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SYNTHESIS AND BIOLOGICAL ACTIVITIES OF *N*-ACETYL-1-THIOMURAMOYL-L-ALANYL-D-ISOGLUTAMINE AND SOME OF ITS LIPOPHILIC DERIVATIVES*

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ABSTRACT

N-Acetyl-1-thiomuramoyl-L-alanyl-D-isoglutamine and some lipophilic analogs were synthesized from benzyl 2-acetamido-2-deoxy-4,6-*O*-isopropylidene-3-*O*-[D-1-(methoxycarbonyl)ethyl]-α-D-glucopyranoside (**1**). *O*-Debenzoylation of **2**, derived from **1** by oxidation, gave 2-acetamido-2-deoxy-4,6-*O*-isopropylidene-3-*O*-[D-1-(methoxycarbonyl)ethyl]-D-glucopyranose (**3**). Condensation of the alkoxytris(dimethylamino)phosphonium chloride (**4**), formed from **3** by the action of carbon tetrachloride and tris(dimethylamino)phosphine, with potassium thioacetate afforded 2-acetamido-1-*S*-acetyl-2-deoxy-4,6-*O*-isopropylidene-3-*O*-[D-1-(methoxycarbonyl)ethyl]-1-thio-β-D-glucopyranose (**8**). Coupling of the acid **9**, obtained from **8** by hydrolysis and subsequent *S*-acetylation, with the methyl ester of L-alanyl-D-isoglutamine gave *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 (**10**), which was converted, *via* *O*-deisopropylidenation, *S*-deacetylation, and de-esterification, into the *N*-acetyl-1-thiomuramoyl dipeptide. Condensation of **11** (derived from **10** by *S*-deacetylation) and of **12** (obtained from **10** by *S*-deacetylation and de-esterification) with various acyl chlorides yielded the corresponding 1-*S*-acyl-*N*-acetylmuramoyl-L-alanyl-D-isoglutamine derivatives, which were converted into the desired, lipophilic 1-thiomuramoyl dipeptides by cleavage of the isopropylidene group. Condensation of **11** with the alkyl bromides yielded the 1-*S*-alkyl derivatives, which were also converted, *via* *O*-deisopropylidenation and de-esterification, into the corresponding 1-*S*-alkylmuramoyl dipeptides. The biological activities were examined in guinea-pigs and mice.

*Studies on Immunoadjuvant Active Compounds, Part XXV. For Part XXIV, see ref. 1. For a preliminary report on part of this work, see ref. 4i.

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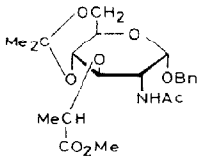
INTRODUCTION

In the course of continuing efforts to elucidate the relationships between the structure of the carbohydrate moiety and the biological activity of *N*-acetylmuramoyl dipeptide (MDP)[2], which is the minimal, adjuvantactive structure of bacterial, cell-wall peptidoglycan, and to obtain glycopeptide adjuvants that exhibit strong activity and lower toxicity, it has been demonstrated that not only is the restricted configuration of the sugar moiety important for the activity[3] but also that chemical modifications[4–7] of the functional groups in the carbohydrate moiety produce various, important effects on the manifestation of the activity. In view of these facts, we now describe the synthesis of *N*-acetyl-1-thiomuramoyl-L-alanyl-D-isoglutamine and its lipophilic derivatives bearing the lipid moiety at C-1 of the sugar skeleton, and their biological activities.

RESULTS AND DISCUSSION

Treatment of benzyl 2-acetamido-2-deoxy-4,6-*O*-isopropylidene-3-*O*-[D-1-(methoxycarbonyl)ethyl]-α-D-glucopyranoside (**1**) with chromium trioxide–pyridine complex[8] in the presence of acetic anhydride in dichloromethane at 45° gave crystalline **2** in 92% yield; this was hydrolyzed with sodium methoxide in methanol, to afford **3** in 93% yield. The action of carbon tetrachloride and tris(dimethylamino)phosphine[9] in dichloromethane at −60° on compound **3** gave, quantitatively, the alkoxytris(dimethylamino)phosphonium chloride (**4**); when evaporated at 40°, **4** was converted into a mixture of compounds **5**, **6**, and **7** (t.l.c.). A sol-



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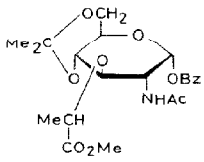

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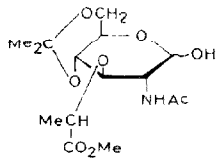

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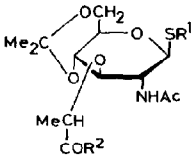
ution of the mixture in dry dichloromethane was evaporated at 40°, and this procedure was repeated several times, whereupon compounds **5** and **7** were completely converted into **6**, which was used for the next reaction without purification.

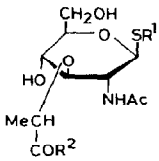
Treatment of **4** or **6** with potassium thioacetate gave 2-acetamido-1-*S*-acetyl-2-deoxy-4,6-*O*-isopropylidene-1-thio-β-D-glucopyranose (**8**) in 87 and 83.5% yields (based on **3**), respectively; significant signals in the n.m.r. spectrum were a three-proton singlet at δ 2.32 (*S*-acetyl) and a one-proton doublet at δ 5.09 ($J_{1,2}$ 10.8 Hz, H-1). Other n.m.r. data are given in the Experimental section, and are consistent with structure **8**.

benzoquinon-6-yl)decanoyl chloride, compound **11** yielded the 4,6-*O*-isopropylidene-1-*S*-(fatty acyl) derivatives (**15, 17, 19,** and **21**), which were converted, by acid hydrolysis, into the corresponding *N*-[2-*O*-(2-acetamido-1-*S*-acyl-2,3-dideoxy-1-thio-β-D-glucopyranose-3-yl)-D-lactoyl]-L-alanyl-D-isoglutamine methyl esters (**32, 34, 36,** and **38**).

In a similar way, condensation of **12**, derived from **10** by hydrolysis, with the fatty acyl chlorides (as already described) afforded compounds **14, 16, 18, 20,** and **22**, which were hydrolyzed by heating with 80% aqueous acetic acid for 2 h at 45°, yielding the desired *N*-[2-*O*-(2-acetamido-1-*S*-acyl-2,3-dideoxy-1-thio-β-D-glucopyranose-3-yl)-D-lactoyl]-L-alanyl-D-isoglutamines (**31, 33, 35, 37,** and **39**).

On the other hand, treatment of the sodium salt of **11**, formed from **10** by addition of sodium methoxide in methanol, with ethyl, decanyl, hexadecanyl, or

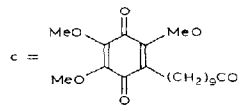




11 R^1 = H, R^2 = a
12 R^1 = H, R^2 = b
13–20; R^1 = $-CO(CH_2)_mMe$
13 m = 8, R^2 = a
14 m = 8, R^2 = b
15 m = 14, R^2 = a
16 m = 14, R^2 = b
17 m = 16, R^2 = a
18 m = 16, R^2 = b
19 m = 28, R^2 = a
20 m = 28, R^2 = b
21 R^1 = c, R^2 = a
22 R^1 = c, R^2 = b
23–26, R^1 = $-(CH_2)_nMe$
23 R^1 = 1, R^2 = a
24 R^1 = 9, R^2 = a
25 R^1 = 15, R^2 = a
26 R^1 = 19, R^2 = a

