Synthesis and Immunoadjuvant Activity of the Conjugates of 1-Thio-*N*-acetyl-muramoyl Dipeptide with Lipid A Subunit Analogs[†]

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A variety of immunomodulators were synthesized by combining biologically active derivatives of 1-thio-muramoyl dipeptide with 4-*O*-phosphono-D-glucosamine derivatives related to bacterial lipid A, and using the (succinoylamino)undecanoyl group as a spacer. Their immunoadjuvant activities in guinea-pigs were examined.

In the course of a synthetic approach¹⁾ to design a new type of immunomodulator by coupling the biologically active derivative of *N*-acetyl-muramoyl dipeptide (MDP)²⁾ with derivatives related to the nonreducing subunit of lipid A,³⁾ it was found that the conjugate of *N*-[2-*O*-(2-acetamido-1,5-anhydro-2,3-dideoxy-D-glucitol-3-yl)-D-lactoyl]-L-alanyl-D-isoglutamine methyl ester (1-deoxy-MDP methyl ester)⁴⁾ with 2-deoxy-4-*O*-phosphono-3-*O*-tetradecanoyl-2-(3-tetradecanoyloxytetradecanamido)-D-glucopyranose (GLA-27)⁵⁾ through acylamido spacer-arms showed strong immunoadjuvant activity.⁶⁾ Especially, the compound carrying the (succinoylamino)undecanoyl group as a spacer expressed the highest degree of immunoadjuvant activity, and more strongly enhanced the colony stimulation factor (CSF)-inductive activity than that of GLA-27 alone.^{1b)}

Recent studies⁷⁾ on MDP analogs showed that potent antitumor and anti-infection activities were effectively expressed by the introduction of lipophilicity at C-6 or at both C-1 and C-6 of the sugar skeleton in 1-thio-MDP.

These compounds did not exhibit such side effects as pyrogenic activity and necrosis-inducing activity,⁸⁾ in contrast to MDP.

Among the lipid A-subunit analogs⁹⁾ synthesized with special reference to fatty acid substituents, GLA-60^{9a)} carrying 2-*N*-linked (3*R*)-3-hydroxytetradecanoyl and 3-*O*-linked (3*R*)-3-tetradecanoyloxytetradecanoyl groups showed the strongest B cell activation and adjuvant activities.¹⁰⁾ In view of these facts, we synthesized a series of novel conjugates (**43** ~ **45**, **52** ~ **54**, **57** and **64** ~ **66**) of 1-thio-MDP with lipid A-subunit analogs, expecting a new type of immunomodulator, and now describe their syntheses and biological activities.

Results and Discussion

The 1-thio-MDP methyl ester derivatives⁷⁾ (**3** ~ **5**) were coupled with *N*-protected amino-undecanoic acid (**2**), in the presence of dicyclohexylcarbodiimide (DCC) and 4-dimethylaminopyridine (DMAP), to give 6-*O*-[11-(*tert*-butoxycarbonylamino)undecanoyl]-1-thio-MDP methyl ester derivatives (**6** ~ **8**) in good

[†] Studies on Immunoadjuvant Active Compounds. Part XXXVII. For Part XXXVI, see ref. 1b.

R	
1	H
2	Boc

Boc: tert-BuOC(O)

R	n
3	H 0 (SAc)
4	H 8
5	H 16
6	BocNH(CH ₂) ₁₀ C(O) 0 (SAc)
7	BocNH(CH ₂) ₁₀ C(O) 8
8	BocNH(CH ₂) ₁₀ C(O) 16
9	TFA·N H ₂ (CH ₂) ₁₀ C(O) 0 (SAc)
10	TFA·N H ₂ (CH ₂) ₁₀ C(O) 8
11	TFA·N H ₂ (CH ₂) ₁₀ C(O) 16

TFA: $\text{F}_3\text{CC(O)OH}$

R	
20	Ac
21	SE

SE: $\text{Me}_3\text{SiCH}_2\text{CH}_2$

R ¹	R ²
22	H H
23	C ₁₄ -OH H
24	C ₁₄ -O-C ₁₄ C ₁₄
25	C ₁₄ -OSEM H
26	C ₁₄ -OSEM C ₁₄ -O-C ₁₄

R ¹	R ²
27	H H
28	H TBDMS
29	(TCEO) ₂ P(O) TBDMS
30	(TCEO) ₂ P(O) H
31	(TCEO) ₂ P(O) HOC(O)(CH ₂) ₂ C(O)

R ¹	R ²
32	H H
33	H TBDMS
34	(TCEO) ₂ P(O) TBDMS
35	(TCEO) ₂ P(O) H
36	(TCEO) ₂ P(O) HOC(O)(CH ₂) ₂ C(O)

C₁₄: $\text{CH}_3(\text{CH}_2)_{12}\text{C(O)}$

C₁₄-OH: $\text{CH}_3(\text{CH}_2)_{10}\text{CHCH}_2\text{C(O)}$

C₁₄-OSEM: $\text{CH}_3(\text{CH}_2)_{10}\text{CHCH}_2\text{C(O)}$

C₁₄-O-C₁₄: $\text{CH}_3(\text{CH}_2)_{10}\text{CHCH}_2\text{C(O)}$

TBDMS: $\text{tert-BuMe}_2\text{Si}$

TCE: Cl_3CCH_2

yields. For the synthesis of the 1-S-[11-(*tert*-butoxycarbonylamino)undecanoyl] derivatives (**14**~**16**), the 1-S-acetyl group in **12** was selectively solvolized with sodium methoxide at -25°C . The reaction mixture was deionized

with cation-exchange resin, and then coupled with **2** in the presence of DCC to afford **13** in a 68% yield. The isopropylidene group of **13** was hydrolyzed, and the resulting **14** was esterified at O-6 to give **15** and **16** in 86% and 65%

	R ¹	R ²	n
37	SE (B)	TCE	0 (SAC)
38	SE (B)	TCE	8
39	SE (B)	TCE	16
40	H	TCE	0 (SAC)
41	H	TCE	8
42	H	TCE	16
43	H	H	0 (SAC)
44	H	H	8
45	H	H	16

	R ¹	R ²	R ³	n
46	SE (B)	SEM	TCE	0 (SAC)
47	SE (B)	SEM	TCE	8
48	SE (B)	SEM	TCE	16
49	H	H	TCE	0 (SAC)
50	H	H	TCE	8
51	H	H	TCE	16
52	H	H	H	0 (SAC)
53	H	H	H	8
54	H	H	H	16

	R ¹	R ²
55	SE (B)	TCE
56	H	TCE
57	H	H

	R ¹	R ²	R ³	R ⁴
58	SE (B)	SEM	TCE	H
59	SE (B)	SEM	TCE	CH ₃ (CH ₂) ₈ C(O)
60	SE (B)	SEM	TCE	CH ₃ (CH ₂) ₁₆ C(O)
61	H	H	TCE	H
62	H	H	TCE	CH ₃ (CH ₂) ₈ C(O)
63	H	H	TCE	CH ₃ (CH ₂) ₁₆ C(O)
64	H	H	H	H
65	H	H	H	CH ₃ (CH ₂) ₈ C(O)
66	H	H	H	CH ₃ (CH ₂) ₁₆ C(O)

yields, respectively.

The *tert*-butoxycarbonyl group of **6~8** and **14~16** was removed with trifluoroacetic acid at 0°C to afford the corresponding trifluoroacetate derivatives (**9~11** and **17~19**).

In view of the known propensity of divalent sulfur compounds for poisoning a catalytic-hydrogenation system, the hydroxyl groups of the lipid A moiety were suitably protected with 2-(trimethylsilyl)ethyl¹¹⁾ and 2-(trimethylsilyl)ethoxymethyl¹²⁾ groups, which are easily removable with Lewis acid.¹³⁾

The formation of 2-(trimethylsilyl)ethyl glycoside was accomplished by the oxazoline method. 2-Acetamido-1,3,4,6-tetra-*O*-acetyl-

2-deoxy-β-D-glucopyranose¹⁴⁾ (**20**) was converted with anhydrous ferric chloride to the oxazoline intermediate, which was then treated with 2-(trimethylsilyl)ethanol in the presence of conc. sulfuric acid at room temperature to give 2-(trimethylsilyl)ethyl 2-acetamido-3,4,6-tri-*O*-acetyl-2-deoxy-β-D-glucopyranoside (**21**) in a good yield.

O-Deacetylation of **21** and 4,6-*O*-isopropylidenation were performed in the usual way, and hydrolytic removal of the *N*-acetyl group with barium hydroxide octahydrate in water-ethanol under reflux gave 2-(trimethylsilyl)ethyl 2-amino-2-deoxy-4,6-*O*-isopropylidene-β-D-glucopyranoside (**22**) in a 92% yield

(based on **21**). Treatment of **22** with (3*R*)-3-hydroxytetradecanoic acid or (3*R*)-3-[2-(trimethylsilyl)ethoxymethoxy]tetradecanoic acid in the presence of 1-ethyl-3-(3-dimethylaminopropyl)carbodiimide hydrochloride (water-soluble carbodiimide, WSC) gave 2-*N*-acylated compounds (**23** and **25**) in 84% and 82% yields, respectively.

The remaining hydroxyl groups of **23** were simultaneously esterified with tetradecanoyl chloride and DMAP in pyridine to afford **24**. Compound **26** was obtained by esterifying **25** with tetradecanoyloxytetradecanoic acid, WSC and DMAP. Compounds **24** and **26** were treated with aqueous acetic acid to remove the 4,6-*O*-isopropylidene group. The *tert*-butyldimethylsilyl group was introduced at O-6, and the resulting **28** and **33** were phosphorylated with bis(2,2,2-trichloroethyl)phosphoro chloridate to afford **29** and **34** in 84% and 78% yields, respectively.

