.6 Hz, 1H, CH$_3$ONC*H*$_2$), 2.49 (t, *J* = 7.4 Hz, 2H, NHCOC*H*$_2$), 1.85-1.80 (m, 2H, HNCOCH$_2$C*H*$_2$), 1.72-1.69 (m, 2H, C*H*$_2$CH$_2$N$_3$); ^{13}C NMR (151 MHz, MeOD): δ 174.37 (C$_q$, Ar*C*=O), 170.31 (C$_q$, ArHN*C*=O), 139.28 (C$_q$, Ar*C*), 136.85 (C$_q$, Ar*C*) 131.73 (C$_q$, Ar*C*), 130.96 (C$_q$, Ar*C*), 130.77 (CH, Ar*C*H), 128.89 (CH, Ar*C*H), 128.57 (CH, Ar*C*H), 125.44 (CH, Ar*C*H), 122.02 (CH, Ar*C*H), 117.18 (CH, Ar*C*H), 94.79 (CH, C-1), 79.69 (CH, C-5), 79.36 (CH, C-3), 71.57 (CH, C-4), 71.30 (CH, C-2), 70.49 (CH$_2$, CH$_2$O*C*H$_2$), 69.62 (CH$_2$, *C*H$_2$OCH$_2$), 62.85 (CH$_2$, C-6), 62.71 (CH$_3$, O*C*H$_3$), 53.11 (CH$_2$, CH$_3$ON*C*H$_2$), 52.19 (CH$_2$,*C*H$_2$N$_3$), 40.97 (CH$_2$, *C*H$_2$NHCO), 37.36 (CH$_2$, HNCO*C*H$_2$), 29.47 (CH$_2$, *C*H$_2$CH$_2$N$_3$), 23.97 (CH$_2$, HNCOCH$_2$*C*H$_2$); ESI-TOF HR-MS: *m/z* 591.2768; calcd. for $C_{27}H_{39}N_6O_9$: 591.2773.

6-(5-Azidopentanamido)-*N*-{2-[2-(*N*-D-mannofuranosyl-*N*-methoxyamino)ethoxy]ethyl}-2-naphthamide (16a)

R_f 0.40 (CH$_2$Cl$_2$/MeOH 5:1); ^{1}H (600 MHz, MeOD): δ 8.32 (d, *J* = 0.9 Hz, 1H, Ar*H*), 8.29 (d, *J* = 1.8 Hz, 1H, Ar*H*), 7.93 (d, *J* = 9.0 Hz, 1H, Ar*H*), 7.87 (dd, *J* = 8.6, 1.7 Hz, 1H, Ar*H*), 7.84 (d, *J* = 8.7 Hz, 1H, Ar*H*), 7.64 (dd, *J* = 8.9, 2.1 Hz, 1H, Ar*H*), 4.59 (d, $J_{1,2}$ = 6.4 Hz, 1H, H-1), 4.30 (dd, $J_{2,3}$ = 4.6 Hz, 1H, H-2), 4.20 (dd, $J_{3,4}$ = 2.2 Hz, 1H, H-3), 3.89-3.88 (m, 2H, H-4, H-5), 3.76 - 3.73 (m, 1H, H-6a), 3.73-3.69 (m, 4H, C*H*$_2$OC*H*$_2$), 3.64 (t, *J* = 5.2 Hz, 2H, C*H*$_2$NHCO), 3.58 (s, 3H, OC*H*$_3$), 3.56-3.55 (m, 1H, H-6b), 3.37 (t, *J* = 6.8 Hz, 2H, C*H*$_2$N$_3$), 3.10 (td, *J* = 13.7, 5.6 Hz, 1H, CH$_3$ONC*H*$_2$), 3.04 (td, *J* = 13.7, 5.8 Hz, 1H, CH$_3$ONC*H*$_2$), 2.49 (t, *J* = 7.4 Hz, 2H, HNCOC*H*$_2$), 1.85-1.80 (m, 2H, HNCOCH$_2$C*H*$_2$), 1.72-1.67 (m, 2H, C*H*$_2$CH$_2$N$_3$); ^{13}C (151 MHz, MeOD): δ 174.37 (C$_q$, Ar*C*=O), 170.30 (C$_q$, ArNH*C*=O), 139.26 (C$_q$, Ar*C*), 136.84 (C$_q$, Ar*C*), 131.78 (C$_q$, Ar*C*), 130.96 (C$_q$, Ar*C*), 130.76 (CH, Ar*C*H), 128.88 (CH, Ar*C*H), 128.56 (CH, Ar*C*H), 125.45 (CH, Ar*C*H), 122.03 (CH, Ar*C*H), 117.20 (CH, Ar*C*H), 100.00 (CH, C-1), 81.84 (CH, C-4), 73.92 (CH, C-2), 73.17 (CH, C-3), 71.34 (CH, C-5), 70.52 (CH$_2$, CH$_2$O*C*H$_2$), 69.41 (CH$_2$, *C*H$_2$OCH$_2$), 64.97 (CH$_2$, C-6), 63.13 (CH$_3$, O*C*H$_3$), 54.17 (CH$_2$, CH$_3$ON*C*H$_2$), 52.19 (CH$_2$, *C*H$_2$N$_3$), 41.04 (CH$_2$, *C*H$_2$NHCO), 37.36 (CH$_2$, HNCO*C*H$_2$), 29.47 (CH$_2$, *C*H$_2$CH$_2$N$_3$), 23.97 (CH$_2$, HNCOCH$_2$*C*H$_2$); ESI-TOF HR-MS: *m/z* 591.2760; calcd. for $C_{27}H_{39}N_6O_9$: 591.2773.

6-(5-Azidopentanamido)-*N*-{2-[2-(*N*-α-D-mannopyranosyl-*N*-methoxy-amino)ethoxy]ethyl}-2-naphthamide (16b)

R_f 0.33 (CH$_2$Cl$_2$/MeOH 5:1); ^{1}H (600 MHz, MeOD): δ 8.33 (s, 1H, Ar*H*), 8.29 (d, *J* = 1.8 Hz, 1H, Ar*H*), 7.94 (d, *J* = 8.8 Hz, 1H, Ar*H*), 7.87 (dd, *J* = 8.6, 1.6 Hz, 1H, Ar*H*), 7.84 (d, *J* = 8.6 Hz, 1H, Ar*H*), 7.63 (dd, *J* = 8.8, 1.9 Hz, 1H, Ar*H*), 4.22 (d, $J_{1,2}$ = 1.3 Hz, 1H, H-1), 4.15 (dd, $J_{1,2}$ = 1.3 Hz, $J_{2,3}$ = 2.6 Hz, 1H, H-2), 3.87

(dd, $J_{3,4}$ = 8.9 Hz, 1H, H-3), 3.79 (d, J = 2.4 Hz, 1H, H-6a), 3.78-3.73 (m, 3H, CH_2OCH_2, H-5), 3.72-3.70 (m, 2H, CH_2OCH_2), 3.66-3.60 (m, 4H, CH_2NHCO, H-4, H-6b), 3.56 (s, 3H, OCH_3), 3.37 (t, J = 6.8 Hz, 2H, C$H_2$$N_3$), 3.02 (td, J = 14.1, 5.2 Hz, 1H, CH_3ONCH_2), 2.86 (td, J = 13.7, 5.4 Hz, 1H, CH_3ONCH_2), 2.48 (t, J = 7.4 Hz, 2H, HNCOCH_2), 1.84-1.79 (m, 2H, HNCOCH_2CH_2), 1.72-1.67 (m, 2H, CH_2C$H_2$$N_3$). ^{13}C (151 MHz, MeOD): δ 174.37 (C_q, Ar*C*=O), 170.33 (C_q, ArNH*C*=O), 139.27 (C_q, Ar*C*), 136.85 (C_q, Ar*C*), 131.74 (C_q, Ar*C*), 130.97 (C_q, Ar*C*), 130.81 (CH, Ar*C*H), 128.90 (CH, Ar*C*H), 128.58 (CH, Ar*C*H), 125.46 (CH, Ar*C*H), 122.01 (CH, Ar*C*H), 117.18 (CH, Ar*C*H), 94.47 (CH, C-1), 77.31 (CH, C-5), 73.15 (CH, C-3), 70.53 (CH, C-2), 70.38 (CH_2, CH_2O*C*H_2), 69.05 (CH_2, *C*H_2OCH_2), 68.92 (CH, C-4*),* 63.24 (CH_2, C-6), 62.71 (CH_3, O*C*H_3), 54.36 (CH_2, CH_3ON*C*H_2), 52.19 (CH_2, *C*$H_2$$N_3$), 41.10 ($CH_2$, *C*$H_2$NHCO), 37.37 ($CH_2$, HNCO*C*$H_2$), 29.48 ($CH_2$, *C*$H_2CH_2$$N_3$), 23.98 ($CH_2$, HNCOC$H_2$*C*$H_2$); ESI-TOF HR-MS: *m/z* 591.2775; calcd. for $C_{27}H_{39}N_6O_9$: 591.2773.

6-(5-Azidopentanamido)-*N*-{2-[2-(*N*-β-D-mannopyranosyl-*N*-methoxy-amino)ethoxy]ethyl}-2-naphthamide (16c)

R_f 0.26 (CH_2Cl_2/MeOH 5:1); ^{1}H (600 MHz, MeOD): δ 8.32 (s, 1H, Ar*H*), 8.29 (d, J = 1.5 Hz, 1H, Ar*H*), 7.93 (d, J = 8.8 Hz, 1H, Ar*H*), 7.87 (d, J = 8.6 Hz, 1H, Ar*H*), 7.84 (d, J = 8.9 Hz, 1H, Ar*H*), 7.64 (d, J = 8.8 Hz, 1H, Ar*H*), 4.10 (d, $J_{1,2}$ = 0.7 Hz, 1H, H-1), 4.02 (d, $J_{1,2}$ = 0.5 Hz, $J_{2,3}$ = 3.2 Hz, 1H, H-2), 3.89-3.86 (m, 1H, H-6a), 3.78-3.72 (m, 2H, CH_2OCH_2), 3.72-3.67 (m, 3H, H-6b, CH_2OCH_2), 3.66-3.61 (m, 3H, H-4, CH_2NHCO), 3.59 (s, 3H, OCH_3), 3.44 (dd, $J_{3,4}$ = 9.3 Hz, 1H, H-3), 3.25-3.22 (m, 1H, H-5), 3.36 (t, J = 6.8 Hz, 2H, C$H_2$$N_3$), 2.48 (t, J = 7.4 Hz, 2H, HNCOCH_2), 1.84-1.79 (m, 2H, HNCOCH_2CH_2), 1.72-1.67 (m, 2H, CH_2C$H_2$$N_3$); ^{13}C (151 MHz, MeOD): δ 174.35 (C_q, Ar*C*=O), 170.31 (C_q, ArNH*C*=O), 139.26 (C_q, Ar*C*), 136.84 (C_q, Ar*C*), 131.73 (C_q, Ar*C*), 130.96 (C_q, Ar*C*), 130.78 (CH, Ar*C*H), 128.88 (CH, Ar*C*H), 128.59 (CH, Ar*C*H), 125.45 (CH, Ar*C*H), 122.03 (CH, Ar*C*H), 117.15 (CH, Ar*C*H), 93.81 (CH, C-1), 80.61 (CH, C-5), 76.23 (CH, C-3), 71.59 (CH, C-2), 70.42 (CH_2, CH_2O*C*H_2), 69.33 (CH_2, *C*H_2OCH_2), 68.60 (CH, C-4), 63.01 (CH_2, C-6), 62.60 (CH_3, O*C*H_3), 54.45 (CH_2, CH_3ON*C*H_2), 52.18 (CH_2, *C*$H_2$$N_3$), 41.04 ($CH_2$, *C*$H_2$NHCO), 37.36 ($CH_2$, HNCO*C*$H_2$), 29.47 ($CH_2$, *C*$H_2CH_2$$N_3$), 23.98 ($CH_2$, HNCOC$H_2$*C*$H_2$).