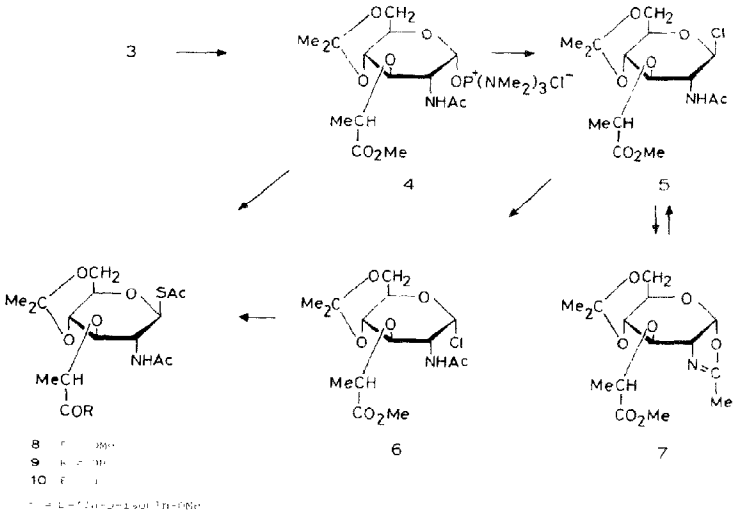
b = L-Ala-D-isoGln

c =



27 R^1 = Ac, R^2 = a
28 R^1 = H, R^2 = a
29 R^1 = H, R^2 = b
30–37; R^1 = $-CO(CH_2)_mMe$
30 m = 8, R^2 = a
31 m = 8, R^2 = b
32 m = 14, R^2 = a
33 m = 14, R^2 = b
34 m = 16, R^2 = a
35 m = 16, R^2 = b
36 m = 28, R^2 = b
37 m = 28, R^2 = b
38 R^1 = c, R^2 = a
39 R^1 = c, R^2 = b
40–47; R^1 = $-(CH_2)_nMe$
40 n = 1, R^2 = a
41 n = 1, R^2 = b
42 n = 9, R^2 = a
43 n = 9, R^2 = b
44 n = 15, R^2 = a
45 n = 15, R^2 = b
46 n = 19, R^2 = a
47 n = 19, R^2 = b

Hydrolysis of the *S*-acetyl and methyl ester groups in compound **8**, and *S*-acetylation of the product in methanol, with acetic anhydride in the presence of triethylamine, gave 2-acetamido-1-*S*-acetyl-3-*O*-(D-1-carboxyethyl)-2-deoxy-4,6-*O*-isopropylidene-1-thio-β-D-glucopyranose (**9**) in 78% yield. Coupling of **9** with L-alanyl-D-isoglutamine methyl ester, using dicyclohexylcarbodiimide (DCC) and *N*-hydroxysuccinimide (HOSu) as the activating agents, afforded **10** in 95% yield; this was used as a convenient intermediate for the synthesis of all of the *N*-acetyl-1-thiomuramoyl dipeptides described herein. Hydrolytic removal of the isopropylidene group in **10** under mild, acidic conditions afforded **27**, which was treated with sodium methoxide in methanol, to give the desired *N*-[2-*O*-(2-acetamido-2,3-



dideoxy-1-thio-β-D-glucopyranose-3-yl)-D-lactoyl]-L-alanyl-D-isoglutamine methyl ester (**28**) in good yield; saponification of the methyl ester group in **28** afforded the 1-thio-MDP (**29**).

In order to synthesize the lipophilic analogs of compounds **28** and **29**, bearing the lipid moiety at C-1 of the sugar skeleton, condensation of **11**, formed by selective hydrolysis of the *S*-acetyl group in **10**, with decanoyl chloride in pyridine–dichloromethane gave **13** in good yield; this was converted, by hydrolytic removal of the isopropylidene group under mild, acidic conditions, into *N*-[2-*O*-(2-acetamido-1-*S*-decanoyl-2,3-dideoxy-1-thio-β-D-glucopyranose-3-yl)-D-lactoyl]-L-alanyl-D-isoglutamine (**30**) in quantitative yield. In the same way, when treated with hexadecanoyl, octadecanoyl, triacontanoyl, or 10-(2,3-dimethoxy-5-methyl-1,4-

TABLE I

ADJUVANT ACTIVITIES OF *N*-ACETYL-1-THIOMURAMOYL-L-ALANYL-D-ISOGLUTAMINE AND ITS LIPOPHILIC ANALOGS ON THE INDUCTION OF DELAYED-TYPE HYPERSENSITIVITY TO ABA-*N*-ACETYLTYROSINE IN GUINEA-PIGS

Compound[a]	*Dose* (μg)	*Skin reaction with ABA-BSA*[b] *(100 μg) (Diam. in mm ± SE)*[c] *at*	
		24 h	*48 h*
27	100	21.0 ±1.0	21.0 ±0.7
	10	20.0 ±0.5	17.5 ±0.7
28	100	22.1 ±0.9	20.9 ±0.7
	10	18.2 ±1.8	16.7 ±1.5
29	100	23.1 +1.2	21.0 ±1.3
	10	20.3 ±1.0	17.0 ±1.0
30	13	12.7 ±1.0	10.8 ±0.4
31	13	19.6 ±0.5	15.5 ±0.3
32	100	20.4 ±0.7	21.0 ±1.4
	15	17.7 ±0.7	18.5 ±0.7
33	100	21.8 ±0.8	23.1 ±1.1
	15	20.8 ±0.4	19.7 ±0.5
34	15	20.0 ±0.5	18.5 ±0.8
35	15	20.5 ±0.6	19.5 ±0.4
36	19	20.3 ±0.8	18.5 ±0.8
37	19	19.4 ±1.3	20.6 ±0.9
38	16	17.1 ±2.4	14.0 ±0.3
39	16	20.0 ±0.8	14.0 ±1.0
41	100	19.1 ±0.7	16.0 ±0.9
43	100	(18.2 ±0.9)	(11.8 ±1.3)
44	100	(13.5 ±0.8)	(12.4 ±0.7)
45	100	(15.6 ±1.9)	(11.6 ±1.4)
47	100	(14.1 ±1.9)	(6.9 ±0.9)
MDP	100	22.7 ±21.3	21.3 ±2.4
	10	20.0 ±0.9	17.8 ±0.7
Control[d]		0	0

[a]The doses of the strong adjuvants were determined as based on their molecular weights. [b]Azobenzenearsonate–*N*-acetyl-L-tyrosine–bovine serum albumin. [c]The data indicate the average diameter ± the standard error (SE) of the skin reaction (induration) of four guinea-pigs; the values in parentheses indicate the size of erythema. [d]ABA-*N*-acetyltyrosine in Freund's incomplete adjuvant.

eicosanyl bromide afforded the corresponding 1-*S*-alkyl derivatives (**23–26**) in good yields. Hydrolytic removal of the isopropylidene group in compounds **23–26** under mildly acidic conditions gave, quantitatively, compounds **40, 42, 44,** and **46**, which were saponified with 0.2M potassium hydroxide in 1,4-dioxane–methanol, to afford the desired *N*-[2-*O*-(alkyl 2-acetamido-2,3-dideoxy-1-thio-β-D-glucopyranoside-3-yl)-D-lactoyl]-L-alanyl-D-isoglutamines (**41, 43, 45,** and **47**) in quantitative yields.

The immunoadjuvant activities of the compounds thus obtained on the induction of the delayed type of hypersensitivity to *N*-acetyl-L-tyrosine-3-azobenzene-4′-arsonate (ABA-*N*-acetyltyrosine) in guinea-pigs were examined[10] (see Table I).

N-Acetyl-1-thiomuramoyl-L-alanyl-D-isoglutamine (**29**), in which the hydroxyl group at C-1 of the carbohydrate moiety in MDP is replaced by a thiol group, and the *N*-acetyl-1-thiomuramoyl dipeptide methyl ester (**28**) showed strong activity at a dose of 100 or 10 μg, comparable to that of MDP. In addition, all of the *N*-acetyl-1-*S*-acyl-MDP analogs were equally as active as MDP as adjuvants, even at lower dosage, whereas the 1-*S*-alkyl derivatives exhibited weak, or negligible, activity. The results clearly indicate that introduction of the fatty acyl group at C-1 of the sugar moiety in *N*-acetyl-1-thiomuramoyl-L-alanyl-D-isoglutamine (**29**) and the methyl ester (**28**) is, for the development of activity, more favorable than that of the fatty alkyl group at C-1 of the sugar skeleton.

The protective activity of compounds **10**, **11**, **28**, **32**, **33**, and **45** in mice infected with *Escherichia coli* (E-77156) was examined[5b]. Compounds **28**, **32**, and **33**, which had potent, adjuvant activity, provided efficient protection, but other compounds were inactive.

We had demonstrated that introduction[4h] of lipophilic character at C-2 in muramoyl-L-alanyl-D-isoglutamine, or at C-6 in *N*-acetyl-6-amino-6-deoxymuramoyl dipeptide, gave compounds that displayed antitumor activity or suppression of tumor growth. Therefore, we examined[4h] the tumor (Meth-A fibrosarcoma) suppression activity of compounds **32** and **33**, as a preliminary experiment, in syngeneic BALB/*c* female mice. Both compounds showed potent, tumor-suppressive activity, a promising result suggesting further development.