The *tert*-butyldimethylsilyl group of **29** and **34** was cleaved with aqueous acetic acid to give **30** and **35**, and these were treated with succinic anhydride in the presence of DMAP in 1,2-dichloroethane at room temperature to afford the 6-*O*-(3-carboxypropanoyl) derivatives (**31** and **36**).

Compounds **31** and **36** were coupled with the 1-thio-MDP methyl ester trifluoroacetate derivatives (**9**~**11** and **17**~**19**), using *N*-hydroxysuccinimide and DCC as the activating reagents, to give the protected coupling products (**37**~**39**, **46**~**48**, **55** and **58**~**60**). Removal of the silyl groups with boron trifluoride etherate in dichloromethane afforded **40**~**42**, **49**~**51**, **56** and **61**~**63**. Finally, the bis(2,2,2-trichloroethyl) groups were cleaved with zinc dust to yield the desired analogs (**43**~**45**, **52**~**54**, **57** and **64**~**66**) as colorless powders, respectively.

The immunoadjuvant activities of the conjugates thus obtained for inducing the delayed type of hypersensitivity (DTH) to *N*-acetyl-L-tyrosine-3-azobenzene-4'-arsonate (ABA-*N*-acetyltyrosine) in guinea-pigs were examined (see Table I).¹⁵ All of the compounds showed strong DTH activity, comparable to that of

Table I. ADJUVANT ACTIVITY OF SOME CONJUGATES ON THE INDUCTION OF DELAYED-TYPE HYPERSENSITIVITY TO ABA-*N*-ACETYLTYROSINE IN GUINEA-PIGS

Compound ^a	Skin reaction with ABA-BSA ^b (100 µg) (diam. in mm ± SE) ^c	
	24 hr	48 hr
43	16.5 ± 1.2	12.3 ± 1.9
44	18.3 ± 0.5	15.8 ± 1.2
45	14.8 ± 2.1	12.8 ± 2.5
52	16.7 ± 1.4	(6.3 ± 1.8)
54	18.3 ± 1.7	12.5 ± 3.0
57	15.8 ± 2.3	10.0 ± 3.4
64	13.8 ± 1.7	(5.3 ± 3.2)
65	14.0 ± 1.5	6.5 ± 2.4
66	22.0 ± 1.2	16.3 ± 1.0
MDP	20.0 ± 0.7	19.0 ± 0.7
Control ^d	0	0

^a Dose: 10 µg.

^b Azobenzene-arsonate-*N*-acetyl-L-tyrosine-bovine serum albumin.

^c The data indicate the average diameter ± standard error (S.E.) of the skin reaction (induration) in four guinea-pigs; the values in parentheses indicate the size of the erythema.

^d ABA-*N*-Acetyltyrosine in Freund's incomplete adjuvant.

MDP at the same dose. In addition, we observed that these conjugates showed potent inducing activities for interleukin-1 and tumoricidal macrophages; especially, compounds **52** and **64** exhibited much higher activity than GLA-60 (data not shown). These results indicate that these conjugates could have practical use as new biological response modifiers.

Experimental

Melting points (mp) were determined with Yanagimoto micromelting-point apparatus and are uncorrected. Specific rotations were determined with a Union PM-201 polarimeter, and IR spectra were recorded with a Jasco A-100 spectrophotometer. ¹H-NMR spectra were recorded with a Jeol JNM-GX270 spectrometer, using tetramethylsilane as an internal standard. Preparative chromatography on silica gel (Wako Co., 200 mesh) was accomplished with the solvent systems specified. Concentrations and evaporations were conducted *in vacuo*.

11-tert-Butoxycarbonylaminoundecanoic acid (**2**). To a

mixture of 11-aminoundecanoic acid (**1**; 3.0 g, 14.9 mmol) and triethylamine (0.02 ml) in a mixture of water–1,4-dioxane (2:1, 30 ml) was added 2-(*tert*-butoxycarbonylthio)-4,6-dimethylpyrimidine (6.15 g, 25.6 mmol), and the mixture was stirred overnight at room temperature. The solvents were removed by lyophilization, and the residue was extracted with dichloromethane. The extract was washed with water, dried (sodium sulfate), and concentrated to a syrup, which was crystallized from hexane to give **2** (4.5 g, quant.), mp 66°C; IR ν_{max} (KBr) cm^{-1} : 3320 (NH), 2950 and 2840 (C–H), and 1680 and 1510 (amide).

Anal. Found: C, 63.59; H, 10.48; N, 4.50. Calcd. for $\text{C}_{16}\text{H}_{31}\text{NO}_4$: C, 63.76; H, 10.37; N, 4.65%.

N-[2-*O*-[2-Acetamido-1-*S*-acetyl-6-*O*-(11-(*tert*-butoxycarbonylamino)undecanoyl]-2,3-dideoxy-1-thio- β -D-glucopyranose-3-yl]-D-lactoyl]-L-alanyl-D-isoglutamine methyl ester (**6**). A mixture of *N*-[2-*O*-[2-acetamido-1-*S*-acetyl-2,3-dideoxy-1-thio- β -D-glucopyranose-3-yl]-D-lactoyl]-L-alanyl-D-isoglutamine methyl ester (**3**; 100 mg, 0.2 mmol), 1,4-dioxane–DMF (3:1, 2 ml), DCC (80 mg, 0.4 mmol), a catalytic amount of DMAP, and compound **2** (12.6 mg, 0.4 mmol) was stirred for 5 hr at room temperature. DC urea was removed by filtration and washed with 1,4-dioxane. The filtrate and washings were combined and concentrated. The residue was extracted with dichloromethane, and the extract was successively washed with 2 M hydrochloric acid and water, dried (sodium sulfate), and concentrated to a syrup, which was chromatographed on a column of silica gel with dichloromethane–methanol (30:1) to give compound **6** (73.5 mg, 48%), mp 146.5~148°C, $[\alpha]_{\text{D}} + 14.9^\circ$ ($c = 1.9$, 2:1 dichloromethane–methanol); IR ν_{max} (film) cm^{-1} : 3300 (OH, NH), 2940 and 2850 (C–H), 1750 (ester), 1700 (AcS), and 1650 and 1550 (amide); NMR (CDCl_3) δ : 1.2~1.5 (m, 31H, lac-Me, ala-Me, *tert*-Boc, and $-\text{CH}_2-$), 1.94 (s, 3H, AcN), 2.34 (s, 3H, AcS), 2.44 (m, 2H, $-\text{COCH}_2-$), 3.11 (m, 2H, $-\text{NHCH}_2-$), 3.68 (s, 3H, MeO), and 5.19 (d, 1H, $J_{1,2} = 10.6 \text{ Hz}$, H-1).

Anal. Found: C, 53.96; H, 7.95; N, 8.46. Calcd. for $\text{C}_{38}\text{H}_{65}\text{N}_5\text{O}_{14}\text{S}$: C, 53.82; H, 7.73; N, 8.26%.

N-[2-*O*-[2-Acetamido-6-*O*-(11-(*tert*-butoxycarbonylamino)undecanoyl]-1-*S*-decanoyl-2,3-dideoxy-1-thio- β -D-glucopyranose-3-yl]-D-lactoyl]-L-alanyl-D-isoglutamine methyl ester (**7**). Compound **7** was obtained as crystals in a 57% yield by a condensation of *N*-[2-*O*-[2-acetamido-1-*S*-decanoyl-2,3-dideoxy-1-thio- β -D-glucopyranose-3-yl]-D-lactoyl]-L-alanyl-D-isoglutamine methyl ester **4** with **2**, according to the method described for **6**; mp 152~155°C, $[\alpha]_{\text{D}} + 6.6^\circ$ ($c = 2.9$, dichloromethane); IR ν_{max} (film) cm^{-1} : 3300 (OH, NH), 2950 and 2860 (C–H), 1750 (ester), 1690 (*S*-acyl), and 1670 and 1540 (amide); NMR (CDCl_3) δ : 0.87 (t, 3H, Me), 1.26~1.44 (m, 45H, lac-Me, ala-Me, *tert*-Boc, and $-\text{CH}_2-$), 1.92 (s, 3H, AcN), 3.67 (s, 3H, MeO), and 5.14 (d, 1H, $J_{1,2} = 10.6 \text{ Hz}$, H-1).

Anal. Found: C, 57.28; H, 8.75; N, 7.20. Calcd. for $\text{C}_{46}\text{H}_{81}\text{N}_5\text{O}_{14}\text{S}$: C, 57.54; H, 8.50; N, 7.29%.

N-[2-*O*-[2-Acetamido-6-*O*-(11-(*tert*-butoxycarbonylamino)undecanoyl]-2,3-dideoxy-1-*S*-octadecanoyl-1-thio- β -D-glucopyranose-3-yl]-D-lactoyl]-L-alanyl-D-isoglutamine methyl ester (**8**). Compound **8** was obtained as crystals in a 70% yield by coupling *N*-[2-*O*-[2-acetamido-2,3-dideoxy-1-*S*-octadecanoyl-1-thio- β -D-glucopyranose-3-yl]-D-lactoyl]-L-alanyl-D-isoglutamine methyl ester **5** with compound **2**, as described for **6**; mp 164.5~169°C, $[\alpha]_{\text{D}} + 8.2^\circ$ ($c = 1.6$, dichloromethane); the IR and NMR spectra were consistent with the structures assigned.

Anal. Found: C, 60.59; H, 9.01; N, 6.33. Calcd. for $\text{C}_{54}\text{H}_{97}\text{N}_5\text{O}_{14}\text{S}$: C, 60.48; H, 9.12; N, 6.53%.

