Syntheses and Evaluation of Biantennary Oligosaccharide Ligands Mimicking Sialyl Lewis X

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Sialyl Lewis X (1) is known to be a ligand of the cell adhesion molecule E-selectin. We have synthesized several biantennary glycoside-terminated ligands mimicking sialyl Lewis X (1), and evaluated their binding activity to E-selectin using HL-60 cells expressing sialyl Lewis X epitope and human umbilical vein endothelial cells (HUVECs). These compounds were found to possess moderate binding activities to E-selectin. Among them, di-fucoside analog (8) which has no sialic acid carboxylate group was more active than 2, which had both the sialylgalactose residue and the fucose residue (IC_{50}, 8: 4.7 mM, 2: 11.7 mM). Furthermore, in the rat pleuritic model *in vivo* induced by carrageenin, 8 was found to reduce neutrophil infiltration at inflammatory lesions.

Key words sialyl Lewis X; E-selectin; anti-inflammatory agent; targeting drug delivery system

The migration of neutrophiles from intravascular spaces to sites of inflammation or tissue injury is initiated by their rolling on the activated vascular endothelium.[2] This adhesion process is mediated by the interaction of cell adhesion molecules, so-called selectins, with their physiological glycoprotein ligands, and leads to pathological inflammation. Three different selectins recognize a common carbohydrate epitope, the sialyl Lewis X (SLe^X, **1**), albeit with different affinities. Therefore, controlled blocking of the SLe^X–selectin binding by SLe^X analogs is a promising new therapeutic approach to battling inflammatory diseases.[3]

SLe^X or its analogs may also be useful as effective homing devices for active-targeting DDS (drug delivery system) to inflammatory lesions, since E-selectin is only expressed on such lesions. In connection with our work on developing DDS, we have found some polysaccharides with molecular sizes above 70 kDa such as carboxymethylchitosan (CMCht), and carboxymethylpullulan (CMPul) to be useful as carriers.[4] Recently, we reported a SLe^X–CMPul conjugate to be a good carrier in active-targeting DDS to inflammatory lesions.[5] To our knowledge, this was the first experimental result to give evidence of SLe^X being useful as a DDS homing device. In the case of SLe^X–selectin interaction, the structure–function relationship study and conformational analysis have led to the rational development of SLe^X mimetics which may be comparable to or even better than the natural ligand as inhibitors of selectins. Several groups have been actively engaged in this effort, and several SLe^X mimetics have been reported.[6] However, considering the complicated synthesis of SLe^X and cost factors, there is still a need for the development of simplified SLe^X mimetics which are more synthetically accessible and yet retain their usefulness as a homing device.

In trying to design novel SLe^X mimetics, we have focused our attention on the space distance and orientation between the negative charge of sialic acid and the three hydroxyl groups of the fucose residue. Several workers have reported that both sialic acid carboxylate and fucose hydroxyl groups were essential for the binding of SLe^X to selectin while the *N*-acetyl glucosamine moiety tolerated some variation.[7] Therefore, by introducing our mimic to the DDS carrier, we designed biantennary glycoside-terminated ligands (**2**, **3**) that replaced the *N*-acetyl glucosamine moiety of SLe^X with a Boc-L-glutamic acid linked adequate spacer arm (C6 carbon chain, and triethylene glycol) as illustrated in Fig. 1. Thus, using the amino group of L-glutamic acid, our mimic could be easily introduced to liposome[8] or polysaccharides,[4] which are useful as DDS carriers.

Here we describe the synthesis and evalution of these SLe^X mimicking compounds.

Synthesis and *in Vitro* Assay The synthesis of **2** is presented in Chart 1. 1-*O*-*p*-Nitrobenzoyl-2,3,4-tri-*O*-benzyl-L-fucopyranose (**10**, $\alpha:\beta=36:64$)[9] was reacted with 6-azidohexanol[10] in the presence of zinc trifluoromethanesulfonate ($Zn(OTf)_2$)/chlorotrimethylsilane (TMSCl)[11] to give α-glycoside (**11**) in 72% yield and β-glycoside (27%). Selective reduction (Lindlar's cat./H_2/*p*-toluenesulfonic acid monohydrate (*p*-TsOH·H_2O)) of the azido group of **11** led to an amine, which was condensed with BocGlu(OBn) followed by selective hydrogenation of benzyl ester,[12] to furnish the fucose–glutamic acid moiety (**14**).

On the other hand, the sialyl-galactose azide (**18**) was prepared from imidate (**17**)[13] by treatment with azidohexanol in the presence of $BF_3\cdot OEt_2$.

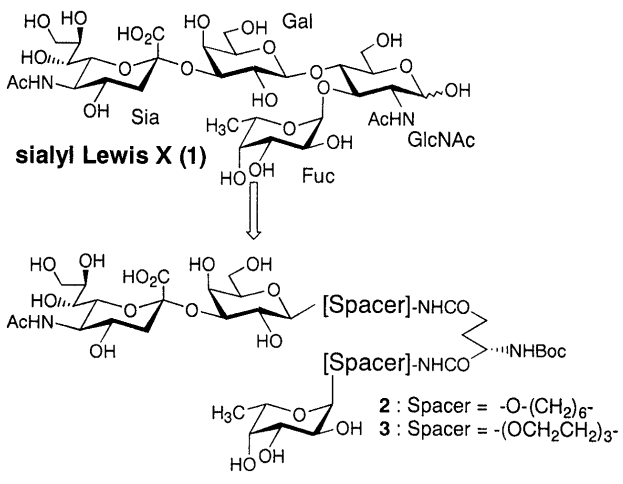

Fig. 1

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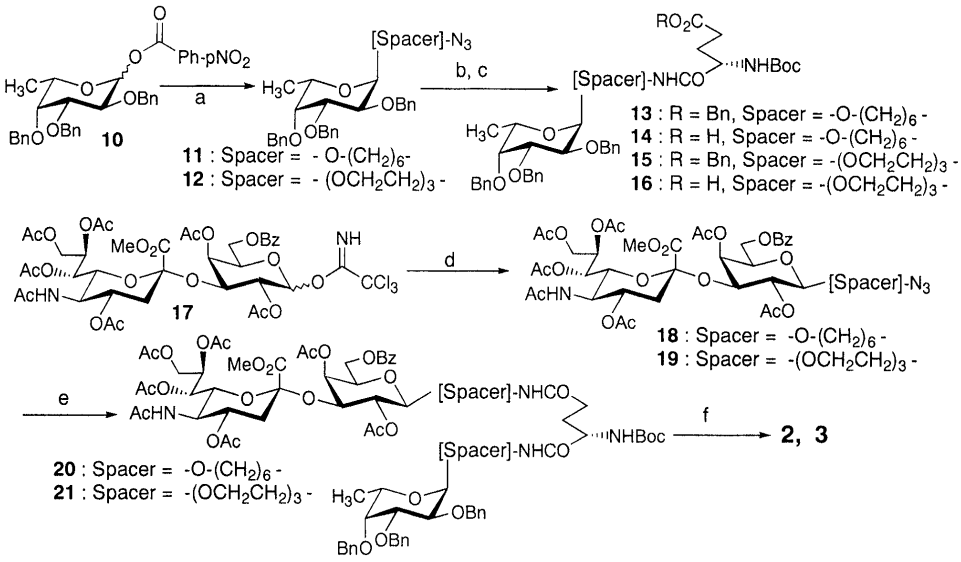


a) R-OH/TMSCl/Zn(OTf)$_2$/CH$_3$CN b) 1) Lindlar's cat./H$_2$/*p*-TsOH·H$_2$O/MeOH 2) BocGlu(OBn)-OSu/*N*-Methylmorpholine/CH$_2$Cl$_2$ c) 1,4-cyclohexadiene/Pd-C/EtOH d) BF$_3$·OEt$_2$/MS4A/CH$_2$Cl$_2$ e) 1) Lindlar's cat./H$_2$/*p*-TsOH·H$_2$O/MeOH 2) **14** or **16**/DCC/HOSu/Et$_3$N/CH$_2$Cl$_2$ f) 1) NaOMe/MeOH 2) aq. NaOH 3) Pd-C/H$_2$/MeOH

Chart 1

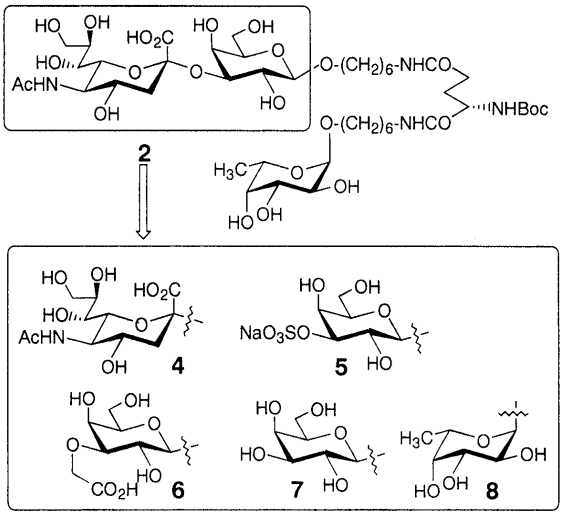
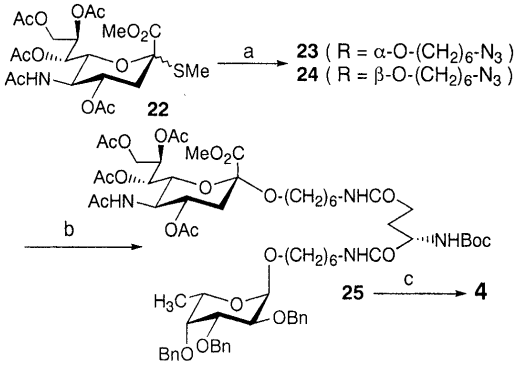


Fig. 2



a) 6-azidohexanol/NIS/TfOH/CH$_3$CN (**23**: 51%, **24**: 24%) b, c) same procedure as **2** and **3**

Chart 2

Reduction of the azido group of **18** followed by coupling with the fucose–glutamic acid moiety (**14**), afforded **20** [1) Lindlar's cat./H$_2$/*p*-TsOH·H$_2$O 2) 1,3-dicyclohexylcarbodiimide (DCC)/*N*-hydroxysuccinimide (HOSu), 47%], which was deprotected in three steps. [1) NaOMe–MeOH 2) aq.NaOH 3) Pd–C/H$_2$/MeOH, 85%].

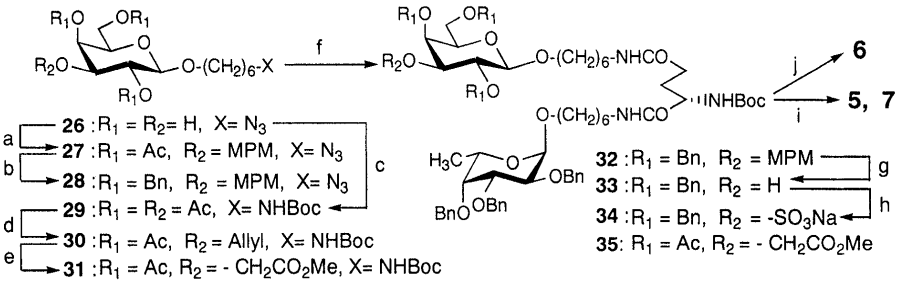
Compound **3**, which had triethylene glycol as a spacer instead of the C6 carbon chain in **2**, was synthesized by the same procedure as **2**.

The binding activity to E-selectin of **1**, **2** and **3** was evaluated using HL-60 cells expressing SLe^X epitope and HUVEC (Human Umbilical Vein Endothelial Cells) (details are given in the Experimental section). Compound **2** was found to possess a potent inhibitory activity to E-selectin (IC$_{50}$ values: **1**, 1.0 mM; **2**, 11.7 mM), whereas compound **3** was completely inactive. The lipophilicity of the spacer is apparently neccessary for the spatial arrangement of the essential sialic acid carboxylate and fucose hydroxyl groups. Next, we synthesized various biantennary ligands with fucose and other sugar derivative residues (**4**—**8**) that simplified the sialylgalactose moieties of **2** (Fig.2).

For the synthesis of **4**, coupling of thioglycoside (**22**)[14] and azidohexanol using *N*-iodosuccinimide (NIS) and trifluoromethanesulfonic acid (TfOH) as the promoter[15] in CH$_3$CN gave α-glycoside (**23**) in 51% yield along with β-glycoside (**24**, 24%), which was converted to **4** according to a procedure similar to that described for **2** (Chart 2).

For the synthesis of sulfated analog (**5**), treatment of 1,2,3,4,6-penta-*O*-acetyl-α-D-galactopyranosyl bromide with azidohexanol in the presence of silver silicate[16] in CH$_2$Cl$_2$ followed by deacetylation gave β-glycoside (**26**) in 75% yield, which was converted to 2,4,6-tri-*O*-benzyl 3-*O*-*p*-methoxybenzyl galactose derivative (**28**) *via* selective alkylation at the *O*-3 position in β-galactoside derivative using dibutyltin oxide/tetrabutylammonium bromide (62%).

Reduction of the azido group of **28** followed by condensation with the fucose–glutamic acid moiety (**14**) gave **32**. The *p*-methoxybenzyl group was removed oxidatively from **32** using 2,3-dichloro-5,6-dicyano-1,4-benzoquinone (DDQ) (72%), and the sulfate group was attached in its place



a) 1) MPMCl/Bu_4NBr/Bu_2SnO/Bz. 2) Ac_2O/Py. (62%) b) 1) NaOMe/MeOH 2) BnBr/NaH/DMF (82%) c) 1) Lindlar's cat./H_2/*p*-TsOH·H_2O/MeOH 2) Boc_2O/Et_3N/MeOH (67%) d) 1) NaOMe/MeOH 2) Allylbromide/Bu_4NBr/Bu_2SnO/Bz. 3) Ac_2O/Py. (69%) e) 1) O_3/MeOH 2) $NaClO_2$/tBuOH–H_2O 3) $TMSCHN_2$/MeOH (60%) f) 1) Lindlar's red. or TFA 2) **14**/DCC/HOSu/Et_3N (**32**: 92%, **35**: 93%) g) DDQ/CH_2Cl_2–H_2O (72%) h) SO_3·NMe_2/DMF (63%) i) Pd-C/H_2/MeOH (**5**: 82%, 7: 84%) j) 1) NaOMe/MeOH 2) aq. NaOH 3) Pd-C/H_2/MeOH (88%)

Chart 3

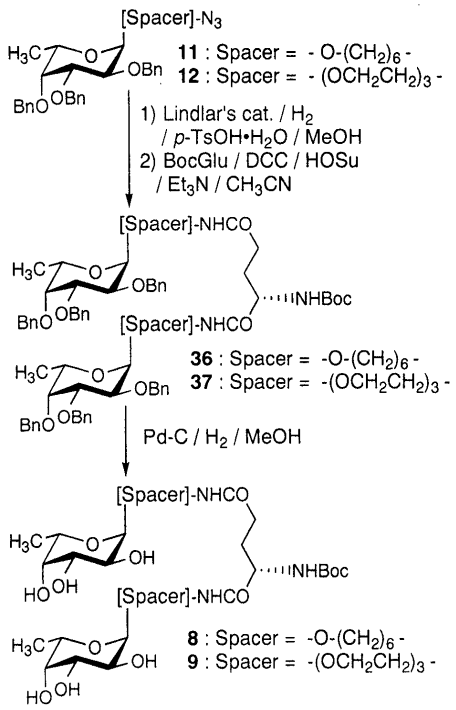


Chart 4

(SO_3·NMe_2/*N*,*N*′-dimethylformamide (DMF), 63%) to furnish **34**. Finally, debenzylation of **34** (Pd–C/H_2/MeOH, 82%) gave the sulfated analog (**5**).