EXPERIMENTAL

General methods. — Melting points were determined with a Yanagimoto micro melting-point apparatus and are uncorrected. Specific rotations were determined with a Union MP-201 polarimeter, and i.r. spectra were recorded with a Jasco IR-1 spectrophotometer. N.m.r. spectra were recorded at 90 MHz with a Hitachi R-22 spectrometer, and the n.m.r. data were confirmed by use of decoupling techniques. Preparative chromatography was performed on silica gel (Waco Co.; 300 mesh) with the solvent systems specified. Evaporations were conducted *in vacuo*

2-Acetamido-1-O-benzoyl-2-deoxy-4,6-O-isopropylidene-3-O-[D-1-(methoxycarbonyl)ethyl]-α-D-glucopyranose (**2**). — To a stirred solution of dry pyridine (0.25 mL) in dry dichloromethane (5 mL) was added chromium trioxide (165 mg), and the mixture was stirred for 15 min at room temperature. A solution of benzyl 2-acetamido-2-deoxy-4,6-*O*-isopropylidene-3-*O*-[D-1-(methoxycarbonyl)ethyl]-α-D-glucopyranoside (**1**; 180 mg) in dichloromethane (3 mL) was added, with stirring, to the mixture at room temperature. A tarry deposit formed at once, and the color of the mixture changed to dark-brown. Acetic anhydride (0.16 mL) was added, and the mixture was stirred for 3 h at 45°, the course of the reaction being monitored by t.l.c. The mixture was chromatographed on a column of silica gel (20 g) with ethyl acetate, to give a crystalline product. Recrystallization

from ether–hexane afforded **2** (170 mg, 92%) as needles; m.p. 135–137°, $[\alpha]_D$ +144° (*c* 1.5, methanol); ν_{max}^{KBr} 3300 (NH), 1730 and 1260 (ester), 1670 and 1520 (amide), 850 (Me_2C), and 700 cm^{-1} (phenyl); n.m.r. data (in chloroform-*d*): δ 1.39, 1.50 (2 s, 6 H, Me_2C), 1.42 (d, 3 H, $J_{Me,CH}$ 7.0 Hz, MeCH), 1.95 (s, 3 H, AcN), 4.56 (q, 1 H, $J_{CH,Me}$ 7.0 Hz, CH), 6.69 (d, 1 H, $J_{1,2}$ 3.4 Hz, H-1), and 7.23–8.04 (m, 5 H, Ph).

Anal. Calc. for $C_{22}H_{29}NO_9$: C, 58.53; H, 6.47; N, 3.10. Found: C, 58.39; H, 6.58; N, 3.02.

2-Acetamido-2-deoxy-4,6-O-isopropylidene-3-O-[D-1-methoxycarbonyl)-ethyl]-D-glucopyranoside (**3**). — To an ice-cooled solution of **2** (4.0 g) in methanol (60 mL) was added sodium methoxide (50 mg), and the mixture was kept for 10 min at room temperature, and then treated with Amberlite IRC-50 (H^+) resin, to remove the base. The product was purified by chromatography on a column of silica gel (50 g) with chloroform and then 50:1 chloroform–methanol. The latter eluate afforded **3** (2.8 g, 91%) as needles; m.p. 180–184°, $[\alpha]_D$ +44.3° (*c* 0.47, chloroform; equil.); ν_{max}^{Nujol} 3350 (OH, NH), 1730 and 1260 (ester), 1655 and 1540 (amide), and 850 cm^{-1} (Me_2C).

Anal. Calc. for $C_{15}H_{25}NO_8$: C, 51.86; H, 7.25; N, 4.03. Found: C, 51.73; H, 7.30; N, 4.08.

2-Acetamido-1-S-acetyl-2-deoxy-4,6-O-isopropylidene-3-O-[D-1-(methoxycarbonyl)ethyl]-1-thio-β-D-glucopyranose (**8**). — *(a) From alkyloxytris(dimethylamino)phosphonium salt* (**4**). To a solution of **3** (5.9 g) in dry dichloromethane (100 mL) and carbon tetrachloride (5.2 g), cooled to −60°, was added dropwise during 15 min, with stirring, an ice-cooled solution of tris(dimethylamino)phosphine (4.0 g) in dry dichloromethane (50 mL). The mixture was stirred for 15 min at −50°; at that time, all of the starting material (**3**) had been converted into the salt. Potassium thioacetate (5.0 g) was added to the mixture at −50°, and it was then stirred for 5 h at −20 to −30°, and the precipitate filtered off, and washed with dichloromethane. The filtrate and washings were combined, washed with water, dried (sodium sulfate), and evaporated. The crystalline residue was recrystallized from ether, to give **8** (5.55 g) as needles. Chromatography of the mother liquor was conducted on a column of silica gel (50 g) with chloroform and then 150:1 chloroform–methanol. The latter eluate afforded a further 420 mg of **8**, raising the total yield to 5.97 g (87%); m.p. 171–173°, $[\alpha]_D$ +8.1° (*c* 0.4, chloroform); ν_{max}^{Nujol} 3330 (NH), 1745, 1720, and 1260 (ester), 1690 (AcS), 1660 and 1520 (amide), and 850 cm^{-1} (Me_2C); n.m.r. data (in chloroform-*d*): δ 1.35 (d, 3 H, $J_{Me,CH}$ 7.2 Hz, MeCH), 1.38, 1.47 (2 s, 6 H, Me_2C), 2.00 (s, 3 H, AcN), 2.32 (s, 3 H, AcS), 3.74 (s, 3 H, MeO), 4.51 (q, 1 H, $J_{CH,Me}$ 7.2 Hz, CH), 5.09 (d, 1 H, $J_{1,2}$ 10.8 Hz, H-1), and 6.78 (d, 1 H, $J_{NH,2}$ 7.0 Hz, NH).

Anal. Calc. for $C_{17}H_{27}NO_8S$: C, 50.36; H, 6.71; N, 3.45. Found: C, 50.28; H, 6.59; N, 3.45.

(b) From 2-acetamido-2-deoxy-4,6-O-isopropylidene-3-O-[D-1-(methoxycarbonyl)ethyl]-α-D-glucopyranosyl chloride (**6**). A solution of **3** (2.5 g) in dry di-

chloromethane (50 mL) containing carbon tetrachloride (2.17 g) was cooled to −50°. The mixture was stirred, and a solution of tris(dimethylamino)phosphine (1.6 g) in dry dichloromethane (25 mL) was added during 15 min at −50°; the stirring was continued for another 1 h at 0°. The mixture was evaporated to a syrup at 40° (bath); at this stage, there were three spots in t.l.c., due to compounds **5**–**7**. Dichloromethane (50 mL) was added to the syrup, and evaporated at 40°; this procedure was repeated several times, until all of compounds **5** and **7** had been converted into **6**.

To a solution of **6** in dry dichloromethane (15 mL) and dry acetone (15 mL) was added *potassium thioacetate* (2.4 g), *and the mixture was stirred overnight at* room temperature, and evaporated to a syrup which was extracted with chloroform. The extract was washed with water, dried (sodium sulfate), and evaporated, to afford a crystalline product. Recrystallization from ether gave **8** (1.94 g) as needles. The mother liquor was purified by chromatography on a column of silica gel (20 g) with 150:1 chloroform–methanol, affording **8** (500 mg); the total yield was 2.44 g (83.5%).