N-[2-*O*-[2-Acetamido-1-*S*-acetyl-6-*O*-(11-aminoundecanoyl)-2,3-dideoxy-1-thio- β -D-glucopyranose-3-yl]-D-lactoyl]-L-alanyl-D-isoglutamine methyl ester trifluoroacetate (**9**). A mixture of **6** (70 mg, 0.08 mmol) and trifluoroacetic acid (2 ml) was stirred for 1 hr at 0°C, and then the solvent was evaporated. The residual syrup was dissolved in 1,4-dioxane, and the dioxane was removed by lyophilization to leave **9** (71 mg, quant.) as an amorphous solid; $[\alpha]_{\text{D}} + 12.5^\circ$ ($c = 1.9$, ethanol); IR ν_{max} (KBr) cm^{-1} : 3300 (OH, NH), 2950 and 2860 (C–H), 1730 (ester), 1690 (AcS), 1660 and 1530 (amide), and 1200, 1180 and 1140 (C–F).

Anal. Found: C, 48.98; H, 6.51; N, 7.96. Calcd. for $\text{C}_{35}\text{H}_{58}\text{N}_5\text{O}_{14}\text{F}_3\text{S}$: C, 48.77; H, 6.78; N, 8.13%.

The trifluoroacetates of other 6-*O*-(11-aminoundecanoyl) compounds (**10** and **11**). Compounds **10** and **11** were obtained quantitatively as amorphous solids, according to the procedure described for **9**.

Compound **10** had $[\alpha]_{\text{D}} + 12.8^\circ$ ($c = 1.6$, methanol); the IR data were similar to those of **9**.

Anal. Found: C, 53.31; H, 7.29; N, 7.05. Calcd. for $\text{C}_{43}\text{H}_{74}\text{N}_5\text{O}_{14}\text{F}_3\text{S}$: C, 53.02; H, 7.66; N, 7.19%.

Compound **11** had mp 135~138°C and $[\alpha]_{\text{D}} + 12.0^\circ$ ($c = 2.0$, 1:1 dichloromethane–methanol); the IR data were similar to those of **9**.

Anal. Found: C, 56.58; H, 8.11; N, 6.28. Calcd. for $\text{C}_{51}\text{H}_{90}\text{N}_5\text{O}_{14}\text{F}_3\text{S}$: C, 56.39; H, 8.35; N, 6.45%.

N-[2-*O*-[2-Acetamido-1-*S*-[11-(*tert*-butoxycarbonylamino)undecanoyl]-2,3-dideoxy-4,6-*O*-isopropylidene-1-thio- β -D-glucopyranose-3-yl]-D-lactoyl]-L-alanyl-D-isoglutamine methyl ester (**13**). To a solution of *N*-[2-*O*-[2-acetamido-1-*S*-acetyl-2,3-dideoxy-4,6-*O*-isopropylidene-1-thio- β -D-glucopyranose-3-yl]-D-lactoyl]-L-alanyl-D-isoglutamine methyl ester (**12**; 500 mg, 0.84 mmol) in methanol was added an excess amount of metallic sodium (50 mg) at -25°C . The mixture was stirred for 20 min at the same temperature, and then treated with Amberlite IR-120 (H^+) resin to deionize the solution. The resin was filtered off and washed with methanol, and the filtrate and washings were combined and concentrated. The resulting product was treated with **2** (360 mg, 1.2 mmol) in the presence of DCC (340 mg, 1.7 mmol) in 1,4-dioxane (5 ml).

The mixture was stirred for 2 hr at room temperature, and DC urea was filtered off and washed with 1,4-dioxane. The filtrate and washings were combined and concentrated to a syrup, which was chromatographed on a column of silica gel with dichloromethane-methanol (50:1) to give **13** as a solid (480 mg, 68%), mp 75~79°C, $[\alpha]_D +11.7^\circ$ ($c=0.5$, dichloromethane); IR $\nu_{\max}$ (film) cm^{-1} : 3300 (NH), 2940 and 2850 (C-H), 1740 (ester), 1700 (S-acyl), 1650 and 1550 (amide), and 860 (Me_2C); NMR (CDCl_3) δ : 1.25~1.49 (m, 37H, lac-Me, ala-Me, *tert*-Boc, $-\text{CH}_2-$, and Me_2C), 1.94 (s, 3H, AcN), 2.51 (m, 2H, $-\text{COCH}_2-$), 3.12 (m, 2H, $-\text{NHCH}_2-$), 3.70 (s, 3H, MeO), and 5.24 (d, 1H, $J_{1,2}=10.6\text{ Hz}$, H-1).

Anal. Found: C, 55.18; H, 7.77; N, 8.06. Calcd. for $\text{C}_{39}\text{H}_{67}\text{N}_5\text{O}_{13}\text{S}$: C, 55.37; H, 7.98; N, 8.28%.

N-[2-*O*-(2-Acetamido-1-*S*-[11-(*tert*-butoxycarbonyl-amino)undecanoyl]-2,3-dideoxy-1-thio- β -D-glucopyranose-3-yl]-D-lactoyl]-L-alanyl-D-isoglutamine methyl ester (**14**). A solution of **13** (450 mg, 0.53 mmol) in 80% aqueous acetic acid (5 ml) was stirred for 2 hr at 45°C, and then concentrated. The residue was dissolved in 1,4-dioxane and the solvent was removed by lyophilization, leaving **14** as a solid (425 mg, quant.), mp 110~111.5°C, $[\alpha]_D +24.6^\circ$ ($c=0.6$, 1:1 dichloromethane-methanol); IR $\nu_{\max}$ (KBr) cm^{-1} : 3300 (OH, NH), 2980 and 2850 (C-H), 1740 (ester), 1700 (S-acyl), and 1650 and 1550 (amide).

Anal. Found: C, 53.38; H, 7.66; N, 8.41. Calcd. for $\text{C}_{36}\text{H}_{63}\text{N}_5\text{O}_{13}\text{S}$: C, 53.65; H, 7.88; N, 8.69%.

N-[2-*O*-(2-Acetamido-1-*S*-[11-(*tert*-butoxycarbonyl-amino)undecanoyl]-6-*O*-decanoyl-2,3-dideoxy-1-thio- β -D-glucopyranose-3-yl]-D-lactoyl]-L-alanyl-D-isoglutamine methyl ester (**15**). Compound **14** (200 mg, 0.25 mmol) in 1,4-dioxane-DMF (10:1, 3 ml) was treated with decanoic acid (55.6 mg, 0.32 mmol) in the presence of DCC (180 mg, 0.89 mmol) and DMAP (a catalytic amount), and the mixture was worked up as described for **6** to give **15** (204 mg, 86%), mp 141~144°C, $[\alpha]_D +7.9^\circ$ ($c=1.9$, dichloromethane); IR $\nu_{\max}$ (film) cm^{-1} : 3300 (OH, NH), 2950 and 2860 (C-H), 1750 (ester), 1690 (S-acyl), and 1670 and 1540 (amide); NMR (CDCl_3) δ : 0.88 (t, 3H, acyl-Me), 1.26~1.44 (m, 45H, lac-Me, ala-Me, *tert*-Boc, and $-\text{CH}_2-$), 1.92 (s, 3H, AcN), 3.62 (s, 3H, MeO), and 5.14 (d, 1H, $J_{1,2}=10.6\text{ Hz}$, H-1).

Anal. Found: C, 57.71; H, 8.76; N, 7.44. Calcd. for $\text{C}_{46}\text{H}_{81}\text{N}_5\text{O}_{14}\text{S}$: C, 57.54; H, 8.50; N, 7.29%.

N-[2-*O*-(2-Acetamido-1-*S*-[11-(*tert*-butoxycarbonyl-amino)undecanoyl]-2,3-dideoxy-6-*O*-octadecanoyl-1-thio- β -D-glucopyranose-3-yl]-D-lactoyl]-L-alanyl-D-isoglutamine methyl ester (**16**). Compound **16** was obtained in a 65% yield as described for **15**.

Compound **16** had mp 150~150.5°C and $[\alpha]_D +14.5^\circ$ ($c=2.5$, 1:1 dichloromethane-methanol); the IR and NMR data were consistent with the structures assigned.

Anal. Found: C, 60.59; H, 9.11; N, 6.41. Calcd. for

$\text{C}_{54}\text{H}_{97}\text{N}_5\text{O}_{14}\text{S}$: C, 60.48; H, 9.12; N, 6.53%.

Trifluoroacetates of 1-S-(11-aminoundecanoyl) derivatives (**17**, **18** and **19**). Compounds **17**, **18** and **19** were each obtained in quantitative yields as described for **9**.

Compound **17** had $[\alpha]_D +13.9^\circ$ ($c=0.2$, ethanol).

Anal. Found: C, 48.36; H, 6.77; N, 8.39. Calcd. for $\text{C}_{33}\text{H}_{56}\text{N}_5\text{O}_{13}\text{F}_3\text{S}$: C, 48.34; H, 6.88; N, 8.54%.

Compound **18** had mp 84~86.5°C and $[\alpha]_D +23.0^\circ$ ($c=4.4$, methanol).

Anal. Found: C, 52.93; H, 7.40; N, 6.99. Calcd. for $\text{C}_{43}\text{H}_{74}\text{N}_5\text{O}_{14}\text{F}_3\text{S}$: C, 53.02; H, 7.66; N, 7.19%.

Compound **19** had mp 156.5~157.5°C and $[\alpha]_D +16.3^\circ$ ($c=1.1$, 1:1 dichloromethane-methanol).

Anal. Found: C, 56.28; H, 8.50; N, 6.03. Calcd. for $\text{C}_{53}\text{H}_{92}\text{N}_5\text{O}_{15}\text{F}_3\text{S}$: C, 56.41; H, 8.22; N, 6.21%.