6-(5-Azidopentanamido)-*N*-{2-[2-(*N*-2-acetamido-2-deoxy-β-D-galactofuranosyl-*N*-methoxyamino)ethoxy]ethyl}-2-naphthamide (17a)

R_f 0.27 (CH_2Cl_2/MeOH 5:1); ^{1}H NMR (600 MHz, MeOD): δ 8.32 (d, J = 0.8 Hz, 1H, Ar*H*), 8.29 (d, J = 1.8 Hz, 1H, Ar*H*), 7.93 (d, J = 9.0 Hz, 1H, Ar*H*), 7.87 (dd, J = 8.6, 1.7 Hz, 1H, Ar*H*), 7.84 (d, J = 8.8 Hz, 1H, Ar*H*), 7.64 (dd, J = 8.8, 2.1 Hz, 1H, Ar*H*), 4.60 (dd, $J_{1,2}$ = 6.8 Hz, $J_{2,3}$ = 8.0 Hz, 1H, H-2), 4.49 (d, $J_{1,2}$ = 6.7 Hz, 1H, H-1), 4.08 (app t, $J_{3,4}$ = 8.2 Hz, 1H, H-3), 3.90 (dd, $J_{4,5}$ = 2.5 Hz, 1H, H-4), 3.72 - 3.67 (m, 4H, CH_2OCH_2), 3.65-3.63 (m, 3H, H-5, CH_2NHCO), 3.62 (s, 3H, OCH_3), 3.60-3.59 (m, 2H, H-6a, H-6b), 3.37 (t, J = 6.8 Hz, 2H, C$H_2$$N_3$), 3.12 (dt, J = 13.6, 5.5 Hz, 1H, CH_3ONCH_2), 3.01 (dt, J = 13.6, 5.9 Hz, 1H, CH_3ONCH_2), 2.48 (t, J = 7.4 Hz, 2H, NHCOCH_2), 1.94 (s, 3H, CH_3CO) 1.84-1.79 (m, 2H, NHCOCH_2CH_2), 1.74-1.67 (m, 2H, CH_2C$H_2$$N_3$); ^{13}C NMR (151 MHz, MeOD): δ 174.38 (C_q, Ar*C*=O), 173.37 (C_q, CH_3*C*=O),

170.29 (C_q, ArNH*C*=O), 139.29 (C_q, Ar*C*), 136.86 (C_q, Ar*C*), 131.78 (C_q, Ar*C*), 130.96 (C_q, Ar*C*), 130.78 (CH, Ar*C*H), 128.91 (CH, Ar*C*H), 128.57 (CH, Ar*C*H), 125.46 (CH, Ar*C*H), 122.05 (CH, Ar*C*H), 117.20 (CH, Ar*C*H), 97.77 (CH, C-1), 83.49 (CH, C-4), 76.20 (CH, C-3), 71.93 (CH, C-5), 70.52 (CH_2, CH_2OCH_2), 69.48 (CH_2, CH_2OCH_2), 64.64 (CH_2, C-6), 63.18 (CH_3, OCH_3), 58.67 (CH, C-2), 54.05 (CH_2, CH_3ONCH_2), 52.20 (CH_2, CH_2N_3), 41.02 (CH_2, CH_2NHCO), 37.38 (CH_2, $NHCOCH_2$), 29.49 (CH_2, $CH_2CH_2N_3$), 23.99 (CH_2, $NHCOCH_2CH_2$), 22.84 (CH_3, CH_3CO); ESI-TOF HR-MS: *m/z* 632.3039; calcd. for $C_{29}H_{42}N_7O_9$: 632.3039.

6-(5-Azidopentanamido)-*N*-{2-[2-(*N*-2-acetamido-2-deoxy-β-D-galactopyranosyl-*N*-methoxyamino)ethoxy]ethyl}-2-naphthamide (17b)

R_f 0.17 (CH_2Cl_2/MeOH 5:1); ^{1}H NMR (600 MHz, MeOD): δ 8.33 (d, *J* = 0.8 Hz, 1H, Ar*H*), 8.29 (d, *J* = 1.8 Hz, 1H, Ar*H*), 7.93 (d, *J* = 8.9 Hz, 1H, Ar*H*), 7.88 (dd, *J* = 8.6, 1.8 Hz, 1H, Ar*H*), 7.84 (d, *J* = 8.6 Hz, 1H, Ar*H*), 7.64 (dd, *J* = 8.9, 2.1 Hz, 1H, Ar*H*), 4.22 (d, $J_{1,2}$ = 9.8 Hz, 1H, H-1), 4.06 (app t, $J_{2,3}$ = 10.1 Hz, 1H, H-2), 3.78 (dd, $J_{3,4}$ = 3.2 Hz, $J_{4,5}$ = 0.5 Hz, 1H, H-4), 3.75 (dd, $J_{5,6a}$ = 6.8 Hz, $J_{6a,6b}$ = 11.4, 1H, H-6a), 3.71-3.64 (m, 5H, H-6b, CH_2OCH_2), 3.62-3.58 (m, 2H, CH_2NHCO), 3.52 (dd, 1H, H-3), 3.48 (s, 3H, OCH_3), 3.43-3.41 (m, 1H, H-5), 3.37 (t, *J* = 6.8 Hz, 2H, CH_2N_3), 3.24 (dt, *J* = 13.7, 6.0 Hz, 1H, CH_3ONCH_2), 3.14 (dt, *J* = 13.8, 6.0 Hz, 1H, CH_3ONCH_2), 2.49 (t, *J* = 7.41 Hz, 2H, $NHCOCH_2$), 1.95 (s, 3H, CH_3CO), 1.84-1.79 (m, 2H, $NHCOCH_2CH_2$), 1.72-1.67 (m, 2H, $CH_2CH_2N_3$);^{13}C NMR (151 MHz, MeOD): δ 174.38 (C_q, Ar*C*=O), 173.83 (C_q, CH_3C=O), 170.30 (C_q, ArNH*C*=O), 139.31 (C_q, Ar*C*), 136.86 (C_q, Ar*C*), 131.79 (C_q, Ar*C*), 130.96 (C_q, Ar*C*), 130.77 (CH, Ar*C*H), 128.90 (CH, Ar*C*H), 128.60 (CH, Ar*C*H), 125.48 (CH, Ar*C*H), 122.05 (CH, Ar*C*H), 117.19 (CH, Ar*C*H), 93.42 (CH, C-1), 78.40 (CH, C-5), 74.74 (CH, C-3), 70.36 (CH_2, CH_2OCH_2), 70.07 (CH_2, CH_2OCH_2) , 69.89 (CH, C-4), 62.71 (CH_2, C-6), 61.94 (CH_3, OCH_3), 52.20 (CH_2, CH_2N_3), 52.05 (CH_2, CH_3ONCH_2), 50.94 (CH, C-2), 41.10 (CH_2, CH_2NHCO), 37.38 (CH_2, $NHCOCH_2$), 29.49 (CH_2, $CH_2CH_2N_3$), 23.99 (CH_2, $NHCOCH_2CH_2$), 22.99 (CH_3, CH_3CO); ESI-TOF HR-MS: *m/z* 632.3043; calcd. for $C_{29}H_{42}N_7O_9$: 632.3039.

6-(5-Azidopentanamido)-*N*-{2-[2-(*N*-β-L-fucopyranosyl-*N*-methoxyamino)ethoxy]ethyl}-2-naphthamide (18)

R_f 0.22 (CH_2Cl_2/MeOH 10:1); ^{1}H NMR (300 MHz, MeOD): δ 8.34 (d, *J* = 0.9 Hz, 1H, Ar*H*), 8.31 (d, *J* = 2.0 Hz, 1H, Ar*H*), 7.94 (d, *J* = 9.2 Hz, 1H, Ar*H*), 7.88 (dd, *J* = 8.6, 1.6 Hz, 1H, Ar*H*), 7.84 (d, *J* = 8.8 Hz, 1H, Ar*H*), 7.64 (dd, *J* = 8.8, 2.1 Hz, 1H, Ar*H*), 4.01 (d, *J* = 9.0 Hz, 1H, H-1), 3.75-3.71 (m, 4H, CH_2OCH_2), 3.67-3.63 (m, 3H, H-3, CH_2NHCO), 3.61 (d, $J_{1,2}$= 9.0 Hz, 1H, H-2), 3.56 (s, 3H, OCH_3), 3.54 (m, 1H, H-5), 3.46 (dd, *J* = 9.3, 3.4 Hz, 1H, H-4), 3.37 (t, *J* = 6.7 Hz, 2H, CH_2N_3), 3.21 (dt, *J* = 14.1, 5.7 Hz, 1H, CH_3ONCH_2), 3.07 (dt, *J* = 14.1, 5.4 Hz, 1H, CH_3ONCH_2), 2.49 (t, *J* = 7.3 Hz, 2H, $HNCOCH_2$), 1.87-1.78 (m, 2H, $HNCOCH_2CH_2$), 1.74-1.67 (m, 2H, $CH_2CH_2N_3$), 1.22 (d, *J* = 6.5 Hz, 3H, H-6); ^{13}C NMR (75 MHz, MeOD): δ 130.31 (CH, Ar*C*H), 128.29 (CH, Ar*C*H), 127.89 (CH, Ar*C*H), 126.46 (CH, Ar*C*H), 121.95 (CH, Ar*C*H), 116.12 (CH, Ar*C*H), 95.46 (CH, C-1), 76.77 (CH, C-4), 73.17 (CH, C-5), 73.15 (CH, C-3), 70.12

(CH_2, CH_2OCH_2), 67.94 (CH, C-2), 62.38 (CH_3, OCH_3), 52.78 (CH_2, CH_3ONCH_2), 52.24 (CH_2, CH_2N_3), 40.90 (CH_2, CH_2NHCO), 37.41 (CH_2, $HNCOCH_2$), 29.71 (CH_2, $CH_2CH_2N_3$), 24.49 (CH_2, $HNCOCH_2CH_2$), 16.66 (CH_3, C-6); ESI-TOF HR-MS: *m/z* 575.2833; calcd. for $C_{27}H_{39}N_6O_8$: 575.2824.

