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SYNTHESIS OF CARBOHYDRATE ANALOGS (POSITIONAL, CONFIGURATIONAL, AND OPTICAL) OF *N*-ACETYLMURAMOYL-L-ALANYL-D-ISOGLUTAMINE, AND THEIR IMMUNOADJUVANT ACTIVITIES*

AKIRA HASEGAWA, YOSHIMI KANEDA, YASUHIKO GOH, KAORU NISHIBORI, MAKOTO KISO,
Department of Agricultural Chemistry, Gifu University, Kakamigahara, Gifu 504 (Japan)
AND ICHIRO AZUMA
Institute of Immunological Science, Hokkaido University, Sapporo 060 (Japan)

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ABSTRACT

2-Acetamido-2-deoxy-4- and -6-*O*-(D-2-propanoyl-L-alanyl-D-isoglutamine)-D-glucopyranose, 2-acetamido-2-deoxy-3-*O*-(D-2-propanoyl-L-alanyl-D-isoglutamine)-D-allopyranose, -D-gulopyranose, -D-galactopyranose, -D-mannopyranose, and -L-idopyranose, and 3-*O*-(D-2-propanoyl-L-alanyl-D-isoglutamine)-D- and -L-glucopyranose were synthesized, in order to clarify the structural requirements for the immunoadjuvant activity of the carbohydrate moiety in *N*-acetylmuramoyl-L-alanyl-D-isoglutamine. Immunoadjuvant activity of the *N*-acetylmuramoyl-dipeptide analogs was examined in guinea-pigs.

INTRODUCTION

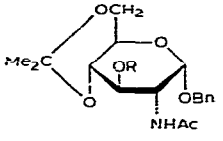
Since the discovery[5] of a simple, dipeptide derivative of muramic acid, namely, *N*-acetylmuramoyl-L-alanyl-D-isoglutamine (MDP), which is the minimal, immunoadjuvant-active structure capable of (*a*) replacing whole bacterial cell in complete Freund's adjuvant, for increasing levels of antibodies against a given antigen, and (*b*) inducing a delayed type of hypersensitivity, a great number of analogs[6] have been synthesized with the objective of elucidating the mechanism of action, and developing a new class of organic compounds useful for clinical applications. In our continuing efforts to delineate the relationship between the activity and the structure of the carbohydrate moiety in MDP, we have demonstrated that not only is the sugar moiety essential for the activity[7] but also that chemical modifications[1,2,8] of the functional groups in the carbohydrate moiety produce various, important effects on the manifestation of activity[2–4]. The present report describes the synthesis of 2-acetamido-2-deoxy-4-and-6-*O*-(D-propanoyl-L-alanyl-D-isoglutamine)-D-glucopyranoses (position-

*Studies on Immunoadjuvant Active Compounds, Part XIII. For Part XII, see ref. 1. For preliminary reports on part of this work, see refs. 2–4.

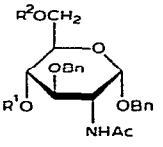
al isomers of MDP, as regards the lactoyl-dipeptide linkage), D-*allo*, D-*gulo*-D-*galacto*, D-*manno*, and L-*ido* types of MDP (configurational isomers of MDP bearing lactoyl-dipeptide at C-3), and D- and L-3-*O*-(D-2-propanoyl-L-alanyl-D-isoglutamine)glucopyranoses, and their immunoadjuvant activities.

RESULTS AND DISCUSSION

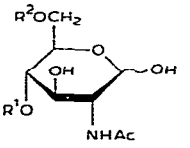
Benzylation of benzyl 2-acetamido-2-deoxy-4,6-*O*-isopropylidene-α-D-glucopyranoside[9] (**1**) gave crystalline[10] **2**, from which benzyl 2-acetamido-3-*O*-benzyl-2-deoxy-α-D-glucopyranoside[11] (**4**) was prepared by hydrolytic removal of the 4,6-*O*-isopropylidene group with 60% aqueous acetic acid. Partial benzylation of **4** according to the procedure of Sinaÿ *et al.*[12], gave benzyl 2-acetamido-3,6- and -3,4-di-*O*-benzyl-2-deoxy-α-D-glucopyranosides (**5** and **8**). Compounds **5** and **8** were condensed with L-2-chloropropanoic acid in the presence of sodium hydride, to give the corresponding 4-*O*- and 6-*O*-(D-1-carboxyethyl) derivatives (**6** and **9**) in high yield. Coupling of **6** and **9** with L-alanyl-D-isoglutamine benzyl ester, using dicyclohexylcarbodiimide and *N*-hydroxysuccinimide (DCC–HOSu) as activating agents in 1,4-dioxane, respectively afforded the corresponding lactoyl-dipeptide derivatives (**7** and **10**) in almost quantitative yield. Hydrogenolytic removal of the protecting groups in compounds **7** and **10**, in 10:10:1 (v/v) methanol–water–acetic acid, with hydrogen in the presence of 10% Pd–C catalyst at 30°, gave the desired, positional isomers (**11** and **12**) of MDP (**45**) as amorphous materials.