2-(Trimethylsilyl)ethyl 2-acetamido-3,4,6-tri-*O*-acetyl-2-deoxy- β -D-glucopyranoside (**21**). To a solution of 2-acetamido-1,3,4,6-tetra-*O*-acetyl-2-deoxy- β -D-glucopyranose (**20**; 4.0 g, 10.3 mmol) in dichloromethane (50 ml) was added anhydrous ferric chloride (3.2 g, 19.7 mmol), and the mixture was vigorously stirred for 1.5 hr at room temperature. Sodium hydrogen carbonate was added to neutralize the solution, and the precipitates were filtered off and washed with dichloromethane. The filtrate and washings were combined, washed with water, dried (sodium sulfate), and concentrated. The resulting oxazoline was treated with 2-(trimethylsilyl)ethanol (1.77 g, 15 mmol) and a catalytic amount of conc. sulfuric acid in dichloromethane (50 ml); stirring was continued overnight at room temperature. The mixture was washed with 1M sodium hydrogen carbonate and water, dried (sodium sulfate) and evaporated. The residue was chromatographed on a column of silica gel with dichloromethane-methanol (400:1). The product was crystallized from hexane to afford **21** (4.3 g, 96%); mp 152~155°C, $[\alpha]_D -7.7^\circ$ ($c=0.8$, dichloromethane); IR $\nu_{\max}$ (film) cm^{-1} : 3280 (NH), 1740 (ester), 1650 and 1560 (amide), and 860 and 830 (Si-C); NMR (CDCl_3) δ : 0.0 (s, 9H, Me_3Si), 0.87 (m, 2H, $\text{SiMe}_3-\text{CH}_2-$), 1.88, 1.96, 1.97, and 2.03 (4s, 12H, Ac), 3.50 (m, 1H, $\text{SiMe}_3-\text{CH}_2-\text{CH}_A-$), 3.64 (m, 1H, H-5), 3.69 (q, 1H, $J_{1,2}=J_{2,3}=J_{2,\text{NH}}=8.8\text{ Hz}$, H-2), 3.90 (m, 1H, $\text{SiMe}_3-\text{CH}_2-\text{CH}_B-$), 4.06 (br. s, 1H, H-6_a), 4.21 (dd, 1H, $J_{\text{gem}}=12\text{ Hz}$, $J_{5,6b}=4.7\text{ Hz}$, H-6_b), 4.67 (d, 1H, $J_{1,2}=8.4\text{ Hz}$, H-1), 5.00 (t, 1H, $J_{3,4}=J_{4,5}=10\text{ Hz}$, H-3), and 5.48 (d, 1H, $J_{2,\text{NH}}=8.8\text{ Hz}$, NH).

Anal. Found: C, 50.84; H, 7.63; N, 3.09. Calcd. for $\text{C}_{19}\text{H}_{33}\text{NO}_9\text{Si}$: C, 50.99; H, 7.43; N, 3.13%.

2-(Trimethylsilyl)ethyl 2-amino-2-deoxy-4,6-*O*-isopropylidene- β -D-glucopyranoside (**22**). To a solution of **21** (4 g, 8.9 mmol) in methanol (100 ml) was added sodium methoxide in methanol (28% soln., 1 ml), and the mixture was stirred for 30 min at room temperature. Amberlite IR-120 (H^+) resin was added to deionize the solution, and then the resin was filtered off and washed with methanol. The

filtrate and washings were combined and concentrated. The resulting 2-(trimethylsilyl)ethyl 2-acetamido-2-deoxy- β -D-glucopyranoside was dissolved in DMF (100 ml) and treated with 2,2-dimethoxypropane (10 ml) in the presence of a catalytic amount of *p*-toluenesulfonic acid monohydrate for 1 hr at room temperature. Sodium methoxide in methanol (28% soln., 1 ml) was added, and then the solvent was evaporated. The residue was treated with barium hydroxide octahydrate (6 g, 19 mmol) in water-ethanol (2:1, 120 ml) for 2 days under refluxing conditions. The product was extracted with chloroform, before the organic layer was washed with water, dried (sodium sulfate) and concentrated to a syrup, which was chromatographed on a column of silica gel with dichloromethane-methanol (100:1). The product was dissolved in 1,4-dioxane and the dioxane was removed by lyophilization, leaving **22** (2.6 g, 92%) as a solid, mp 110~111°C, $[\alpha]_D -59.0^\circ$ ($c=0.5$, dichloromethane); IR $\nu_{\max}$ (film) cm^{-1} : 3400 (OH, NH), 1580 (NH₂), and 850 (Si-C, Me₂C); NMR (CDCl₃) δ : 0.0 (s, 9H, Me₃Si), 1.00 (m, 2H, Me₃Si-CH₂-), 1.40 and 1.48 (2s, 6H, Me₂C), 1.92 (br. s, 3H, OH and NH₂), 2.71 (t, 1H, $J=8$ Hz, H-2), 3.23 (m, 1H, H-5), 3.4~3.6 (m, 3H, Me₃Si-CH₂-CH_a-, H-3 and 4), 3.78 (t, 1H, $J_{\text{gem}}=J_{5,6a}=10$ Hz, H-6_a), 3.89 (dd, 1H, $J_{\text{gem}}=10$ Hz, $J_{5,6b}=5.5$ Hz, H-6_b), 3.95 (m, 1H, Me₃Si-CH₂-CH_b-), and 4.21 (d, 1H, $J_{1,2}=8$ Hz, H-1).

Anal. Found: C, 52.47; H, 9.21; N, 4.12. Calcd. for C₁₄H₂₉NO₅Si: C, 52.63; H, 9.15; N, 4.38%.

2-(Trimethylsilyl)ethyl 2-deoxy-2-[(3R)-3-hydroxytetradecanamido]-4,6-O-isopropylidene- β -D-glucopyranoside (**23**). A mixture of **22** (2.6 g, 8.1 mmol), (3R)-3-hydroxytetradecanoic acid (2.4 g, 9.8 mmol) and WSC (4.0 g, 20.9 mmol) in dichloromethane (100 ml) was stirred overnight at room temperature, before the reaction mixture was chromatographed on a column of silica gel with dichloromethane-methanol (150:1) to give **23** (3.7 g, 84%) as a syrup: $[\alpha]_D -40^\circ$ ($c=1.8$, dichloromethane); IR $\nu_{\max}$ (film) cm^{-1} : 3300 (OH, NH), 2950 and 2850 (C-H), 1640 and 1550 (amide), and 860 (Si-C, Me₂C); NMR (CDCl₃) δ : 0.0 (s, 9H, Me₃Si), 0.9 (m, 5H, Me₃Si-CH₂- and Me), 1.2~1.6 (m, 20H, -CH₂-), 1.41 and 1.50 (2s, 6H, Me₂C), 2.2~2.5 (m, 2H, -COCH₂-), 4.83 (d, 1H, $J_{1,2}=8.4$ Hz, H-1), and 6.50 (br. s, 1H, NH).

Anal. Found: C, 61.33; H, 10.78; N, 2.36. Calcd. for C₂₈H₅₅NO₇Si: C, 61.61; H, 10.61; N, 2.57%.

2-(Trimethylsilyl)ethyl 2-deoxy-4,6-O-isopropylidene-3-O-tetradecanoyl-2-[(3R)-3-tetradecanoyloxytetradecanamido]- β -D-glucopyranoside (**24**). To a solution of **23** (3.5 g, 6.9 mmol) and DMAP (1.3 g, 10.7 mmol) in pyridine (50 ml) was added tetradecanoyl chloride (5.1 g, 20.7 mmol), before the mixture was stirred overnight at room temperature. Methanol was added to decompose the excess reagent, and the solvent was evaporated. The residue was extracted with chloroform, and the extract was washed with 2M hydrochloric acid and water, dried (sodium sulfate) and concentrated to a syrup, which was

chromatographed on a column of silica gel with dichloromethane. The title compound **24** (6.2 g, 92%) was dissolved in 1,4-dioxane and lyophilized to leave a solid, mp 47.5°C, $[\alpha]_D -17.1^\circ$ ($c=3.7$, dichloromethane); IR $\nu_{\max}$ (film) cm^{-1} : 3400 (OH, NH), 2930 and 2850 (C-H), 1740 (ester), 1650 and 1520 (amide), and 860 and 840 (Si-C, Me₂C); NMR (CDCl₃) δ : 0.0 (s, 9H, Me₃Si), 0.9 (m, 11H, Me₃Si-CH₂- and Me), 1.1~1.7 (m, 64H, -CH₂-), 1.36 and 1.46 (2s, 6H, Me₂C), 2.2~2.5 (m, 6H, -COCH₂-), 3.35 (m, 1H, H-5), 3.55 (m, 1H, Me₃Si-CH₂-CH_a-), 3.65~4.05 (m, 5H, H-2, 4, 6, and Me₃Si-CH₂-CH_b-), 4.53 (d, 1H, $J_{1,2}=8$ Hz, H-1), 5.05 (m, 2H, H-3 of C₁₄-O-C₁₄ and H-3), and 6.05 (br., 1H, NH).

Anal. Found: C, 69.39; H, 11.28; N, 1.26. Calcd. for C₅₆H₁₀₇NO₉Si: C, 69.59; H, 11.16; N, 1.45%.

2-(Trimethylsilyl)ethyl 2-deoxy-4,6-O-isopropylidene-2-[(3R)-3-[2-(trimethylsilyl)ethoxymethoxy]tetradecanamido]- β -D-glucopyranoside (**25**). To a solution of **22** (2.0 g, 6.3 mmol) in dichloromethane (30 ml) was added (3R)-3-[2-(trimethylsilyl)ethoxymethoxy]tetradecanoic acid $[\alpha]_D -1.4^\circ$ ($c=0.8$, dichloromethane), 2.98 g, 8.0 mmol} and WSC (2.4 g, 12.6 mmol). The mixture was stirred overnight at room temperature, and processed as described for **23**, to give **25** (3.6 g, 82%), mp 74~75.5°C, $[\alpha]_D -32.8^\circ$ ($c=1.3$, dichloromethane); IR $\nu_{\max}$ (KBr) cm^{-1} : 3490 (OH), 3300 (NH), 2930 and 2850 (C-H), 1640 and 1550 (amide), and 860 and 840 (Si-C, Me₂C); NMR (CDCl₃) δ : 0.0 (s, 18H, Me₃Si), 0.85 (m, 7H, Me₃Si-CH₂- and Me), 1.2~1.6 (m, 20H, -CH₂-), 3.2~4.0 (m, 11H, Me₃Si-CH₂-CH₂-, H-2, 3, 4, 5, 6 and H-3 of C₁₄-OSEM), 4.7 (m, 4H, -OCH₂O-, H-1, and OH), and 6.44 (d, 1H, $J=5.5$ Hz, NH).