For the synthesis of **6**, **26** was converted to **30** *via* selective allylation at the *O*-3 position in the β-galactoside derivative using dibutyltin oxide/tetrabutylammonium bromide. Ozonolysis of **30** followed by reductive work up, oxidation of the formyl group and methylation with trimethylsilyldiazomethane ($TMSCHN_2$) gave **31** (60%), which was converted to **6** according to a procedure similar to that described for **2**.

Compound **7**, which has no acidic functional group, was easily prepared from **32** by hydrogenolysis.

For the synthesis of **8**, reduction of the azido group of **11** followed by condensation with Boc-L-Glu, gave the hexabenzyl ether derivative (**36**), which was then reduced to **8** using Pd–C/H_2 (Chart 4).

The biological activity of **4**—**8** was tested with the binding assay using HL-60 cells and HUVEC. These compounds except **7** were found to have moderate binding activities to E-selectin (IC_{50} value: **4**, 7.4 mM; **5**, 11.4 mM; **6**, 12.8 mM; **8**, 4.7 mM). Interestingly, compound **8** having no sialic acid carboxylate, was the most active among these analogs. The two fucose residues, not sialic acid carboxylate, were essential for the binding to E-selectin. The difucose analog (**9**) which has the more hydrophilic triethylene glycol arms as a spacer was found to be inactive like **3** (Chart 4). The spatial arrangement of the two fucose residues was important and the conformation for the desired interaction was dependent on the length and lipophilicity of the spacer arms.

***In Vivo* Assay** The *in vivo* effect of compound **8** was examined on the rat pleuritic model induced by carrageenin.[17]

As controls, SLe^X (**1**) and **9** were also examined. Each compound was administered intravenously at 0, 2 and 4 h after carrageenin challenge (dose: 5 mg/kg×3 times). After 24 h, the number of neutrophils and the amount of exudate proteins were measured.

As shown in Fig. 3, **8** reduced neutrophil infiltration at the inflammatory lesion, while **9**, which had no binding activity at the *in vitro* assay, was inactive. Thus, the results of the *in vivo* assay almost paralleled to those of the *in vitro* assay. Compound **8** was shown to reach the inflammatory lesion *in vivo* and offer the possibility of being a useful homing device aimed at inflammatory lesions.

On the other hand, **8** was almost inactive for reducing exudate proteins, similar to **9**. These results showed that **8** did not affect the vascular permeability, namely, no anti-inflammatory effect of **8** was observed at this dose.

In conclusion, we demonstrated the practical synthesis of biantennary oligosaccharide ligands mimicking SLe^X. These ligands have moderate binding activitites to E-selectin. Compound **8** was found to reduce neutrophil infiltration at inflammatory lesions *in vivo*. The spatial arrangement of the two fucose residues, and the length and lipophilicity of the spacer were important for binding to E-selectin. These ligands should be useful as effective homing devices for active targeting DDS of inflammatory lesions. As cell-surface receptor–ligand interactions are often multivalent, inhibitors prepared in multivalent form should increase the inhibition potency.[18] As for application to DDS, Boc groups of **8**, **25**, **36** could be easily deprotected without cleavage of the glycosidic bond (trifluoroacetic acid (TFA)/0 °C). Utilizing the amino groups, we are presently trying to introduce these branched oligosaccharides to CMPul (M.W. 150 kDa).

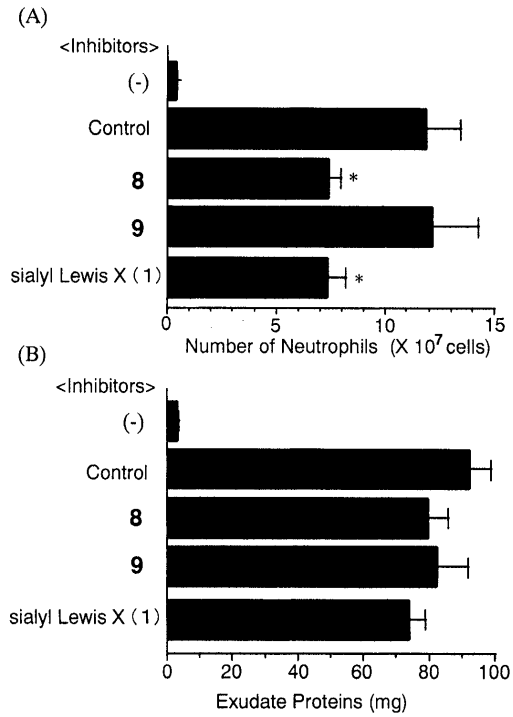


Fig. 3. Protective Effect of SLe^x or SLe^x-Mimetic on (A) Neutrophil Infiltration and (B) Exudate Proteins in Rat Pleuritic Model Induced by Carrageenin

Each column represents mean±S.D. of four to six rats. $*p<0.01$, significantly different from control; (−), sham treatment.

Experimental

All melting points were determined on a Yanagimoto micromelting point apparatus and were not corrected. ^{1}H-NMR spectra were measured on a Varian VXR-500S (500 MHz) spectrometer, unless otherwise specified. ^{1}H-NMR (300 MHz) and ^{13}C-NMR (75 MHz) were measured on a Varian VXR-300S. Chemical shifts (δ) are given in ppm relative to tetramethylsilane as an internal standard. IR spectra were measured on a Shimadzu FT-IR-4300. Optical rotations were determined with a Perkin-Elmer 430 polarimeter. FAB-Mass spectra were recorded on a Hitachi M-90 instrument.

6-Azidohexyl 2,3,4-tri-*O*-Benzyl-α-L-fucopyranoside (11) To a stirred solution of 1-*O*-*p*-nitrobenzoyl 2,3,4-tri-*O*-benzyl-L-fucopyranose[8] (**10**, $\alpha:\beta$ =36 : 64, 100 mg, 0.171 mmol), 6-azidohexanol[9] (37 mg, 0.257 mmol), and $Zn(OTf)_2$ (94 mg, 0.257 mmol) in acetonitrile (2.0 ml) at 0 °C was added TMSCl (33 μl, 0.257 mmol). After being stirred at 0 °C for 1 h, the mixture was diluted with ethyl acetate, washed with brine and saturated aqueous $NaHCO_3$, dried over $MgSO_4$ and concentrated *in vacuo*. The crude product was purified by preparative TLC [hexane/ethyl acetate (6/1, v/v)] to give **11** (60 mg, 64%), and β-glycoside (21 mg, 22%).

11: Colorless oil, $[\alpha]_D^{25}$ −39.0° (c=1.08, $CHCl_3$). IR ($CHCl_3$) cm^{-1}: 2098. ^{1}H-NMR ($CDCl_3$) δ: 1.10 (3H, d, J=6.6 Hz, H-6 of Fuc), 1.34—1.42 (4H, m), 1.54—1.68 (4H, m), 3.24 (2H, t, J=7.0 Hz, $-CH_2N_3$), 3.43 (1H, m), 3.61 (1H, m), 3.66 (1H, d, J=2.9 Hz, H-4 of Fuc), 3.86 (1H, q, J=6.6 Hz, H-5 of Fuc), 3.93 (1H, dd, J=10.0, 2.9 Hz, H-3 of Fuc) 4.03 (1H, dd, J=10.0, 3.7 Hz, H-2 of Fuc), 4.78 (1H, d, J=3.7 Hz, H-1 of Fuc), 4.66, 4.98 (each 1H, d, J=11.5 Hz, $-CH_2Ph$), 4.74, 4.78, 4.81, 4.87 (each 1H, d, J=12.0 Hz, $-CH_2Ph$), 7.24—7.41 (15H, m). ^{13}C-NMR (75 MHz, $CDCl_3$) δ: 16.5 (C-6 of Fuc) 25.6, 26.4, 28.6, 29.2, 51.2 ($-CH_2N_3$), 66.0 (C-5 of Fuc), 67.8 ($-O\underline{C}H_2(CH_2)N_3$), 73.05 ($-OCH_2Ph$), 73.11 ($-OCH_2Ph$), 74.7 ($-OCH_2Ph$), 76.4, 77.7, 79.2, 97.4 (C-1 of Fuc), 127.3, 127.4, 127.8, 128.06, 128.16, 128.22, 138.5, 138.7, 138.9. *Anal.* Calcd for $C_{33}H_{41}N_3O_5$: C, 70.82; H, 7.38; N, 7.51. Found: C, 71,05; H, 7.38; N, 7.23.

β-Glycoside: Colorless oil, $[\alpha]_D^{25}$ +6.2° (c=0.99, $CHCl_3$). IR ($CHCl_3$) cm^{-1}: 2098. ^{1}H-NMR ($CDCl_3$) δ: 1.17 (3H, d, J=6.3 Hz, H-6 of Fuc), 1.32—1.47 (4H, m), 1.52—1.58 (2H, m), 1.58—1.70 (2H, m), 3.21 (2H, t, J=7.1 Hz, $-CH_2N_3$), 3.44 (1H, q, J=6.6 Hz, H-5 of Fuc), 3.46 (1H, m), 3.50 (1H, dd, J=9.8, 3.2 Hz, H-3 of Fuc), 3.55 (1H, br d, H-4 of Fuc), 3.79 (1H, dd, J=9.8, 7.6 Hz, H-2 of Fuc), 3.93 (1H, dt, J=9.3, 6.3 Hz), 4.30 (1H, d, J=7.6 Hz, H-1 of Fuc) 4.70, 4.72, 4.79, 4.97 (each 1H, d, J=12.0 Hz, $-CH_2Ph$), 4.74, 4.93 (each 1H, d, J=11.0 Hz, $-CH_2Ph$), 7.24—7.38 (15H, m). *Anal.* Calcd for $C_{33}H_{41}N_3O_5$: C, 70.82; H, 7.38; N, 7.51. Found: C, 71,01; H, 7.30; N, 7.41.

γ-Benzyl N^2-*tert*-Butyloxycarbonyl-N^1-[1-(2,3,4-tri-*O*-benzyl-α-L-fucopyranosyloxy)-hexyl-6-amino]-L-glutamate-α-amide (13) To a stirred solution of **11** (5.50 g, 10.0 mmol) in methanol (100 ml) was added *p*-TsOH·H_2O (1.90 g, 10.0 mmol), and the mixture was hydrogenated over Lindlar's catalyst (2.40 g) at 3.5×10^4 kg/m^2 (50 psi) for 3 h. The catalyst was filtered off and the solvent was removed *in vacuo*, giving 6-aminohexyl 2,3,4-tri-*O*-benzyl-α-L-fucopyranoside *p*-toluenesulfonate (6.80 g) as a pale brown viscous oil.

To a solution of 6-aminohexyl 2,3,4-tri-*O*-benzyl-α-L-fucopyranoside *p*-toluenesulfonate (6.80 g) in dichloromethane (50 ml) were added Boc-Glu(OBzl)-OSu (4.78 g, 11.0 mmol) and *N*-methylmorpholine (1.21 ml, 11.0 mmol). After being stirred at 0 °C for 1 h, the mixture was diluted with dichloromethane, washed successively with H_2O, 10% aqueous citric acid, and H_2O, dried over $MgSO_4$ and concentrated *in vacuo*. The crude product was purified by silica gel column chromatography (SiO_2 330 g, hexane : ethyl acetate : methanol=70 : 30 : 1, v/v) to give **13** (6.70 g, 79%). mp 100—103 °C (colorless needle from $CHCl_3$–hexane). $[\alpha]_D^{25}$ −29.6° (c=1.09, $CHCl_3$). IR (KBr) cm^{-1}: 1728, 1686, 1659. ^{1}H-NMR ($CDCl_3$) δ: 1.10 (3H, d, J=6.6 Hz, H-6 of Fuc), 1.27—1.38 (4H, m), 1.43—1.49 (2H, m), 1.43 (9H, s), 1.54—1.62 (2H, m), 1.91 (1H, m), 2.12 (1H, m), 2.42 (1H, m), 2.55 (1H, m), 3.18—3.22 (2H, m), 3.42 (1H, dt, J=10.0, 6.8 Hz), 3.57 (1H, dt, J=10.0, 6.8 Hz), 3.86 (1H, q, J=6.6 Hz, H-5 of Fuc), 3.93 (1H, dd, J=10.0, 2.9 Hz, H-3 of Fuc), 4.02 (1H, dd, J=10.0, 3.7 Hz, H-2 of Fuc), 4.10 (1H, m), 4.65, 4.98 (1H, each d, J=11.5 Hz, $-CH_2Ph$), 4.67, 4.74, 4.81, 4.88 (1H, each d, J=11.7 Hz, $-CH_2Ph$), 4.77 (1H, d, J=3.7 Hz, H-1 of Fuc) 5.11, 5.14 (1H, each d, J=12.7 Hz, $-CH_2Ph$), 5.23 (1H, br s), 6.13 (1H, br s), 7.41—7.25 (20H, m). ^{13}C-NMR (75 MHz, $CDCl_3$) δ: 16.6 (C-6 of Fuc), 25.7, 26.5, 28.0, 28.2 ($-NHCO_2C(\underline{C}H_3)_3$), 29.2, 29.3, 30.4, 39.3 ($-\underline{C}H_2NHCO$), 53.8 (α-C of Glu), 66.0 (C-5 of Fuc), 66.4 ($-CO_2\underline{C}H_2Ph$), 67.9 ($-O\underline{C}H_2(CH_2)_5N_3$), 73.06 ($-O\underline{C}H_2Ph$), 73.14 ($-O\underline{C}H_2Ph$), 74.7 ($-O\underline{C}H_2Ph$), 76.4, 77.7, 79.3, 79.9 ($-NHCO_2\underline{C}(CH_3)$), 97.4 (C-1 of Fuc), 127.3, 127.5, 127.8, 128.07, 128.14, 128.18, 128.21, 128.25, 128.3, 128.5, 135.6, 138.5, 138.7, 138.9, 155.6 ($-NH\underline{C}O_2C(CH_3)_3$), 171.2, 173.1. *Anal.* Calcd for $C_{50}H_{64}N_2O_{10}$: C, 70.40; H, 7.56; N, 3.28. Found: C, 70.35; H, 7.50 ; N, 3.46 .