2-Acetamido-1-S-acetyl-3-O-(D-1-carboxyethyl)-2-deoxy-4,6-O-isopropylidene-1-thio-β-D-glucopyranose (**9**). — To a solution of **8** (200 mg) in 1,4-dioxane (10 mL) was added 0.2M potassium hydroxide (12 mL), and the mixture was stirred for 10 min at room temperature, and then treated with Amberlite IRC-50 (H^+) resin to remove the base. The resin was filtered off and washed with methanol, and the filtrate and washings were combined, and evaporated. To an ice-cooled solution of the residue in methanol (4 mL) were added, with stirring, acetic anhydride (0.8 mL) and triethylamine (1 mL), and the mixture was kept for 30 min at room temperature, and then evaporated. The residue was chromatographed on a column of silica gel (20 g) with chloroform and then 50:1 chloroform–methanol. The latter eluate gave compound **9** (150 mg, 78%) as needles, after recrystallization from ether; m.p. 193–200° (dec.), $[\alpha]_D$ +10.5° (*c* 0.3, chloroform); ν_{max}^{Nujol} 3280 (NH), 2700–2480 (CO$_2$H), 1720 (C=O), 1700 (AcS), 1620 and 1560 (amide), and 850 cm^{-1} (Me_2C); n.m.r. data (in 1:1 chloroform-*d*–methanol-d_4): δ 1.35 (d, 3 H, $J_{Me,CH}$ 6.8 Hz, MeCH), 1.39, 1.50 (2 s, 6 H, Me_2C), 1.95 (s, 3 H, AcN), 2.33 (s, 3 H, AcS), 4.36 (q, 1 H, $J_{CH,Me}$ 6.8 Hz, CH), and 5.16 (d, 1 H, $J_{1,2}$ 10.5 Hz, H-1).

Anal. Calc. for $C_{16}H_{25}NO_8S$: C, 49.09; H, 6.44; N, 3.58. Found: 49.13; H, 6.51; N, 3.48.

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 (**10**). — To a solution of **9** (160 mg) in dry 1,4-dioxane (5 mL) were added, with stirring, HOSu (100 mg) and DCC (210 mg), and the mixture was stirred for 30 min at room temperature; *at that time, the starting material had been converted into the activated* ester. The 1,3-dicyclohexylurea formed was filtered off and washed with dry 1,4-dioxane (2 mL). The filtrate and washings were combined, and L-alanyl-D-isoglutamine methyl ester trifluoroacetate (200 mg) and triethylamine (5 drops) were added to the solution, which was stirred for 40 min at room temperature, and then

evaporated. The residue was chromatographed on a column of silica gel (20 g) with (*a*) chloroform, (*b*) 100:1, and (*c*) 50:1 chloroform–methanol. Eluant *c* afforded **10** (235 mg, 95%) as crystals; m.p. 148–151°, $[\alpha]_D$ +11°(*c* 0.2, chloroform); ν_{max}^{Nujol} 3340 and 3230 (NH), 1730 and 1250 (ester), 1690 (AcS), 1670, 1650, 1630, 1540, and 1520 (amide), and 850 cm^{-1} (Me_2C); n.m.r. data (in 1:1 chloroform-*d*–methanol-d_4): δ 1.30–1.42 (m, 6 H, 2 MeCH), 1.39, 1.50 (2 s, 6 H, Me_2C), 1.93 (s, 3 H, AcN), 2.35 (s, 3 H, AcS), 3.76 (s, 3 H, MeO), and 5.19 (d, 1 H, $J_{1,2}$ 10.5 Hz, H-1).

Anal. Calc. for $C_{25}H_{40}N_4O_{11}S$: C, 49.66; H, 6.67; N, 9.27. Found: C, 49.76; H, 6.65; N, 9.30.

N-*[2-O-(2-Acetamido-2,3-dideoxy-4,6-O-isopropylidene-1-thio-β-*D*-glucopyranose-3-yl)-*D*-lactoyl]-*L*-alanyl-*D*-isoglutamine methyl ester* (**11**). — To a solution of **10** (85 mg) in methanol (5 mL) was added sodium methoxide (10 mg), and the mixture was kept for 1.5 h at room temperature, treated with Amberlite IRC-50 (H^+) resin to remove the base, and evaporated. The residue was chromatographed on a column of silica gel (10 g) with chloroform and then 40:1 chloroform–methanol. The latter eluate gave **11** (76 mg, 96%) as crystals; m.p. 105–107° (dec.), $[\alpha]_D$ +33° (*c* 0.5, chloroform); ν_{max}^{Nujol} 3240 (NH), 1730 and 1250 (ester), 1650 and 1530 (amide), and 850 cm^{-1} (Me_2C).

Anal. Calc. for $C_{23}H_{38}N_4O_{10}S$: C, 49.10; H, 6.81; N, 9.96. Found: C, 48.89; H, 6.85; N, 9.83.

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* (**27**). — A solution of **10** (100 mg) in 80% aqueous acetic acid (5 mL) was heated, with stirring, for 45 min at 45°, cooled, and evaporated to a syrup which crystallized from ether, to give **27** (94 mg, quantitative); m.p. 158–162° (dec.), $[\alpha]_D$ +31° (*c* 0.2, 1:1 chloroform–methanol); ν_{max}^{Nujol} 3370–3200 (OH, NH), 1730 and 1250 (ester), 1700 (AcS), and 1640 and 1540–1510 cm^{-1} (amide).

Anal. Calc. for $C_{22}H_{36}N_4O_{11}S$: C, 46.80; H, 6.43; N, 9.92. Found: C, 46.75; H, 6.35; N, 9.68.

N-*[2-O-(2-Acetamido-2,3-dideoxy-1-thio-β-*D*-glucopyranose-3-yl)-*D*-lactoyl]-*L*-alanyl-*D*-isoglutamine methyl ester* (**28**). — To a solution of **27** (20 mg) in methanol (2 mL) was added sodium methoxide (5 mg), and the mixture was kept for 10 min at room temperature, treated with Amberlite IR-120 (H^+) resin, and evaporated. The product crystallized from ether, to give **28** (17.5 mg, 95%); m.p. 117–125° (dec.), $[\alpha]_D$ +12° (*c* 0.2, methanol); ν_{max}^{KBr} 3370 (OH, NH), 1720 and 1250 (ester), and 1650 and 1530 cm^{-1} (amide).

Anal. Calc. for $C_{20}H_{34}N_4O_{11}S$: C, 45.97; H, 6.56; N, 10.72. Found: C, 45.86; H, 6.69; N, 10.88.

N-*[2-O-(2-Acetamido-2,3-dideoxy-1-thio-β-*D*-glucopyranose-3-yl)-*D*-lactoyl]-*L*-alanyl-*D*-isoglutamine* (**29**). — To a solution of **27** (25 mg) in methanol (2 mL) was added sodium methoxide (5 mg), and, after 3 min, 0.2M potassium hydroxide (1 mL) was added to the mixture, the course of the reaction being monitored

by t.l.c. The mixture was treated with Amberlite IR-120 (H^+) resin, and evaporated, to give amorphous **29** (22.4 mg; quantitative), which showed a single spot in t.l.c.; m.p. 157–166° (dec.), $[\alpha]_D$ +10.3° (*c* 0.2, methanol); ν_{max}^{KBr} 3360 (OH, NH), 2760–2630 (CO_2H), 1715 (C=O), and 1650 and 1520 cm^{-1} (amide).

Anal. Calc. for $C_{19}H_{32}N_4O_{10}S$: C, 44.87; H, 6.34; N, 11.02. Found: C, 44.45; H, 6.53; N, 10.99.

*N-[2-O-(2-Acetamido-1-S-decanoyl-2,3-dideoxy-4,6-O-isopropylidene-1-thio-β-*D*-glucopyranose-3-yl)-*D*-lactoyl]-*L*-alanyl-*D*-isoglutamine methyl ester* (**13**). — To an ice-cooled solution of **11** (80 mg) in dry pyridine (1 mL) and dichloromethane (2 mL) was added dropwise, with stirring, a solution of decanoyl chloride (52 mg) in dichloromethane (1 mL), and the mixture was stirred for 40 min at 5–10°; methanol (0.5 mL) was added to the mixture, which was evaporated, and the residue extracted with chloroform. The extract was washed with water, dried (sodium sulfate), and evaporated to a syrup which was chromatographed on a column of silica gel (15 g) with (*a*) 150:1, (*b*) 70:1, and (*c*) 30:1 chloroform–methanol. Eluant *c* gave amorphous **13** (65 mg, 64%); $[\alpha]_D$ +6.6° (*c* 0.33, chloroform); ν_{max}^{KBr} 3230 (NH), 2900 and 2830 (Me, methylene), 1730 and 1250 (ester), 1700 (S-acyl), 1650 and 1530 (amide), and 850 cm^{-1} (Me_2C); n.m.r. data (in chloroform-*d*): δ 0.88 (near t, J_{Me,CH_2} 6.0 Hz, $MeCH_2$), 1.23–1.48 (m, 26 H, 4 Me, 7 CH_2), 1.92 (s, 3 H, AcN), 3.68 (s, 3 H, MeO), 5.22 (d, 1 H, $J_{1,2}$ 10.5 Hz, H-1), and 6.20, 7.02, 7.12, and 7.64 (5 H, 3 NH, NH_2).