6-(5-Azidopentanamido)-*N*-{2-[2-(*N*-β-D-lactopyranosyl-*N*-methoxyamino)ethoxy]ethyl}-2-naphthamide (19)

R_f 0.13 ($CHCl_3$/MeOH 3:1); ^{1}H NMR (600 MHz, MeOD): δ 8.34 (d, *J* = 0.9 Hz, 1H, Ar*H*), 8.30 (d, *J* = 1.9 Hz, 1H, Ar*H*), 7.94 (d, *J* = 8.9 Hz, 1H, Ar*H*), 7.88 (dd, *J* = 8.6, 1.7 Hz, 1H, Ar*H*), 7.85 (d, *J* = 8.7 Hz, 1H, Ar*H*), 7.65 (dd, *J* = 8.9, 2.1 Hz, 1H, Ar*H*), 4.35 (d, $J_{1',2'}$ = 7.6 Hz, 1H, H-1´), 4.13 (d, $J_{1,2}$ = 8.6 Hz, 1H, H-1), 3.86 (dd, *J* = 12.3, 2.5 Hz, 1H, H-6a), 3.83-3.82 (m, 2H, H-4´, H-6b), 3.79 (dd, *J* = 11.5, 7.4 Hz, 1H, H-6´a), 3.75-3.69 (m, 5H, H-6´b, CH_2OCH_2), 3.66-3.64 (m, 2H, CH_2NHCO), 3.60 (s, 3H, OCH_3), 3.59-3.51 (m, 5H, H-2, H-3, H-4, H-2´, H-5'), 3.49 (dd, *J* = 9.7, 3.3 Hz, 1H, H-3´), 3.38-3.36 (m, 3H, CH_2N_3, H-5), 3.23 (dt, *J* = 13.8, 5.4 Hz, 1H, CH_3ONCH_2), 3.14 (dt, *J* = 13.8, 5.7 Hz, 1H, CH_3ONCH_2), 2.49 (t, *J* = 7.4 Hz, 2H, $NHCOCH_2$), 1.85-1.80 (m, 2H, $NHCOCH_2CH_2$), 1.70 (m, 2H, $CH_2CH_2N_3$); ^{13}C NMR (151 MHz, MeOD): δ 174.42 (C_q, Ar*C*=O), 170.34 (C_q, ArHN*C*=O), 139.30 (C_q, Ar*C*), 136.87 (C_q, Ar*C*), 131.72 (C_q, Ar*C*), 130.97 (C_q, Ar*C*), 130.81 (CH, Ar*C*H), 128.93 (CH, Ar*C*H), 128.60 (CH, Ar*C*H), 125.46 (CH, Ar*C*H), 122.08 (CH, Ar*C*H), 117.22 (CH, Ar*C*H), 105.11 (CH, C-1´), 94.55 (CH, C-1), 80.32 (CH, C-4), 78.19 (CH, C-5), 77.57 (CH, C-3), 77.07 (CH, C-5'), 74.83 (CH, C-3´), 72.56 (CH, C-2´), 71.25 (CH, C-2), 70.54 (CH_2, CH_2OCH_2), 70.35 (CH, C-4´), 69.67 (CH_2, CH_2OCH_2), 62.78 (CH_3, OCH_3), 62.54 (CH_2, C-6´), 62.01 (CH_2, C-6), 53.23 (CH_2, CH_3ONCH_2), 52.20 (CH_2, CH_2N_3), 40.98 (CH_2, CH_2NHCO), 37.38 (CH_2, $HNCOCH_2$), 29.48 (CH_2, $CH_2CH_2N_3$), 23.99 (CH_2, $HNCOCH_2CH_2$); ESI-TOF HR-MS: *m/z* 753.3299; calcd. for $C_{33}H_{49}N_6O_{14}$: 753.3301.

6-(5-Azidopentanamido)-*N*-{2-[2-(*N*-2-acetamido-2-deoxy-β-D-lactopyranosyl-*N*-methoxyamino) ethoxy]ethyl}-2-naphthamide (20)

R_f 0.13 ($CHCl_3$/MeOH 3:1); ^{1}H NMR (600 MHz, MeOD): δ 8.34 (d, *J* = 0.5 Hz, 1H, Ar*H*), 8.30 (d, *J* = 1.6 Hz, 1H, Ar*H*), 7.93 (d, *J* = 8.9 Hz, 1H, Ar*H*), 7.88 (dd, *J* = 8.6, 1.7 Hz, 1H, Ar*H*), 7.85 (d, *J* = 8.6 Hz, 1H, Ar*H*), 7.64 (dd, *J* = 8.8, 2.0 Hz, 1H, Ar*H*), 4.36 (d, $J_{1',2'}$ = 7.6 Hz, 1H, H-1´), 4.30 (d, $J_{1,2}$ = 9.8 Hz, 1H, H-1), 3.91 (d, *J* = 9.7 Hz, 1H, H-2), 3.87 (dd, *J* = 12.3, 2.3 Hz, 1H, H-6a), 3.83-3.80 (m, 2H, H-4´, H-6b), 3.76 (dd, *J* = 11.5, 7.4 Hz, 1H, H-6´a), 3.73-3.64 (m, 6H, CH_2OCH_2, CH_2NHCO, H-6´b), 3.61 (t, *J* = 6.0 Hz, 1H, CH_2NHCO), 3.59-3.56 (m, 3H, H-3, H-4, H-5´), 3.53 (dd, $J_{2',3'}$ = 9.8 Hz, 1H, H-2´), 3.49 (s, 3H, OCH_3), 3.48 (dd, $J_{3',4'}$= 3.3 Hz, 1H, H-3´), 3.37 (t, *J* = 6.8 Hz, 2H, CH_2N_3), 3.23-3.10 (m, 2H, CH_3ONCH_2), 2.49 (t, *J* = 7.4 Hz, 2H, $HNCOCH_2$), 1.94 (s, 3H, CH_3CO), 1.84-1.80 (m, 2H, $HNCOCH_2CH_2$), 1.72-1.67 (m, 2H, $CH_2CH_2N_3$); ^{13}C NMR (151 MHz, MeOD): δ 174.40 (C_q, Ar*C*=O), 173.16 (C_q, CH_3C=O), 170.31 (C_q, ArNH*C*=O), 139.30 (C_q, Ar*C*), 136.87 (C_q, Ar*C*), 131.76 (C_q, Ar*C*), 130.98 (C_q, Ar*C*), 130.80 (CH, Ar*C*H), 128.93 (CH, Ar*C*H), 128.61 (CH, Ar*C*H), 125.47 (CH, Ar*C*H), 122.07 (CH, Ar*C*H), 117.22 (CH, Ar*C*H), 105.07 (CH, C-1´), 92.92 (CH, C-1), 80.66 (CH, C-4), 78.32 (CH, C-5), 77.17 (CH, C-5´), 75.64 (CH, C-3),

74.85 (CH, C-3´), 72.60 (CH, C-2´), 70.40 (CH_2, CH_2OCH_2), 70.36 (CH, C-4´), 70.01 (CH_2, CH_2OCH_2), 62.55 (CH_2, C-6´), 62.11 (CH_3, OCH_3), 62.01 (CH_2, C-6), 53.49 (CH, C-2), 52.26 (CH_2, CH_3ONHCH_2), 52.21 (CH_2, CH_2N_3), 41.08 (CH_2, CH_2NHCO), 37.38 (CH_2, $HNCOCH_2$), 29.49 (CH_2, $CH_2CH_2N_3$), 23.99 (CH_2, $HNCOCH_2CH_2$), 22.98 (CH_3, CH_3CO); ESI-TOF HR-MS: *m/z* 794.3565; calcd. for $C_{35}H_{52}N_7O_{14}$: 794.3567.

6-(5-Azidopentanamido)-*N*-{2-[2-(*N*-β-D-chitobiopyranosyl-*N*-methoxyamino)ethoxy]ethyl}-2-naphthamide (21)

R_f 0.13 ($CHCl_3$/MeOH 3:1); ^{1}H NMR (600 MHz, MeOD) δ 8.33 (s, 1H, Ar*H*), 8.31 (d, *J* = 1.3 Hz, 1H, Ar*H*), 7.94 (d, *J* = 8.9 Hz, 1H, Ar*H*), 7.88 (dd,*J* = 8.6, 1.7 Hz, 1H, Ar*H*), 7.85 (d,*J* = 8.6 Hz, 1H, Ar*H*), 7.65 (dd, *J* = 8.9, 2.0 Hz, 1H, Ar*H*), 4.46 (d, $J_{1´,2´}$ = 8.5 Hz, 1H, H-1´), 4.24 (d, $J_{1,2}$ = 9.9 Hz, 1H, H-1), 3.89 (dd, $J_{5,6a}$ = 2.2 Hz, $J_{6a,6b}$ = 11.9 Hz, 1H, H-6a), 3.89 (t, $J_{2,3}$ = 9.8 Hz 1H, H-2), 3.77 (dd, $J_{5,6b}$ = 1.6 Hz, $J_{6a,6b}$ = 12.1 Hz, 1H, H-6b), 3.71-3.66 (m, 3H, $(CH_3)ONCH_2CH_2O$,H-2´), 3.64-3.56 [m, 6H,$OCH_2CH_2NHC(O)$, H-6a´, H-6b´], 3.52 (t, $J_{2,3}$ = $J_{3,4}$= 9.1 Hz, 1H, H-3), 3.49 (s, 3H, OCH_3), 3.47 (d, $J_{4,3}$ = 8.6 Hz, 1H, H-4), 3.43 (dd, $J_{2´,3´}$ = 8.5 Hz, $J_{3´,4´}$ = 10.3 Hz, 1H, H-3´), 3.37 (t, *J* = 6.7 Hz, 2H, CH_2N_3), 3.35-3.33 (m, 1H, H-4´), 3.28 (d, $J_{5´,6´}$= 10.0 Hz, 1H, H-5´), 3.25-3.18 (m, 1H, H-5), 3.15 [dt, *J* = 13.6, 6.0 Hz, 1H, $(CH_3)ONCH_2$], 2.49 [t, *J* = 7.4 Hz, 2H, $HNC(O)CH_2$], 2.00 (s, 3H, CH_3CO), 1.93 (s, 3H, NHAc), 1.85-1.79 [m, 2H, $HNC(O)CH_2CH_2$] , 1.72-1.67 (m, 2H, $CH_2CH_2NH_2$); ^{13}C NMR (151 MHz, MeOD) δ 130.82 (CH, Ar*C*H), 128.97 (CH, Ar*C*H), 128.61 (CH, Ar*C*H), 125.46 (CH, Ar*C*H), 122.07 (CH, Ar*C*H), 117.23 (CH, Ar*C*H), 103.22 (CH, C-1´), 92.85 (CH, C-1), 81.21 (CH, C-4), 78.21 (CH, C-4´), 78.16 (CH, C-5), 75.91 (CH, C-3´), 75.66 (CH, C-3), 71.96 (CH, C-5´), 70.43 (CH_2, CH_2OCH_2), 69.97 (CH_2, CH_2OCH_2), 62.56 (CH_2, C-6´), 62.07 (CH_3, OCH_3), 61.86 (CH_2, C-6), 57.31 (CH, C-2´), 53.33 (CH, C-2), 52.39 [CH_2, $(CH_3)ONCH_2$], 52.21 (CH_2, CH_2N_3), 41.03 (CH_2, $CH_2NHC=O$), 37.39 [CH_2, $HNC(O)CH_2$], 29.49 (CH_2, $CH_2CH_2N_3$), 24.19 (CH_3, CH_3CO), 23.99 [CH_2, $HNC(O)CH_2CH_2$], 22.98 (CH_3, CH_3CO); ESI-TOF HR-MS: *m/z* 835.3820; calcd. for $C_{37}H_{55}N_8O_{14}$: 835.3832.