Anal. Found: C, 60.17; H, 10.63; N, 1.98. Calcd. for C₃₄H₆₉NO₈Si₂: C, 60.40; H, 10.39; N, 2.09%.

2-(Trimethylsilyl)ethyl 2-deoxy-4,6-O-isopropylidene-3-O-[(3R)-3-tetradecanoyloxytetradecanoyl]-2-[(3R)-3-[2-(trimethylsilyl)ethoxymethoxy]tetradecanamido]- β -D-glucopyranoside (**26**). To a solution of **25** (3.5 g, 5.2 mmol) in dichloromethane (50 ml) were added (3R)-3-tetradecanoyloxytetradecanoic acid (3 g, 6.3 mmol), WSC (2.0 g, 10.5 mmol) and DMAP (100 mg, 0.8 mmol), the mixture being stirred overnight at room temperature. The product was purified by chromatography on a column of silica gel with dichloromethane to afford **26** (5.8 g, quant.) as a syrup, $[\alpha]_D -12.6^\circ$ ($c=0.8$, dichloromethane); IR $\nu_{\max}$ (film) cm^{-1} : 3300 (NH), 2930 and 2850 (C-H), 1740 (ester), 1650 and 1550 (amide), and 860 and 840 (Si-C, Me₂C); NMR (CDCl₃) δ : 0.0 (s, 18H, Me₃Si), 0.85 (m, 13H, Me₃Si-CH₂- and Me), 1.1~1.7 (m, 62H, -CH₂-), 1.40 and 1.49 (2s, 6H, Me₂C), 2.2~2.5 (m, 6H, -COCH₂-), 3.2~4.0 (m, 10H, Me₃Si-CH₂-CH₂-, H-2, 4, 5, 6, and H-3 of C₁₄-OSEM), 4.45~4.65 (m, 3H, -OCH₂O- and H-1), 5.05 (m, 2H, H-3 of C₁₄-O-C₁₄ and H-3), and 6.28 (d, 1H, $J=9.1$ Hz, NH).

Anal. Found: C, 67.06; H, 11.03; N, 1.38. Calcd. for

$C_{62}H_{12}NO_{11}Si_2$: C, 66.92; H, 10.96; N, 1.26%.

2-(Trimethylsilyl)ethyl 2-deoxy-3-O-tetradecanoyl-2-[(3R)-3-tetradecanoyloxytetradecanamido]- β -D-glucopyranoside (**27**). A mixture of **24** (6.2 g, 6.4 mmol) and 80% aqueous acetic acid (300 ml) was stirred for 3 hr at 50°C. The mixture was concentrated and chromatographed on a column of silica gel with dichloromethane-methanol (100:1) to give **27** (5.2 g, 88%), which was lyophilized from 1,4-dioxane solution, mp 82~84°C, $[\alpha]_D -12.3^\circ$ ($c=0.5$, dichloromethane); IR ν_{max} (KBr) cm^{-1} : 3450 (OH), 3300 (NH), 2900 and 2850 (C-H), 1720 (ester), 1650 and 1550 (amide), and 850 and 830 (Si-C); NMR ($CDCl_3$) δ : 0.0 (s, 9H, Me_3Si), 0.87 (m, 11H, Me_3Si-CH_2- and Me), 1.1~1.7 (m, 64H, $-CH_2-$), 2.1~2.5 (m, 6H, $-COCH_2-$), 3.42 (m, 1H, H-5), 3.52 (m, 1H, $Me_3Si-CH_2-CH_a-$), 3.71 (t, 1H, $J_{3,4}=J_{4,5}=9.3$ Hz, H-4), 3.75~3.95 (m, 4H, H-2, 6, and $Me_3Si-CH_2-CH_b-$), 4.61 (d, 1H, $J_{1,2}=8.3$ Hz, H-1), 5.09 (m, 2H, H-3 of $C_{14}-O-C_{14}$ and H-3), and 6.06 (d, 1H, $J=7.8$ Hz, NH).

Anal. Found: C, 68.48; H, 11.43; N, 1.72. Calcd. for $C_{53}H_{103}NO_9Si$: C, 68.71; H, 11.21; N, 1.51%.

2-(Trimethylsilyl)ethyl 6-O-tert-butyltrimethylsilyl-2-deoxy-3-O-tetradecanoyl-2-[(3R)-3-tetradecanoyloxytetradecanamido]- β -D-glucopyranoside (**28**). To a solution of **27** (3.0 g, 3.2 mmol) in pyridine (30 ml) was added tert-butyltrimethylsilyl chloride (1 g, 6.7 mmol), before stirring overnight at room temperature. Methanol was added, and the solvent was then evaporated. The residue was chromatographed on a column of silica gel with dichloromethane-methanol (400:1) and lyophilized from 1,4-dioxane solution to afford **28** (3.3 g, 98%), mp 58.5°C, $[\alpha]_D -16.4^\circ$ ($c=3.8$, dichloromethane); IR ν_{max} (film) cm^{-1} : 3450 (OH), 3300 (NH), 2900 and 2850 (C-H), 1720 and 1700 (ester), 1650 and 1550 (amide), and 830 (Si-C); NMR ($CDCl_3$) δ : 0.0 (s, 15H, Me-Si), 0.9 (m, 20H, Me_3Si-CH_2- , tert-Bu, and Me), 1.1~1.7 (m, 64H, $-CH_2-$), 2.1~2.5 (m, 6H, $-COCH_2-$), 3.28 (br. s, 1H, OH), 3.43 (m, 1H, H-5), 3.55 (m, 1H, $Me_3Si-CH_2-CH_a-$), 3.70 (t, 1H, $J_{3,4}=J_{4,5}=9.2$ Hz, H-4), 3.8~4.0 (m, 4H, H-2, 6, and $Me_3Si-CH_2-CH_b-$), 4.52 (d, 1H, $J_{1,2}=8.4$ Hz, H-1), 5.04 (m, 2H, H-3 of $C_{14}-O-C_{14}$ and H-3), and 5.83 (d, 1H, $J=8.8$ Hz, NH).

Anal. Found: C, 67.89; H, 11.48; N, 1.56. Calcd. for $C_{59}H_{117}NO_9Si_3$: C, 68.09; H, 11.33; N, 1.35%.

2-(Trimethylsilyl)ethyl 4-O-[bis(2,2,2-trichloroethyl)-phosphono]-6-O-tert-butyltrimethylsilyl-2-deoxy-3-O-tetradecanoyl-2-[(3R)-3-tetradecanoyloxytetradecanamido]- β -D-glucopyranoside (**29**). A mixture of **28** (0.5 g, 0.5 mmol), dichloromethane (5 ml), *N,N*-diisopropylethylamine (0.6 g, 4.6 mmol), a catalytic amount of DMAP, and bis(2,2,2-trichloroethyl) phosphorochloridate (0.75 g, 2 mmol) was stirred for 1 hr at room temperature. The reaction mixture was extracted with chloroform, and the extract was successively washed with 2 M hydrochloric acid and water,

dried (sodium sulfate) and concentrated. The residual syrup was chromatographed on a column of silica gel with dichloromethane-methanol (400:1) to give **29** (0.56 g, 84%) and lyophilized from 1,4-dioxane solution, mp 50~52.5°C, $[\alpha]_D -1.9^\circ$ ($c=1.3$, dichloromethane); IR ν_{max} (film) cm^{-1} : 3300 (NH), 2900 and 2830 (C-H), 1740 (ester), 1650 and 1550 (amide), 910 (P-O- CH_2), 860 and 840 (Si-C), and 720 (C-Cl); NMR ($CDCl_3$) δ : 0.0 (s, 15H, Me-Si), 0.9 (m, 20H, Me_3Si-CH_2- , tert-Bu, and Me), 1.0~1.75 (m, 64H, $-CH_2-$), 2.2~2.5 (m, 6H, $-COCH_2-$), 4.45~4.75 (m, 6H, H-1, 4, and CCl_3CH_2-), 5.00 (m, 1H, H-3 of $C_{14}-O-C_{14}$), 5.37 (t, 1H, $J_{2,3}=J_{3,4}=9$ Hz, H-3), and 5.90 (d, 1H, $J=8.4$ Hz, NH).

Anal. Found: C, 54.78; H, 8.94; N, 1.08. Calcd. for $C_{63}H_{120}NO_{12}Cl_6PSi_2$: C, 54.69; H, 8.74; N, 1.01%.