N^2-*tert*-Butyloxycarbonyl-N^1-[1-(2,3,4-tri-*O*-benzyl-α-L-fucopyranosyloxy) hexyl-L-glutamate-α-amide (14) To a solution of **13** (360 mg, 0.422 mmol) in ethanol (5 ml) was added 10%Pd–C (360 mg) and 1,4-cyclohexadiene (650 μl, 6.87 mmol).[12] After being stirred for 1 h at room temperature, the catalyst was filtered off. The filtrates were concentrated *in vacuo* to give **14** (282 mg, 88%) as colorless powder. ^{1}H-NMR ($CDCl_3$) δ: 1.11 (3H, d, J=6.6 Hz, H-6 of Fuc), 1.29—1.36 (4H, m), 1.42 (9H, s), 1.45—1.52 (2H, m), 1.54—1.64 (2H, m), 1.91, 2.03, 2.35, 2.47 (each 1H, m), 3.16—3.28 (2H, m), 3.41 (1H, ddd, J=9.9, 6.6, 6.6 Hz), 3.57 (1H, ddd, J=9.9, 7.1, 7.1 Hz), 3.67 (1H, br d, H-4 of Fuc), 3.86 (1H, q, J=6.6 Hz, H-5 of Fuc), 3.93 (1H, dd, J=10.3, 3.0 Hz, H-3 of Fuc), 4.02 (1H, dd, J=10.3, 3.8 Hz, H-2 of Fuc), 4.21 (1H, m, α-H of Glu), 4.67, 4.74, 4.80, 4.84 (each 1H, d, J=11.8 Hz, $-CH_2Ph$), 4.63, 4.96 (each 1H, d, J=11.8 Hz, $-CH_2Ph$), 4.77 (1H, J=3.8 Hz, H-1 of Fuc), 5.49 (1H, br s), 6.73 (1H, br s), 7.24—7.39 (15H, m).

6-Azidohexyl *O*-(Methyl 5-Acetamido-4,7,8,9-tetra-*O*-acetyl-3,5-dideoxy-D-*glycero*-α-D-*galacto*-2-nonulopyranosylonate)-(2→3)-2,4-di-*O*-acetyl-6-*O*-benzoyl-β-D-galactopyranoside (18) To a solution of 6-azidohexanol[10] (131 mg, 0.912 mmol) and **17**[13] (300 mg, 0.304 mmol) in dichloromethane (10 ml), was added activated molecular sieves 4A (MS4A, 4 g). The mixture was stirred at room temperature for 2 h and then cooled to 0 °C. To the solution was added $BF_3\cdot OEt_2$ (112 μl, 0.912 mmol), and the stirring was continued at 0 °C for 1 h. The reaction mixture was diluted with dichloromethane, and filtered though a Celite bed. The filtrates were washed with saturated aqueous $NaHCO_3$, dried over $MgSO_4$ and concentrated *in vacuo*. The crude product was purified by silica gel column chromatography (SiO_2 20 g, dichloromethane : methanol=50 : 1, v/v) to give **18** (230 mg, 79%) as a colorless powder. $[\alpha]_D^{27}$ −17.8° (c=1.03, $CHCl_3$). IR ($CHCl_3$) cm^{-1}: 2100, 1744, 1690. ^{1}H-NMR ($CDCl_3$) δ: 1.33—1.42 (4H, m), 1.54—1.67 (4H, m), 1.72 (1H, dd, J=12.7, 12.3 Hz, H-3 of Sia), 1.85, 2.00, 2.04, 2.08, 2.13, 2.21 (each 3H, s), 2.59 (1H, dd, J=12.7, 4.6 Hz, H-3 of Sia), 3.25 (2H, t, J=7.0 Hz), 3.54 (1H, dt, J=9.5, 6.6 Hz), 3.63 (1H, dd, J=10.7, 2.4 Hz, H-6 of Sia), 3.76 (3H, s), 3.88 (1H, dt, J=9.5, 6.3 Hz), 3.99 (1H, dd, J=7.1, 6.3 Hz, H-5 of Gal), 4.00 (1H, dd, J=12.5, 5.6 Hz, H-9 of Sia), 4.04 (1H, m, H-5 of Sia), 4.21 (1H, dd, J=11.0, 7.1 Hz, H-6 of Gal), 4.35 (1H, dd, J=12.5, 2.4 Hz, H-9 of Sia), 4.43 (1H, dd, J=11.0, 6.3 Hz, H-6 of Gal), 4.58 (1H, dd, J=10.0, 3.1 Hz, H-3 of Gal), 4.61 (1H, d, J=8.0 Hz, H-1 of Gal), 4.88 (1H, m, H-4 of Sia), 5.03—5.08 (3H, m), 5.37 (1H, dd, J=9.5,

2.4 Hz, H-7 of Sia), 5.56 (1H, ddd, J=9.5, 5.6, 2.7 Hz, H-8 of Sia), 7.23 (2H, m), 7.56 (1H, m), 8.03 (2H, m). *Anal.* Calcd for $C_{43}H_{58}N_4O_{21}\cdot H_2O$: C, 52.44; H, 6.14; N, 5.69. Found: C, 52.43; H, 6.10; N, 5.52.

N^2-*tert*-Butyloxycarbonyl-N^5-[6-{*O*-(methyl 5-acetamido-4,7,8,9-tetra-*O*-acetyl-3,5-dideoxy-D-*glycero*-α-D-*galacto*-2-nonulopyranosylonate)-(2→3)-2,4-di-*O*-acetyl-6-*O*-benzoyl-β-D-galactopyranosyl}hexyl]-N^1-[6-(2,3,4-tri-*O*-benzyl-α-L-fucopyranosyloxy)-hexyl]-L-glutamin-α-amide (20) To a stirred solution of **18** (130 mg, 0.136 mmol) in methanol (10 ml) was added p-TsOH·H_2O (26 mg, 0.136 mmol), and the mixture was hydrogenated over Lindlar's catalyst (100 mg) at 3.5×10^4 kg/m^2 (50 psi) for 2 h. The catalyst was filtered off and the solvent was removed *in vacuo*, giving 6-aminohexyl *O*-(methyl 5-acetamido-4,7,8,9-tetra-*O*-acetyl-3,5-dideoxy-D-*glycero*-α-D-*galacto*-2-nonulopyranosylonate)-(2→3)-2,4-di-*O*-acetyl-6-*O*-benzoyl-β-D-gatoctopyranoside p-toluenesulfonate (123 mg) as a pale brown viscous oil.

To a solution of **14** (125 mg) in dichloromethane (1.5 ml) were added HOSu (17.5 mg, 0.152 mmol), and DCC (31.0 mg, 0.150 mmol). After being stirred at 0 °C for 3 h, 6-aminohexyl *O*-(methyl 5-acetamido-4,7,8,9-tetra-*O*-acetyl-3,5-dideoxy-D-*glycero*-α-D-*galacto*-2-nonulopyranosylonate)-(2→3)-2,4-di-*O*-acetyl-6-*O*-benzoyl-β-D-gatoctopyranoside p-toluenesulfonate (123 mg), and triethylamine (25 μl, 0.179 mmol) were added, and the stirring was continued at 0 °C for 12 h. After the precipitates were filtered off, the filtrates were diluted with dichloromethane, washed with saturated aqueous $NaHCO_3$, dried over $MgSO_4$ and concentrated *in vacuo*. The crude product was purified by silica gel column chromatography (SiO_2 20 g, toluene : acetone=7 : 4, v/v) to give **20** (93 mg, 41%) as a colorless powder.

$[\alpha]_D^{27}$ −26.2° (c=1.06, $CHCl_3$). IR ($CHCl_3$) cm^{-1}: 1744, 1690, 1676. ^{1}H-NMR ($CDCl_3$) δ: 1.11 (3H, d, J=6.3 Hz, H-6 of Fuc), 1.40—1.30 (8H, m), 1.43 (9H, s), 1.46—1.52 (4H, m), 1.58—1.65 (4H, m), 1.72 (1H, dd, J= 12.7, 12.5 Hz, H-3 of Sia), 1.85, 2.01, 2.05, 2.09, 2.11, 2.13, 2.22 (each 3H, s), 2.26 (1H, m), 2.35 (1H, m), 2.59 (1H, dd, J=12.7, 4.6 Hz, H-3 of Sia), 3.15—3.31 (4H, m), 3.42 (1H, dt, J=10.0, 6.3 Hz), 3.53 (1H, dt, J=9.5, 6.8 Hz), 3.57 (1H, dt, J=10.0, 7.1 Hz), 3.64 (1H, dd, J=10.7, 2.7 Hz, H-6 of Sia), 3.67 (1H, d-like, H-4 of Fuc), 3.77 (3H, s), 3.89—3.84 (2H, m, H-5 of Fuc, H-5 of Sia), 3.93 (1H, dd, J=10.0, 2.7 Hz, H-3 of Fuc), 3.98—4.10 (5H, m), 4.21 (1H, dd, J=11.0, 7.1 Hz, H-6 of Gal), 4.35 (1H, dd, J=12.4, 2.4 Hz, H-9 of Sia), 4.44 (1H, dd, J=11.0, 6.6 Hz, H-6 of Gal), 4.59 (1H, dd, J=10.2, 3.4 Hz, H-3 of Gal), 4.61 (1H, d, J=8.1 Hz, H-1 of Gal), 4.65, 4.74, 4.87, 4.97 (each 1H, d, J=12.0 Hz, –CH_2Ph), 4.66, 4.81 (each 1H, d, J=12.0 Hz, –CH_2Ph), 4.77 (1H, d, J=3.4 Hz, H-1 of Fuc), 4.89 (1H, m, H-4 of Sia), 5.05 (1H, dd, J=10.2, 8.1 Hz, H-2 of Gal), 5.06 (1H, d, J=3.4 Hz, H-4 of Gal), 5.09 (1H, br d, J=11.0 Hz, NH), 5.36 (1H, dd, J=9.3, 2.7 Hz, H-7 of Sia), 5.57 (1H, ddd, J=9.3, 5.8, 2.4 Hz, H-8 of Sia), 5.73 (1H, br s), 6.07 (1H, br s), 6.67 (1H, m), 7.14—7.45 (17H, m), 7.57 (1H, m), 8.03 (2H, d, J=7.1 Hz). *Anal.* Calcd for $C_{86}H_{116}N_4O_{30}$: C, 61.27; H, 6.94; N, 3.32. Found: C, 61.01; H, 7.01; N, 3.13.

N^5-[6-{*O*-(5-Acetamido-3,5-dideoxy-D-*glycero*-α-D-*galacto*-2-nonulopyranosylonic acid)-(2→3)-β-D-galactopyranosyl}hexyl]-N^2-*tert*-butyloxycarbonyl-N^1-(6-α-L-fucopyranosyloxyhexyl)-L-glutamin-α-amide (2) To a solution of **20** (118 mg, 0.070 mmol) in methanol (2 ml) at 0 °C was added 3% sodium methoxide in methanol (400 μl), and stirring was continued at 0 °C for 2 h. The mixture was neutralized with Dowex 50W-X8 (hydrogen form), filtered, and concentrated *in vacuo*.

The residue was dissolved in 1,4-dioxane, and then to the solution was added 0.1 N sodium hydroxide (2.0 ml). After being stirred at room temperature for 1 h, the mixture was neutralized with Dowex 50W-X8 (hydrogen form), filtered, and concentrated *in vacuo*. Column chromatography of the residue on Sephadex LH-20 (90 g, methanol) gave N^5-[6-{*O*-(5-Acetamido-3,5-dideoxy-D-*glycero*-α-D-*galacto*-2-nonulopyranosylonic acid)-(2→3)-β-D-galactopyranosyl}hexyl]-N^2-*tert*-butyloxycarbonyl-N^1-[6-(α-L-2,3,4-tri-*O*-benzyl-α-L-fucopyranosyloxy)-hexyl]-L-glutamin-α-amide (89 mg, 97%) as a colorless powder. $[\alpha]_D^{27}$ −23.1° (c=1.05, $CHCl_3$). IR ($CHCl_3$) cm^{-1}: 3422, 1697. ^{1}H-NMR (CD_3OD) δ: 1.13 (3H, d, J=6.3 Hz, H-6 of Fuc), 1.43 (9H, s), 1.30—1.46 (8H, m), 1.46—1.54 (4H, m), 1.55—1.65 (4H, m), 1.79—1.88 (2H, m), 2.00 (3H, s), 2.00 (1H, m), 2.23—2.27 (2H, m), 2.81 (1H, br d, J=12.9 Hz, H-3 of Sia), 3.12—3.24 (4H, m), 3.26—4.04 (21H, m), 4.26 (1H, d, J=7.8 Hz, H-1 of Gal), 4.61, 4.90 (each 1H, d, J=11.2 Hz), 4.66, 4.75(each 1H, d, J=11.7 Hz, –CH_2Ph), 4.77 (2H, s), 4.79 (1H, d, J=2.4 Hz, H-1 of Fuc), 7.24—7.41 (15H, m).