1 R = H
2 R = Bn
3 R = Ms
Ac = MeCO
Bn = $PhCH_2$
Ms = $MeSO_2$



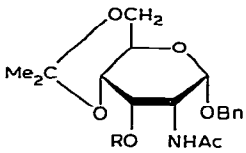
4 $R^1 = R^2 = H$
5 $R^1 = H, R^2 = Bn$
6 $R^1 = a, R^2 = Bn$
7 $R^1 = b, R^2 = Bn$
8 $R^1 = Bn, R^2 = H$
9 $R^1 = Bn, R^2 = a$
10 $R^1 = Bn, R^2 = b$
a = $-CH(Me)CO_2H$
b = —CH(Me)CO-L-Ala-D-isoGln-OBn



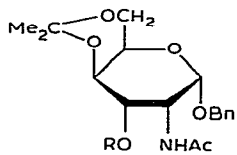
11 $R^1 = c, R^2 = H$
12 $R^1 = H, R^2 = c$
c = —CH(Me)CO-L-Ala-D-isoGln

On the other hand, the benzyl 4,6-*O*-isopropylidenehexopyranosides (**13**, **16**, and **19**) and the benzyl 5,6-*O*-isopropylidenehexofuranosides (**25** and **37**) were first prepared as convenient intermediates for the synthesis of the configurational isomers of the carbohydrate moiety of MDP (**45**). Treatment of benzyl 2-acetamido-2-deoxy-4,6-*O*-isopropylidene-α-D-glucopyranoside[9] (**1**) with methanesulfonyl chloride in pyridine afforded the 3-*O*-mesyl derivative **3**, which was converted, in 79% yield, into benzyl 2-acetamido-2-deoxy-4,6-*O*-isopropylidene-α-D-allopyranoside (**13**) by

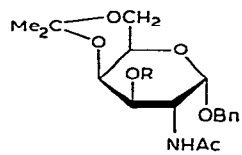
heating with sodium acetate in aqueous 95% 2-methoxyethanol. The kinetically controlled isopropylidenation[13] of benzyl 2-acetamido-2-deoxy-α-D-gulopyranoside[14] and benzyl 2-acetamido-2-deoxy-α-D-galactopyranoside[15], with 2,2-dimethoxypropane, *N*,*N*-dimethylformamide, and *p*-toluenesulfonic acid, gave the corresponding 4,6-*O*-isopropylidene derivatives (**16** and (**19**) in good yield. Acetylation of **19** with



13 R = H
14 R = a
15 R = b



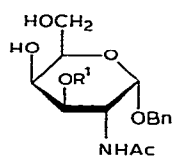
16 R = H
17 R = a
18 R = b



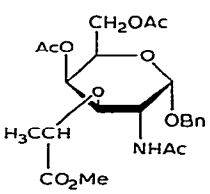
19 R = H
20 R = Ac
21 R = a

acetic anhydride–pyridine afforded a 92% yield of a crystalline monoacetate, **20**, whose n.m.r. spectrum indicated that the observed low-field position of the H-3 signal provided clear evidence for the presence of an acetoxyl group at C-3. Treatment of benzyl 2-acetamido-2-deoxy-α-D-mannofuranoside[16] with the 2,2-dimethoxypropane reagent at room temperature gave a crystalline 5,6-*O*-isopropylidene derivative (**25**) in 95% yield.

Benzyl 2-acetamido-2-deoxy-5,6-*O*-isopropylidene-β-D-glucofuranoside[17] (**28**), derived from 2-acetamido-2-deoxy-D-glucopyranose in one step, served as the starting material for the synthesis of the L-*ido* analog of MDP. Benzoylation of **28**, to give **29**, followed by *O*-deisopropylidenation of **29**, gave benzyl 2-acetamido-3-*O*-benzoyl-



22 R1 = a
23 R2 = b



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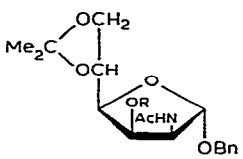
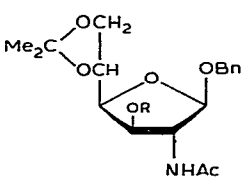
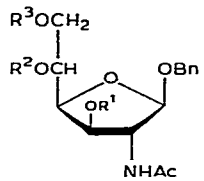


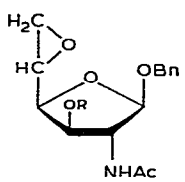
25 R = H
26 R = a
27 R = b



28 R = H
29 R = Bz
Bz = PhCO

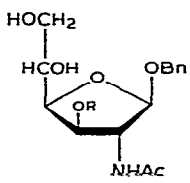


30 R1 = Bz, R2 = R3 = H
31 R1 = R3 = Bz, R2 = H
32 R1 = R3 = Bz, R2 = Ms

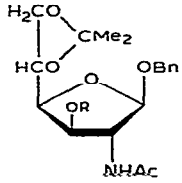


33 R = H
34 R = THP
THP = tetrahydropyran-2-yl

2-deoxy-β-D-glucofuranoside (**30**) in good yield. Selective benzoylation, with benzoyl chloride in pyridine at −20°, of the primary hydroxyl group on C-6 in **30** afforded the 3,6-dibenzoate (**31**) in 84% yield; **31** was mesylated with methanesulfonyl chloride in pyridine to afford the 5-*O*-mesyl derivative **32**. Compound **32** in dry chloroform was treated with methanolic sodium methoxide at −20°, to give crystalline benzyl 2-acetamido-5,6-anhydro-2-deoxy-α-L-idofuranoside (**33**) in 82% yield; this was treated with dihydropyran, to afford the 3-*O*-(tetrahydropyran-2-yl) derivative (**34**). Hydrolysis of the epoxide ring in **34**, under mild, alkaline conditions, gave benzyl 2-acetamido-2-deoxy-3-*O*-(tetrahydropyran-2-yl)-α-L-idofuranoside (**35**); the reaction did not involve inversion of C-5, because of predominant attack[18] at C-6 in **34** by hydroxyl ion. Partial hydrolysis of **35** with 70% aqueous acetic acid, and subsequent isopropylidenation with 2,2-dimethoxypropane, gave the desired 5,6-*O*-isopropylidene derivative (**37**) in good yield.