2-(Trimethylsilyl)ethyl 4-O-[bis(2,2,2-trichloroethyl)-phosphono]-2-deoxy-3-O-tetradecanoyl-2-[(3R)-3-tetradecanoyloxytetradecanamido]- β -D-glucopyranoside (**30**). A mixture of **29** (0.5 g, 0.4 mmol) and 80% aqueous acetic acid (50 ml) was stirred for 3 hr at 50°C, before the solvent was evaporated. The residue was chromatographed on a column of silica gel with dichloromethane-methanol (300:1) to give **30** (0.46 g, 75%), which was lyophilized from 1,4-dioxane solution, mp 69~71°C, $[\alpha]_D -5.4^\circ$ ($c=0.3$, dichloromethane); IR ν_{max} (film) cm^{-1} : 3450 (OH), 3300 (NH), 2920 and 2850 (C-H), 1740 (ester), 1650 and 1550 (amide), 920 (P-O- CH_2), 860 and 840 (Si-C), and 720 (C-Cl); NMR ($CDCl_3$) δ : 0.0 (s, 9H, Me_3Si), 0.9 (m, 11H, Me_3Si-CH_2- and Me), 1.1~1.75 (m, 64H, $-CH_2-$), 2.2~2.5 (m, 6H, $-COCH_2-$), 2.64 (br., 1H, OH), 3.5 (m, 2H, $Me_3Si-CH_2-CH_a-$ and H-5), 3.70 (q, 1H, $J_{1,2}=J_{2,3}=J_{2,NH}=8$ Hz, H-2), 3.94 (br. s, 3H, $Me_3Si-CH_2-CH_b-$ and H-6), 4.65 (m, 5H, CCl_3CH_2- and H-4), 4.80 (d, 1H, $J_{1,2}=8.4$ Hz, H-1), 4.99 (m, 1H, H-3 of $C_{14}-O-C_{14}$), 5.48 (t, 1H, $J_{2,3}=J_{3,4}=9.1$ Hz, H-3), and 6.09 (br. s, 1H, NH).

Anal. Found: C, 54.10; H, 8.66; N, 0.95. Calcd. for $C_{57}H_{106}NO_{12}Cl_6PSi$: C, 53.94; H, 8.42; N, 1.10%.

2-(Trimethylsilyl)ethyl 4-O-[bis(2,2,2-trichloroethyl)-phosphono]-6-O-(3-carboxypropanoyl)-2-deoxy-3-O-tetradecanoyl-2-[(3R)-3-tetradecanoyloxytetradecanamido]- β -D-glucopyranoside (**31**). To a solution of **30** (0.23 g, 0.2 mmol) and DMAP (a catalytic amount) in 1,2-dichloroethane (4 ml) was added succinic anhydride (0.16 g, 1.6 mmol). The mixture was stirred for 1.5 hr at room temperature, and extracted with chloroform. The extract was washed with 2 M hydrochloric acid and water, dried (sodium sulfate), and concentrated. The residue was chromatographed on a column of silica gel with dichloromethane-methanol (200:1) to afford **31** as a syrup (0.21 g, 84%), $[\alpha]_D -1.7^\circ$ ($c=0.8$, dichloromethane); IR ν_{max} (film) cm^{-1} : 3350 (NH), 2900 and 2800 (C-H), 300~2400 (COOH), 1750 and 1720 (C=O), 1650 and 1540 (amide), 920 (P-O- CH_2), 860 (Si-C), and 730 (C-Cl); NMR ($CDCl_3$) δ : 0.0 (s, 9H, Me_3Si), 0.9 (m, 11H, Me_3Si-CH_2- and Me), 1.0~1.75 (m, 64H, $-CH_2-$), 2.2~2.5 (m, 6H, $-COCH_2-$), 2.68 (s, 4H, $-CO(CH_2)_2CO-$), 3.55 (m,

1H, Me₃-CH₂-CH₂-), 3.75 (m, 2H, H-2 and 5), 3.90 (m, 1H, Me₃Si-CH₂-CH₂-), 4.27 (dd, 1H, $J_{\text{gem}} = 12$ Hz, $J_{5,6a} = 4.4$ Hz, H-6_a), 4.60 (m, 6H, H-4, 6_b, and CCl₃CH₂-), 4.73 (d, 1H, $J_{1,2} = 8.4$ Hz, H-1), 4.99 (m, 1H, H-3 of C₁₄-O-C₁₄), 5.41 (t, 1H, $J_{2,3} = J_{3,4} = 10$ Hz, H-3), and 6.09 (d, 1H, $J = 8.4$ Hz, NH).

Anal. Found: C, 53.38; H, 8.34; N, 1.13. Calcd. for C₆₁H₁₁₀NO₁₅Cl₆PSi₃: C, 53.51; H, 8.10; N, 1.02%.

2-(Trimethylsilyl)ethyl 4-O-[bis(2,2,2-trichloroethyl)-phosphono]-6-O-tert-butyltrimethylsilyl-2-deoxy-3-O-[(3R)-3-tetradecanoyloxytetradecanoyl]-2-[(3R)-3-[2-(trimethylsilyl)ethoxymethoxy]tetradecanamido]-β-D-glucopyranoside (**34**). A mixture of **26** (5.8 g, 5.2 mmol) and 80% aqueous acetic acid (300 ml) was treated in the usual manner to give **32** (4.5 g, 78%), $[\alpha]_D -3.3^\circ$ ($c = 0.12$, dichloromethane); IR ν_{max} (film) cm⁻¹: 3300 (OH, NH), 2930 and 2850 (C-H), 1740 (ester), 1650 and 1550 (amide), and 860 and 840 (Si-C); NMR (CDCl₃) δ : 0.0 (s, 18H, Me₃Si), 0.85 (m, 13H, Me₃Si-CH₂- and Me), 1.1~1.7 (m, 62H, -CH₂-), 2.1~2.5 (m, 6H, -COCH₂-), 3.3~3.9 (m, 10H, Me₃Si-CH₂-CH₂-, H-2, 4, 5, and H-3 of C₁₄-OSEM), 4.5~4.7 (m, 3H, -OCH₂O- and H-1), 4.99 (t, 1H, $J_{2,3} = J_{3,4} = 9.1$ Hz, H-3), 5.05 (m, 1H, H-3 of C₁₄-O-C₁₄), and 6.28 (d, 1H, $J = 9.2$ Hz, NH).

Anal. Found: C, 66.11; H, 11.15; N, 1.20. Calcd. for C₅₉H₁₁₇NO₁₁Si₂: C, 66.06; H, 10.99; N, 1.31%.

Compound **32** (2.7 g, 2.5 mmol) was treated with *tert*-butyldimethylsilyl chloride (0.75 g, 5 mmol) in pyridine (30 ml) overnight at room temperature, as described for **28**, to give **33** (2.85 g, 94%), mp 38~39°C, $[\alpha]_D -6.2^\circ$ ($c = 0.45$, dichloromethane); IR ν_{max} (film) cm⁻¹: 3400 (OH), 3300 (NH), 2930 and 2850 (C-H), 1740 (ester), 1650 and 1550 (amide), and 860 and 840 (Si-C); NMR (CDCl₃) δ : 0.0 (s, 24H, Me-Si), 0.7~1.0 (m, 22H, Me₃Si-CH₂- and Me), 1.1~1.7 (m, 62H, -CH₂-), 2.2~2.6 (m, 6H, -COCH₂-), 3.3~3.9 (m, 11H, Me₃Si-CH₂-CH₂-, H-2, 4, 5, 6, H-3 of C₁₄-OSEM, and OH), 4.45 (d, 1H, $J_{1,2} = 8.4$ Hz, H-1), 4.61 (2d, 2H, $J_{\text{gem}} = 7$ Hz, -OCH₂O-), 4.95 (t, 1H, $J_{2,3} = J_{3,4} = 9.1$ Hz, H-3), 5.08 (m, 1H, H-3 of C₁₄-O-C₁₄), and 6.09 (d, 1H, $J = 9.2$ Hz, NH).

Anal. Found: C, 65.52; H, 11.01; N, 0.93. Calcd. for C₆₅H₁₃₁NO₁₁Si₃: C, 65.77; H, 11.12; N, 1.18%.

Compound **33** (0.3 g, 0.3 mmol) was phosphorylated with bis(2,2,2-trichloroethyl) phosphorochloridate (0.38 g, 1 mmol), *N,N*-diisopropylethylamine (0.2 g, 1.5 mmol), and a catalytic amount of DMAP in dichloromethane (10 ml), as described for **29**, to afford **34** (0.32 g, 83%), $[\alpha]_D +5.0^\circ$ ($c = 1.2$, dichloromethane); IR ν_{max} (film) cm⁻¹: 3300 (NH), 2930 and 2850 (C-H), 1740 (ester), 1650 and 1540 (amide), 910 (P-O-CH₂), 860 and 840 (Si-C), and 800~720 (C-Cl); NMR (CDCl₃) δ : 0.0 (m, 24H, Me-Si), 0.8~1.0 (m, 22H, Me₃Si-CH₂- and Me), 1.1~1.7 (m, 62H, -CH₂-), 2.25 and 2.6 (2m, 6H, -COCH₂-), 3.45~3.75 and 3.8~4.0 (2m, 9H, Me₃Si-CH₂-CH₂-, H-2, 5, 6, and H-3 of C₁₄-OSEM), 4.41 (q, 1H, $J_{3,4} = J_{4,5} = J_{4,P} = 9.2$ Hz, H-4), 4.5~4.75 (m, 6H, -OCH₂O- and CCl₃CH₂-),

4.93 (d, 1H, $J_{1,2} = 8.4$ Hz, H-1), 5.19 (m, 1H, H-3 of C₁₄-O-C₁₄), 5.50 (t, 1H, $J_{2,3} = J_{3,4} = 9.2$ Hz, H-3), and 6.36 (d, 1H, $J = 8.1$ Hz, NH).

Anal. Found: C, 54.41; H, 8.53; N, 1.03. Calcd. for C₆₉H₁₃₄NO₁₄Cl₆PSi₃: C, 54.17; H, 8.83; N, 0.92%.