To a solution of N^5-[6-{*O*-(5-Acetamido-3,5-dideoxy-D-*glycero*-α-D-*galacto*-2-nonulopyranosylonic acid)-(2→3)-β-D-galactopyranosyl}hexyl]-N^2-*tert*-butyloxycarbonyl-N^1-[6-(α-L-2,3,4-tri-*O*-benzyl-α-L-fucopyranosyloxy)-hexyl]-L-glutamin-α-amide (70 mg) in methanol (7 ml) was added palladium on activated carbon (Pd 10%, 70 mg). Hydrogenation was carried out at 3.5×10^4 kg/m^2 (50 psi) hydrogen pressure for 12 h. The catalyst was filtered off and the filtrates were concentrated *in vacuo*. Column chromatography of the residue on Sephadex LH-20 (90 g, methanol) gave **2** (49 mg, 88%) as a colorless powder. $[\alpha]_D^{27}$ −31.0° (c=0.51, MeOH). IR (KBr) cm^{-1}: 3422, 1697. ^{1}H-NMR (D_2O) δ: 1.22 (3H, d, J=6.6 Hz, H-6 of Fuc), 1.30—1.43 (8H, m), 1.44 (9H, s), 1.49—1.57 (4H, m), 1.60—1.67 (4H, m), 1.81 (1H, dd, J=12.5, 12.2 Hz, H-3 of Sia), 1.89 (1H, m), 2.04 (3H, s), 2.05 (1H, m), 2.30—2.39 (2H, t-like), 2.77 (1H, dd, J=12.5, 4.6 Hz, H-3 of Sia), 3.14—3.22 (4H, m), 3.25 (1H, m), 3.53 (1H, m), 3.54 (1H, dd, J=10.0, 8.1 Hz, H-2 of Gal), 3.96 (1H, d, J=3.0 Hz, H-4 of Gal), 4.07 (1H, m), 4.09 (1H, dd, J=10.0, 3.0 Hz, H-3 of Gal), 4.46 (1H, d, J=8.1 Hz, H-1 of Gal), 4.87 (1H, d, J=3.7 Hz, H-1 of Fuc). *Anal.* Calcd for $C_{45}H_{80}N_4O_{23}\cdot 3H_2O$: C, 49.17; H, 7.89; N, 5.10. Found: C, 48.92; H, 7.58; N, 5.18.

8-Azido-3,6-dioxaoctyl 2,3,4-tri-*O*-benzyl-α-L-fucopyranoside (12) Following the procedure described for **11**, 1-*O*-p-nitrobenzoyl 2,3,4-tri-*O*-benzyl-L-fucopyranose (**10**) was converted to **12** (**12**: 61%, β-glycoside: 21%).

12: Colorless oil, $[\alpha]_D^{23}$ −34.1° (c=1.00, $CHCl_3$). IR ($CHCl_3$) cm^{-1}: 2108. ^{1}H-NMR ($CDCl_3$) δ: 1.10 (3H, d, J= 6.6 Hz, H-6 of Fuc), 3.34 (2H, t, J= 5.0 Hz, –CH_2N_3), 3.58—3.78 (11H, m), 3.93 (1H, q, J= 6.6 Hz, H-5 of Fuc), 3.94 (1H, dd, J=10.3, 2.9 Hz, H-3 of Fuc), 4.03 (1H, dd, J=10.3, 3.7 Hz, H-2 of Fuc), 4.65, 4.98 (each 1H, d, J= 11.5 Hz, –CH_2Ph), 4.69, 4.74, 4.80, 4.86 (each 1H, d, J=12.0 Hz, –CH_2Ph), 4.86 (1H, d, J=3.7 Hz, H-1 of Fuc), 7.24—7.41 (15H, m, aromatic). *Anal.* Calcd for $C_{33}H_{41}N_3O_7$: C, 66.99; H, 6.98; N, 7.10. Found: C, 67.35; H, 7.03; N, 6.84.

β-Glycoside: Colorless oil, $[\alpha]_D^{20}$ +8.5° (c=0.65, $CHCl_3$). IR ($CHCl_3$) cm^{-1}: 2108. ^{1}H-NMR ($CDCl_3$) δ: 1.17 (3H, d, J=6.3 Hz, H-6 of Fuc), 3.31 (2H, t, J= 5.1 Hz, CH_2N_3), 3.44 (1H, q-like, H-5 of Fuc), 3.50 (1H, dd, J=9.5, 2.9 Hz, H-3 of Fuc), 3.55 (1H, br d, H-4 of Fuc), 3.56—3.61 (4H, m), 3.63—3.66 (2H, m), 3.67—3.76 (3H, m), 3.80 (1H, dd, J=9.5, 7.6 Hz, H-2 of Fuc), 4.03 (1H, m), 4.36 (1H, d, J=7.6 Hz, H-1 of Fuc), 4.69, 4.79 (each 1H, d, J=12.0 Hz, –CH_2Ph), 4.71, 4.97 (each 1H, d, J=11.7 Hz, –CH_2Ph), 4.75, 4.96 (each 1H, d, J=11.0 Hz, –CH_2Ph), 7.24—7.40 (15H, m, aromatic). *Anal.* Calcd for $C_{33}H_{41}N_3O_7\cdot 0.2H_2O$: C, 66.58; H, 7.01; N, 7.06. Found: C, 66.53; H, 6.88; N, 7.09.

γ-Benzyl N^2-*tert*-Butyloxycarbonyl-N^1-[8-(2,3,4-tri-*O*-benzyl-α-L-fucopyranosyloxy)-3,6-dioxaoctyl]-L-glutamate-α-amide (15) Following the procedure described for **13**, **12** was converted to **15** (92%). Colorless oil, $[\alpha]_D^{27}$ −44.8° (c=0.92, $CHCl_3$). ^{1}H-NMR ($CDCl_3$) δ: 1.10 (3H, d, J=6.6 Hz, H-6 of Fuc), 1.91 (1H, m), 2.12 (1H, m), 2.41 (1H, ddd, J=16.6, 7.8, 6.3 Hz), 2.49 (1H, ddd, J=16.6, 7.5, 7.5 Hz), 3.38—3.44 (2H, m), 3,50 (2H, t, J=5.1 Hz), 3.54 (1H, t, J=4.6 Hz), 3.70—3.59 (5H, m), 3.73 (1H, m), 3.91 (1H, q, J=6.6 Hz, H-5 of Fuc), 3.94 (1H, dd, J=10.0, 2.9 Hz, H-3 of Fuc), 4.03 (1H, dd, J=10.0, 3.7 Hz, H-2 of Fuc), 4.15 (1H, m, α-H of Glu), 4.64, 4.97 (each 1H, d, J=11.5 Hz, –CH_2Ph), 4.69, 4.86 (each 1H, d, J=12.2 Hz, –CH_2Ph), 4.73, 4.81 (each 1H, d, J=12.0 Hz, –CH_2Ph), 4.84 (1H, d, J=3.7 Hz, H-1 of Fuc), 5.10 (2H, s), 5.28 (1H, br d, NH), 6.76 (1H, br t, NH), 7.24—7.41 (20H, m). *Anal.* Calcd for $C_{33}H_{41}N_3O_7$: C, 67.85; H, 7.29; N, 3.19. Found: C, 67.98; H, 7.31; N, 3.19.

N^2-*tert*-Butyloxycarbonyl-N^1-[8-(2,3,4-tri-*O*-benzyl-α-L-fucopyranosyloxy)-3,6-dioxaoctyl]-L-glutamic-α-amide (16) Following the procedure described for **17**, **15** was converted to **16** (90%). Colorless syrup, ^{1}H-NMR (300 MHz, CD_3OD) δ: 1.14 (3H, d, J=6.6 Hz, H-6 of Fuc), 1.43 (9H, s), 1.85, 2.00 (each 1H, m), 2.36 (2H, t-like), 3.42—3.80 (12H, m), 3.81 (1H, br s, H-4 of Fuc), 3.95 (2H, m), 3.98 (1H, q, J=6.6 Hz, H-5 of Fuc), 4.05 (1H, m, α-H of Glu), 4.61, 4.90 (each 1H, d, J=11.4 Hz, –CH_2Ph), 4.72 (1H, J=4.0 Hz, H-1 of Fuc), 4.77 (2H, s), 7.20—7.43 (15H, m).

8-Azido-3,6-dioxaoctyl *O*-(Methyl 5-Acetamido-4,7,8,9-tetra-*O*-acetyl-3,5-dideoxy-D-*glycero*-α-D-*galacto*-2-nonulopyranosylonate)-(2→3)-2,4-di-*O*-acetyl-6-*O*-benzoyl-β-D-galactopyranoside (19) Following the procedure described for **18**, **16** was converted to **19** (73%). Colorless powder, $[\alpha]_D^{27}$ −17.0° (c=1.03, $CHCl_3$). IR ($CHCl_3$) cm^{-1}: 2100, 1744, 1690. ^{1}H-NMR ($CDCl_3$) δ: 1.86, 2.01, 2.06, 2.09, 2.12, 2.13, 2.22 (each 3H, s), 1.72 (1H, dd, J=12.7, 12.3 Hz, H-3 of Sia), 2.59 (1H, dd, J=12.7, 4.6 Hz, H-3 of Sia), 3.39 (2H, t, J=5.0 Hz), 3.61—3.71 (9H, m), 3.77 (3H, s), 3.77 (1H, m), 3.98—4.08 (4H, m), 4.22 (1H, dd, J=11.2, 6.8 Hz, H-6 of Gal), 4.35 (1H, dd, J=12.7, 2.7 Hz, H-9 of Sia), 4.43 (1H, dd, J=11.2, 6.6 Hz, H-6 of Gal), 4.61 (1H, dd, J=10.3, 3.4 Hz, H-3 of Gal), 4.69 (1H, d, J=8.1 Hz, H-1 of Gal), 4.89 (1H, m, H-4 of Sia), 5.06 (1H, d-like, H-4 of Gal), 5.07 (1H, dd, J=10.3, 8.1 Hz, H-2 of Gal), 5.09 (1H, br d, J=11.5 Hz, NH), 5.38 (1H, dd, J=9.3, 2.7 Hz, H-7 of Sia), 5.56 (1H, ddd, J=9.3, 5.4, 2.7 Hz, H-8 of Sia), 7.44 (1H, m), 7.57 (1H, m), 8.03 (2H, d, J=7.0 Hz). *Anal.* Calcd for $C_{43}H_{58}N_4O_{23}\cdot 0.5H_2O$: C, 51.24; H, 5.90; N, 5.56. Found: C, 51.19; H, 5.78; N, 5.38.

N^2-*tert*-Butyloxycarbonyl-N^5-[8-{*O*-(methyl 5-acetamido-4,7,8,9-tetra-

***O*-acetyl-3,5-dideoxy-D-*glycero*-α-D-*galacto*-2-nonulopyranosylonate)-(2→3)-2,4-di-*O*-acetyl-6-*O*-benzoyl-β-D-galactopyranosyl}-3,6-dioxaoctyl]-N^1-[8-(2,3,4-tri-*O*-benzyl-α-L-fucopyranosyloxy)-3,6-dioxaoctyl]-L-glutamin-α-amide (21)** Following the procedure described for **20**, **19** was converted to **21** (73%). Colorless powder, $[\alpha]_D^{25}$ −38.5° (c=0.50, $CHCl_3$). IR ($CHCl_3$) cm^{-1}: 3428, 1742, 1673 . ^{1}H-NMR (300 MHz, $CDCl_3$) δ: 1.10 (3H, d, J=6.6 Hz, H-6 of Fuc), 1.42 (9H, s), 1.71 (1H, dd, J=12.6, 12.6 Hz, H-3 of Sia), 1.85, 2.01, 2.05, 2.09, 2.11, 2.13, 2.22 (each 3H, s), 2.30 (2H, m), 2.60 (1H, dd, J=12.6, 4.2 Hz, H-3 of Sia), 3.30—3.54 (4H, m), 3.46—3.84 (20H, m), 3.77 (3H, s), 3.91 (1H, q, J=6.6 Hz, H-5 of Fuc), 3.93—4.10 (7H, m), 4.11 (1H, m), 4.21 (1H, dd, J=11.0, 7.0 Hz, H-6 of Gal), 4.35 (1H, dd, J=12.3, 2.7 Hz, H-9 of Sia), 4.44 (1H, dd, J=11.0, 6.6 Hz, H-6 of Gal), 4.61 (1H, dd, J=10.2, 3.3 Hz, H-3 of Gal), 4.64, 4.97 (each 1H, d, J=11.7 Hz, $-CH_2Ph$), 4.67 (1H, d, J=7.5 Hz, H-1 of Gal), 4.68, 4.74, 4.81, 4.86 (each 1H, d, J=12.0 Hz, $-CH_2Ph$), 4.84 (1H, d, J=4.0 Hz, H-1 of Fuc), 4.89 (1H, m, H-4 of Sia), 5.06 (1H, dd, J=10.2, 7.5 Hz, H-2 of Gal), 5.06 (1H, br s, H-4 of Gal), 5.17 (1H, br d, J=10.8 Hz, NH), 5.38 (1H, dd, J=9.3, 2.7 Hz, H-7 of Sia), 5.56 (1H, ddd, J=9.3, 5.4, 2.7 Hz, H-8 of Sia), 5.70 (1H, m), 5.76 (1H, t-like), 7.18—7.48 (17H, m), 7.57 (1H, m), 8.03 (2H, m). *Anal.* Calcd for $C_{86}H_{116}N_4O_{34}\cdot 1.5H_2O$: C, 58.13; H, 6.75; N, 3.15. Found: C, 58.13; H, 6.62 ; N, 3.19.

N^5-[8-{*O*-(5-Acetamido-3,5-dideoxy-D-*glycero*-α-D-*galacto*-2-nonulopyranosyl)-(2→3)-β-D-galactopyranosyl}-3,6-dioxaoctyl]-N^2-*tert*-butyloxycarbonyl-N^1-[8-α-L-fucopyranosyloxy-3,6-dioxaoctyl]-L-glutamin-α-amide (3) Following the procedure described for **2**, **21** was converted to **3** (93%). Colorless powder, $[\alpha]_D^{27}$ −18.2° (c=0.53, MeOH). ^{1}H-NMR (D_2O) δ: 1.19 (3H, d, J=6.6 Hz, H-6 of Sia), 1.41 (9H, s), 1.79 (1H, dd, J=12.3, 12.0 Hz, H-3 of Sia), 1.88 (1H, m), 2.01 (3H, s), 2.02 (1H, m), 2.34 (2H, m), 2.73 (1H, dd, J=12.3, 4.6 Hz, H-3 of Sia), 3.35—3.43 (3H, m), 3.52—3.98 (37H, m), 4.03—4.10 (2H, m), 4.47 (1H, d, J=7.8 Hz, H-1 of Gal), 4.87 (1H, d, J=3.7 Hz, H-1 of Fuc). *Anal.* Calcd for $C_{45}H_{80}N_4O_{27}\cdot 2H_2O$: C, 47.20; H, 7.39; N, 4.89. Found: C, 47.11; H, 7.46 ; N, 4.79.