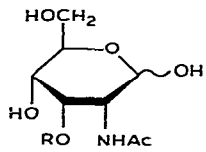
35 R = THP
36 R = H



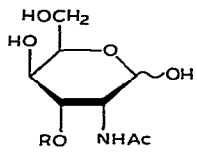
37 R = H
38 R = a
39 R = b

Compounds **13**, **16**, **25**, and **37** thus obtained, were condensed with L-2-chloropropanoic acid in the presence of sodium hydride in dry 1,4-dioxane, to afford the corresponding 3-*O*-(D-1-carboxyethyl) derivatives (**14**, **17**, **26**, and **38**), respectively, in good yields. However, in the condensation of **19** with L-2-chloropropionic acid, the 4,6-*O*-isopropylidene group in compound **21**, thus formed, was very labile, even under only mildly acidic conditions, and therefore, benzyl 2-acetamido-3-*O*-(D-1-carboxyethyl)-2-deoxy-α-D-galactopyranoside (**22**), lacking the isopropylidene group because of hydrolysis during chromatography on a column of silica gel, was obtained in 55% yield. Treatment of the crude **21** with Amberlite IR-120 (H^+) ion-exchange resin in methanol, and subsequent acetylation, afforded, in 79% yield, **24**, the n.m.r. spectrum of which was consistent with the structure depicted.

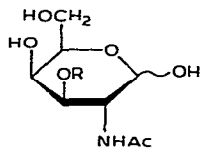
Coupling of compounds **14**, **17**, **22**, **26**, and **38** with L-alanyl-D-isoglutamine benzyl ester was conducted with dicyclohexylcarbodiimide and *N*-hydroxysuccinimide as the activating agents, to afford the corresponding lactoyl-dipeptide derivatives (**15**, **18**, **23**, **27**, and **39**) in almost quantitative yields. Hydrolysis of the isopropylidene group in **15**, **18**, **27**, and **39** under mild, acidic conditions, and subsequent hydrogenolytic removal of the protecting groups (in methanol–water-acetic acid, with hydrogen in the presence of 10% Pd–C catalyst at 30–35°) gave the desired, configurational isomers (**40**–**44**) of *N*-acetylmuramoyl-L-alanyl-D-isoglutamine (**45**; MDP) in high yields.



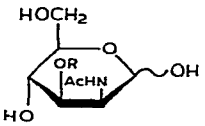
40 R = c



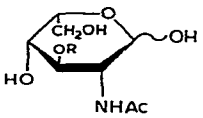
41 R = c



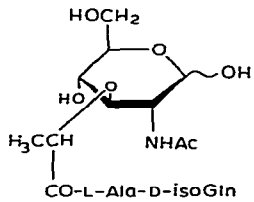
42 R = c



43 R = c



44 R = c



45
(MDP)

Condensation of 1,2:5,6-di-*O*-isopropylidene-α-D-glucofuranose or 1,2:5,6-di-*O*-isopropylidene-α-L-glucofuranose[19] with L-2-chloropropanoic acid as already described, gave the corresponding 3-*O*-(D-1-carboxyethyl) derivatives (**46** and **48**), respectively, in good yields. Coupling of the acids **46** and **48** with the L-alanyl-D-isoglutamine derivative yielded the corresponding dipeptides (**47** and **49**). Compounds **47** and **49** were first treated with 90% aqueous trifluoroacetic acid, to convert them

TABLE I

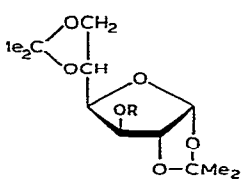
ADJUVANT ACTIVITY OF CARBOHYDRATE ANALOGS OF *N*-ACETYLMURAMOYL-L-ALANYL-D-ISOGLUTAMINE (MDP) ON THE INDUCTION OF DELAYED-TYPE HYPERSENSITIVITY TO ABA-*N*-ACETYLTYROSINE IN GUINEA-PIGS

Compound[a]	*Skin reaction with ABA-BSA*[b] *(100 μg) (diam. in mm + SE)*[c] *at*	
	24 h	*48 h*
11	(4.8 ±1.4)	0
12	(9.7 ±0.2)	(1.4 ±0.9)
40	(7.6 ±1.6)	0
41	(8.5 ±0.6)	0
42	24.6 ±0.9	21.3 ±0.8
43	19.7 ±2.4	19.8 ±1.9
44	(10.7 ±2.1)	(4.5 ±2.0)
51	17.7 ±0.8	14.4 ±0.9
53	(7.8 ±1.2)	0
45 (MDP)	21.0 ±1.8	19.0 ±2.0
Control[d]	0	0