6-(5-Aminopentanamido)-*N*-{2-[2-(*N*-2-acetamido-2-deoxy-β-D-glucopyranosyl-*N*-methoxyamino) ethoxy]ethyl}-2-naphthamide (22)

R_f 0.38 ($CHCl_3$/MeOH/H_2O 10:10:3); ^{1}H NMR (600 MHz, MeOD): δ 8.34 (d, *J* = 1.6 Hz, 1H, Ar*H*), 8.30 (d, *J* = 1.9 Hz, 1H, Ar*H*), 7.94 (d, *J* = 8.9 Hz, 1H, Ar*H*), 7.88 (dd, *J* = 8.6, 1.8 Hz, 1H, Ar*H*), 7.84 (d, *J* = 8.6 Hz, 1H, Ar*H*), 7.64 (dd, *J* = 8.9, 2.1 Hz, 1H, Ar*H*), 4.27 (d, $J_{1,2}$ = 9.8 Hz, 1H, H-1), 3.83 (dd, $J_{5,6a}$ = 2.3 Hz, $J_{6a,6b}$ = 12.1 Hz, 1H, H-6a), 3.81 (t, $J_{2,3}$ = 9.9 Hz, 1H, H-2), 3.71-3.59 (m, 7H, CH_2OCH_2, CH_2NHCO, H-6b), 3.49 (s, 3H, OCH_3), 3.42 (dd, $J_{3,4}$ = 8.8 Hz, 1H, H-3), 3.28 (t, $J_{4,5}$ = 8.8 Hz, 1H, H-4), 3.23-3.19 (m, 2H, CH_3ONCH_2, H-5), 3.14 (dt, *J* = 13.7, 6.0 Hz, 1H, CH_3ONCH_2), 2.91 (t, *J* = 7.4 Hz, 2H, CH_2NH_2), 2.52 (t, *J* = 7.1 Hz, 2H, $HNCOCH_2$), 1.94 (s, 3H, CH_3CO), 1.84-1.79 (m, 2H, $HNCOCH_2CH_2$), 1.75-1.69 (m, 2H, $CH_2CH_2NH_2$); ^{13}C NMR (151 MHz, MeOD): δ 139.27 (C_q, Ar*C*), 136.84 (C_q, Ar*C*), 131.82 (C_q, Ar*C*), 131.00 (C_q, Ar*C*) 130.82 (CH, Ar*C*H), 128.87 (CH, Ar*C*H), 128.61 (CH, Ar*C*H), 125.53 (CH, Ar*C*H), 121.97

(CH, Ar*C*H), 117.17 (CH, Ar*C*H), 92.97 (CH, C-1), 79.76 (CH, C-5), 77.63 (CH, C-3), 71.89 (CH, C-4), 70.40 (CH_2, CH_2OCH_2), 69.96 (CH_2, CH_2OCH_2), 62.95 (CH_2, C-6), 62.09 (CH_3, OCH_3), 54.16 (CH, C-2), 52.32 (CH_2, CH_3ONCH_2), 41.09 (CH_2, CH_2NHCO), 40.93 (CH_2, CH_2NH_2), 37.14 (CH_2, $HNCOCH_2$), 29.51 (CH_2, $CH_2CH_2NH_2$), 24.12 (CH_2, $HNCOCH_2CH_2$), 22.97 (CH_3, CH_3CO); ESI-TOF HR-MS: *m/z* 606.3148; calcd. for $C_{29}H_{44}N_5O_9$: 606.3120.

6-(5-Aminopentanamido)-*N*-{2-[2-(*N*-β-D-galactofuranosyl-*N*-methoxyamino)ethoxy]ethyl}-2-naphthamide (23)

R_f 0.05 ($CHCl_3$/MeOH/H_2O 10:10:3); ^{1}H NMR (600 MHz, MeOD): δ 8.32 (d, *J* = 1.0 Hz, 1H, Ar*H*), 8.30 (d, *J* = 1.7 Hz, 1H, Ar*H*), 7.93 (d, *J* = 8.9 Hz, 1H, Ar*H*), 7.87 (dd, *J* = 8.6, 1.7 Hz, 1H, Ar*H*), 7.84 (d, *J* = 8.7 Hz, 1H, Ar*H*), 7.64 (dd, *J* = 8.9, 2.1 Hz, 1H, Ar*H*), 4.49 (d, $J_{1,2}$ = 5.6 Hz, 1H, H-1), 4.20 (dd, $J_{2,3}$ = 6.8 Hz, 1H, H-2), 4.08 (dd, $J_{3,4}$ = 8.3 Hz, 1H, H-3), 3.84 (dd, $J_{4,5}$ = 2.7 Hz, 1H, H-4), 3.72 (t, *J* = 5.8 Hz, 2H, CH_2OCH_2), 3.71 (t, *J* = 5.9 Hz, 2H, CH_2OCH_2), 3.65-3.63 (m, 3H, CH_2NHCO, H-5), 3.59 (s, 3H, OCH_3), 3.13 (dt, *J* = 13.7, 5.6 Hz, 1H, CH_3ONCH_2), 3.04 (dt, *J* = 13.7, 5.7 Hz, 1H, CH_3ONCH_2), 2.78 (t, *J* = 7.3 Hz, 1H, CH_2NH_2), 2.49 (t, *J* = 7.4 Hz, 2H, $HNCOCH_2$), 2.19 (t, *J* = 7.6 Hz, 1H, CH_2NH_2), 1.82-1.76 (m, 2H, $HNCOCH_2CH_2$), 1.66-1.59 (m, 2H, $CH_2CH_2NH_2$); ^{13}C NMR (151 MHz, MeOD): δ 174.59 (C_q, Ar*C*=O), 170.31 (C_q, ArHN*C*=O), 139.42 (C_q, Ar*C*), 136.85 (C_q, Ar*C*), 131.81 (C_q, Ar*C*), 130.97 (C_q, Ar*C*), 130.79 (CH, Ar*C*H), 128.88 (CH, Ar*C*H), 128.58 (CH, Ar*C*H), 125.49 (CH, Ar*C*H), 122.02 (CH, Ar*C*H), 117.18 (CH, Ar*C*H), 99.90 (CH, C-1), 83.23 (CH, C-4), 78.60 (CH, C-2), 77.49 (CH, C-3), 72.15 (CH, C-5), 70.52 (CH_2, CH_2OCH_2), 69.44 (CH_2, CH_2OCH_2), 64.60 (CH_2, C-6), 63.06 (CH_3, OCH_3), 53.91 (CH_2, CH_3ONCH_2), 41.72 (CH_2, CH_2NH_2), 41.04 (CH_2, CH_2NHCO), 37.55 (CH_2, $NHCOCH_2$), 31.47 (CH_2, $CH_2CH_2NH_2$), 23.89 (CH_2, $NHCOCH_2CH_2$); ESI-TOF HR-MS: *m/z* 565.2870; calcd for $C_{27}H_{41}N_4O_9$: 565.2868.

6-(5-Aminopentanamido)-*N*-{2-[2-(*N*-β-D-galactopyranosyl-*N*-methoxyamino)ethoxy]ethyl}-2-naphthamide (24)

R_f 0.06 ($CHCl_3$/MeOH/H_2O 10:10:3); ^{1}H-NMR (600 MHz, MeOD): δ 8.33 (d, *J* = 0.9 Hz, 1H, Ar*H*), 8.30 (d, *J* = 1.7 Hz, 1H, Ar*H*), 7.94 (d, *J* = 9.0 Hz, 1H, Ar*H*), 7.88 (dd, *J* = 8.6, 1.8 Hz, 1H, Ar*H*), 7.84 (d, *J* = 8.6 Hz, 1H, Ar*H*), 7.64 (dd, *J* = 8.8, 2.1 Hz, 1H, Ar*H*), 4.06 (d, $J_{1,2}$ = 9.1 Hz, 1H, H-1), 3.79 (dd, $J_{3,4}$ = 3.4 Hz, $J_{4,5}$ = 0.8 Hz, 1H, H-4), 3.75-3.72 (m, 6H, CH_2OCH_2, H-6a, H-6b), 3.68 (t, $J_{1,2}$ = $J_{2,3}$= 9.3 Hz, 1H, H-2), 3.68-3.65 (m, 2H, CH_2NHCO), 3.57 (s, 3H, OCH_3), 3.47 (dd, $J_{2,3}$ = 9.3 Hz, $J_{3,4}$= 3.4 Hz, 1H, H-3), 3.44 (dd, $J_{5,6}$ = 5.3 Hz, $J_{4,5}$ = 1.0 Hz, 1H, H-5), 3.26-3.22 (dt, *J* = 14.0, 5.8 Hz, 1H, CH_3ONCH_2), 3.14-3.10 (dt, *J* = 14.0, 5.4 Hz, 1H, CH_3ONCH_2), 2.80 (t, *J* = 7.3 Hz, 2H, CH_2NH_2), 2.49 (t, *J* = 7.3 Hz, 2H, $HNCOCH_2$), 1.82-1.67 (m, 2H, $HNCOCH_2CH_2$), 1.67-1.61 (m, 2H, $CH_2CH_2NH_2$); ^{13}C-NMR (151 MHz, MeOD): δ 174.48 (C_q, Ar*C*=O), 170.31 (C_q, ArNH*C*=O), 139.31 (C_q, Ar*C*), 136.85 (C_q, Ar*C*), 131.79 (C_q, Ar*C*), 130.97 (C_q, Ar*C*), 130.80 (CH, Ar*C*H), 128.88 (CH, Ar*C*H), 128.59 (CH, Ar*C*H), 125.49 (CH, Ar*C*H), 122.02 (CH, Ar*C*H), 117.17 (CH, Ar*C*H), 95.39 (CH, C-1), 78.40 (CH, C-5), 76.23 (CH, C-3), 70.53 (CH, C-4), 70.50

(CH_2, CH_2OCH_2), 69.72 (CH_2, CH_2OCH_2), 69.00 (CH, C-2), 62.67 (CH_2, C-6), 62.43 (CH_3, OCH_3), 52.58 (CH_2, CH_3ONCH_2), 41.57 (CH_2, CH_2NH_2), 40.98 (CH_2, CH_2NHCO), 37.47 (CH_2, $NHCOCH_2$), 31.47 (CH_2, $CH_2CH_2NH_2$), 23.81 (CH_2, $NHCOCH_2CH_2$); ESI-TOF HR-MS: *m/z* 565.2866; calcd. for $C_{27}H_{41}N_4O_9$: 565.2868.