Methyl (6-Azidohexyl 5-Acetamido-4,7,8,9-tetra-*O*-acetyl-3,5-dideoxy-2-*O*-D-*glycero*-α-D-*galacto*-2-nonulopyranosid)onate (23) and Methyl (6-azidohexyl 5-acetamido-4,7,8,9-tetra-*O*-acetyl-3,5-dideoxy-2-*O*-D-glycero-β-D-galacto-2-nonulopyranosid)onate (24) To a solution of 6-azidohexanol[10] (344 mg, 2.40 mmol) and **22**[14] (609 mg, 1.20 mmol), was added activated molecular sieves 3A (MS3A, 1.5 g). The mixture was stirred at room temperature for 2 h and then cooled to −40 °C. To the solution were added NIS (542 mg, 2.40 mmol), and TfOH (22 μl, 0.240 mmol), and the stirring was continued at −40 °C for 1 h. The reaction mixture was diluted with dichloromethane, and then filtered. The filtrates were washed with saturated aqueous $Na_2S_2O_5$, and saturated aqueous $NaHCO_3$, successively, dried over $MgSO_4$ and concentrated *in vacuo*.

The crude product was purified by silica gel column chromatography (SiO_2 200 g, dichloromethane : methanol=75 : 1, v/v) to give **23** (370 mg, 51%) and **24** (170 mg, 24%).

23: mp 84—85.5 °C (colorless needles from $CHCl_3$–hexane). $[\alpha]_D^{27}$ −13.8° (c=1.15, $CHCl_3$). IR (KBr) cm^{-1}: 2100, 1749, 1690, 1659. ^{1}H-NMR ($CDCl_3$) δ: 1.34—1.43 (4H, m), 1.52—1.64 (4H, m), 1.88, 2.03, 2.04, 2.14, 2.15 (each 3H, s), 1.95 (1H, dd, J=12.9, 12.4 Hz, H-3 of Sia), 2.58 (1H, dd, J=12.9, 4.6 Hz, H-3 of Sia), 3.22 (1H, dt, J=9.3, 6.6 Hz), 3.27 (2H, t, J=7.1 Hz), 3.76 (1H, dt, J=9.3, 6.3 Hz), 3.80 (3H, s), 4.04—4.12 (3H, m, H-5, 6, 9 of Sia), 4.31 (1H, dd, J=12.5, 2.7 Hz, H-9 of Sia), 4.84 (1H, ddd, J=12.4, 9.8, 4.6 Hz, H-4 of Sia), 5.12 (1H, br d, J=8.1 Hz, NH), 5.33 (1H, dd, J=8.5, 2.0 Hz, H-7 of Sia), 5.39 (1H, ddd, J=8.5, 5.6, 2.7 Hz, H-8 of Sia). ^{13}C-NMR (75 MHz, $CDCl_3$) δ: 20.6, 20.7, 20.9, 22.9, 25.2, 26.2, 28.6, 29.1, 37.9 (C-3 of Sia), 49.1 (C-5 of Sia), 51.1 ($-O-(CH_2)_5-\underline{C}H_2N_3$), 52.4 ($-CO_2\underline{C}H_3$), 62.3 (C-9 of Sia), 64.6 ($-O-\underline{C}H_2-(CH_2)_5-N_3$), 67.3 (C-7 of Sia), 68.7 (C-8 of Sia), 69.1 (C-4 of Sia), 72.3 (C-6 of Sia), 98.6 (C-2 of Sia), 168.4, 170.0, 170.2, 170.5, 170.9. *Anal.* Calcd for $C_{26}H_{40}N_4O_{13}$: C, 50.64; H, 6.54; N, 9.09. Found: C, 50.46; H, 6.52; N, 8.91.

24: Colorless syrup, $[\alpha]_D^{27}$ −12.3° (c=1.01, $CHCl_3$). IR (KBr) cm^{-1}: 2099, 1746, 1688. ^{1}H-NMR ($CDCl_3$) δ: 1.38—1.50 (4H, m), 1.52—1.69 (4H, m), 1.86 (1H, dd, J=12.9, 11.7 Hz, H-3 of Sia), 1.88, 2.02, 2.03, 2.07, 2.15 (each 3H, s), 2.46 (1H, dd, J=12.9, 4.8 Hz, H-3 of Sia), 3.30 (2H, t, J=7.0 Hz), 3.34 (1H, dt, J=9.6, 6.3 Hz), 3.59 (1H, dt, J=9.6, 6.3 Hz), 3.80 (3H, s), 3.93 (1H, dd, J=10.5, 2.4 Hz, H-6 of Sia), 4.10 (1H, dd, J=12.5, 7.8 Hz, H-9 of Sia), 4.11 (1H, ddd, J=10.5, 10.5, 10.5 Hz, H-5 of Sia), 4.81 (1H, dd, J=12.5, 2.4 Hz, H-9 of Sia), 5.19 (1H, ddd, J=7.8, 3.3, 2.4 Hz, H-8 of Sia), 5.26 (1H, ddd, J=11.7, 10.5, 4.8 Hz, H-4 of Sia), 5.37 (1H, br d, J=10.5 Hz, NH), 5.39 (1H, dd, J=3.3, 2.4 Hz, H-8 of Sia). *Anal.* Calcd for $C_{26}H_{40}N_4O_{13}\cdot 0.5H_2O$: C, 49.92; H, 6.61; N, 8.96. Found: C, 50.03; H, 6.39; N, 8.91.

N^2-*tert*-Butyloxycarbonyl-N^5-[6-*O*-(methyl 5-acetamido-4,7,8,9-tetra-*O*-acetyl-3,5-dideoxy-D-glycero-α-D-galacto-2-nonulopyranosylonate)-oxyhexyl]-N^1-[6-(2,3,4-tri-*O*-benzyl-α-L-fucopyranosyloxy)hexyl]-L-glutamin-α-amide (25) Following the procedure described for **20**, **23** was converted to **25**. (58%) Colorless powder, $[\alpha]_D^{27}$ −23.3° (c=1.09, $CHCl_3$). IR ($CHCl_3$): 1745, 1672 cm^{-1}. ^{1}H-NMR ($CDCl_3$) δ: 1.10 (3H, d, J=6.6 Hz, H-6 of Fuc), 1.29—1.40 (8H, m), 1.43 (9H, s), 1.45—1.71 (8H, m), 1.88, 2.03, 2.04, 2.14, 2.15 (each 3H, s), 1.94 (1H, dd, J=12.7, 12.5 Hz, H-3 of Sia), 2.26 (1H, ddd, J=13.6, 7.6, 5.4 Hz), 2.35 (1H, ddd, J=13.6, 8.0, 5.6 Hz), 2.57 (1H, dd, J=12.7, 4.6 Hz, H-3 of Sia), 3.16—3.30 (5H, m), 3.42 (1H, dt, J=10.0, 6.6 Hz), 3.57 (1H, dt, J=10.0, 6.8 Hz), 3.67 (1H, br d, H-4 of Fuc), 3.74 (1H, dt, J=9.5, 6.6 Hz), 3.79 (3H, s), 3.86 (1H, q, J=6.6 Hz, H-5 of Fuc), 3.93 (1H, dd, J=10.3 , 2.9 Hz, H-3 of Fuc), 4.02 (1H, dd, J=10.3, 3.7 Hz, H-2 of Fuc), 4.04—4.11 (4H, m, H-5, 6, 9 of Sia, α-H of Glu), 4.32 (1H, dd, J=12.5, 2.0 Hz, H-9 of Sia), 4.65, 4.97 (each 1H, d, J=11.5 Hz, $-CH_2Ph$), 4.67, 4.81 (each 1H, d, J=12.2 Hz, $-CH_2Ph$), 4.74, 4.88 (each 1H, d, J=11.7 Hz, $-CH_2Ph$), 4.77 (1H, d, J=3.7 Hz, H-1 of Fuc), 4.84 (1H, m, H-4 of Sia), 5.15 (1H, br s), 5.32 (1H, dd, J=8.5, 1.7 Hz, H-7 of Sia), 5.39 (1H, ddd, J=9.3, 5.6, 2.7 Hz, H-8 of Sia), 5.74 (1H, br s), 6.18 (1H, br s), 6.69 (1H, br s), 7.25—7.41 (15H, m). *Anal.* Calcd for $C_{69}H_{98}N_4O_{22}\cdot 0.5H_2O$: C, 61.64; H, 7.42; N, 4.17. Found: C, 61.63; H, 7.41; N, 4.24.

N^5-[6-(5-acetamido-3,5-dideoxy-D-*glycero*-α-D-*galacto*-2-nonulopyranosylonic acid)oxyhexyl-]-N^2-*tert*-butyloxycarbonyl-N^1-[6-α-L-fucopyranosyloxyhexyl]-L-glutamin-α-amide (4) Following the procedure described for **2**, **25** was converted to **4**. (87%) Colorless powder, $[\alpha]_D^{22}$ −41.1° (c=0.62, MeOH). IR (KBr) cm^{-1}: 3470, 1652. ^{1}H-NMR (CD_3OD) δ: 1.20 (3H, d, J=6.6 Hz, H-6 of Fuc), 1.29—1.44 (8H, m), 1.44 (9H, s), 1.46—1.59 (6H, m), 1.59—1.70 (3H, m), 1.83 (1H, m), 2.00 (1H, m), 2.01 (3H, s), 2.25 (2H, t, J=7.1 Hz), 2.74 (1H, m, H-3 of Sia), 3.12—3.25 (5H, m), 3.40—3.47 (2H, m), 3.49—3.57 (2H, m), 3.60—3.75 (7H, m), 3.77 (1H, m), 3.81—3.87 (2H, m), 3.93 (1H, q, J=6.6 Hz, H-5 of Fuc), 3.98 (1H, m), 4.73 (1H, d, J=2.9 Hz, H-1 of Fuc). *Anal.* Calcd for $C_{39}H_{70}N_4O_{18}\cdot 1.2H_2O$: C, 51.78; H, 8.07; N, 6.19. Found: C, 51.53; H, 7.86 ; N, 6.45.

6-Azidohexyl β-D-galactopyranoside (26) To a solution of acetobromo-α-D-galactose (4.00 g, 9.73 mmol) and 6-azidohexanol[10] (2.08 g, 14.5 mmol) was added activated MS4A (4 g). The mixture was stirred at room temperature for 2 h, and then cooled to −20 °C. To the solution was added silver-silicate[16] (10 g), and the stirring was continued at 0 °C for 2 h. The reaction mixture was diluted with dichloromethane, and filtered though a Celite bed. The mixture was washed with H_2O, dried over $MgSO_4$ and concentrated *in vacuo*.

To a solution of the residue in methanol (20 ml) was added 28% sodium methoxide in methanol (500 μl), and the stirring was continued at room temperature for 10 min. The mixture was neutralized with Dowex 50W-X8 (hydrogen form), filtered, and concentrated *in vacuo*. The crude product was purified by silica gel column chromatography (SiO_2 200 g, dichloromethane : methanol=15 : 2, v/v) to give **26** (1.83 g, 62%) as colorless powder. $[\alpha]_D^{25}$ −14.5° (c=1.01, MeOH). IR (KBr) cm^{-1}: 3400, 2102. ^{1}H-NMR (CD_3OD) δ: 1.37—1.47 (4H, m), 1.56—1.67 (4H, m), 3.28 (2H, t, J=7.1 Hz), 3.45 (1H, dd, J=10.0, 3.4 Hz), 3.47—3.52 (2H, m), 3.55 (1H, dt, J=9.5, 6.8 Hz), 3.72 (1H, dd, J=11.2, 5.6 Hz, H-6 of Gal), 3.75 (1H, dd, J=11.2, 6.6 Hz, H-6 of Gal), 3.82 (1H, m), 3.90 (1H, dt, J=9.5, 6.8 Hz), 4.10 (1H, d, J=7.6 Hz, H-1 of Gal).

6-Azidohexyl 2,4,6-tri-*O*-acetyl-3-*O*-*p*-methoxybenzyl-β-D-galactopyranoside (27) To a solution of **26** (3.26 g, 10.7 mmol) in methanol (50 ml) was added dibutyltin oxide (2.93 g, 11.8 mmol). After being stirred at reflux for 2 h, the solvent was removed by evaporation.

To a solution of the residue in benzene (50 ml) were added *p*-methoxybenzylchloride (MPMCl, 4.35 ml, 32.1 mmol) and tetra-butylammonium bromide (3.45 g, 10.7 mmol), and the stirring was continued at reflux for 2 h. The mixture was cooled to room temperature, and concentrated to give the crude alcohol, which was acetylated with acetic anhydride (12 ml) and pyridine (20 ml) at room temperature for 12 h. After usual aqueous work-up, the crude product was purified by silica gel column chromatography (SiO_2 450 g, hexane : ethyl acetate=3 : 1, v/v) to give **27** (3.66 g, 62%) as a colorless syrup.

$[\alpha]_D^{24}$ +29.1° (c=1.04, $CHCl_3$). IR ($CHCl_3$) cm^{-1}: 2100, 1745. ^{1}H-NMR ($CDCl_3$) δ: 1.28—1.41 (4H, m), 1.50—1.64 (4H, m), 2.03, 2.08, 2.15(each 3H, s), 3.25 (2H, t, J=7.1 Hz), 3.43 (1H, dt, J=9.5, 7.1 Hz), 3.50 (1H, dd, J=10.0, 3.4 Hz, H-3 of Gal), 3.78 (1H, t-like, H-5 of Gal), 3.81 (3H, s), 3.86 (1H, dt, J=9.8, 6.3 Hz), 4.15 (1H, dd, J=12.9, 6.6 Hz, H-6 of Gal), 4.18 (1H, dd, J=12.9, 6.6 Hz, H-6 of Gal), 4.33 (1H, d, J=8.1 Hz, H-1 of Gal), 4.33, 4.62 (each 1H, d, J=12.0 Hz, $-CH_2Ph$), 5.08 (1H, dd, J=10.0, 8.1 Hz, H-2 of Gal), 5.48 (1H, br d, H-4 of Gal), 6.86 (2H, A_2B_2, J=8.8 Hz), 7.19

(2H, A_2B_2, J=8.8 Hz). *Anal.* Calcd for $C_{26}H_{37}N_3O_{10}\cdot 0.5H_2O$: C, 55.71; H, 6.83; N, 7.50. Found: C, 55.82 H, 6.67; N, 7.40.