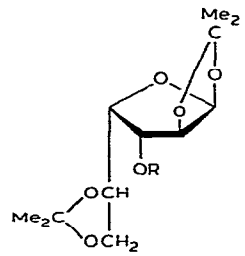
[a]Dose: 100 μg. [b]Azobenzenearsonate–*N*-acetyl-L-tyrosine–bovine serum albumin. [c]The data indicate the average diameter ± the standard error (SE) of the skin reaction (induration) of four guinea-pigs; the values in parentheses indicate the size of the erythema. [d]ABA-*N*-acetyltyrosine in Freund's incomplete adjuvant.

into the pyranoid structures (**50** and **52**), and then hydrogenated in ethanol, with hydrogen in the presence of 10% Pd–C catalyst, to give the desired, optical isomers (**51** and **53**) of the MDP analog.

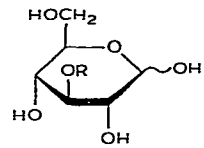
The immunoadjuvant activities of the compounds thus obtained on the induction of the delayed type of hypersensitivity to *N*-acetyl-L-tyrosine-3-azobenzene-4′-arsonate (ABA-*N*-acetyltyrosine) in guinea-pigs were examined[6c] (see Table I). Compounds **11** and **12** abolished the activity almost completely, indicating that, for manifestation of the activity, the position of the linkage between the lactoyldipeptide and the sugar moiety must be at C-3. On the other hand, among the configurational isomers (**40–44**) of the carbohydrate moiety in MDP, the D-*galacto* and D-*manno* analogs (**42** and **43**) showed strong activity, comparable to that of MDP, whereas other isomers had no adjuvant activity. 3-*O*-(D-2-Propanoyl-L-alanyl-D-isoglutamine)-D-glucopyranose (**51**), bearing a hydroxyl group (instead of an



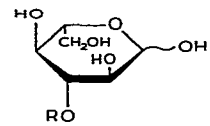
46 R = a
47 R = b



48 R = a
49 R = b



50 R = b
51 R = c



52 R = b
53 R = c

acetamido group on C-2 of the carbohydrate moiety in MDP), showed potent activity, almost comparable to that of MDP; however the corresponding L-glucopyranose analog (**53**) exhibited complete loss of activity. These results suggest that, for activity, the configurations of C-3 and C-5 are highly critical, but that those of C-2 and C-4 are unimportant. In addition, it was indicated that the substituent on C-2 is not restricted to the acetamido group, which can be replaced by other groups.

EXPERIMENTAL

General methods. — Melting points were determined with a Yanagimoto micro melting-point apparatus and are uncorrected. Specific rotations were determined with a Union PM-201 polarimeter, and i.r. spectra were recorded with a Jasco IRA-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*.

Benzyl 2-acetamido-3-O-benzyl-2-deoxy-4,6-O-isopropylidene-α-D-glucopyranoside (**2**). — To a stirred solution of[9] **1** (1.35 g) in *N*,*N*-dimethylformamide (10 mL)

kept at 0° were added benzyl bromide (3 mL), barium oxide (3.0 g), and barium hydroxide octahydrate (1.2 g). The mixture was stirred for 2 h at 0°, and then for 15 h at room temperature. Chloroform (50 mL) was added, and the mixture was filtered, the residue being washed with more chloroform. The filtrate and washings were combined, washed with water, dried (sodium sulfate), and evaporated to a syrup which was chromatographed on a column of silica gel (25 g) with benzene and then 100:1 benzene–methanol. The latter eluate afforded 1.3 g (78%) of the product; recrystallization from ethanol–hexane gave needles, m.p. 135–137°, $[\alpha]_D^{20}$ +149° (*c* 0.3, chloroform) (lit.[10] m.p. 127–128°, $[\alpha]_D^{25}$ +112°).

Anal. Calc. for $C_{25}H_{31}NO_6$: C, 68.00; H, 7.08; N, 3.17. Found: C, 67.92; H, 7.03; N, 3.22.

*Benzyl 2-acetamido-3-*O*-benzyl-2-deoxy-α-*D*-glucopyranoside* (**4**). — A solution of **2** (3.2 g) in 60% aqueous acetic acid (30 mL) was heated for 2 h at 40–45°, and evaporated, and the residue crystallized from ethyl acetate–hexane to give **4** (2.8 g, 95%) as needles, m.p. 184–185°, $[\alpha]_D^{20}$ +154° (*c* 1.0, methanol) (lit.[11] m.p. 179°, $[\alpha]_D^{20}$ +155°).

Anal. Calc. for $C_{22}H_{27}NO_6$: C, 65.42; H, 6.78; N, 3.49. Found: C, 65.38; H, 6.61; N, 3.39.