Anal. Calc. for $C_{33}H_{56}N_4O_{11}S$: C, 55.29; H, 7.87; N, 7.82. Found: C, 55.36; H, 7.93; N, 7.62.

Other N-*[2-O-(2-acetamido-1-S-acyl-2,3-dideoxy-4,6-O-isopropylidene-1-thio-β-*D*-glucopyranose-3-yl)-*D*-lactoyl]-*L*-alanyl-*D*-isoglutamine methyl esters* (**15, 17, 19,** and **21**) were synthesized according to the method described for **13**.

*N-[2-O-(2-Acetamido-2,3-dideoxy-1-S-hexadecanoyl-4,6-O-isopropylidene-1-thio-β-*D*-glucopyranose-3-yl)-*D*-lactoyl]-*L*-alanyl-*D*-isoglutamine methyl ester* (**15**). — Compound **15** was obtained in 82% yield; m.p. 98–100°, $[\alpha]_D$ +14° (*c* 0.2, chloroform); ν_{max}^{KBr} 3340–3230 (NH), 2910 and 2830 (Me, CH_2), 1740 and 1260 (ester), 1700 (S-acyl), 1650 and 1520 (amide), and 850 cm^{-1} (Me_2C); n.m.r. data (in chloroform-*d*): δ 0.89 (near t, 3 H, J_{Me,CH_2} 6.0 Hz, $MeCH_2$), 1.40, 1.50 (2 s, 6 H, Me_2C), 1.96 (s, 3 H, AcN), 3.70 (s, 3 H, MeO), and 5.19 (d, 1 H, $J_{1,2}$ 10.5 Hz, H-1).

Anal. Calc. for $C_{39}H_{68}N_4O_{11}S$: C, 58.47; H, 8.56; N, 6.99. Found: C, 58.53; H, 8.63; N, 6.75.

*N-[2-O-(2-Acetamido-2,3-dideoxy-1-S-octadecanoyl-4,6-O-isopropylidene-1-thio-β-*D*-glucopyranose-3-yl)-*D*-lactoyl]-*L*-alanyl-*D*-isoglutamine methyl ester* (**17**). — Compound **17** was obtained as an amorphous mass in 58% yield; $[\alpha]_D$ +6.6° (*c* 0.5, chloroform); ν_{max}^{KBr} 3230 (NH), 2910 and 2830 (Me, methylene), 1730 and 1250 (ester), 1700 (S-acyl), 1640 and 1530 (amide), and 850 cm^{-1} (Me_2C); n.m.r. data (in chloroform-*d*): δ 0.87 (near t, 3 H, J_{Me,CH_2} 6.0 Hz, $MeCH_2$), 1.93,

(s, 3 H, AcN), 3.69 (s, 3 H, MeO), 5.24 (d, 1 H, $J_{1,2}$ 10.5 Hz, H-1), and 6.22, 7.05, 7.16, and 7.65 (5 H, 3 NH, NH_2).

Anal. Calc. for $C_{41}H_{72}N_4O_{11}S$: C, 59.39; H, 8.75; N, 6.76. Found: C, 59.18; H, 8.80; N, 6.75.

N-*[2-O-(2-Acetamido-2,3-dideoxy-4,6-O-isopropylidene-1-thio-1-S-triacontanoyl-β-*D-glucopyranose-3-yl)-D-*lactoyl]-*L-*alanyl-*D-*isoglutamine methyl ester* (**19**). — Compound **19** was obtained as an amorphous mass in 48% yield; $[\alpha]_D$ +13.1° (*c* 0.6, chloroform); ν_{max}^{KBr} 3230 (NH), 2880 and 2820 (Me, methylene), 1730 and 1250 (ester), 1700 (S-acyl), 1640 and 1520 (amide), and 850 cm^{-1} (Me_2C); n.m.r. data (in chloroform-*d*): δ 0.88 (near t, J_{Me,CH_2} 6.0 Hz, $MeCH_2$), 1.92 (s, 3 H, AcN), 3.69 (s, 3 H, MeO), 5.20 (d, 1 H, $J_{1,2}$ 10.0 Hz, H-1), and 6.11, 6.88, 7.02, 7.15, and 7.60 (5 H, 3 NH, NH_2).

Anal. Calc. for $C_{53}H_{96}N_4O_{11}S$: C, 63.82; H, 9.70; N, 5.62. Found: C, 63.69; H, 9.94; N, 5.55.

N-*[2-O-{2-Acetamido-2,3-dideoxy-1-S-[10-(2,3-dimethoxy-5-methyl-1,4-benzoquinone-6-yl)decanoyl]-4,6-O-isopropylidene-1-thio-β-*D-*glucopyranose-3-yl}-*D-*lactoyl]-*L-*alanyl-*D-*isoglutamine methyl ester* (**21**). — Compound **21** was obtained as an amorphous mass in 63% yield; $[\alpha]_D$ +9.7° (*c* 0.58, chloroform); ν_{max}^{KBr} 3250 (NH), 2950, 2920, and 2830 (Me, methylene), 1730 and 1260 (ester), 1700 (S-acyl), 1650 and 1530 (amide), and 850 cm^{-1} (Me_2C); n.m.r. data (in chloroform-*d*): δ 1.15–1.50 (m, 31 H, 5 Me, 8 methylene), 2.00 (s, 3 H, AcN), 3.67 (s, 3 H, MeO), 3.99 (s, 6 H, 2 MeO), 5.22 (d, 1 H, $J_{1,2}$ 10.5 Hz, H-1), and 6.12, 7.08, and 7.56 (5 H, 3 NH, NH_2).

Anal. Calc. for $C_{42}H_{64}N_4O_{15}S$: C, 56.23; H, 7.19; N, 6.25. Found: C, 56.31; H, 7.38; N, 6.25.

N-*[2-O-(2-Acetamido-1-S-decanoyl-2,3-dideoxy-4,6-O-isopropylidene-1-thio-β-*D-*glucopyranose-3-yl)-*D-*lactoyl]-*L-*alanyl-*D-*isoglutamine* (**14**). — To a solution of **10** (100 mg) in methanol (10 mL) was added sodium methoxide (15 mg), and, after 10 min, 0.2M potassium hydroxide (10 mL) was added to the solution. The mixture was treated with Amberlite IRC-50 (H^+) resin to remove the bases, and evaporated, to give amorphous **12**. To an ice-cooled solution of **12** in dry pyridine (1 mL) and dichloromethane (2 mL) was added dropwise, with stirring, a solution of decanoyl chloride (60 mg) in dry dichloromethane (1 mL), and the mixture was stirred for 1.5 h at 0°; methanol (0.5 mL) was added to the mixture, which was evaporated, and the residue extracted with chloroform. The extract was washed with water, dried (sodium sulfate), and evaporated to a syrup which was chromatographed on a column of silica gel (20 g) with (*a*) 50:1 and (*b*) 10:1 chloroform–methanol. Eluant *b* afforded **14** (93 mg, 80%) as crystals; m.p. 143°, $[\alpha]_D$ +1.9° (*c* 0.2, 1:1 chloroform–methanol); ν_{max}^{KBr} 3350 (NH), 1720 (C=O), 1690 (S-acyl), 1650 and 1540 (amide), and 850 cm^{-1} (Me_2C).

Anal. Calc. for $C_{32}H_{54}N_4O_{11}S$: C, 54.68; H, 7.74; N, 7.97. Found: C, 54.36; H, 7.83; N, 7.84.

Other N-*[2-O-(2-Acetamido-1-S-acyl-2,3-dideoxy-4,6-O-isopropylidene-1-*

*thio-β-*D*-glucopyranose-3-yl)-*D*-lactoyl]-*L*-alanyl-*D*-isoglutamines* (**16**, **18**, **20**, and **22**) were prepared according to the method described for **14**.