6-Azidohexyl 2,4,6-tri-*O*-benzyl-3-*O*-*p*-methoxybenzyl-β-D-galactopyranoside (28) To a solution of **27** (2.60 g, 4.71 mmol) in methanol (30 ml) was added 28% sodium methoxide in methanol (300 μl), and the stirring was continued at room temperature for 2 h. The mixture was neutralized with Dowex 50W-X8 (hydrogen form), filtered, and concentrated *in vacuo*.

To a solution of the residue in DMF (30 ml) was added sodium hydride (60%, 1.13 g, 28.3 mmol) at 0 °C. After being stirred at 0 °C for 30 min, benzylbromide (2.24 ml, 18.8 mmol) was added, and then the stirring was continued at room temperature for 20 h. To this mixture were added methanol (10 ml) and 28% sodium methoxide in methanol (7 ml), and the mixture was stirred at room temperature for 30 min. The mixture was poured into brine, and extracted with ethyl acetate. The extracts were dried over $MgSO_4$ and concentrated *in vacuo*.

The crude product was purified by silica gel column chromatography (SiO_2 150 g, hexane : ethyl acetate=3 : 1, v/v) to give **28** (2.67 g, 82%) as a colorless syrup.

$[\alpha]_D^{23}$ −6.5° (c=1.02, $CHCl_3$). IR ($CHCl_3$) cm^{-1}: 2100. ^{1}H-NMR ($CDCl_3$) δ: 1.33—1.45 (4H, m), 1.52—1.68 (4H, m), 1.55 (9H, s), 3.20 (2H, t, J= 7.1 Hz), 3.44—3.62 (5H, m), 3.77 (1H, dd, J=10.3, 7.8 Hz, H-2 of Gal), 3.80 (3H, s), 3.85 (1H, br d, H-4 of Gal), 3.91 (1H, dt, J=9.5, 6.3 Hz), 4.32 (1H, d, J=7.8 Hz, H-1 of Gal), 4.40, 4.44, 4.60, 4.93 (each 1H, d, J=11.7 Hz), 4.64, 4.67 (each 1H, d, J=11.5 Hz), 4.76, 4.90 (each 1H, d, J=11.0 Hz), 6.84 (2H, A_2B_2, J=8.5 Hz), 7.21—7.38 (17H, m). *Anal.* Calcd for $C_{41}H_{49}N_3O_7$: C, 70.77; H, 7.10; N, 6.04. Found: C, 70.56; H, 7.10; N, 6.03.

N^2-*tert*-Butyloxycarbonyl-N^1-[6-(2,3,4-tri-*O*-benzyl-α-L-fucopyranosyloxy)hexyl]-N^5-[6-(2,4,6-tri-*O*-benzyl-3-*O*-*p*-methoxybenzyl-β-D-galactopyranosyl)hexyl]-L-glutamin-α-amide (32) Following the procedure described for **20**, **28** was converted to **32**. (92%) mp 129—130 °C (colorless needles from $CHCl_3$–hexane). $[\alpha]_D^{25}$ −13.9° (c=1.03, $CHCl_3$). IR ($CHCl_3$) cm^{-1}: 3300, 1688, 1643. ^{1}H-NMR ($CDCl_3$) δ: 1.10 (3H, d, J=6.6 Hz, H-6 of Fuc), 1.27—1.52 (12H, m), 1.42 (9H, s), 1.54—1.66 (2H, m), 1.66—1.73 (2H, m), 1.93 (1H, m), 2.03 (1H, m), 2.23 (1H, ddd, J=14.9, 7.6, 5.1 Hz), 2.32 (1H, ddd, J=14.9, 8.1, 5.1 Hz), 3.13—3.26 (4H, m), 3.41 (1H, dt, J=9.8, 6.6 Hz), 3.44—3.53 (3H, m), 3.54—3.61 (3H, m), 3.66 (1H, br d), 3.77 (1H, dd, J=9.8, 7.6 Hz, H-2 of Gal), 3.80 (3H, s), 3.85 (1H, d-like), 3.86 (1H, q, J=6.6 Hz, H-5 of Fuc), 3.91 (1H, dt, J=9.8, 6.6 Hz), 3.93 (1H, dd, J=10.0, 2.9 Hz, H-3 of Fuc), 4.02 (1H, dd, J=10.0, 3.7 Hz, H-2 of Fuc), 4.05 (1H, m, aH of Glu), 4.32 (1H, d, J=7.8 Hz, H-1 of Gal), 4.40, 4.44, 4.60, 4.74, 4.90, 4.97 (each 1H, d, J=11.7 Hz, –CH_2Ph), 4.63, 4.65 (each 1H, d, J=11.5 Hz, –CH_2Ph), 4.66 (2H, d), 4.77 (1H, d, J=3.9 Hz, H-1 of Fuc), 4.80 (1H, d, J=12.2 Hz, –CH_2Ph), 4.87, 4.95 (each 1H, d, –CH_2Ph), 5.68 (1H, br s), 5.84 (1H, br s), 6.57 (1H, t-like), 6.84 (2H, A_2B_2, J=8.5 Hz), 7.22—7.41 (32H, m). *Anal.* Calcd for $C_{84}H_{106}N_3O_{16}$: C, 71.36; H, 7.56; N, 2.97. Found: C, 71.15; H, 7.59; N, 2.89.

N^2-*tert*-Butyloxycarbonyl-N^1-[6-(2,3,4-tri-*O*-benzyl-α-L-fucopyranosyloxy)hexyl]-N^5-[6-(2,4,6-tri-*O*-benzyl-β-D-galactopyranosyl)hexyl]-L-glutamin-α-amide (33) To a solution of **32** (790 mg, 0.561 mmol) in dichloromethane (10 ml) were added H_2O (0.5 ml) and DDQ (191 mg, 0.841 mmol), and the stirring was continued at room temperature for 30 min. The mixture was diluted with dichloromethane, washed with saturated aqueous $NaHCO_3$, dried over $MgSO_4$ and concentrated *in vacuo*. The crude product was purified by silica gel column chromatography (SiO_2 45 g, toluene : acetone : methanol=600 : 100 : 7, v/v) to give **33** (520 mg, 72%). mp 125—127 °C (colorless needles from $CHCl_3$–hexane). $[\alpha]_D^{25}$ −14.5° (c=1.02, $CHCl_3$). IR ($CHCl_3$) cm^{-1}: 3317, 1688, 1643. ^{1}H-NMR ($CDCl_3$) δ: 1.10 (3H, d, J=6.3 Hz, H-6 of Fuc), 1.28—1.52 (12H, m), 1.42 (9H, s), 1.54—1.66 (4H, m), 1.91 (1H, m), 2.03 (1H, m), 2.20—2.36 (3H, m), 3.14—3.27 (4H, m), 3.41 (1H, dt, J=9.8, 6.6 Hz), 3.48 (1H, dt, J=9.5, 6.8 Hz), 3.55 (1H, dd, J=9.8, 7.6 Hz, H-2 of Gal), 3.57 (1H, dt, J=9.8, 7.1 Hz), 3.60—3.66 (2H, m), 3.66 (1H, br d), 3.83—3.89 (2H, m), 3.92 (1H, q, J=6.3 Hz, H-5 of Fuc), 3.93 (1H, dd, J=10.3, 2.9 Hz, H-3 of Fuc), 4.02 (1H, dd, J=10.3, 3.7 Hz, H-2 of Fuc), 4.05 (1H, m, α-H of Glu), 4.32 (1H, d, J=7.6 Hz, H-1 of Gal), 4.43, 4.49 (each 1H, d, J=12.0 Hz, –CH_2Ph), 4.63, 4.74 (each 1H, d, J=11.7 Hz, –CH_2Ph), 4.65, 4.67, 4.95, 4.97 (each 1H, d, J=11.2 Hz, –CH_2Ph), 4.66, 4.80 (each 1H, d, J=12.2 Hz, –CH_2Ph), 4.77 (1H, d, J=3.7 Hz, H-1 of Fuc), 4.78 (1H, d), 5.69 (1H, br d), 5.89 (1H, br s), 6.58 (1H, br s), 7.24—7.41 (25H, m). *Anal.* Calcd for $C_{76}H_{98}N_3O_{15}\cdot 1.5H_2O$: C, 69.12; H, 7.71; N, 3.18. Found: C, 69.09; H, 7.60; N, 3.33.

N^2-*tert*-Butyloxycarbonyl-N^1-[6-(2,3,4-tri-*O*-benzyl-α-L-fucopyranosyloxy)hexyl]-N^5-[6-(2,4,6-tri-*O*-benzyl-3-*O*-sulfo-β-D-galactopyranosyl)hexyl]-L-glutamin-α-amide sodium salt (34) To a solution of **33** (386 mg, 0.300 mmol) in DMF (4 ml) was added $SO_3\cdot NMe_2$ (210 mg, 1.51 mmol), and the stirring was continued at 55 °C for 30 min. The mixture was concentrated, and then to the residue were added methanol (5 ml) and Dowex 50W-X8 (sodium form). After being stirred at room temperature for 30 min, the mixture was filtered, then the filtrates were concentrated *in vacuo*. The crude product was purified by silica gel column chromatography (SiO_2 45 g, toluene : acetone : methanol=600 : 100 : 7, v/v), and gel filtration (Sephadex LH20, 100 g, methanol to give **34** (265 mg, 63%) as a colorless powder. ^{1}H-NMR (CD_3OD) δ: 1.13 (3H, d, J=6.6 Hz, H-6 of Fuc), 1.25—1.53 (12H, m), 1.42 (9H, s), 1.54—1.64 (4H, m), 1.82 (1H, m), 1.99 (1H, m), 2.18—2.29 (3H, m), 3.07—3.23 (4H, m), 3.39 (1H, dt, J=10.0, 6.3 Hz), 3.42 (1H, dd, J=9.8, 6.1 Hz, H-6 of Gal), 3.51 (1H, dd, J=9.8, 6.3 Hz, H-6 of Gal), 3.51 (1H, m), 3.61 (1H, m), 3.62 (1H, dd, J=9.5, 7.6 Hz, H-2 of Gal), 3.68 (1H, dd, J=6.3, 6.1 Hz, H-5 of Gal), 3.79 (1H, br s), 3.84 (1H, J=9.8, 6.1 Hz), 3.90 (1H, q, J=6.6 Hz, H-5 of Fuc), 3.92—3.95 (2H, m), 3.97 (1H, m, α-H of Glu), 4.31 (1H, br d, H-4 of Fuc), 4.39 (1H, d, J=7.6 Hz, H-1 of Gal), 4.39, 4.43 (each 1H, d, J=12.0 Hz, –CH_2Ph), 4.60, 4.89, 4.89 (each 1H, d, J=11.2 Hz, –CH_2Ph), 4.64 (1H, d, J=11.5 Hz, –CH_2Ph), 4.66 (1H, d), 4.72—4.88 (5H, m), 5.03 (1H, d, J=11.5 Hz), 7.19—7.38 (26H, m), 7.38—7.41 (2H, d-like), 7.40—7.46 (2H, d-like).

N^2-*tert*-Butyloxycarbonyl-N^1-(6-α-L-fucopyranosyloxyhexyl)-N^5-[1-(3-*O*-sulfo-β-D-galactopyranosyl)hexyl]-L-glutamin-α-amide sodium salt (5) To a solution of **34** (240 mg, 0.172 mmol) in methanol (20 ml) was added palladium on activated carbon (Pd 10%, 200 mg). Hydrogenation was carried out at 3.5×10^4 kg/m^2 (50 psi) hydogen pressure for 12 h. The catalyst was filtered off and the filtrates were concentrated *in vacuo*. The crude product was purified by silica gel column chromatography (SiO_2 20 g, dichloromethane : methanol : H_2O=65 : 35 : 10 v/v (lower phase)), and gel filtration (Sephadex LH20, 100 g, methanol) to give **5** (120 mg, 82%) as a colorless powder. $[\alpha]_D^{23}$ −42.8° (c=0.31, MeOH). IR (KBr) cm^{-1}: 3454, 1700, 1654. ^{1}H-NMR (CD_3OD) δ: 1.20 (3H, d, J=6.8 Hz, H-6 of Fuc), 1.32—1.46 (8H, m), 1.44 (9H, s), 1.46—1.56 (4H, m), 1.58—1.68 (4H, m), 1.83 (1H, m), 2.00 (1H, m), 2.20—2.31 (2H, t-like), 3.12—3.25 (2H, m), 3.44 (1H, dt, J=9.8, 6.3 Hz), 3.54 (1H, m), 3.63—3.71 (3H, m), 3.69 (1H, dd, J=9.3, 7.8 Hz, H-2 of Gal), 3.71—3.76 (3H, m), 3.90 (1H, dt, J=9.5, 6.8 Hz), 3.94 (1H, q, J=6.8 Hz, H-5 of Fuc), 3.98 (1H, m, α-H of Glu), 4.23 (1H, dd, J=9.3, 3.2 Hz, H-3 of Gal), 4.23 (1H, br s, H-4 of Gal), 4.31 (1H, d, J=7.8 Hz, H-1 of Gal), 4.73 (1H, d, J=3.2 Hz, H-1 of Fuc). FAB-MS: *m/z* 878 $[M+Na]^+$ *Anal.* Calcd for $C_{34}H_{62}N_3O_{18}SNa\cdot 1.5H_2O$: C, 46.25; H, 7.42; N, 4.76. Found: C, 46.11; H, 7.27; N, 4.78.

6-(*tert*-Butyloxycarbonylamino)-hexyl 2,3,4,6-tetra-*O*-acetyl-β-D-galactopyranoside (29) To a stirred solution of **26** (2.18 g, 7.14 mmol) in methanol (80 ml) was added *p*-TsOH·H_2O (1.36 g, 7.15 mmol), and the mixture was hydrogenated over Lindlar's catalyst (3.00 g) at 3.5×10^4 kg/m^2 (50 psi) for 4 h. The catalyst was filtered off and the solvent was removed *in vacuo*, giving 6-aminohexyl 2,3,4,6-tetra-*O*-acetyl-β-D-galactopyranoside *p*-toluenesulfonate (2.93 g) as a viscous pale brown oil.