*Benzyl 2-acetamido-3,6-di-*O*-benzyl-2-deoxy-α-*D*-glucopyranoside* (**5**) *and benzyl 2-acetamido-3,4-di-*O*-benzyl-2-deoxy-α-*D*-glucopyranoside* (**8**). — To a stirred solution of **4** (1.4 g) in *N*,*N*-dimethylformamide (14 mL) kept at −5° were added benzyl bromide (1.2 g), barium oxide (3.5 g), and barium hydroxide octahydrate (1.1 g). The mixture was stirred for 4 h at −5°, and then water (3 mL) was added. The mixture was filtered, and the solid was washed with chloroform. The filtrate and washings were combined, washed with water, dried (sodium sulfate), and evaporated to a syrup which was chromatographed on a column of silica gel (20 g) with (*a*) chloroform, (*b*) 200:1, and (*c*) 100:1 chloroform–methanol. Eluant (*a*) gave 2-acetamido-3,4,6-tri-*O*-benzyl-2-deoxy-α-D-glucopyranoside (500 mg, 25%), m.p. 174–176°, $[\alpha]_D^{20}$ +108° (*c* 0.5, chloroform) (lit.[12] m.p. 175–176°, $[\alpha]_D^{20}$ +111°); eluant (*b*) afforded the 3,6-di-*O*-benzyl derivative **5** (66 mg, 39%), m.p. 146–147°, $[\alpha]_D^{22}$ +110° (*c* 0.5, chloroform) (lit.[11] m.p. 145–155.5°, $[\alpha]_D^{20}$ +114°); and eluant (*c*) yielded the 3,4-di-*O*-benzyl derivative **8** (140 mg, 8.2%), m.p. 207–208°, $[\alpha]_D^{20}$ +127.5° (*c* 0.5, chloroform) (lit.[11] m.p. 204–205°, $[\alpha]_D^{20}$ +121°).

*Benzyl 2-acetamido-3,6-di-*O*-benzyl-4-*O*-(*D*-1-carboxyethyl)-2-deoxy-α-*D*-glucopyranoside* (**6**). — To a stirred solution of **5** (500 mg) in dry 1,4-dioxane (10 mL) was added sodium hydride in oil suspension (150 mg; 50% of sodium hydride by weight) at room temperature. The mixture was kept for 1 h at 95°, and then L-2-chloropropanoic acid (980 mg) was added, with stirring, at 65°. The mixture was stirred for 3 h at 65°, and cooled; 2M hydrochloric acid was carefully added to the cooled mixture until pH 8 was reached, and the mixture was evaporated. Chloroform (100 mL) and water (100 mL) were added to the residue, and then 2M hydrochloric acid was added, with stirring, at 0°, to pH 3. The chloroform layer was separated, and the aqueous solution was thoroughly extracted with chloroform. The chloroform

layer and extracts were combined, washed with water, dried (sodium sulfate), and evaporated to a syrup which was chromatographed on a column of silica gel (10 g) with chloroform, and then 100:1 chloroform–methanol. The latter eluate gave compound **6** (430 mg, 75%). Recrystallization from ethyl acetate–hexane gave needles, m.p. 139–140°, $[\alpha]_D^{20}$ +127° (*c* 1.0, chloroform); ν_{max}^{Nujol} 3260 (NH), 2730–2600 (COOH), 1720 (C=O), 1640 and 1540 (amide), and 740, 730, and 690 cm^{-1} (phenyl); n.m.r. data (in chloroform-*d*): δ 1.40 (d, 3 H, $J_{CH_3,CH}$ 7.0 Hz, MeCH), 1.76 (s, 3 H, AcN), 4.86 (d, 1 H, $J_{1,2}$ 3.8 Hz, H-1), 5.43 (d, 1 H, $J_{2,NH}$ 9.5 Hz, NH), 7.18 (s, 1 H, COOH), and 7.26 (s, 15 H, 3 Ph).

Anal. Calc. for $C_{32}H_{37}NO_8$: C, 68.19; H, 6.62; N, 2.49. Found: C, 68.15; H, 6.58; N, 2.55.

*Benzyl 2-acetamido-3,4-di-*O*-benzyl-6-*O*-(*D*-1-carboxyethyl)-2-deoxy-α-*D*-glucopyranoside* (**9**). — To a stirred solution of **8** (1.0 g) in dry 1,4-dioxane (10 mL) was added the sodium hydride reagent (300 mg), and the mixture was kept, with stirring, for 1.5 h at 95°, and then cooled. L-2-Chloropropanoic acid (430 mg) was added to the stirred mixture, which was then kept for 1 h at 65°, and processed as described for the preparation of compound **6**. Chromatographic fractionation of the product on silica gel (20 g), with elution with 50:1 chloroform–methanol, afforded compound **9** (1.0 g, 87%) as crystals. Recrystallization from ethyl acetate–hexane gave needles, m.p. 165–166°, $[\alpha]_D^{22}$ +85° (*c* 0.3, chloroform); ν_{max}^{Nujol} 3270 (NH), 2730–2600 (COOH), 1730 (C=O), 1640 and 1540 (amide), and 740, 720, and 690 cm^{-1} (phenyl); n.m.r. data (in chloroform-*d*): δ 1.45 (d, 3 H, $J_{CH_3,CH}$ 7.0 Hz, MeCH), 1.80 (s, 3 H, AcN), 4.85 (d, 1 H, $J_{1,2}$ 3.6 Hz, H-1), 5.55 (d, 1 H, $J_{2,NH}$ 9.0 Hz, NH), 7.35 (s, 15 H, 3 Ph), and 8.75 (s, 1 H, COOH).

Anal. Calc. for $C_{32}H_{37}NO_8$: C, 68.19; H, 6.62; N, 2.49. Found: C, 68.30; H, 6.55; N, 2.38.