*N-[2-O-(2-Acetamido-2,3-dideoxy-1-S-hexadecanoyl-4,6-O-isopropylidene-1-thio-β-*D*-glucopyranose-3-yl)-*D*-lactoyl]-*L*-alanyl-*D*-isoglutamine* (**16**). — Compound **16** was obtained as crystals in 72% yield; m.p. 103–106°, $[\alpha]_D$ +21° (*c* 0.15, 1:1 chloroform–methanol); ν_{max}^{KBr} 3350–3250 (NH), 2900 and 2820 (Me, methylene), 1710 (C=O), 1700 (S-acyl), 1660 and 1520 (amide), and 850 cm^{-1} (Me_2C).

Anal. Calc. for $C_{38}H_{69}N_4O_{11}S$: C, 57.99; H, 8.45; N, 7.12. Found: C, 57.65; H, 8.69; N, 7.05.

*N-[2-O-(Acetamido-2,3-dideoxy-4,6-O-isopropylidene-1-S-octadecanoyl-1-thio-β-*D*-glucopyranose-3-yl)-*D*-lactoyl]-*L*-alanyl-*D*-isoglutamine* (**18**). — Compound **18** was obtained as crystals in 53% yield; m.p. 215–217°, $[\alpha]_D$ +11.7° (*c* 0.48, 1:1 chloroform–methanol); ν_{max}^{KBr} 3340 (NH), 2900 and 2830 (Me, methylene), 1710 (C=O), 1700 (S-acyl), 1650 and 1540 (amide), and 850 cm^{-1} (Me_2C).

Anal. Calc. for $C_{40}H_{70}N_4O_{11}S$: C, 58.95; H, 8.66; N, 6.87. Found: C, 58.71; H, 8.75; N, 6.88.

*N-[2-O-(2-Acetamido-2,3-dideoxy-4,6-O-isopropylidene-1-thio-1-S-triacontanoyl-β-*D*-glucopyranose-3-yl)-*D*-lactoyl]-*L*-alanyl-*D*-isoglutamine* (**20**). — Compound **20** was obtained as crystals in 48% yield; m.p. 207–209°, $[\alpha]_D$ +15.2° (*c* 0.55, 1:1 chloroform–methanol); ν_{max}^{KBr} 3350–3240 (NH), 2920 and 2820 (Me, methylene), 1710 (C=O), 1700 (S-acyl), 1650 and 1540 (amide), and 860 cm^{-1} (Me_2C).

Anal. Calc. for $C_{57}H_{104}N_4O_{11}S$: C, 63.51; H, 9.64; N, 5.70. Found: C, 63.24; H, 9.59; N, 5.78.

*N-[2-O-{2-Acetamido-2,3-dideoxy-1-S-[10-(2,3-dimethoxy-5-methyl-1,4-benzoquinon-6-yl)decanoyl]-4,6-O-isopropylidene-1-thio-β-*D*-glucopyranose-3-yl}-*D*-lactoyl]-*L*-alanyl-*D*-isoglutamine* (**22**). — Compound **22** was obtained as crystals in 47% yield; m.p. 193–194°, $[\alpha]_D$ +6.0° (*c* 0.47, 1:2 chloroform–methanol); ν_{max}^{KBr} 3350 (NH), 2910 and 2830 (Me, methylene), 1720 (C=O), 1700 (S-acyl), 1640 and 1530 (amide), and 850 cm^{-1} (Me_2C).

Anal. Calc. for $C_{41}H_{67}N_4O_{15}S$: C, 55.77; H, 7.08; N, 6.35. Found: C, 55.68; H, 7.33; N, 6.31.

*N-[2-O-(Ethyl 2-acetamido-2,3-dideoxy-4,6-O-isopropylidene-1-thio-β-*D*-glucopyranoside-3-yl)-*D*-lactoyl]-*L*-alanyl-*D*-isoglutamine methyl ester* (**23**). — To a solution of **10** (100 mg) in dry methanol (5 mL) was added sodium methoxide (10 mg); the mixture was kept for 30 min at room temperature, and then ethyl bromide (50 mg) was added. The mixture was stirred for 6 h at room temperature and evaporated, and the residue was chromatographed on a column of silica gel (15 g) with (*a*) 150:1, (*b*) 50:1, and (*c*) 30:1 chloroform–methanol. Eluant *c* afforded compound **23** (64 mg, 65.5%) as crystals; m.p. 106–108°, $[\alpha]_D$ +41.8° (*c* 0.46, chloroform); ν_{max}^{KBr} 3200 (NH), 1720 and 1250 (ester), 1650 and 1525 (amide), and 850 cm^{-1} (Me_2C); n.m.r. data (in chloroform-*d*): δ 1.15–1.52 (m, 17 H, 5 Me,

CH_2), 2.02 (s, 3 H, AcN), 3.81 (s, 3 H, MeO), and 5.48 (d, 1 H, $J_{1,2}$ 6.0 Hz, H-1).

Anal. Calc. for $C_{25}H_{42}N_4O_{10}S$: C, 50.83; H, 7.17; N, 9.49. Found: C, 50.76; H, 7.36; N, 9.28.

Other N-*[2-O-(alkyl 2-acetamido-2,3-dideoxy-4,6-O-isopropylidene-1-thio-β-*D-*glucopyranoside-3-yl)-*D-*lactoyl]-*L-*alanyl-*D-*isoglutamine methyl esters* (**24**–**26**) were prepared according to the method described for **23**.

N-*[2-O-(Decyl 2-acetamido-2,3-dideoxy-4,6-O-isopropylidene-1-thio-β-*D-*glucopyranoside-3-yl)-*D-*lactoyl]-*L-*alanyl-*D-*isoglutamine methyl ester* (**24**). — Compound **24** was obtained as crystals in 75% yield; m.p. 75–78°, $[\alpha]_D$ +35.7° (*c* 0.49, chloroform); ν_{max}^{KBr} 3350–3200 (NH), 2900 and 2820 (Me, methylene), 1720 and 1250 (ester), 1640 and 1520 (amide), and 850 cm^{-1} (Me_2C); n.m.r. data (in chloroform-*d*): δ 0.89 (near t, 3 H, J_{Me,CH_2} 6.0 Hz, $MeCH_2$), 1.22–1.51 (m, 30 H, 4 Me, 9 CH_2), 2.02 (s, 3 H, AcN), 3.82 (s, 3 H, MeO), and 5.42 (d, 1 H, $J_{1,2}$ 6.0 Hz, H-1).

Anal. Calc. for $C_{33}H_{58}N_4O_{10}S$: C, 56.39; H, 8.32; N, 7.97. Found: C, 56.32; H, 8.39; N, 7.86.

N-*[2-O-(Hexadecyl 2-acetamido-2,3-dideoxy-4,6-O-isopropylidene-1-thio-β-*D-*glucopyranoside-3-yl)-*D-*lactoyl]-*L-*alanyl-*D-*isoglutamine methyl ester* (**25**). — Compound **25** was obtained as crystals in 93% yield; m.p. 93–96° (dec.), $[\alpha]_D$ +35.0° (*c* 0.3, chloroform); ν_{max}^{KBr} 3360–3250 (NH), 1730 and 1260 (ester), 1645 and 1530 (amide), and 850 cm^{-1} (Me_2C); n.m.r. data (in chloroform-*d*): δ 0.88 (near t, 3 H, J_{Me,CH_2} 6.0 Hz, $MeCH_2$), 1.24–1.50 (m, 42 H, 4 Me, 15 CH_2), 2.01 (s, 3 H, AcN), 3.68 (s, 3 H, MeO), and 5.42 (d, 1 H, $J_{1,2}$ 6.0 Hz, H-1).

Anal. Calc. for $C_{39}H_{70}N_4O_{10}S$: C, 59.51; H, 8.97; N, 7.12. Found: C, 59.33; H, 8.86; N, 7.15.

N-*[2-O-(Eicosyl 2-acetamido-2,3-dideoxy-4,6-O-isopropylidene-1-thio-β-*D-glucopyranoside-3-yl)-*D-*lactoyl]-*L-*alanyl-*D-*isoglutamine methyl ester* (**26**). — Compound **26** was obtained as crystals in 85% yield; m.p. 75–78°, $[\alpha]_D$ +34.5° (*c* 0.43, chloroform); ν_{max}^{KBr} 3330–3220 (NH), 2900 and 2800 (Me, CH_2), 1720 and 1250 (ester), 1640 and 1530 (amide), and 850 cm^{-1} (Me_2C); n.m.r. data (in chloroform-*d*): δ 0.88 (near t, 3 H, J_{Me,CH_2} 6.0 Hz, $MeCH_2$), 1.20–1.51 (m, 50 H, 4 Me, 19 CH_2), 2.02 (s, 3 H, AcN), 3.73 (s, 3 H, MeO), and 5.40 (d, 1 H, $J_{1,2}$ 6.0 Hz, H-1).