2-(Trimethylsilyl)ethyl 4-O-[bis(2,2,2-trichloroethyl)-phosphono]-6-O-(3-carboxypropanoyl)-2-deoxy-3-O-[(3R)-3-tetradecanoyloxytetradecanoyl]-2-[(3R)-3-[2-(trimethylsilyl)ethoxymethoxy]tetradecanamido]-β-D-glucopyranoside (**36**). Compound **34** (0.43 g, 0.3 mmol) was treated with 80% aqueous acetic acid (100 ml) at 50°C, as described for **30**, to give **35** (0.34 g, 86%), $[\alpha]_D +2.1^\circ$ ($c = 0.75$, dichloromethane); IR ν_{max} (film) cm⁻¹: 3450 (OH), 3300 (NH), 2930 and 2850 (C-H), 1740 (ester), 1650 and 1550 (amide), 910 (P-O-CH₂), 860 and 840 (Si-C), and 800~720 (C-Cl); NMR (CDCl₃) δ : 0.0 (m, 18H, Me₃Si), 0.7~0.95 (m, 13H, Me₃Si-CH₂- and Me), 1.1~1.8 (m, 62H, -CH₂-), 2.25 and 2.55 (2m, 6H, -COCH₂-), 2.83 (br. s, 1H, OH), 3.4~3.65 and 3.75~3.95 (2m, 9H, Me₃Si-CH₂-CH₂-, H-2, 5, 6, and H-3 of C₁₄-OSEM), 4.45~4.65 (m, 7H, H-4, -OCH₂O-, and CCl₃CH₂-), 4.92 (d, 1H, $J_{1,2} = 8$ Hz, H-1), 5.12 (m, 1H, H-3 of C₁₄-O-C₁₄), 5.47 (t, 1H, $J_{2,3} = J_{3,4} = 9.2$ Hz, H-3), and 6.43 (d, 1H, $J = 8$ Hz, NH).

Anal. Found: C, 53.58; H, 8.34; N, 0.75. Calcd. for C₆₃H₁₂₀NO₁₄Cl₆PSi₂: C, 53.46; H, 8.55; N, 0.99%.

A mixture of compound **35** (0.5 g, 0.4 mmol) and succinic anhydride (0.35 g, 3.5 mmol) in the presence of DMAP in 1,2-dichloroethane (8 ml) was processed as described for **12** to afford **17** (0.5 g, 95%), $[\alpha]_D +7.4^\circ$ ($c = 1.1$, dichloromethane); IR ν_{max} (film) cm⁻¹: 3300 (NH), 2930 and 2850 (C-H), 3400~2400 (COOH), 1740 and 1720 (ester), 1650 and 1550 (amide), 910 (P-O-CH₂), 860 and 840 (Si-C), and 800~720 (C-Cl); NMR (CDCl₃) δ : 0.0 (s, 18H, Me₃Si), 0.8~1.0 (m, 13H, Me₃Si-CH₂- and Me), 1.1~1.7 (m, 62H, -CH₂-), 2.2~2.35 (m, 4H, -COCH₂-), 2.66 [br. s, 6H, -COCH₂- and -CO(CH₂)₂-CO-], 3.35~3.49 (m, 8H, Me₃Si-CH₂-CH₂-, H-2, 5, 6_a, and H-3 of C₁₄-OSEM), 4.23 (dd, 1H, $J_{\text{gem}} = 12.1$ Hz, $J_{5,6} = 5.2$ Hz, H-6_b), 4.52 (q, 1H, $J_{3,4} = J_{4,5} = J_{4,P} = 8.8$ Hz, H-4), 4.50~4.75 (m, 6H, -OCH₂O- and CCl₃CH₂-), 4.88 (d, 1H, $J_{1,2} = 8.4$ Hz, H-1), 5.21 (m, 1H, H-3 of C₁₄-O-C₁₄), 5.50 (t, 1H, $J_{2,3} = J_{3,4} = 8.8$ Hz, H-3), and 6.55 (d, 1H, $J = 8.1$ Hz, NH).

Anal. Found: C, 52.88; H, 7.95; N, 0.72. Calcd. for C₆₇H₁₂₄NO₁₇Cl₆PSi₂: C, 53.10; H, 8.25; N, 0.92%.

6-O-[11-[3-[2-(Trimethylsilyl)ethyl 4-O-[bis(2,2,2-trichloroethyl)phosphono]-2-deoxy-3-O-tetradecanoyl]-2-[(3R)-3-tetradecanoyloxytetradecanamido]-β-D-glucopyranoside-6-yl-oxycarbonyl]propionamido]undecanoyl]-1-S-acetyl-1-thio-MDP methyl ester (**37**). To a solution of **31** (150 mg, 0.11 mmol) in 1,4-dioxane (2 ml) were added DCC (50 mg, 0.25 mmol) and *N*-hydroxysuccinimide (15.5 mg, 0.13 mmol). The mixture was stirred for 30 min at room temperature, DC urea was filtered off and washed

Table II.

Compound No.	Yield (%)	mp (°C)	[α] _D	Molecular formula	Found (Calcd.) % of		
					C	H	N
38	46	151	+6.7° (<i>c</i> = 1.2, 1 : 1 CH ₂ Cl ₂ –CH ₃ OH)	C ₁₀₂ H ₁₈₁ Cl ₆ N ₆ O ₂₆ PSSi	55.18 (55.40)	8.49 (8.25)	3.67 (3.80)
39	70	152.5	+7.1° (<i>c</i> = 1.1, 1 : 1 CH ₂ Cl ₂ –CH ₃ OH)	C ₁₁₀ H ₁₉₇ Cl ₆ N ₆ O ₂₆ PSSi	56.71 (56.84)	8.81 (8.54)	3.34 (3.62)
46	42	139	+2.9° (<i>c</i> = 1.1, CH ₂ Cl ₂)	C ₁₀₀ H ₁₇₉ Cl ₆ N ₆ O ₂₈ PSSi ₂	53.40 (53.49)	7.96 (8.04)	3.50 (3.74)
47	46	166	+3.8° (<i>c</i> = 0.9, CH ₂ Cl ₂)	C ₁₀₈ H ₁₉₅ Cl ₆ N ₆ O ₂₈ PSSi ₂	54.99 (55.02)	8.21 (8.34)	3.57 (3.56)
48	36	184	+1.2° (<i>c</i> = 1.8, CH ₂ Cl ₂)	C ₁₁₆ H ₂₁₁ Cl ₆ N ₆ O ₂₈ PSSi ₂	56.49 (56.41)	8.40 (8.61)	3.56 (3.40)
55	60	148	+9.9° (<i>c</i> = 1.7, 1 : 1 CH ₂ Cl ₂ –CH ₃ OH)	C ₁₀₂ H ₁₈₁ Cl ₆ N ₆ O ₂₆ PSSi	55.60 (55.40)	7.95 (8.25)	3.93 (3.80)
58	44	130	+7.4° (<i>c</i> = 1.6, CH ₂ Cl ₂)	C ₉₈ H ₁₇₇ Cl ₆ N ₆ O ₂₇ PSSi ₂	53.56 (53.42)	8.27 (8.10)	3.97 (3.81)
59	61	144 ~ 145	+5.2° (<i>c</i> = 1.5, CH ₂ Cl ₂)	C ₁₀₈ H ₁₉₅ Cl ₆ N ₆ O ₂₈ PSSi ₂	55.12 (55.02)	8.36 (8.34)	3.48 (3.56)
60	54	146.5	+3.3° (<i>c</i> = 1.3, CH ₂ Cl ₂)	C ₁₁₆ H ₂₁₁ Cl ₆ N ₆ O ₂₈ PSSi ₂	56.11 (56.41)	8.73 (8.61)	3.19 (3.40)
41	85	101	+18.3° (<i>c</i> = 1.1, 1 : 1 CH ₂ Cl ₂ –CH ₃ OH)	C ₉₇ H ₁₆₉ Cl ₆ N ₆ O ₂₆ PS	54.95 (55.18)	8.21 (8.07)	3.77 (3.98)
42	70	105	+16.5° (<i>c</i> = 1.6, 1 : 1 CH ₂ Cl ₂ –CH ₃ OH)	C ₁₀₅ H ₁₈₅ Cl ₆ N ₆ O ₂₆ PS	56.81 (56.72)	8.56 (8.39)	3.98 (3.78)
49	88	95 ~ 96	+8.3° (<i>c</i> = 0.7, 1 : 1 CH ₂ Cl ₂ –CH ₃ OH)	C ₈₉ H ₁₅₃ Cl ₆ N ₆ O ₂₇ PS	53.20 (53.05)	7.91 (7.65)	4.25 (4.17)
50	90	70 ~ 72	+10.6° (<i>c</i> = 0.6, 1 : 1 CH ₂ Cl ₂ –CH ₃ OH)	C ₉₇ H ₁₆₉ Cl ₆ N ₆ O ₂₇ PS	54.48 (54.77)	7.84 (8.01)	4.02 (3.95)
51	75	73 ~ 75	+6.1° (<i>c</i> = 1.0, 1 : 1 CH ₂ Cl ₂ –CH ₃ OH)	C ₁₀₅ H ₁₈₅ Cl ₆ N ₆ O ₂₇ PS	56.02 (56.32)	8.20 (8.33)	3.60 (3.75)
56	90	85 ~ 88	+19.1° (<i>c</i> = 1.6, 1 : 1 CH ₂ Cl ₂ –CH ₃ OH)	C ₉₇ H ₁₆₉ Cl ₆ N ₆ O ₂₆ PS	55.01 (55.18)	8.21 (8.07)	4.06 (3.98)
61	85	103	+15.7° (<i>c</i> = 0.5, 1 : 1 CH ₂ Cl ₂ –CH ₃ OH)	C ₈₇ H ₁₅₁ Cl ₆ N ₆ O ₂₆ PS	53.11 (52.96)	7.63 (7.71)	4.02 (4.26)
62	85	80 ~ 82	+13.1° (<i>c</i> = 0.8, 1 : 1 CH ₂ Cl ₂ –CH ₃ OH)	C ₉₇ H ₁₆₉ Cl ₆ N ₆ O ₂₇ PS	54.93 (54.77)	8.30 (8.01)	3.78 (3.95)
63	92	107 ~ 108	+9.7° (<i>c</i> = 1.5, 1 : 1 CH ₂ Cl ₂ –CH ₃ OH)	C ₁₀₅ H ₁₈₅ Cl ₆ N ₆ O ₂₇ PS	56.36 (56.32)	8.39 (8.33)	3.50 (3.75)
44	75	172	+15.6° (<i>c</i> = 0.9, 1 : 1 CH ₂ Cl ₂ –CH ₃ OH)	C ₉₃ H ₁₆₇ N ₆ O ₂₆ PS	60.63 (60.43)	9.34 (9.11)	4.81 (4.55)
45	80	170 ~ 171	+17.6° (<i>c</i> = 0.9, 1 : 1 CH ₂ Cl ₂ –CH ₃ OH)	C ₁₀₁ H ₁₈₃ N ₆ O ₂₆ PS	61.61 (61.87)	9.64 (9.41)	4.52 (4.29)
52	90	161 ~ 163	+10.1° (<i>c</i> = 0.4, 1 : 1 CH ₂ Cl ₂ –CH ₃ OH)	C ₈₅ H ₁₅₁ N ₆ O ₂₇ PS	58.32 (58.27)	8.51 (8.69)	4.61 (4.80)
53	89	167 ~ 167.5	+13.2° (<i>c</i> = 0.5, 1 : 1 CH ₂ Cl ₂ –CH ₃ OH)	C ₉₃ H ₁₆₇ N ₆ O ₂₇ PS	60.10 (59.91)	9.19 (9.03)	4.50 (4.51)
54	80	173 ~ 174	+7.3° (<i>c</i> = 0.3, 1 : 1 CH ₂ Cl ₂ –CH ₃ OH)	C ₁₀₁ H ₁₈₃ N ₆ O ₂₇ PS	61.53 (61.37)	9.50 (9.33)	4.24 (4.25)
57	82	174 ~ 175	+18.2° (<i>c</i> = 1.2, 1 : 1 CH ₂ Cl ₂ –CH ₃ OH)	C ₉₃ H ₁₆₇ N ₆ O ₂₆ PS	60.19 (60.43)	9.30 (9.11)	4.26 (4.55)
64	87	177 ~ 178	+17.3° (<i>c</i> = 0.5, 1 : 1 CH ₂ Cl ₂ –CH ₃ OH)	C ₈₃ H ₁₄₉ N ₆ O ₂₆ PS	58.51 (58.30)	8.60 (8.78)	4.99 (4.91)
65	90	137 ~ 138	+14.3° (<i>c</i> = 0.5, 1 : 1 CH ₂ Cl ₂ –CH ₃ OH)	C ₉₃ H ₁₆₇ N ₆ O ₂₇ PS	59.68 (59.91)	8.73 (9.03)	4.76 (4.51)
66	88	168 ~ 169	+10.1° (<i>c</i> = 1.0, 1 : 1 CH ₂ Cl ₂ –CH ₃ OH)	C ₁₀₁ H ₁₈₃ N ₆ O ₂₇ PS	61.66 (61.37)	9.14 (9.33)	4.44 (4.25)