6-(5-Aminopentanamido)-*N*-{2-[2-(*N*-β-D-glucopyranosyl-*N*-methoxyamino)ethoxy]ethyl}-2-naphthamide (25)

R_f 0.15 ($CHCl_3$/MeOH/H_2O 10:10:2); ^{1}H NMR (600 MHz, MeOD): δ 8.34 (d, *J* = 1.1 Hz, 1H, Ar*H*), 8.30 (d, *J* = 1.9 Hz, 1H, Ar*H*), 7.95 (d, *J* = 9.1 Hz, 1H, Ar*H*), 7.88 (dd, *J* = 8.6, 1.8 Hz, 1H, Ar*H*), 7.84 (d, *J* = 8.8 Hz, 1H, Ar*H*), 7.64 (dd, *J* = 8.9, 2.1 Hz, 1H, Ar*H*), 4.09 (d, $J_{1,2}$ = 8.8 Hz, 1H, H-1), 3.82 (dd, $J_{5,6a}$ = 2.2 Hz, $J_{6a,6b}$ = 12.1 Hz, 1H, H-6a), 3.74 (t, *J* = 5.7 Hz, 2H, CH_2OCH_2), 3.72 (dd, *J* = 5.3, 1.4 Hz, 2H, CH_2OCH_2), 3.66-3.63 (m, 3H, CH_2NHCO, H-6b), 3.59 (s, 3H, OCH_3), 3.42 (t, $J_{2,3}$ = 8.9 Hz, 1H, H-2), 3.37 (app t, $J_{3,4}$ = 8.8 Hz, 1H, H-3), 3.27 (dd, $J_{4,5}$ = 9.8 Hz, 1H, H-4), 3.24-3.20 (m, 1H, CH_3ONCH_2), 3.20-3.19 (m, 1H, H-5), 3.13 (dt, *J* = 14.0, 5.7 Hz, 1H, CH_3ONCH_2), 2.98 (t, *J* = 7.3 Hz, 2H, CH_2NH_2), 2.54 (t, *J* = 7.0 Hz, 2H, $HNCOCH_2$), 1.85-1.80 (m, 2H, $HNCOCH_2CH_2$), 1.79-1.74 (m, 2H, $CH_2CH_2NH_2$); ^{13}C NMR (151 MHz, MeOD): δ 130.42 (CH, Ar*C*H), 128.58 (CH, Ar*C*H), 128.37 (CH, Ar*C*H), 124.89 (CH, Ar*C*H), 121.20 (CH, Ar*C*H), 116.70 (CH, Ar*C*H), 94.38 (CH, C-1), 79.23 (CH, C-5), 78.82 (CH, C-3), 71.04 (CH, C-2, C-4), 69.89 (CH_2, CH_2OCH_2), 68.89 (CH_2, CH_2OCH_2), 63.10 (CH_2, C-6), 62.44 (CH_3, OCH_3), 53.00 (CH_2, CH_3ONCH_2), 40.57 (CH_2, CH_2NHCO), 40.19 (CH_2, CH_2NH_2), 36.7 (CH_2, $NHCOCH_2$), 31.47 (CH_2, $CH_2CH_2NH_2$), 23.04 (CH_2, $NHCOCH_2CH_2$); ESI-TOF HR-MS: *m/z* 565.2866; calcd. for $C_{27}H_{41}N_4O_9$: 565.2868.

6-(5-Aminopentanamido)-*N*-{2-[2-(*N*-D-mannofuranosyl-*N*-methoxyamino)ethoxy]ethyl}-2-naphthamide (26)

R_f 0.24 $CHCl_3$/MeOH/H_2O 10:10:3); ^{1}H NMR (600 MHz, MeOD): δ 8.33 (d, *J* = 1.4 Hz, 1H, Ar*H*), 8.30 (d, *J* = 1.9 Hz, 1H, Ar*H*), 7.94 (d, *J* = 9.1 Hz, 1H, Ar*H*), 7.88 (dd, *J* = 8.6, 1.7 Hz, 1H, Ar*H*), 7.84 (d, *J* = 8.7 Hz, 1H, Ar*H*), 7.64 (dd, *J* = 8.8, 2.0 Hz, 1H, Ar*H*), 4.59 (d, $J_{1,2}$ = 6.4 Hz, 1H, H-1), 4.30 (dd, $J_{2,3}$ = 4.6 Hz, 1H, H-2), 4.20 (dd, $J_{3,4}$ = 2.1 Hz, 1H, H-3), 3.89-3.86 (m, 2H, H-4, H-5), 3.75 (dd, $J_{5,6a}$ = 2.7 Hz, 1H, H-6a), 3.73-3.69 (m, 4H, CH_2OCH_2), 3.65 (t, *J* = 5.1 Hz, 2H, CH_2NHCO), 3.58 (s, 3H, OCH_3), 3.56-3.54 (m, 1H, H-6b), 3.10 (dt, *J* = 13.5, 5.5 Hz, 1H, CH_3ONCH_2), 3.04 (dt, *J* = 13.8, 5.7 Hz, 1H, CH_3ONCH_2), 2.91 (t, *J* = 7.4 Hz, 2H, CH_2NH_2), 2.52 (t, *J* = 7.1 Hz, 2H, $HNCOCH_2$), 1.85-1.78 (m, 2H, $HNCOCH_2CH_2$), 1.74-1.68 (m, 2H, $CH_2CH_2NH_2$); ESI-TOF HR-MS: *m/z* 565.2869; calcd. for $C_{27}H_{41}N_4O_9$: 565.2868.

6-(5-Aminopentanamido)-*N*-{2-[2-(*N*-α-D-mannopyranosyl-*N*-methoxyamino)ethoxy]ethyl}-2-naphthamide (27)

R_f 0.39 ($CHCl_3$/MeOH/H_2O 10:10:3); ^{1}H NMR (600 MHz, MeOD): δ 8.17 (s, 1H, Ar*H*), 8.29 (s, 1H, Ar*H)*, 7.93 (d, *J* = 8.9 Hz, 1H, Ar*H*), 7.86 (dd, *J* = 8.6, 1.7 Hz, 1H, Ar*H*), 7.83 (d, *J* = 8.7 Hz, 1H, Ar*H*), 7.63

(dd, *J* = 8.8, 2.0 Hz, 1H, Ar*H*), 4.21 (d, $J_{1,2}$ = 1.6 Hz, 1H, H-1), 4.14 (dd, $J_{2,3}$ = 3.4 Hz, 1H, H-2), 3.86 (dd, $J_{3,4}$ = 8.9 Hz, 1H, H-3), 3.78 (d, *J* = 2.4 Hz, 1H, H-6a), 3.76-3.73 (m, 3H, CH_2OCH_2, H-5), 3.70-3.69 (m, 2H, CH_2OCH_2), 3.65-3.60 (m, 4H, CH_2NHCO, H-4, H-6b), 3.55 (s, 3H, OCH_3), 2.85 (dt, *J* = 13.6, 5.4 Hz, 1H, CH_3ONHCH_2), 2.73 (t, *J* = 7.2 Hz, 2H, CH_2NH_2), 2.60 (dt, *J* = 14.4, 6.6 Hz, 1H, CH_3ONHCH_2), 2.47 (t, *J* = 7.4 Hz, 2H, HNCOCH_2), 1.80-1.75 (m, 2H, $HNCOCH_2CH_2$), 1.63-1.58 (m, 2H, C$H_2CH_2NH_2$); ^{13}C NMR (151 MHz, MeOD): δ 174.62 (C_q, Ar*C*=O), 170.33 (C_q, ArHN*C*=O), 139.31 (C_q, Ar*C*), 136.86 (C_q, Ar*C*), 131.76 (C_q, Ar*C*), 130.97 (C_q, Ar*C*), 130.81 (CH, Ar*C*H), 128.88 (CH, Ar*C*H), 128.59 (CH, Ar*C*H), 125.48 (CH, Ar*C*H), 122.01 (CH, Ar*C*H), 117.16 (CH, Ar*C*H), 94.47 (CH, C-1), 77.31 (CH, C-5), 73.16 (CH, C-3), 70.53 (CH, C-2), 70.39 (CH_2, CH_2OCH_2), 69.06 (CH_2, CH_2OCH_2), 68.93 (CH, C-4), 63.24 (CH_2, C-6), 62.71 (CH_3, OCH_3), 54.37 (CH_2, CH_3ONCH_2), 41.89 (CH_2, CH_2NH_2), 41.11 (CH_2, CH_2NHCO), 37.63 (CH_2, $HNCOCH_2$), 32.47 (CH_2, $CH_2CH_2NH_2$), 23.98 (CH_2, $HNCOCH_2CH_2$); ESI-TOF HR-MS: *m/z* 565.2870; calcd. for $C_{27}H_{41}N_4O_9$: 565.2868.