To a solution of 6-aminohexyl 2,3,4,6-tetra-*O*-acetyl-β-D-galactopyranoside *p*-toluenesulfonate (2.93 g) in methanol (20 ml) were added triethylamine (1 ml) and Boc_2O (2.03 g, 9.30 mmol). After being stirred at room temperature for 12 h, the mixture was concentrated to give the crude alcohol, which was acetylated with acetic anhydride (15 ml) and pyridine (25 ml) at room temperature for 12 h. After usual aqueous work-up, the crude product was purified by silica gel column chromatography (SiO_2 150 g, hexane : ethyl acetate=2 : 1, v/v) to give **29** (2.63 g, 67%) as a colorless syrup. $[\alpha]_D^{25}$ −10.1° (c=0.57, $CHCl_3$). IR ($CHCl_3$) cm^{-1}: 1747, 1709. ^{1}H-NMR (CD_3OD) δ: 1.28—1.38 (4H, m), 1.42—1.55 (2H, m), 1.44 (9H, s), 1.55—1.62 (2H, m), 1.99, 2.05, 2.15 (each 3H, s), 3.04—3.15 (2H, m), 3.47 (1H, dt, J=9.5, 6.8 Hz), 3.89 (1H, m), 3.90 (1H, dd, J=7.1, 6.3 Hz, H-5 of Gal), 4.13 (1H, dd, J=11.2, 7.1 Hz, H-6 of Gal), 4.19 (1H, dd, J=11.2, 6.3 Hz, H-6 of Gal), 4.45 (1H, d, J=8.1 Hz, H-1 of Gal), 4.53 (1H, br s, NH), 5.02 (1H, dd, J=10.5, 3.4 Hz, H-3 of Gal), 5.20 (1H, dd, J=10.5, 8.1 Hz, H-2 of Gal), 5.39 (1H, br d, J=3.2 Hz, H-4 of Gal). *Anal.* Calcd for $C_{25}H_{41}NO_{12}$: C, 54.84; H, 7.55; N, 2.56. Found: C, 54.58; H, 7.54; N, 2.56.

6-(*tert*-Butyloxycarbonylamino)-hexyl 2,4,6-tri-*O*-acetyl-3-*O*-allyl-β-D-galactopyranoside (30) To a solution of **29** (1.68 g, 3.07 mmol) in methanol (30 ml) was added 28% sodium methoxide in methanol (500 μl), and the stirring was continued at room temperature for 10 min. The mixture was neutralized with Dowex 50W-X8 (hydrogen form), filtered, and concentrated *in vacuo*.

To a solution of the residue in methanol (20 ml) was added dibutyltin oxide (844 mg, 3.39 mmol). After being stirred at reflux for 2 h, the solvent was removed by evaporation.

To a solution of the residue in benzene (20 ml) were added allylbromide (0.80 ml, 9.24 mmol) and tetra-butylammonium bromide (990 mg, 3.07

mmol), and the stirring was continued at reflux for 2 h. The reaction mixture was cooled to room temperature, and concentrated to give the crude alcohol, which was acetylated with acetic anhydride (6 ml) and pyridine (10 ml) at room temperature for 12 h. After the usual aqueous work-up, the crude product was purified by silica gel column chromatography (SiO_2 120 g, hexane : ethyl acetate=2 : 1, v/v) to give **30** (1.15 g, 69%) as a colorless syrup. $[\alpha]_D^{24}$ +7.1° (c=0.57, $CHCl_3$). IR ($CHCl_3$) cm^{-1}: 1745, 1708. ^{1}H-NMR ($CDCl_3$) δ: 1.28—1.38 (4H, m), 1.42—1.50 (2H, m), 1.44 (9H, s), 1.52—1.62 (2H, m), 2.05, 2.07, 2.08, 2.14 (each 3H, s), 3.04—3.14 (2H, m), 3.45 (1H, dt, J=9.5, 6.8 Hz), 3.51 (1H, dd, J=10.0, 3.4 Hz, H-3 of Gal), 3.80 (1H, dd, J=6.6, 6.6, 1.0 Hz, H-5 of Gal), 3.87 (1H, dt, J=9.5, 6.4 Hz), 3.91 (1H, dddd, J=13.2, 6.1, 1.2, 1.2 Hz, –C<u>H</u>$_2$CH=CH$_2$), 4.12 (1H, dddd, J=13.2, 5.1, 1.5, 1.5 Hz, –C<u>H</u>$_2$CH=CH$_2$), 4.16 (2H, d, J=6.6 Hz, H-6 of Gal), 4.38 (1H, d, J=8.1 Hz, H-1 of Gal), 4.54 (1H, br s, NH), 5.08 (1H, dd, J=10.0, 8.1 Hz, H-2 of Gal), 5.17 (1H, dddd, J=10.5, 1.7, 1.5, 1.2 Hz, –CH=C<u>H</u>$_2$), 5.24 (1H, dddd, J=15.6, 1.7, 1.5, 1.2 Hz, –CH=C<u>H</u>$_2$), 5.41 (1H, dd, J=3.4, 1.0 Hz), 5.78 (1H, dddd, J=15.6, 10.5, 6.1, 5.1 Hz, –C<u>H</u>=CH$_2$). *Anal.* Calcd for $C_{26}H_{43}NO_{11}$: C, 57.23; H, 7.94; N, 2.57 Found: C, 56.93; H, 7.93; N, 2.65.

6-(*tert*-Butyloxycarbonylamino)-hexyl 2,4,6-tri-*O*-acetyl-3-*O*-methoxy carbonylmethyl-β-D-galactopyranoside (31) A solution of **30** (645 mg, 1.18 mmol) in methanol (45 ml) was cooled to −78 °C. Ozone was passed though the solution until a faint blue color appeared, and then oxygen was passed in until the solution became colorless. Dimethylsulfide (5 ml) was added to the mixture, and the mixture was stirred for 1 h at room temperature. After removal of the solvent *in vacuo*, the residue was chomatographed on a silica gel column (SiO_2 45 g, hexane : ethyl acetate : methanol=200 : 300 : 5, v/v) to give a crude aldehyde (577 mg).

To a solution of the crude aldehyde (577 mg) in 2-methyl 2-propanol (20 ml) and 2-methyl-2-butene (5 ml) was added a solution of $NaClO_2$ (2.67 g, 29.5 mmol) and $NaH_2PO_4 \cdot 2H_2O$ (3.68 g, 23.6 mmol) in H_2O (10 ml), and the stirring was continued at room temperature for 4 h. The mixture was diluted with ethyl acetate, washed with H_2O, dried over $MgSO_4$ and concentrated *in vacuo* to give the crude carboxylic acid, which was methylated with $TMSCHN_2$ in methanol–hexane at room temperaturure for 1 h. After the usual work-up, the crude product was purified by silica gel column chromatography (SiO_2 120 g, hexane : ethyl acetate : methanol=300 : 200 : 5 v/v) to give **31** (410 mg, 60%) as a colorless syrup. $[\alpha]_D^{25}$ +12.4° (c=1.07, $CHCl_3$). IR ($CHCl_3$) cm^{-1}: 1747, 1708. ^{1}H-NMR ($CDCl_3$) δ: 1.28—1.40 (4H, m), 1.44 (9H, s), 1.42—1.50 (2H, m), 1.52—1.64 (2H, m), 2.07, 2.14, 2.15 (each 3H, s), 3.05—3.14 (2H, m), 3.46 (1H, dt, J=9.5, 6.6 Hz), 3.66 (1H, dd, J=10.0, 3.4 Hz, H-3 of Gal), 3.73 (3H, s), 3.79 (1H, dd, J=6.8, 6.6, 1.0 Hz, H-5 of Gal), 3.87 (1H, dt, J=9.5, 6.3 Hz), 4.10, 4.11 (each 1H, d, J=17.1 Hz), 4.15 (1H, dd, J=11.5, 6.6 Hz, H-6 of Gal), 4.18 (1H, dd, J=11.5, 6.6 Hz, H-6 of Gal), 4.43 (1H, d, J=8.1 Hz, H-1 of Gal), 4.53 (1H, br s, NH), 5.10 (1H, dd, J=10.0, 8.1 Hz, H-2 of Gal), 5.44 (1H, dd, J=3.4, 1.0 Hz, H-4 of Gal). *Anal.* Calcd for $C_{26}H_{43}NO_{13}$: C, 54.06; H, 7.50; N, 2.42. Found: C, 53.79; H, 7.44; N, 2.44.

***N*^{2}-*tert*-Butyloxycarbonyl-*N*^{5}-[6-(2,4,6-tri-*O*-acetyl-3-*O*-methoxycarbonylmethyl-β-D-galactopyranosyl)hexyl]-*N*^{1}-[6-(2,3,4-tri-*O*-benzyl-α-L-fucopyranosyloxy) hexyl]-L-glutamin-α-amide (35)** To a solution of **31** (109 mg, 0.188 mmol) in dichloromethane (1 ml) was added TfOH (1 ml) at 0 °C. After being stirring at 0 °C for 30 min, the mixture was concentrated to give 6-aminohexyl 2,4,6-tri-*O*-acetyl-3-*O*-methoxycarbonyl methyl-β-D-galactopyranoside, trifluoroacetic acid salt (105 mg) as a colorless syrup, which was converted to **35** by the procedure described for **20** (93%). Colorless powder, $[\alpha]_D^{24}$ −11.3° (c=0.99, $CHCl_3$). IR (KBr) cm^{-1}: 1749, 1688, 1645. ^{1}H-NMR ($CDCl_3$) δ: 1.10 (3H, d, J=6.6 Hz, H-6 of Fuc), 1.28—1.40 (8H, m), 1.43 (9H, s), 1.45—1.52 (4H, m), 1.53—1.65 (4H, m), 1.93 (1H, m), 2.04 (1H, m), 2.06, 2.14, 2.14 (each 3H, s), 2.27 (1H, m), 2.35 (1H, m), 3.16—3.29 (4H, m), 3.42 (1H, dt, J=9.8, 6.6 Hz), 3.45 (1H, dt, J=9.5, 6.6 Hz), 3.57 (1H, dt, J=9.8, 7.1 Hz), 3.66 (1H, dd, J=9.8, 3.4 Hz, H-3 of Gal), 3.67 (1H, br s, H-4 of Fuc), 3.72 (3H, s), 3.78 (1H, dd, J=6.6, 6.6 Hz, H-5 of Gal), 3.86 (1H, q, J=6.6 Hz, H-5 of Fuc), 3.88 (1H, m), 3.93 (1H, dd, J=10.0, 3.4 Hz, H-3 of Fuc), 4.02 (1H, dd, J=10.0, 3.7 Hz, H-2 of Fuc), 4.07 (1H, m, α-H of Glu), 4.10, 4.14 (each 1H, d, J=17.1 Hz), 4.14 (1H, dd, J=11.2, 6.6 Hz, H-6 of Gal), 4.18 (1H, dd, J=11.2, 6.6 Hz, H-6 of Gal), 4.42 (1H, d, J=8.1 Hz, H-1 of Gal), 4.77 (1H, d, J=3.7 Hz, H-1 of Fuc), 4.65, 4.97 (each 1H, d, J=11.5 Hz, –CH_2Ph), 4.67, 4.87 (each 1H, d, J=12.0 Hz, –CH_2Ph), 4.74, 4.77 (each 1H, d, J=11.7 Hz, –CH_2Ph), 5.09 (1H, dd, J=9.8, 8.1 Hz, H-2 of Gal), 5.44 (1H, br d, J=3.4 Hz, H-4 of Gal), 5.70 (1H, br d), 6.05 (1H, br s), 6.63 (1H, br s), 7.41—7.24 (15H, m).

***N*^{2}-*tert*-Butyloxycarbonyl-*N*^{1}-[6-(α-L-fucopyranosyloxy)hexyl]-*N*^{5}-[6-(3-*O*-carboxymethyl-β-D-galactopyranosyl)hexyl]-L-glutamin-α-amide (6)** Following the procedure described for **2**, **35** was converted to **6**. (88%) Colorless powder, $[\alpha]_D^{25}$ −50.3° (c=0.38, MeOH). IR (KBr) cm^{-1}: 3420, 1701, 1653. ^{1}H-NMR (CD_3OD) δ: 1.23 (3H, d, J=6.6 Hz, H-6 of Fuc), 1.30—1.44 (8H, m), 1.45 (9H, s), 1.49—1.59 (4H, m), 1.59—1.70 (4H, m), 1.89 (1H, m), 2.05 (1H, m), 2.30—2.40 (2H, m), 3.13—3.23 (3H, m), 3.26 (1H, m), 3.53 (1H, m), 3.53 (1H, dd, J=10.0, 3.2 Hz, H-3 of Gal), 3.66 (1H, dd, J=10.0, 7.8 Hz, H-2 of Gal), 3.66—3.73 (3H, m), 3.75—3.83 (4H, m), 3.86 (1H, dd, J=10.3, 3.2 Hz, H-3 of Fuc), 3.94 (2H, m), 4.07 (1H, q, J=6.6 Hz, H-5 of Fuc), 4.14 (1H, d, J=3.2 Hz, H-4 of Gal), 4.24, 4.28 (each 1H, d, J=16.6 Hz), 4.43 (1H, d, J=7.8 Hz, H-1 of Gal), 4.88 (1H, d, J=3.9 Hz, H-1 of Fuc). *Anal.* Calcd for $C_{36}H_{65}N_3O_{17}$: C, 53.26; H, 8.07; N, 5.18. Found: C, 53.19; H, 8.27; N, 5.08.

***N*^{2}-*tert*-Butyloxycarbonyl-*N*^{1}-[6-(α-L-fucopyranosyloxy)hexyl]-*N*^{5}-(6-β-D-galactopyranosyloxyhexyl)-L-glutamin-α-amide (7)** Following the procedure described for **5**, **32** was converted to **7** (84%). $[\alpha]_D^{25}$ −52.1° (c=0.34, MeOH). IR (KBr) cm^{-1}: 3350, 1699, 1655. ^{1}H-NMR (CD_3OD) δ: 1.20 (3H, d, J=6.6 Hz, H-6 of Fuc), 1.31—1.46 (8H, m), 1.44 (9H, s), 1.47—1.56 (4H, m), 1.59—1.68 (4H, m), 1.83 (1H, m), 2.00 (1H, m), 2.23—2.29 (2H, m), 3.14—3.25 (4H, m), 3.44 (1H, m), 3.45 (1H, dd, J=9.8, 3.2 Hz), 3.47—3.59 (2H, m, H-2,5 of Gal), 3.54 (1H, ddd, J=9.5, 6.6, 6.6 Hz), 3.63—3.69 (2H, m), 3.83 (1H, br d, H-4 of Gal), 3.70—3.77 (4H, m), 3.90 (1H, ddd, J=9.5, 6.8, 6.8 Hz), 3.93 (1H, q, J=6.6 Hz, H-5 of Fuc), 3.98 (1H, m, α-H of Glu), 4.20 (1H, d, J=7.3 Hz, H-1 of Gal), 4.73 (1H, d, J=2.7 Hz, H-1 of Fuc). *Anal.* Calcd for $C_{34}H_{63}N_3O_{15}$: C, 54.17; H, 8.64; N, 5.60. Found: C, 54.11; H, 8.71; N, 5.71.