with dichloromethane. The filtrate and washings were combined and concentrated to give the activated compound, which was used for the next coupling reaction without further purification.

The compound just obtained and triethylamine (1 drop) were added to a solution of **9** (60 mg, 0.07 mmol) in DMF (1 ml) at room temperature. The mixture was stirred for 1 hr, and then concentrated to a syrup, which was chromatographed on a column of silica gel with dichloromethane-methanol (20:1) to afford the coupling compound **37** (82.9 mg, 57%), mp 145~146°C, $[\alpha]_D^{25} +5.9^\circ$ ($c=1.0$, 1:1 dichloromethane-methanol); IR $\nu_{\max}$ (film) cm^{-1} : 3280 (OH, NH), 2940 and 2850 (C-H), 1740 (ester), 1690 (AcS), 1570 and 1550 (amide), 920 (P-O-CH₂), 840 (Si-C), and 800 and 720 (C-Cl); NMR (CDCl₃ + CD₃OD) δ : 0.0 (s, 9H, Me₃Si), 0.88 (m, 11H, Me₃Si-CH₂- and acyl-Me), 1.41~1.25 (m, 86H, -CH₂-, lac-Me, and ala-Me), 1.92 (s, 3H, AcN), 2.35 (s, 3H, AcS), 2.71 (m, 2H, -OCOCH₂-), 3.68 (s, 3H, MeO), 4.62 (m, 4H, CCl₃CH₂-), 4.71 (d, 1H, $J_{1,2}=8.4\text{ Hz}$, H-1 of GLA-27 unit), 5.03 (m, 1H, H-3 of C₁₄-O-C₁₄), 5.11 (d, 1H, $J_{1,2}=10.6\text{ Hz}$, H-1 of MDP unit), and 5.38 (t, 1H, $J_{2,3}=J_{3,4}=9.5\text{ Hz}$, H-3 of GLA-27 unit).

Anal. Found: C, 53.49; H, 8.11; N, 3.81. Calcd. for C₉₄H₁₆₅N₆O₂₆Cl₆PSSi: C, 53.78; H, 7.92; N, 4.00%.

Other coupling products (38, 39, 46~48, 55 and 58~60). The title compounds were synthesized by coupling **10**, **11** and **18** with **31**, and **9~11** and **17~19** with **36**, respectively, as described for **37**. The IR and NMR spectra were consistent with the structures assigned, their yields and physical data being recorded in Table II.

6-O-[11-[3-[4-O-[Bis(2,2,2-trichloroethyl)phosphono]-2-deoxy-3-O-tetradecanoyl-2-[(3R)-3-tetradecanoyloxy-tetradecanamido]-D-glucopyranose-6-yl-oxycarbonyl]-propionamido]undecanoyl]-1-S-acetyl-1-thio-MDP methyl ester (40). Compound **37** (90 mg, 0.04 mmol) was dissolved in dichloromethane (1 ml), boron trifluoride etherate (0.1 ml) was added at 0°C, and the mixture was then stirred overnight at room temperature. Methanol and water were added, and the reaction mixture was neutralized with Amberlite IR-410 (OH⁻) resin. The resin was filtered off and washed with dichloromethane. The filtrate and washings were combined and concentrated to give the title compound **40** (84 mg, quant.) which was crystallized from water, mp 97~98.5°C, $[\alpha]_D^{25} +12.5$ ($c=2.1$, 1:1 dichloromethane-methanol); IR $\nu_{\max}$ (film) cm^{-1} : 3400 (OH, NH), 2940 and 2850 (C-H), 1720 (ester), 1680 (AcS), 1660 and 1530 (amide), 900 (P-O-CH₂), and 800 and 720 (C-Cl); NMR (CDCl₃ + CD₃OD) δ : 0.9 (t, 9H, acyl-Me), 1.29~1.62 (m, 86H, -CH₂-, lac-Me, and ala-Me), 1.92 (s, 3H, AcN), 2.36 (s, 3H, AcS), 2.59 (t, 2H, -COCH₂-), 3.08 (t, 2H, -CH₂NHCO-), 3.69 (s, 3H, MeO), 5.03~5.15 (m, 3H, H-1 of GLA-27 unit, H-3 of C₁₄-O-C₁₄, and H-1 of MDP unit), and 5.44 (t, 1H, $J_{2,3}=J_{3,4}=9.2\text{ Hz}$, H-3 of GLA-27 unit).

Anal. Found: C, 53.20; H, 7.90; N, 4.22. Calcd. for C₈₉H₁₅₃N₆O₂₆Cl₆PS: C, 53.48; H, 7.71; N, 4.20%.

Other desilylated derivatives (41, 42, 49~51, 56 and 61~63). Compounds **41**, **42**, **49~51**, **56** and **61~63** were obtained as crystals, by desilylating of **38**, **39**, **46~48**, **55** and **58~60** according to the method described for **40**. The IR and NMR data were consistent with the structures assigned, and their yields and physical data are recorded in Table II.

6-O-[11-[3-[2-Deoxy-4-O-phosphono-3-O-tetradecanoyl-2-[(3R)-3-tetradecanoyloxytetradecanamido]-D-glucopyranose-6-yl-oxycarbonyl]propionamido]undecanoyl]-1-S-acetyl-1-thio-MDP methyl ester (43). To a solution of **40** (80 mg, 0.04 mmol) in acetic acid (1.5 ml) was added zinc dust (900 mg) at 45°C. After 20 min, the solid was filtered off, and washed with acetic acid. The filtrate and washings were combined and concentrated. The residual syrup was dissolved in methanol, and the solution was deionized with Amberlite IR-120 (H⁺) resin. The resin was filtered off, and washed with methanol and dichloromethane. The filtrate and washings were combined and concentrated to yield **43** (69 mg, 92%), which was crystallized from water. **43** gave a positive test for the phosphate group, using the phosphomolybdate spray reagent,¹⁶⁾ mp 171°C, $[\alpha]_D^{25} +13.1^\circ$ ($c=1.4$, 1:1 dichloromethane-methanol); IR $\nu_{\max}$ (KBr) cm^{-1} : 3350 (OH, NH), 2940 and 2860 (C-H), 1740 (ester), 1680 (S-acyl), and 1660 and 1550 (amide).

Anal. Found: C, 58.66; H, 8.88; N, 4.96. Calcd. for C₈₅H₁₅₁N₆O₂₆PS: C, 58.80; H, 8.77; N, 4.84%.

Other deprotected conjugates (44, 45, 52~54, 57 and 64~66). These compounds were respectively prepared by de-trichloroethylation of **41**, **42**, **49~51**, **56** and **61~63**, as described for **43**, and showed similar IR data. Their yields and physical data are shown in Table II.

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