*Benzyl 2-acetamido-3,6-di-*O*-benzyl-2-deoxy-4-*O*-(*D*-2-propanoyl-*L*-alanyl-*D*-isoglutamine benzyl ester)-α-*D*-glucopyranoside* (**7**). — To a solution of **6** (250 mg) in dry 1,4-dioxane (2.5 mL) were added *N*-hydroxysuccinimide (HOSu) (76 mg) and dicyclohexylcarbodiimide (DCC) (136 mg), the mixture was stirred for 1 h at room temperature, and then the 1,3-dicyclohexylurea formed was removed by filtration. L-Alanyl-D-isoglutamine benzyl ester trifluoroacetate (220 mg) and triethylamine (53 mg) were added to the filtrate, and the mixture was stirred for 1 h at room temperature, and then evaporated. The residue was purified by chromatography on a column of silica gel (10 g) with (*a*) 100:1, and (*b*) 50:1 chloroform–methanol. Eluant (*b*) afforded compound **7** (340 mg, 90%), amorphous, $[\alpha]_D^{20}$ +67° (*c* 1.0, chloroform); ν_{max}^{KBr} 3280 (NH), 1720 and 1240 (ester), 1670, 1640, and 1530 (amide), and 730, 710, and 690 cm^{-1} (phenyl); n.m.r. data (in 1:1 chloroform-*d*–methanol-d_4): δ 1.26 and 1.30 (2 d, 6 H, $J_{CH_3,CH}$ 7.0 Hz, 2 MeCH), 1.73 (s, 3 H, AcN), 4.82 (d, 1 H, $J_{1,2}$ 3.8 Hz, H-1), 5.05 (s, 2 H, CO_2CH_2Ph), 6.18 (d, 1 H, $J_{2,NH}$ 9.5 Hz, NH), 7.25 (s, 20 H, 4 Ph), and 7.45 and 7.80 (2 d, 2 H, $J_{NH,CH}$ 7.5 Hz, 2 NH).

Anal. Calc. for $C_{47}H_{56}N_4O_{11}$: C, 66.18; H, 6.62; N, 6.57. Found: C, 66.09; H, 6.58; N, 6.51.

*Benzyl 2-acetamido-3,4-di-*O*-benzyl-2-deoxy-6-*O*-(*D*-2-propanoyl-*L*-alanyl-*D*-isoglutamine benzyl ester)-α-*D*-glucopyranoside* (**10**). — Coupling of **9** (250 mg) with L-alanyl-D-isoglutamine benzyl ester trifluoroacetate (220 mg) in dry 1,4-dioxane (2.5 mL) by using HOSu (75 mg), DCC (136 mg), and triethylamine (53 mg) was performed as described in the previous section. The product was purified by chromatography on a column of silica gel (10 g) with 50:1 chloroform–methanol, to afford compound **10** (310 mg, 82%), m.p. 204–205° (dec.), $[\alpha]_D^{20}$ +64° (*c* 0.9, 1:1 chloroform–methanol); ν_{max}^{Nujol} 3300 and 3230 (NH), 1720 (ester), 1650 and 1550 (amide), and 740 and 690 cm^{-1} (phenyl); n.m.r. data (in 1:1 chloroform-*d*–methanol-d_4): δ 1.27 and 1.32 (2 d, 2 H, $J_{CH_3,CH}$ 6.8 Hz, 2 MeCH), 1.81 (s, 3 H, AcN), 4.85 (d, 1 H, $J_{1,2}$ 3.6 Hz, H-1), 5.10 (s, 2 H, CO_2CH_2Ph), 6.60 (d, 1 H, $J_{2,NH}$ 9.5 Hz, NH), 7.30 (s, 20 H, 4 Ph), and 7.70 and 7.82 (2 d, 2 H, $J_{NH,CH}$ 7.5 Hz, 2 NH).

Anal. Calc. for $C_{47}H_{56}N_4O_{11}$: C, 66.18; H, 6.62; N, 6.57. Found: C, 66.23; H, 6.55; N, 6.52.

*2-Acetamido-2-deoxy-4-*O*-(*D*-2-propanoyl-*L*-alanyl-*D*-isoglutamine)-*D*-glucopyranose* (**11**). — To a solution of **7** (200 mg) in methanol (20 mL) and acetic acid (2 mL) was added 10% Pd–C catalyst (200 mg), and hydrogen was bubbled through for 3 h while the solution was stirred at 30–35°. Water (20 mL) was added to the solution, and hydrogen was again bubbled through the mixture for 1 h at 30°. The catalyst was filtered off, and successively washed with methanol and water. The filtrate and washings were combined, and evaporated below 40°, to give amorphous **11** (110 mg; quantitative), $[\alpha]_D^{20}$ +55° (*c* 0.3, methanol; equil.); ν_{max}^{KBr} 3300 (OH, NH), and 1650 and 1540 cm^{-1} (amide).

Anal. Calc. for $C_{19}H_{32}N_4O_{11}$: C, 46.33; H, 6.55; N, 11.38. Found: C, 45.98; H, 6.90; N, 11.01.