Anal. Calc. for $C_{43}H_{78}N_4O_{10}S$: C, 61.25; H, 9.33; N, 6.65. Found: C, 61.02; H, 9.35; N, 6.58.

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* (**30**). — A solution of **13** (50 mg) in 80% aqueous acetic acid (2 mL) was heated for 2 h at 45°, cooled, and evaporated to a crystalline mass. Recrystallization from ether gave **30** (45.6 mg, quantitative); m.p. 176.5°, $[\alpha]_D$ +41.3° (*c* 0.44, 1:1 chloroform–methanol); ν_{max}^{KBr} 3360–3230 (OH, NH), 2900 and 2820 (Me, methylene), 1730 and 1250 (ester), 1700 (S-acyl), and 1640 and 1530 cm^{-1} (amide).

Anal. Calc. for $C_{30}H_{52}N_4O_{11}S$: C, 53.23; H, 7.74; N, 8.28. Found: C, 53.31; H, 7.89; N, 8.33.

Other N-*[2-O-(2-acetamido-1-S-acyl-2,3-dideoxy-1-thio-β-*D*-glucopyranose-3-yl)-*D*-lactoyl]-*L*-alanyl-*D*-isoglutamine methyl esters* (**32**, **34**, **36**, and **38**), N-*[2-O-(2-acetamido-1-S-acyl-2,3-dideoxy-1-thio-β-*D*-glucopyranose-3-yl)-*D*-lactoyl]-*L*-alanyl-*D*-isoglutamines* (**31**, **33**, **35**, **37**, and **39**), *and* N-*[2-O-(alkyl 2-acetamido-2,3-dideoxy-1-thio-β-*D*-glucopyranoside-3-yl)-*D*-lactoyl]-*L*-alanyl-*D*-isoglutamine methyl esters* (**40**, **42**, **44**, and **46**) were prepared from the corresponding 4,6-*O*-isopropylidene derivatives, according to the procedure described for **30**.

N-*[2-O-(2-Acetamido-1-S-decanoyl-2,3-dideoxy-1-thio-β-*D*-glucopyranose-3-yl)-*D*-lactoyl]-*L*-alanyl-*D*-isoglutamine* (**31**). — Compound **31** was obtained as crystals in 93% yield; m.p. 118–120° (dec.), $[\alpha]_D$ +4.2° (*c* 0.34, 1:2 chloroform–methanol); ν_{max}^{KBr} 3350–3300 (OH, NH), 2900 and 2830 (Me, methylene), 1720 (C=O), 1690 (S-acyl), and 1650 and 1540 cm^{-1} (amide).

Anal. Calc. for $C_{29}H_{50}N_4O_{11}S$: C, 52.55; H, 7.60; N, 8.45. Found: C, 52.32; H, 7.80; N, 8.31.

N-*[2-O-(2-Acetamido-2,3-dideoxy-1-S-hexadecanoyl-1-thio-β-*D*-glucopyranose-3-yl)-*D*-lactoyl]-*L*-alanyl-*D*-isoglutamine methyl ester* (**32**). — Compound **32** was obtained as crystals in 91% yield; m.p. 181–183°, $[\alpha]_D$ +31.5° (*c* 0.2, 1:1 chloroform–methanol); ν_{max}^{KBr} 3320–3260 (OH, NH), 2920 and 2840 (Me, methylene), 1740 and 1260 (ester), 1690 (S-acyl), and 1650 and 1530 cm^{-1} (amide).

Anal. Calc. for $C_{36}H_{64}N_4O_{11}S$: C, 56.82; H, 8.48; N, 7.36. Found: C, 56.71; H, 8.66; N, 7.30.

N-*[2-O-(2-Acetamido-2,3-dideoxy-1-S-hexadecanoyl-1-thio-β-*D*-glucopyranose-3-yl)-*D*-lactoyl]-*L*-alanyl-*D*-isoglutamine* (**33**). — Compound **33** was obtained as crystals in quantitative yield; m.p. 125–129° (dec.), $[\alpha]_D$ +31.3° (*c* 0.2, 1:1 chloroform–methanol); ν_{max}^{KBr} 3380–3260 (OH, NH), 2920 and 2840 (Me, methylene), 1720 (C=O), 1700 (S-acyl), and 1650 and 1540 cm^{-1} (amide).

Anal. Calc. for $C_{35}H_{62}N_4O_{11}S$: C, 56.28; H, 8.33; N, 7.50. Found: C, 56.07; H, 8.42; N, 7.43.

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* (**34**). — Compound **34** was obtained as an amorphous mass in quantitative yield; $[\alpha]_D$ +29.0° (*c* 0.64, 1:1 chloroform–methanol); ν_{max}^{KBr} 3350–3230 (OH, NH), 2900 and 2820 (Me, methylene), 1730 and 1250 (ester), 1690 (S-acyl), and 1650 and 1530 cm^{-1} (amide).

Anal. Calc. for $C_{38}H_{68}N_4O_{11}S$: C, 57.84; H, 8.69; N, 7.10. Found: C, 57.55; H, 8.73; N, 7.01.

N-*[2-O-(2-Acetamido-2,3-dideoxy-1-S-octadecanoyl-1-thio-β-*D*-glucopyranose-3-yl)-*D*-lactoyl]-*L*-alanyl-*D*-isoglutamine* (**35**). — Compound **35** was obtained as crystals in 95% yield; m.p. 230–232° (dec.), $[\alpha]_D$ +11.2° (*c* 0.5, 1:2 chloroform–methanol); ν_{max}^{KBr} 3350–3300 (OH, NH), 2920 and 2840 (Me, methylene), 1710 (C=O), 1700 (S-acyl), and 1655 and 1540 cm^{-1} (amide).

Anal. Calc. for $C_{37}H_{66}N_4O_{11}S$: C, 57.34; H, 8.58; N, 7.23. Found: C, 57.21; H, 8.66; N, 7.41.

N-*[2-O-(2-Acetamido-2,3-dideoxy-1-thio-1-S-triacontanoyl-β-*D*-gluco-*

pyranose-3-yl)-D-*lactoyl]*-L-*alanyl*-D-*isoglutamine methyl ester* (**36**). — Compound **36** was isolated as crystals; m.p. 182°, $[\alpha]_D$ +33.3° (*c* 0.6, 1:1 chloroform–methanol); ν_{max}^{KBr} 3400–3230 (OH, NH), 2920 and 2830 (Me, methylene), 1730 and 1250 (ester), 1700 (S-acyl), and 1630 and 1530 cm^{-1} (amide).

Anal. Calc. for $C_{50}H_{92}N_4O_{11}S$: C, 62.73; H, 9.69; N, 5.85. Found: C, 62.65; H, 9.83; N, 5.81.

N-*[2-O-(2-Acetamido-2,3-dideoxy-1-thio-1-S-triacontanoyl-β-*D-*glucopyranose-3-yl)*-D-*lactoyl]*-L-*alanyl*-D-*isoglutamine* (**37**). — Compound **37** was obtained as crystals in 93% yield; m.p. 212–214° (dec.), $[\alpha]_D$ +7.0° (*c* 0.49, 1:2 chloroform–methanol); ν_{max}^{KBr} 3350–3300 (OH, NH), 2920 and 2840 (Me, methylene), 1720 (C=O), 1700 (S-acyl), and 1650 and 1540 cm^{-1} (amide).

Anal. Calc. for $C_{49}H_{90}N_4O_{11}S$: C, 62.39; H, 9.62; N, 5.94. Found: C, 62.38; H, 9.68; N, 5.96.

N-*[2-O-{2-Acetamido-2,3-dideoxy-1-S-[10-(2,3-dimethoxy-5-methyl-1,4-benzoquinon-6-yl)-decanoyl]-1-thio-β-*D-*glucopyranose-3-yl}*-D-*lactoyl]*-L-*alanyl*-D-*isoglutamine methyl ester* (**38**). — Compound **38** was obtained as an amorphous mass in quantitative yield; $[\alpha]_D$ +9.7° (*c* 0.6, chloroform); ν_{max}^{KBr} 3400–3280 (OH, NH), 2930 and 2850 (Me, methylene), 1740 and 1270 (ester), 1690 (S-acyl), and 1650 and 1550 cm^{-1} (amide).