***N*^{1},*N*^{5}-bis{6-(2,3,4-tri-*O*-benzyl-α-L-fucopyranosyloxy)hexyl}-*N*^{2}-*tert*-butyloxycarbonyl-L-glutamin-α-amide (36)** To a solution of Boc-L-Glu (74 mg, 0.30 mmol) in acetonitrile (20 ml) were added HOSu (76 mg, 0.660 mmol), and DCC (136 mg, 0.660 mmol). After being stirred at 0 °C for 3 h, 6-aminohexyl 2,3,4-tri-*O*-benzyl-α-L-fucopyranoside *p*-toluenesulfonate (466 mg, 0.66 mmol), and triethylamine (134 μl, 0.66 mmol) were added, and the stirring was continued at 0 °C for 12 h. After the precipitates were filtered off, the filtrates were diluted with dichloromethane, washed with saturated aqueous $NaHCO_3$, dried over $MgSO_4$ and concentrated *in vacuo*. The crude product was purified by silica gel column chromatography (SiO_2 60 g, chloroform : methanol=100 : 1, v/v) to give **36** (288 mg, 97%). mp 149—151 °C (colorless needles from $CHCl_3$–hexane). $[\alpha]_D^{25}$ −34.0° (c=1.45, $CHCl_3$). IR ($CHCl_3$) cm^{-1}: 1689, 1647. ^{1}H-NMR ($CDCl_3$) δ: 1.10 (6H, d, J=6.6 Hz, H-6 of Fuc), 1.28—1.37 (8H, m), 1.43 (9H, s), 1.45—1.52 (4H, m), 1.55—1.62 (4H, m), 1.90 (1H, m), 2.08 (1H, m), 2.21—2.39 (2H, m), 3.16—3.29 (4H, m), 3.38—3.46 (2H, m), 3.54—3.61 (2H, m), 3.66 (2H, d, J=2.9 Hz, H-4 of Fuc), 3.86 (2H, q, J=6.6 Hz, H-5 of Fuc), 3.93—3.96 (2H, m, H-3 of Fuc), 4.00—4.05 (2H, m, H-2 of Fuc), 4.77 (1H, d, J=3.4 Hz, H-1 of Fuc), 4.78 (1H, d, J=3.4 Hz, H-1 of Fuc), 4.65, 4.67, 4.74, 4.81, 4.98 (each 2H, d, J=12.0 Hz, –CH_2Ph), 4.87, 4.88 (each 1H, d, J=12.0 Hz, –CH_2Ph), 7.41—7.24 (30H, m). *Anal.* Calcd for $C_{76}H_{99}N_3O_{14}$: C, 71.39; H, 7.80; N, 3.29. Found: C, 71.21; H, 7.89; N, 3.49.

***N*^{1},*N*^{5}-bis[6-(α-L-fucopyranosyloxy)hexyl]-*N*^{2}-*tert*-butyloxycarbonyl-L-glutamin-α-amide (8)** Following the procedure described for **5**, **36** was converted to **8** (90%). Colorless powder, $[\alpha]_D^{25}$ −80.9° (c=0.78, MeOH). IR (KBr) cm^{-1}: 3368, 1695, 1653. ^{1}H-NMR (CD_3OD) δ: 1.21 (6H,d, J=6.6 Hz, H-6 of Fuc), 1.34—1.44 (8H, m), 1.44 (9H, s), 1.47—1.57 (4H, m), 1.57—1.68 (4H, m), 1.84 (1H, m), 2.00 (1H, m), 2.25 (2H, m), 3.94 (2H, q, J=6.6 Hz, H-4 of Fuc), 4.73 (2H, d, J=2.7 Hz, H-1 of Fuc). ^{13}C-NMR (75 MHz, CD_3OD) δ: 16.7 (C-6 of Fuc), 26.9, 27.6, 27.7, 28.7, 29.5, 30.3, 30.5, 33.3, 40.2 (–<u>C</u>H$_2$NHCO), 40.4 (–<u>C</u>H$_2$NHCO), 55.7, 67.4, 69.1, 70.0, 71.7, 73.6, 80.6 (–NHCO$_2$<u>C</u>(CH$_3$)$_3$), 100.4 (C-1 of Fuc), 157.7 (–NH<u>C</u>O$_2$C(CH$_3$)$_3$), 174.5, 174.8. *Anal.* Calcd for $C_{34}H_{63}N_3O_{14} \cdot H_2O$: C, 54.03; H, 8.67; N, 5.56. Found: C, 54.30; H, 8.64; N, 5.60.

***N*^{1},*N*^{5}-bis[6-(2,3,4-tri-*O*-benzyl-α-L-fucopyranosyloxy)-3,6-dioxaoctyl]-*N*^{2}-*tert*-butyloxycarbonyl-L-glutamin-α-amide (37)** Following the procedure described for **36**, **12** was converted to **37** (72%). Colorless syrup, $[\alpha]_D^{25}$ −32.3° (c=0.30, $CHCl_3$). IR ($CHCl_3$) cm^{-1}: 3436, 1708, 1647. ^{1}H-NMR ($CDCl_3$) δ: 1.10 (6H, d, J=6.6 Hz, H-6 of Fuc), 1.41 (9H, s), 1.85—2.05 (2H, m), 2.13—2.34 (2H, m), 3.32—3.46 (4H, m), 3.46—3.78 (20H, m), 3.89 (2H, m, H-5 of Fuc), 3.94 (2H, dd, J=10.2, 2.7 Hz, H-3 of Fuc), 4.03 (2H, dd, J=10.2, 3.0 Hz, H-2 of Fuc), 4.08 (1H, m), 4.70, 4.80 (each 1H, d, J=12.2 Hz, –CH_2Ph), 4.85 (2H, d, J=3.0 Hz, H-1 of Fuc), 4.64, 4.78, 4.78, 4.85, 4.86, 4.97 (each 2H, d, J=11.4 Hz, –CH_2Ph), 5.63 (1H, d, J=7.2 Hz), 6.72 (1H, t-like), 7.10 (1H, br s), 7.24—7.42 (30H, m).

***N*^{1},*N*^{5}-bis-(8-α-L-fucopyranosyloxy-3,6-dioxaoctyl)-*N*^{2}-*tert*-butyloxycarbonyl-L-glutamin-α-amide (9)** Following the procedure described for **5**, **37** was converted to **9** (95%). Colorless syrup, $[\alpha]_D^{25}$ −48.5° (c=0.30, $CHCl_3$). IR ($CHCl_3$) cm^{-1}: 3400, 1663. ^{1}H-NMR (CD_3OD) δ: 1.21 (6H, d,

J=6.6 Hz, H-6 of Fuc), 1.44 (9H, s), 1.89 (1H, m), 2.00 (1H, m), 2.29 (2H, m), 3.50—3.76 (24H, m), 3.76—3.86 (2H, m), 4.00 (2H, q, J=6.6 Hz, H-5 of Fuc), 4.03 (1H, m), 4.80 (2H, d, J=3.0 Hz, H-1 of Fuc). FAB-MS: *m/z* 802 $[M+H]^+$, 824$[M+Na]^+$. *Anal.* Calcd for $C_{34}H_{63}N_3O_{18}$: C, 50.93; H, 7.92; N, 5.24. Found: C, 50.74; H, 8.12; N, 5.04.

Cell Adhesion Assay HUVECs (5×10^4 cells per well) were seeded in gelatin-coated 96-well plates and cultured in E-GM UV medium (Kurabo Industries Ltd.) until the confluent monolayers. HUVEC monolayers were stimulated for 4 h with 20 U/ml recombinant human interleukin 1β (Gemzyme Co.) and then rinsed once with MEM medium (Gibco-BRL) containing 0.4% BSA. Fifty microliters of E-GM UV medium containing soluble inhibitors was added per well and the sample were left for 1 h at 37 °C. HL-60 promyelocytic leukemia cells were metabolically radiolabeled overnight with ^{3}H-thymidine and added to the wells (1.5×10^4 cells per well in 50 ml of E-GM UV medium). After being incubated for 30 min at 37 °C, the microplates were rinsed twice with MEM medium containing 0.4% BSA. The remaining adhering HL-60 cells were lysed with 1% SDS. The radioactivities of the lysates were measured with a liquid scintillation counter (Aloka LSC-3500).

Protective Effect of SLe^X and Its Mimetics in Rat Pleutic Model Induced by Carrageenin Female 10-week-old Lewis rats (weight: 180—200 g) were used in these studies (Japan SLC, Inc.). Pleurisy was induced by intrapleural injection of 200 μl of 2% carrageenin. When used, SLe^X or its mimetics (5 mg/kg, PBS (−) solution) was administered intravenously 0, 2, 4 h after the carrageenin injection. Control rats received PBS (−) only. The exudates into pleural cavity were collected 2 h after the last administration. The number of neutrophils were measured by hemocytometer, and the amount of exudate proteins were measured by using BCA protein assay reagent™ (PIERCE Co.).

References and Notes

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2) *a)* Phillips M. L., Nudelman E., Gaeta F. C. A., Perez M., Singhal A. K., Hakomori S., Paulson J. C., *Science*, **250**, 1130—1132 (1990); *b)* Walz G., Aruffo A., Kolanus W., Bevilacqua M., Seed B., *ibid.*, **250**, 1132—1135 (1990); *c)* Springer T. A., Lasky L. A., *Nature* (London), **349**, 196—197 (1991); *d)* Bevilacqua M., Pober J. S., Mendrick D. L., Cotran R. S., Gimbrone M. A., Jr., *Proc. Natl. Acad. Sci. U.S.A.*, **84**, 9238—9242 (1987); *e)* Bevilacqua M., Stengelin S., Gimbrone M. A., Jr., Seed B., *Science*, **243**, 1160—1165 (1989); *f)* Lowe J. B., Stoolman L. M., Nair R. P., Larsen R. D., Berhend T. L., Marks R. M. *Cell*, **63**, 475—484 (1990).

3) Uchiyama T., Vassiley V. P., Kajimoto T., Wong W., Huang H., Lin C.-C., Wong C.-H., *J. Am. Chem. Soc.*, **117**, 5995—5996 (1995).

4) *a)* Inoue K., Ito T., Okuno S., Aono K., Patent WO/923480; *b)* Nogusa H., Yano T., Okuno S., Hamana H., Inoue K., *Chem. Pharm. Bull.*, **43**, 1931—1936 (1995).

5) *a)* Sakagami M., Horie K., Nakamoto K., Kawaguchi K., Hamana H., *Bioorg. Med. Chem. Lett.*, **8**, 2783—2786 (1998); *b)* Horie K., Sakagami M., Kuramochi K., Hanasaki K., Hamana H., Ito T., *Pharm. Res.*, **16**, 314—320 (1999).

6) *a)* Simanek E. E., McGarvey G. J., Jablonowski J. A., Wong C.-H., *Chem. Rev.*, **98**, 833—862 (1998); *b)* Kogan T. P., Dupre B., Keller K. M., Scott I. L., Bui H., Market R. V., Beck P. J., Voytus J. A., Revelle B. M., Scott D., *J. Med. Chem.*, **38**, 4976—4984 (1995); *c)* Wu S. H., Shimazaki M., Lin C.-C., Moore W. J., Weitz-Schmidt G., Wong C. H., *Angew. Chem. Int. Ed. Engl.*, **35**, 88—90 (1996); *d)* Tsukida T., Hiramatsu Y., Tsujishita H., Kiyoi T., Yoshida M., Kurokawa K., Moriyama H., Ohmoto H., Wada Y., Saito T., Kondo H., *J. Med. Chem.*, **40**, 3534—3541 (1997); *e)* Kolb H. C., Ernst B., *Chem. Eur. J.*, **3**, 1571—1578 (1997).

7) *a)* Tyrrell D., James P., Rao N., Foxall C., Abbas S., Dasgupta F., Nashed M., Hasegawa A., Kiso M., Asa D., Kidd J., Brandley B. K., *Proc. Natl. Acad. Sci. U.S.A.*, **88**, 10372—10376 (1991); *b)* Brandley B. K., Kiso M., Abbas S., Nikrad P., Srivasatava O., Foxall C., Oda Y., Hasegawa A. *Glycobiology*, **3**, 633—639 (1993).

8) *a)* Sasaki A., Murahashi N., Yamada H., Morikawa A., *Biol. Pharm. Bull.*, **17**, 680—685 (1994); *b)* Murahashi N., Ishihara H., Sasaki A., Sakagami M., Hamana H., *ibid.*, **20**, 259—266 (1997).

9) Juszynski D., Flowers H. M., *Carbohydr. Res.*, **18**, 219—226 (1968).

10) 6-Azidohexanol was prepared from 6-chlorohexanol (NaN_3/DMF, 98%).

11) Higashi K., Susaki H., *Chem. Pharm. Bull.*, **40**, 2019—2022 (1992).

12) Bajwa J. S., *Tetrahedron Lett.*, **33**, 2299—2302 (1992).

13) Murase T., Kameyama A., Kartha K. P. R., Ishida H., Kiso M., Hasegawa A., *J. Carbohydrate Chemistry*, **8**, 265—283 (1989).

14) Hasegawa A., Ohki T., Nagahama T., Ishida H., Kiso M., *Carbohydr. Res.*, **212**, 277—281 (1991).

15) Hasegawa A., Nagahama T., Ohki T., Hotta K., Ishida H., Kiso M., *J. Carbohydrate Chemistry*, **10**, 493—498 (1991).

16) Paulsen H., Lockhoff O., *Chem. Ber.*, **114**, 3102—3104 (1981).

17) Oh-ishi S., Tsuji N., Hayashi I., *Jpn. J. Phamacol.*, **50**, 11—18 (1989).

18) *a)* DeFrees S. A., Philips L., Guo L., Zalipsky S., *J. Am. Chem. Soc.*, **118**, 6101—6104 (1996); *b)* Miyauchi H., Tanaka M., Koike H., Kawamura N., Hayashi M., *Bioorg. Med. Chem. Lett.*, **7**, 985—988 (1997); *c)* Thoma G., Magnani L., Ohlein R., Ernst B., Schwarzenbach F., Duthaler R. O., *J. Am. Chem. Soc.*, **119**, 7414—7415 (1997).