*2-Acetamido-2-deoxy-6-*O*-(*D*-2-propanoyl-*L*-alanyl-*D*-isoglutamine)-*D*-glucopyranose* (**12**). — Hydrogenation of **10** (200 mg) in methanol (20 mL), acetic acid (2 mL), and water (20 mL), in the presence of 10% Pd–C catalyst (200 mg), was conducted according to the procedure described in the previous section, affording amorphous **12** (115 mg; quantitative), $[\alpha]_D^{20}$ +24° (*c* 1.1, methanol; equil.); ν_{max}^{KBr} 3300–3250 (OH, NH), and 1650 and 1540 cm^{-1} (amide).

Anal. Calc. for $C_{19}H_{32}N_4O_{11}$: C, 46.33; H, 6.55; N, 11.38. Found: C, 46.29; H, 6.80; N, 11.15.

*Benzyl 2-acetamido-2-deoxy-4,6-*O*-isopropylidene-3-*O*-mesyl-α-*D*-glucopyranoside* (**3**). — To an ice-cooled solution of **1** (5.0 g) in dry pyridine (30 mL) was added methanesulfonyl chloride (2.5 g), and the mixture was kept for 4 h at 0°, and evaporated; the residue was extracted with chloroform, and the extract successively washed with 2M hydrochloric acid, M sodium carbonate, and water, dried (sodium sulfate), and evaporated to a syrup. The product was purified by chromatography on a column of silica gel (50 g) with ethyl acetate. Recrystallization from ether–hexane afforded **3** (4.95 g, 81%) as needles, m.p. 112°, $[\alpha]_D^{20}$ +108° (*c* 1.0, chloroform); ν_{max}^{Nujol} 3280 (NH), 1650 and 1540 (amide), 1180 (SO_2), 850 (Me_2C), and 740–690 cm^{-1} (phenyl).

Anal. Calc. for $C_{19}H_{27}NO_8S$: C, 53.13; H, 6.34; N, 3.26. Found: C, 53.28; H, 6.51; N, 3.52.

Benzyl 2-acetamido-2-deoxy-4,6-O-isopropylidene-α-D-allopyranoside (**13**). — To a solution of **3** (2.0 g) in 95% aqueous 2-methoxyethanol (100 mL) was added sodium acetate (4.0 g), and the mixture was boiled under reflux, with stirring, for 38 h, cooled, and evaporated. The residue was extracted with chloroform, and the extract successively washed with 2M hydrochloric acid, M sodium carbonate, and water, dried (sodium sulfate), and evaporated, to give a syrup that crystallized from ether. The product was obtained as needles, yield 1.3 g (79%), m.p. 163°, $[\alpha]_D^{20}$ +128.5° (*c* 1.0, chloroform); ν_{max}^{Nujol} 3460 (OH), 3380 (NH), 1650 and 1500 (amide), 860 (Me_2C), and 740–695 cm^{-1} (phenyl).

Anal. Calc. for $C_{18}H_{25}NO_6$: C, 61.52; H, 7.17; N, 3.99. Found: C, 61.55; H, 7.20; N, 4.13.

Benzyl 2-acetamido-3-O-(D-1-carboxyethyl)-2-deoxy-4,6-O-isopropylidene-α-D-allopyranoside (**14**). — To a stirred solution of **13** (400 mg) in dry 1,4-dioxane (3 mL) was added the sodium hydride reagent (200 mg), and the mixture was stirred for 1.5 h at 95°, and cooled. L-2-Chloropropanoic acid (250 mg) and dry 1,4-dioxane (1 mL) were added to the stirred mixture, and it was stirred for 2 h at 65°; t.l.c. then showed the reaction to be complete. The mixture was processed, as already described, to give crystalline **14** (340 mg, 71%), m.p. 161–162°, $[\alpha]_D^{20}$ +56° (*c* 1.0, chloroform); ν_{max}^{Nujol} 3430 (NH), 2720–2630 (COOH), 1730 (C=O), 1640 and 1520 (amide), 850 (Me_2C), and 730 and 690 cm^{-1} (phenyl); n.m.r. data (in chloroform-*d*): δ 1.35, 1.45 (2 s, 6 H, Me_2C), 1.38 (d, 3 H, $J_{CH_3,CH}$ 7.0 Hz, MeCH), 1.89 (s, 3 H, AcN), 4.15 (q, 1 H, J_{CH,CH_3} 7.0 Hz, MeCH), 4.55 (2 d, 2 H, J_{gem} 12.0 Hz, benzyl methylene), 4.75 (d, 1 H, $J_{1,2}$ 4.0 Hz, H-1), 6.13 (d, 1 H, $J_{2,NH}$ 8.0 Hz, NH), 7.25 (s, 5 H, Ph), and 8.30 (broad s, 1 H, COOH).

Anal. Calc. for $C_{21}H_{29}NO_8$: C, 59.56; H, 6.90; N, 3.31. Found: C, 59.39; H, 6.86; N, 3.25.

Benzyl 2-acetamido-2-deoxy-4,6-O-isopropylidene-3-O-(D-2-propanoyl-L-alanyl-D-isoglutamine benzyl ester)-α-D-allopyranoside (**15**). — Coupling of **14** (250 mg) with L-alanyl-D-isoglutamine benzyl ester trifluoroacetate (300 mg) in dry 1,4-dioxane (3 mL) by using HOSu (103 mg), DCC (185 mg), and triethylamine (70 mg) was performed as described for the preparation of **8**, to give amorphous **15** (400 mg, 95%); $[\alpha]_D^{20}$ +25° (*c* 1.0, chloroform); ν_{max}^{KBr} 3380–3300 (NH), 1720 and 1250 (ester), 1640 and 1500 (amide), and 730 and 680 cm^{-1} (phenyl).