Anal. Calc. for $C_{39}H_{60}N_4O_{15}S$: C, 54.66; H, 7.06; N, 6.54. Found: C, 54.29; H, 7.34; N, 6.51.

N-*[2-O-{2-Acetamido-2,3-dideoxy-1-S-[10-(2,3-dimethoxy-5-methyl-1,4-benzoquinon-6-yl)-decanoyl]-1-thio-β-*D-*glucopyranose-3-yl}*-D-*lactoyl]*-L-*alanyl*-D-*isoglutamine* (**39**). — Compound **39** was obtained as crystals in 93% yield; m.p. 170–171°, $[\alpha]_D$ +24.2° (*c* 0.6, 1:2 chloroform–methanol); ν_{max}^{KBr} 3380–3250 (OH, NH), 2920 and 2840 (Me, methylene), 1710 (C=O), 1700 (S-acyl), and 1650 and 1540 cm^{-1} (amide).

Anal. Calc. for $C_{38}H_{58}N_4O_{15}S$: C, 54.14; H, 6.94; N, 6.65. Found: C, 54.02; H, 6.74; N, 6.66.

N-*[2-O-(Ethyl 2-acetamido-2,3-dideoxy-1-thio-β-*D-*glucopyranoside-3-yl)-*D-*lactoyl]*-L-*alanyl*-D-*isoglutamine methyl ester* (**40**). — Compound **40** was obtained as crystals in 94% yield; m.p. 110–113°, $[\alpha]_D$ +51.5° (*c* 0.4, methanol); ν_{max}^{KBr} 3300–3230 (OH, NH), 1720 and 1240 (ester), and 1650 and 1540 cm^{-1} (amide).

Anal. Calc. for $C_{22}H_{38}N_4O_{10}S$: C, 47.99; H, 6.96; N, 10.18. Found: C, 47.70; H, 7.25; N, 10.05.

N-*[2-O-(Decyl 2-acetamido-2,3-dideoxy-1-thio-β-*D-*glucopyranoside-3-yl)-*D-*lactoyl]*-L-*alanyl*-D-*isoglutamine methyl ester* (**42**). — Compound **42** was isolated as crystals; m.p. 98–102° (dec.), $[\alpha]_D$ +41.1° (*c* 0.44, methanol); ν_{max}^{KBr} 3300–3230 (OH, NH), 2900 and 2830 (Me, methylene), 1720 and 1240 (ester), and 1650 and 1530 cm^{-1} (amide).

Anal. Calc. for $C_{30}H_{54}N_4O_{10}S$: C, 54.36; H, 8.21; N, 8.45. Found: C, 54.21; H, 8.36; N, 8.33.

N-*[2-O-(Hexadecyl 2-acetamido-2,3-dideoxy-1-thio-β-*D-*glucopyranoside-3-*

yl)-D-*lactoyl]*-L-*alanyl*-D-*isoglutamine methyl ester* (**44**). — Compound **44** was obtained as crystals in quantitative yield; m.p. 179–182° (dec.), $[\alpha]_D$ +44.0° (*c* 0.2, 1:1 chloroform–methanol); ν_{max}^{KBr} 3360–3240 (OH, NH), 2900 and 2830 (Me, methylene), 1720 and 1230 (ester), and 1670–1635 and 1530 cm^{-1} (amide).

Anal. Calc. for $C_{36}H_{66}N_4O_{10}S$: C, 57.88; H, 8.91; N, 7.51. Found: C, 57.65; H, 8.96; N, 7.43.

N-*[2-O-(Eicosyl 2-acetamido-2,3-dideoxy-1-thio-β*-D-*glucopyranoside-3-yl)*-D-*lactoyl]*-L-*alanyl*-D-*isoglutamine methyl ester* (**46**). — Compound **46** was obtained as crystals in quantitative yield; m.p. 110–115° (dec.), $[\alpha]_D$ +38.8° (*c* 0.4, methanol); ν_{max}^{KBr} 3340–3230 (OH, NH), 2900 and 2820 (Me, methylene), 1720 and 1250 (ester), and 1645 and 1530 cm^{-1} (amide).

Anal. Calc. for $C_{40}H_{74}N_4O_{10}S$: C, 59.82; H, 9.29; N, 6.98. Found: C, 59.79; H, 9.38; N, 6.75.

N-*[2-O-(Ethyl 2-acetamido-2,3-dideoxy-1-thio-β*-D-*glucopyranoside-3-yl)*-D-*lactoyl]*-L-*alanyl*-D-*isoglutamine* (**41**). — To a solution of **40** (38 mg) in 1,4-dioxane (3 mL) was added 0.2M potassium hydroxide (3 mL), and the solution was kept for 20 min at room temperature, and then treated with Amberlite IR-120 (H^+) resin to remove the base; the resin was filtered off, and washed with methanol. The filtrate and washings were combined, and evaporated, to afford **41** (36 mg, quantitative), which showed a single spot in t.l.c.; m.p. 116–118°, $[\alpha]_D$ +53.6° (*c* 0.4, methanol); ν_{max}^{KBr} 3350–3250 (OH, NH), 1710 (C=O), and 1640 and 1530 cm^{-1} (amide).

Anal. Calc. for $C_{21}H_{36}N_4O_{10}S$: C, 47.00; H, 6.76; N, 10.44. Found: C, 46.75; H, 6.91; N, 10.38.

Other N-*[2-O-(alkyl 2-acetamido-2,3-dideoxy-1-thio-β*-D-*glucopyranoside-3-yl)*-D-*lactoyl]*-L-*alanyl*-D-*isoglutamines* (**43, 45,** and **47**) were prepared from the corresponding methyl esters (**42, 44,** and **46**) as described for **41**.

N-*[2-O-(Decyl 2-acetamido-2,3-dideoxy-1-thio-β*-D-*glucopyranoside-3-yl)*-D-*lactoyl]*-L-*alanyl*-D-*isoglutamine* (**43**). — Compound **43** was obtained as crystals in quantitative yield; m.p. 105–110° (dec.), $[\alpha]_D$ +38.0° (*c* 0.4, methanol); ν_{max}^{KBr} 3350–3220 (OH, NH), 2900 and 2820 (Me, methylene), 1710 (C=O), and 1650 and 1530 cm^{-1} (amide).

Anal. Calc. for $C_{29}H_{52}N_4O_{10}S$: C, 53.63; H, 8.08; N, 8.64. Found: C, 53.49; H, 8.21; N, 8.65.

N-*[2-O-(Hexadecyl 2-acetamido-2,3-dideoxy-1-thio-β*-D-*glucopyranoside-3-yl)*-D-*lactoyl]*-L-*alanyl*-D-*isoglutamine* (**45**). — Compound **45** was obtained as crystals in 91% yield; m.p. 110–115° (dec.), $[\alpha]_D$ +24.0° (*c* 0.2, methanol); ν_{max}^{KBr} 3380–3260 (OH, NH), 2900 and 2820 (Me, methylene), 1710 (C=O), and 1650 and 1530 cm^{-1} (amide).

Anal. Calc. for $C_{35}H_{64}N_4O_{10}S$: C, 57.35; H, 8.80; N, 7.64. Found: C, 57.09; H, 8.95; N, 7.55.

N-*[2-O-(Eicosyl 2-acetamido-2,3-dideoxy-1-thio-β*-D-*glucopyranoside-3-yl)*-D-*lactoyl]*-L-*alanyl*-D-*isoglutamine* (**47**). — Compound **47** was obtained as crys-

tals in 86% yield; m.p. 132–137° (dec.), $[\alpha]_D$ +38.0° (*c* 0.3, methanol); ν_{max}^{KHr} 3360–3230 (OH, NH), 2900 and 2820 (Me, methylene), 1720 (C=O), and 1650 and 1530 cm^{-1} (amide).

Anal. Calc. for $C_{39}H_{72}N_4O_{10}S$: C, 59.36; H, 9.20; N, 7.10. Found: C, 59.35; H, 9.26; N, 7.15.

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