6-(5-Aminopentanamido)-*N*-{2-[2-(*N*-2-acetamido-2-deoxy-β-D-galactofuranosyl-*N*-methoxyamino) ethoxy]ethyl}-2-naphthamide (28)

R_f 0.30 ($CHCl_3$/MeOH/H_2O 10:10:3); ^{1}H NMR (600 MHz, MeOD): δ 8.32 (s, 1H, Ar*H*), 8.30 (s, 1H, Ar*H*), 7.94 (d, *J* = 9.0 Hz, 1H, Ar*H*), 7.87 (dd, *J* = 8.6, 1.7 Hz, 1H, Ar*H*), 7.84 (d, *J* = 8.6 Hz, 1H, Ar*H*), 7.64 (dd, *J* = 8.9, 1.8 Hz, 1H, Ar*H*), 4.60 (dd, $J_{1,2}$ = 7.0 Hz, $J_{2,3}$ = 7.8 Hz, 1H, H-2), 4.49 (d, $J_{1,2}$ = 6.7 Hz, 1H, H-1), 4.08 (app t, $J_{3,4}$ = 8.2 Hz, 1H, H-3), 3.90 (dd, $J_{4,5}$ = 2.5 Hz, 1H, H-4), 3.72-3.68 (m, 4H, CH_2OCH_2), 3.66-3.64 (m, 3H, CH_2NHCO, H-5), 3.62 (s, 3H, OCH_3), 3.61-3.60 (m, 2H, H-6a, H-6b), 3.13 (dt, *J* = 13.4, 5.6 Hz, 1H, CH_3ONCH_2), 3.02 (dt, *J* = 13.6, 6.1 Hz, 1H, CH_3ONCH_2), 2.90 (t, *J* = 7.4 Hz, 2H, CH_2NH_2), 2.52 (t, *J* = 7.2 Hz, 2H, HNCOCH_2), 1.94 (s, 3H, CH_3CO), 1.84-1.78 (m, 2H, $HNCOCH_2CH_2$), 1.74-1.69 (m, 2H, C$H_2CH_2NH_2$); ^{13}C NMR (151 MHz, MeOD): δ 130.79 (CH, Ar*C*H), 128.88 (CH, Ar*C*H), 128.58 (CH, Ar*C*H), 125.49 (CH, Ar*C*H), 122.02 (CH, Ar*C*H), 117.18 (CH, Ar*C*H), 97.85 (CH, C-1), 83.31 (CH, C-4), 75.94 (CH, C-3), 71.64 (CH, C-5), 70.06 (CH_2, CH_2OCH_2), 69.92 (CH_2, CH_2OCH_2), 64.52 (CH_2, C-6), 63.04 (CH_3, OCH_3), 58.74 (CH, C-2), 51.74 (CH_2, CH_3ONCH_2), 41.72 (CH_2, CH_2NH_2) 41.04 (CH_2, CH_2NHCO), 37.55 (CH_2, $HNCOCH_2$), 29.27 (CH_2, $CH_2CH_2NH_2$), 23.89 ($HNCOCH_2CH_2$), 22.71 (CH_3, CH_3CO); ESI-TOF HR-MS: *m/z* 606.3128; calcd. for $C_{29}H_{44}N_5O_9$: 606.3120.

6-(5-Aminopentanamido)-*N*-{2-[2-(*N*-2-acetamido-2-deoxy-β-D-galactopyranosyl-*N*-methoxyamino) ethoxy]ethyl}-2-naphthamide (29)

R_f 0.23 ($CHCl_3$/MeOH/H_2O 10:10:3); ^{1}H NMR (600 MHz, MeOD): δ 8.34 (d, *J* = 1.7 Hz, 1H, Ar*H*), 8.30 (d, *J* = 1.7 Hz, 1H, Ar*H*), 7.93 (d, *J* = 9.0 Hz, 1H, Ar*H*), 7.88 (dd, *J* = 8.6, 1.8 Hz, 1H, Ar*H*), 7.84 (d, *J* = 8.6 Hz, 1H, Ar*H*), 7.64 (dd, *J* = 8.9, 2.1 Hz, 1H, Ar*H*), 4.22 (d, $J_{1,2}$ = 9.8 Hz, 1H, H-1), 4.05 (t, $J_{2,3}$ = 10.0 Hz, 1H, H-2), 3.78 (d, $J_{3,4}$ = 3.3 Hz, $J_{4,5}$ = 0.8 Hz, 1H, H-4), 3.75 (dd, $J_{5,6a}$ = 6.8 Hz, $J_{6a,6b}$ = 11.5 Hz, 1H, H-6a), 3.71-3.66 (m, 5H, CH_2OCH_2, H-6b), 3.62-3.60 (m, 2H, CH_2NHCO), 3.52 (dd, $J_{3,4}$ = 3.2 Hz, 1H, H-3), 3.48 (s, 3H, OCH_3), 3.43-3.41 (m, 1H, H-5), 3.24 (dt, *J* = 13.7, 6.0 Hz, 1H, CH_3ONCH_2), 3.14 (dt, *J* = 13.6, 6.2

Hz, 1H, CH_3ONCH_2), 2.81 (t, *J* = 7.3 Hz, 2H, CH_2NH_2), 2.49 (t, *J* = 7.3 Hz, 2H, $HNCOCH_2$), 1.95 (s, 3H, CH_3CO), 1.82-1.77 (m, 2H, $HNCOCH_2CH_2$), 1.68-1.63 (m, 2H, $CH_2CH_2NH_2$); ^{13}C NMR (151 MHz, MeOD): δ 130.49 (CH, Ar*C*H), 128.59 (CH, Ar*C*H), 128.31 (CH, Ar*C*H), 125.22 (CH, Ar*C*H), 121.72 (CH, Ar*C*H), 116.87 (CH, Ar*C*H), 93.14 (CH, C-1), 78.13 (CH, C-5), 74.45 (CH, C-3), 70.07 (CH_2, CH_2OCH_2), 69.78 (CH_2, CH_2OCH_2), 69.58 (CH, C-4), 62.42 (CH_2, C-6), 61.64 (CH_3, OCH_3), 51.74 (CH_2, CH_3ONCH_2), 50.65 (CH, C-2), 41.25 (CH_2, CH_2NHCO), 40.81 (CH_2, CH_2NH_2), 37.16 (CH_2, $HNCOCH_2$), 31.11 (CH_2, $CH_2CH_2NH_2$), 23.50 (CH_2, $HNCOCH_2CH_2$), 22.71 (CH_3, CH_3CO); ESI-TOF HR-MS: *m/z* 606.3150; calcd. for $C_{29}H_{44}N_5O_9$: 606.3120.

6-(5-Aminopentanamido)-*N*-{2-[2-(*N*-β-L-fucopyranosyl-*N*-methoxyamino)ethoxy]ethyl}-2-naphthamide (30)

R_f 0.07 ($CHCl_3$/MeOH/H_2O 10:10:3); ^1H NMR (600 MHz, MeOD): δ 8.33 (d, *J* = 1.1 Hz, 1H, Ar*H*), 8.30 (d, *J* = 1.3 Hz, 1H, Ar*H*), 7.94 (d, *J* = 9.0 Hz, 1H, Ar*H*), 7.88 (dd, *J* = 8.6, 1.8 Hz, 1H, Ar*H*), 7.84 (d, *J* = 8.6 Hz, 1H, Ar*H*), 7.64 (dd, *J* = 8.8, 2.0 Hz, 1H, Ar*H*), 4.01 (d, $J_{1,2}$ = 9.1 Hz, 1H, H-1), 3.74-3.71 (m, 4H, CH_2OCH_2), 3.66-3.60 (m, 3H, CH_2NHCO, H-3), 3.64 (d, $J_{1,2}$ = 9.3 Hz, 1H, H-2), 3.56 (s, 3H, OCH_3), 3.56-3.55 (m, 1H, H-5), 3.46 (dd, *J* = 9.3, 3.6 Hz, 1H, H-4), 3.20 (dt, *J* = 14.1, 5.7 Hz, 1H, CH_3ONCH_2), 3.08 (dt, *J* = 14.0, 5.4 Hz, 1H, CH_3ONCH_2), 2.82 (t, *J* = 7.3 Hz, 2H, CH_2NH_2), 2.50 (t, *J* = 7.3 Hz, 2H, $NHCOCH_2$), 1.82-1.77 (m, 2H, $NHCOCH_2CH_2$), 1.70-1.64 (m, 2H, $CH_2CH_2NH_2$), 1.22 (d, *J* = 6.4 Hz, 3H, H-6); ^{13}C NMR (151 MHz, MeOD): δ 130.81 (CH, Ar*C*H), 128.88 (CH, Ar*C*H), 128.60 (CH, Ar*C*H), 125.50 (CH, Ar*C*H), 122.02 (CH, Ar*C*H), 117.17 (CH, Ar*C*H), 95.33 (CH, C-1), 76.43 (CH, C-4), 73.68 (CH, C-5), 73.27 (CH, C-3), 70.50 (CH_2, CH_2OCH_2), 69.77 (CH_2, CH_2OCH_2), 68.79 (CH, C-2), 62.42 (CH_3, OCH_3), 52.69 (CH_2, CH_3ONCH_2), 41.48 (CH_2, CH_2NH_2), 40.99 (CH_2, CH_2NHCO), 37.46 (CH_2, $HNCOCH_2$), 23.82 (CH_2, $HNCOCH_2CH_2$), 16.74 (CH_3, C-6). 31.90 (CH_2, $CH_2CH_2NH_2$); ESI-TOF HR-MS: *m/z* 549.2929; calcd. for $C_{27}H_{41}N_4O_8$: 549.2919.

6-(5-Aminopentanamido)-*N*-{2-[2-(*N*-β-D-lactopyranosyl-*N*-methoxyamino)ethoxy]ethyl}-2-naphthamide (31)

R_f 0.02 ($CHCl_3$/MeOH/H_2O 10:10:2); ^1H NMR (600 MHz, MeOD): δ 8.34 (d, *J* = 1.4 Hz, 1H, Ar*H*), 8.30 (d, *J* = 1.9 Hz, 1H, Ar*H*), 7.95 (d, *J* = 8.9 Hz, 1H, Ar*H*), 7.88 (dd, *J* = 8.6, 1.8 Hz, 1H, Ar*H*), 7.84 (d, *J* = 8.7 Hz, 1H, Ar*H*), 7.64 (dd, *J* = 8.8, 2.1 Hz, 1H, Ar*H*), 4.34 (d, $J_{1',2'}$ = 7.6 Hz, 1H, H-1´), 4.11 (d, $J_{1,2}$ = 8.5 Hz, 1H, H-1), 3.85 (dd, $J_{5,6a}$ = 2.5 Hz, $J_{6a,6b}$ = 12.2 Hz, 1H, H-6a), 3.82 - 3.81 (m, 2H, H-4´, H-6b), 3.78 (dd, $J_{5',6'a}$ = 7.4 Hz, $J_{6'a',6'b}$ = 11.4 Hz, 1H, H-6´a), 3.75 - 3.69 (m, 5H, CH_2OCH_2, H-6´b), 3.66 - 3.64 (m, 2H, CH_2NHCO), 3.60 (s, 3H, OCH_3), 3.59-3.50 (m, 5H, H-2, H-3, H-4, H-2´, H-5'), 3.48 (dd, $J_{2',3'}$ = 9.7, $J_{3',4'}$ = 3.3 Hz, 1H, H-3´), 3.34 - 3.32 (m, 1H, H-5), 3.22 (dt, *J* = 13.8, 5.4 Hz, 1H, CH_3ONCH_2), 3.14 (dt, *J* = 13.7, 5.8 Hz, 1H, CH_3ONCH_2), 2.96 (t, *J* = 7.4 Hz, 2H, CH_2NH_2), 2.53 (t, *J* = 7.0 Hz, 2H, $HNCOCH_2$), 1.85-1.80 (m, 2H, $HNCOCH_2CH_2$), 1.77-1.73 (m, 2H, $CH_2CH_2NH_2$); ^{13}C NMR (151 MHz, MeOD): δ 130.87 (CH, Ar*C*H), 128.90 (CH, Ar*C*H), 128.61 (CH, Ar*C*H), 125.55 (CH, Ar*C*H), 122.03 (CH, Ar*C*H), 117.20

(CH, Ar*C*H), 105.11 (CH, C-1´), 94.63 (CH, C-1), 80.35 (CH, C-4), 78.20 (CH, C-5), 77.62 (CH, C-3), 77.08 (CH, C-5'), 74.86 (CH, C-3´), 72.59 (CH, C-2´), 71.25 (CH, C-2), 70.51 (CH_2, CH_2OCH_2), 70.33 (CH, C-4´), 69.66 (CH_2, CH_2OCH_2), 62.77 (CH_3, OCH_3), 62.51 (CH_2, C-6´), 62.07 (CH_2, C-6), 41.02 (CH_2, CH_2NHCO), 40.69 (CH_2, CH_2NH_2), 37.04 (CH_2, $HNCOCH_2$), 23.30 (CH_2, $HNCOCH_2CH_2$); ESI-TOF HR-MS: *m/z* 727.3396; calcd. for $C_{33}H_{51}N_4O_{14}$: 727.3396.

6-(5-Aminopentanamido)-*N*-{2-[2-(*N*-2-acetamido-2-deoxy-β-D-lactopyranosyl-*N*-methoxyamino)ethoxy]ethyl}-2-naphthamide (32)

R_f 0.17 ($CHCl_3$/MeOH/H_2O 10:10:3); ^{1}H NMR (600 MHz, MeOD): δ 8.35 (d, *J* = 0.8 Hz, 1H, Ar*H*), 8.31 (d, *J* = 1.7 Hz, 1H, Ar*H*), 7.94 (d, *J* = 8.8 Hz, 1H, Ar*H*), 7.88 (dd, *J* = 5.8, 1.7 Hz, 1H, Ar*H*), 7.85 (d, *J* = 8.7 Hz, 1H, Ar*H*), 7.65 (dd, *J* = 8.9, 2.0 Hz, 1H, Ar*H*), 4.35 (d, $J_{1´,2´}$ = 7.6 Hz, 1H, H-1´), 4.29 (d, $J_{1,2}$ = 10.0 Hz, 1H, H-1), 3.91-3.85 (m, 2H, H-2, H-6a), 3.82-3.77 (m, 2H, H-4´, H-6b), 3.76 (dd, *J* = 11.5, 7.4 Hz, 1H, H-6´a), 3.70-3.66 (m, 6H, CH_2OCH_2, CH_2NHCO, H-6´b), 3.61 (t, *J* = 5.2 Hz, 1H, CH_2NHCO), 3.59-3.55 (m, 3H, H-3, H-4, H-5´) 3.52 (dd, $J_{2´,3´}$ = 9.5 Hz, 1H, H-2´), 3.49 (s, 3H, OCH_3), 3.48 (dd, $J_{3´,4´}$ = 3.4 Hz, 1H, H-3´), 3.34-3.33 (m, 1H, H-5), 3.23-3.13 (m, 2H, CH_3ONHCH_2), 2.99 (t, *J* = 7.3 Hz, 1H, CH_2NH_2), 2.54 (t, *J* = 7.0 Hz, 2H, $HNCOCH_2$), 2.19 (t, *J* = 7.6 Hz, 1H, CH_2NH_2), 1.94 (s, 3H, CH_3CO), 1.84-1.74 (m, 2H, $HNCOCH_2CH_2$), 1.66-1.57 (m, 2H, $CH_2CH_2NH_2$); ^{13}C NMR (151 MHz, MeOD): δ 174.03 (C_q, Ar*C*=O), 173.15 (C_q, CH_3C=O), 170.25 (C_q, ArNH*C*=O), 139.26 (C_q, Ar*C*), 136.84 (C_q, Ar*C*), 131.86 (C_q, Ar*C*), 131.01 (C_q, Ar*C*), 130.86 (CH, Ar*C*H), 128.91 (CH, Ar*C*H), 128.63 (CH, Ar*C*H), 125.55 (CH, Ar*C*H), 121.99 (CH, Ar*C*H), 117.21 (CH, Ar*C*H), 105.07 (CH, C-1´), 92.92 (CH, C-1), 80.66 (CH, C-4), 78.32 (CH, C-5), 77.17 (CH, C-5´), 75.62 (CH, C-3), 74.86 (CH, C-3´), 72.60 (CH, C-2´), 70.42 (CH_2, CH_2OCH_2), 70.36 (CH, C-4´), 70.02 (CH_2, CH_2OCH_2), 62.55 (CH_2, C-6´), 62.11 (CH_2, C-6), 62.01 (CH_3, OCH_3), 53.49 (CH, C-2), 52.26 (CH_2, CH_3ONCH_2), 41.09 (CH_2, CH_2NHCO), 40.53 (CH_2, CH_2NH_2), 36.93 (CH_2, $HNCOCH_2$), 36.54 (CH_2, CH_2NH_2), 28.22 (CH_2, $CH_2CH_2NH_2$), 26.91 (CH_2, $CH_2CH_2NH_2$), 23.23 (CH_2, $HNCOCH_2CH_2$), 22.97 (CH_3, CH_3O); ESI-TOF HR-MS: *m/z* 768.3638; calcd for $C_{35}H_{54}N_5O_{14}$: 768.3662.

6-(5-Aminopentanamido)-*N*-{2-[2-(*N*-β-D-chitobiopyranosyl-*N*-methoxyamino)ethoxy]ethyl}-2-naphthamide (33)

R_f 0.03 ($CHCl_3$/MeOH/H_2O 10:10:2); ^{1}H NMR (600 MHz, MeOD) δ 8.34 (s, 1H, Ar*H*), 8.31 (d, *J* =1.6 Hz, 1H, Ar*H*), 7.94 (d, *J*= 8.8 Hz, 1H, Ar*H*), 7.88 (dd, *J* = 8.7, 1.7 Hz, 1H, Ar*H*), 7.85 (d, *J* =8.6 Hz, 1H, Ar*H*), 7.65 (dd, *J* = 8.8, 2.0 Hz, 1H, Ar*H*), 4.45 (d, $J_{1´,2´}$ = 8.5 Hz, 1H, H-1´), 4.25 (d, $J_{1,2}$ = 9.9 Hz, 1H, H-1), 3.89 (dd, *J* = 11.8, 2.0 Hz, 1H, H-6a), 3.88 (t, $J_{1,2}$ = 9.8 Hz, 1H, H-2), 3.76 (dd, *J* = 12.1, 1.7 Hz, 1H, H-6b), 3.71-3.65 [m, 7H, CH_2OCH_2, $CH_2NHC(O)$, H-2´], 3.63 (dd, *J* = 11.8, 6.5 Hz, 1H, H-6´b), 3.60-3.55 (m, 1H, H-6´a), 3.51 (d, $J_{2,3}$ = 8.6 Hz, 1H, H-3), 3.49 (s, 3H, OCH_3), 3.47 (d, *J* = 9.5 Hz, 1H, H-4), 3.43 (dd, *J* =10.5, 8.3 Hz, 1H, H-3´), 3.34-3.32 (m, 1H, H-4´), 3.28 (d, *J* = 9.8 Hz, 1H, H-5´), 3.24-3.21 (m, 1H, H-5), 3.20-3.14 (m, 2H, CH_3ONCH_2), 2.96 (t, *J* = 7.3 Hz, 2H, CH_2NH_2), 2.54 [t, *J* = 7.0 Hz, 2H, $HNC(O)CH_2$], 2.00 (s, 3H, NHAc´), 1.94 (s, 3H, NHAc), 1.80-1.80 [m, 2H, $HNC(O)CH_2CH_2$], 1.78-1.74 (m, 2H,

C*H*$_2$CH$_2$NH$_2$); ^{13}C NMR (151 MHz, MeOD) δ 130.65 (CH, Ar*C*H), 128.29 (CH, Ar*C*H), 128.42 (CH, Ar*C*H), 125.00 (CH, Ar*C*H), 121.70 (CH, Ar*C*H), 116.71 (CH, Ar*C*H), 102.91 (CH, C-1´), 92.48 (CH, C-1), 81.19 (CH, C-4), 78.00 (CH, C-4´), 78.00 (CH, C-5), 75.94 (CH, C-3´), 75.41 (CH, C-3), 71.55 (CH, C-5´), 69.96 (CH$_2$, *C*H$_2$O*C*H$_2$), 62.37 (CH$_2$, C-6´), 61.78 (CH$_3$, O*C*H$_3$), 61.77 (CH$_2$, C-6), 57.03 (CH, C-2´), 53.11 (CH, C-2), 52.04 (CH$_2$, CH$_3$ON*C*H$_2$), 40.78 (CH$_2$, *C*H$_2$NHC=O), 40.60 (CH$_2$, *C*H$_2$NH$_2$), 37.00 [CH$_2$, HNC(O)*C*H$_2$], 23.23 (CH$_3$, NHAc), 23.18 (CH$_3$, NHAc´), 28.26 (CH$_2$, *C*H$_2$CH$_2$NH$_2$) 23.13 [CH$_2$, HNC(O)CH$_2$*C*H$_2$]; ESI-TOF HR-MS: *m/z* 809.3935; calcd for $C_{37}H_{57}N_6O_{14}$: 809.3927.

Stability of amino-functionalized neoglycoconjugates

To study stability at different pH over time, neoglycoconjugate **24** (0.85 mg, 1.5 μmol) was dissolved in aqueous solutions to obtain 40 mM final concentrations. Following solutions of different pH were tested (0.5 M HCOOH pH 2, 0.1 M NH_4OAc/HOAc pH 4, 0.1 M NH_4OAc/HOAc pH 6, 0.1 M NH_4HCO_3/NH_4OH pH 8.5 and 0.1 M NH_4HCO_3/NH_4OH pH 10 and 0.25 M NH_4OH pH 12). The solutions were incubated at rt for 48 h. Aliquots from these solutions were taken after 0, 3, 19, 24, and 48 h, diluted with 90% MeCN and injected into HPLC-MS with a HILIC column at flow rate 0.75 mL/min and linear gradient of 75 to 40% B in A over 20 min (A= H_2O + 0.1% HCOOH; B= MeCN + 0.1% HCOOH). Peak containing the glycoconjugate of *m/z* 565.25 were used to estimate the neoglycoconjugate stability.

To evaluate stability under storage conditions, 0.1 mM solutions of compounds **10**, **22**, and **24** were prepared in water and stored at 4° C and at -20 °C. Aliquots were taken every 7 days and every month from the 4°C and -20°C storage conditions, respectively, diluted with MeCN and analyzed by HPLC with a Tosoh Amide-80 column with flow rate 1 mL/min and a linear gradient of 75 to 40% B in A over 40 min (A= 10 mM ammonium formate, pH 7; B= 95% MeCN) and equipped with a fluorescence detector. Peak areas were used to monitor the stability of the compounds under storage conditions.

Preparation of recombinant enzymes

Recombinant glycosyltransferases (*Ce* FUT-6, POMGnT1, α2,3-SiaT, α2,6-SiaT and β1,3-GalT) were expressed, either in *Pichia pastoris* or *E. coli*, and purified in the laboratory:

Expression in *Pichia pastoris*: Recombinant strain was first inoculated in 10 mL of MGYCZ medium (1% (w/v) yeast extract, 2% (w/v) peptone, 1% (w/v) casamino acid, 1.34% (w/v) yeast nitrogen base, 1% (v/v) glycerol, 0.00004% (w/v) biotin). The culture was grown at 30 °C until an $O.D._{600}$ of 10 was reached. The cells were re-suspended in 50 mL of MMYC (composition as for MGYC with MeOH 1% (v/v) instead of glycerol) and the protein expression was carried out at 16 °C with shaking at 220 rpm for 3 days (5 days for human POMGnT1). The enzyme was concentrated using an Amicon® ultrafiltration device, purified using Ni-NTA beads (QIAGEN), eluted with imidazole and stored at 4 °C.

Expression of recombinant glycosyltransferases in *E. coli*: Recombinant strain was first inoculated in 2 mL of LB medium supplemented with kanamycin. The culture was grown at 37 °C with shaking at 200 rpm overnight and then enlarged to 10 mL until an $O.D._{600}$ of 0.6-0.8 was reached. The expression of the enzyme was induced with IPTG and carried out with shaking at 200 rpm at 37 °C for 3 h for α2,3-SiaT and β1,3-GalT (at 25 °C for 5 h for α2,6-SiaT). The *E. coli* cells were lysed and the enzyme was purified using either Ni-NTA beads (QIAGEN) and eluted with imidazole for α2,3-SiaT and α2,6-SiaT or an amylose resin column (NEB) and eluted with maltose. The purified enzymes were stored at 4 °C.

(i) *Caenorhabditis elegans* FUT-6, which is known to possess both Lewis-type α1,3-fucosyltransferase activity towards LacNAc and LacdiNAc antennae as well as to the core region of a restricted range of N-glycans, was expressed in *Pichia pastoris*.

(ii) Human POMGnT1 β1,2-*N*-acetylglucosaminyltransferase was expressed in *Pichia pastoris. In vitro* activity was verified by using a synthetic O-mannosyl peptide as substrate (Akasaka-Manya K.*et al.* 2011).

(iii) α2,3-SiaT was expressed in *E. coli* from a synthetic clone in pET30a encoding a fusion of *Neisseria* CMP-sialic acid synthase with *Neisseria* α2,3-sialyltransferase. *In vitro* activity was verified by using dabsyl-GalGal as substrate.

(iv) α2,6-SiaTwas expressed in *E. coli* from a synthetic clone in pET30a encoding a fusion of *Neisseria* CMP-sialic acid synthase with *Photobacterium damsela* α2,6-sialyltransferase (Pd2,6ST). *In vitro* activity was verified by using dabsyl-GalGal as substrate.

(v) β1,3-GalT, which is known to transfer Gal to both α- and β-anomers of GalNAc, was expressed in *E. coli* from a clone (pCJL-136; pCW*malE*-C/NdeI-EcoRI+$cgtB_{OH4384}$-90bp/NdeI-EcoRI) encoding *Campylobacter jejuni* CgtB as a fusion with *E. coli* maltose-binding protein (MalE).

In solution modification of neoglycoconjugates by glycosyltransferases

Synthesis of type 2 LN (LacNAc) epitope: Compound **22** (*i.e,* the β-GlcNAc-linker; 0.45 µg, 0.75 nmol) was dissolved in 80 mM MES buffer pH 6.5 and mixed with UDP-Gal (30.5 µg, 50 nmol), β1,4-GalT (3 µg) and $MnCl_2$ (20 mM). The reaction mixture was incubated at 37 °C overnight.

Synthesis of 3'-sialyl-LN epitope: Compound **22** (0.45 µg, 0.75 nmol), UDP-Gal (30.5 µg, 50 nmol), β1,4-GalT (3 µg), CMP-Neu5Ac (31.8 µg, 50 nmol), α2,3-SiaT (3 µL) and $MnCl_2$ (20 mM) in 80 mM MES buffer pH 6.5 was prepared and incubated at 37 °C overnight.

Synthesis of sLeX epitope: Compound **22** (0.45 µg, 0.75 nmol) was first incubated with UDP-Gal (30.5 µg, 50 nmol), β1,4-GalT (3 µg), CMP-Neu5Ac (31.8 µg, 50 nmol), α2,3-SiaT (3µL) and $MnCl_2$ (20 mM) in 80 mM MES buffer pH 6.5 at 37 °C overnight. Afterwards, GDP-Fuc (31.7 µg, 50 nmol) and FUT-6 (3 µL) were added and the reaction mixture was incubated at 37 °C overnight to prepare the tetrasaccharide sLeX epitope.

Synthesis of LeX epitope: A solution of compound **22** (0.45 µg, 0.75 nmol), UDP-Gal (30.5 µg, 50 nmol), β-1,4GalT (3 µg), GDP-Fuc (31.7 µg, 50 nmol), FUT-6 (3 µL) and $MnCl_2$ (20 mM) in 80 mM MES buffer pH 6.5 was prepared and incubated at 37 °C overnight.

Synthesis of α-Gal antigen epitope: Compound **22** (0.45 µg, 0.75 nmol) was mixed with UDP-Gal (61 µg, 100 nmol), β1,4-GalT (3 µg), α1,3-GalT (3 µg), $MnCl_2$ (20 mM) in 80 mM MES buffer pH 6.5. The reaction was incubated at 37 °C overnight.

Synthesis of N-glycan antennal structure: Compound **27** (0.45 µg, 0.75 nmol) was first incubated with UDP-GlcNAc (30.4 µg, 50nmol), POMGnT1 (3 µL) and $MnCl_2$ (20 mM) in 80 mM MES buffer pH 6.5 at 37 °C overnight. Afterwards, UDP-Gal (30.5 µg, 50 nmol) and β1,4GalT (3 µg) were added and the reaction mixture was incubated at 37 °C overnight.

Synthesis of T-antigen*:* Compound **29** (0.45 µg, 0.75 nmol) was incubated with UDP-Gal (30.5 µg, 50 nmol), β1,3-GalT (3 µL) and $MnCl_2$ (20 mM) in 80mM MES buffer pH 6.5 at 37 °C overnight.

Synthesis of STn-antigen*:* Compound **29** (0.45 µg, 0.75 nmol) was incubated with CMP-Neu5Ac (31.8 µg, 50 nmol), α2,6-SiaT (3 µL) and $MnCl_2$ (20 mM) in 80 mM MES buffer pH 6.5 at 37 °C overnight

Supplementary References

Akasaka-Manya K, Manya H, Mizuno M, Inazu T, Endo T. 2011. Effects of length and amino acid sequence of O-mannosyl peptides on substrate specificity of protein O-linked mannose β1,2-N-acetylglucosaminyltransferase 1 (POMGnT1). *Biochemical and Biophysical Research Communications*, 410:632-636.

Galili U. 2005. The [alpha]-gal epitope and the anti-Gal antibody in xenotransplantation and in cancer immunotherapy. *Immunol Cell Biol*, 83:674-686.

Gill HS, Tinianow JN, Ogasawara A, Flores JE, Vanderbilt AN, Raab H, Scheer JM, Vandlen R, Williams S-P, Marik J. 2009. A Modular Platform for the Rapid Site-Specific Radiolabeling of Proteins with 18F Exemplified by Quantitative Positron Emission Tomography of Human Epidermal Growth Factor Receptor 2. *Journal of Medicinal Chemistry*, 52:5816-5825.

Kimura Y, Ushijima T, Maeda M, Hama Y, Kimura M, Okihara K, Sugimoto H, Yamada H. 2006. Tumor Antigen Occurs in N-Glycan of Royal Jelly Glycoproteins: Honeybee Cells Synthesize T-Antigen Unit in N-Glycan Moiety. *Bioscience, Biotechnology, and Biochemistry*, 70:2583-2587.

Mohler DL, Shen G. 2006. The synthesis of tethered ligand dimers for PPAR[gamma]-RXR protein heterodimers. *Organic & Biomolecular Chemistry*, 4:2082-2087.

Persson K, Ly HD, Dieckelmann M, Wakarchuk WW, Withers SG, Strynadka NCJ. 2001. Crystal structure of the retaining galactosyltransferase LgtC from Neisseria meningitidis in complex with donor and acceptor sugar analogs. *Nat Struct Mol Biol*, 8:166-175.

Svennerholm L. 1962. The chemical structure of normal human brain and Tay-Sachs gangliosides. *Biochemical and Biophysical Research Communications*, 9:436-441.

Yamamoto T, Nakashizuka M, Terada I. 1998. Cloning and Expression of a Marine Bacterial β-Galactoside α2,6-Sialyltransferase Gene from Photobacterium damsela JT0160. *Journal of Biochemistry*, 123:94-100.

Yu H, Huang S, Chokhawala H, Sun M, Zheng H, Chen X. 2006. Highly Efficient Chemoenzymatic Synthesis of Naturally Occurring and Non-Natural α-2,6-Linked Sialosides: A P. damsela α-2,6-Sialyltransferase with Extremely Flexible Donor–Substrate Specificity. *Angewandte Chemie International Edition*, 45:3938-3944.