Anal. Calc. for $C_{36}H_{48}N_4O_{11}$: C, 60.66; H, 6.79; N, 7.86. Found: C, 60.49; H, 6.58; N, 7.83.

Benzyl 2-acetamido-2-deoxy-4,6-O-isopropylidene-α-D-gulopyranoside (**16**). — A suspension of benzyl 2-acetamido-2-deoxy-α-D-gulopyranoside[14] (1.52 g) in 1,4-dioxane (20 mL) was stirred at room temperature while 2,2-dimethoxypropane (5 mL) and *p*-toluenesulfonic acid monohydrate (20 mg) were added; stirring was continued for 1 h. The mixture was treated with Amberlite IRA-410 (OH^-) ion-exchange resin to remove the acid and the resin was filtered off, and washed with 1,4-dioxane. The

filtrate and washings were combined, and evaporated to a crystalline solid. Recrystallization from ethyl acetate gave **16** (1.56 g, 91%) as needles, m.p. 165–166°, $[\alpha]_D^{20}$ +139° (*c* 1.0, methanol); ν_{max}^{Nujol} 3320–3270 (OH, NH), 1620 and 1540 (amide), 850 (Me_2C), and 740 and 680 cm^{-1} (phenyl).

Anal. Calc. for $C_{18}H_{25}NO_6$: C, 61.52; H, 7.17; N, 3.99. Found: C, 61.39; H, 7.19; N, 4.25.

*Benzyl 2-acetamido-3-O-(*D*-1-carboxyethyl)-2-deoxy-4,6-O-isopropylidene-α-*D*-gulopyranoside* (**17**). — To a stirred solution of **16** (1.05 g) in dry 1,4-dioxane (8 mL) was added the sodium hydride reagent (300 mg), and the mixture was stirred for 1 h at 95°, and then cooled to 65°. L-2-Chloropropanoic acid (520 mg) was added to the stirred mixture, and the mixture was stirred for 3 h at 65°; it was then processed as already described. The product crystallized from ethyl acetate–hexane, to give **17** (1.01 g, 80%) as needles, m.p. 167–169°, $[\alpha]_D^{20}$ +90° (*c* 1.0, methanol); ν_{max}^{Nujol} 3420 (NH), 2730–2550 (COOH), 1740 (C=O), 1630 and 1530 (amide), 855 (Me_2C), and 740 and 690 cm^{-1} (phenyl); n.m.r. data (in chloroform-*d*): δ 1.43 (s, 6 H, Me_2C), 1.45 (d, 3 H, $J_{CH_3,CH}$ 6.0 Hz, MeCH), 1.98 (s, 3 H, AcN), 4.98 (d, 1 H, $J_{1,2}$ 4.0 Hz, H-1), 6.38 (d, 1 H, $J_{2,NH}$ 9.0 Hz, NH), 7.32 (s, 5 H, Ph), and 9.04 (s, 1 H, COOH).

Anal. Calc. for $C_{21}H_{29}NO_8$: C, 59.56; H, 6.90; N, 3.31. Found: C, 59.33; H, 6.85; N, 3.30.

*Benzyl 2-acetamido-2-deoxy-4,6-O-isopropylidene-3-O-(*D*-2-propanoyl-*L*-alanyl-*D*-isoglutamine benzyl ester)-α-*D*-gulopyranoside* (**18**). — Coupling of **17** (230 mg) with the dipeptide (250 mg) in dry 1,4-dioxane (2.5 mL) in the presence of HOSu (90 mg), DCC (150 mg), and triethylamine (70 mg) as already described gave the product. It was purified by chromatography on a column of silica gel (10 g) with chloroform, and then 30:1 chloroform–methanol. With the latter eluate, amorphous compound **18** (370 mg, 96%) was obtained; $[\alpha]_D^{20}$ +10° (*c* 1.0, chloroform); ν_{max}^{KBr} 3330–3280 (NH), 1740 (C=O), 1660 and 1520 (amide), 850 (Me_2C), and 740, 720 and 700 cm^{-1} (phenyl); n.m.r. data (in chloroform-*d*): δ 1.15 and 1.30 (2 d, 6 H, $J_{CH_3,CH}$ 7.0 Hz, 2 MeCH), 1.42 (s, 6 H, Me_2C), 1.95 (s, 3 H, AcN), 4.95 (d, 1 H, $J_{1,2}$ 3.8 Hz, H-1), 5.10 (s, 2 H, CO_2CH_2Ph), and 7.32 (s, 10 H, 2 Ph).

Anal. Calc. for $C_{36}H_{48}N_4O_{11}$: C, 60.66; H, 6.79; N, 7.86. Found: C, 60.52; H, 6.65; N, 7.82.

*Benzyl 2-acetamido-2-deoxy-4,6-O-isopropylidene-α-*D*-galactopyranoside* (**19**). — To a stirred solution of benzyl 2-acetamido-2-deoxy-α-D-galactopyranoside[15] (1.25 g) in *N*,*N*-dimethylformamide (10 mL) were added 2,2-dimethoxypropane (1.5 mL) and *p*-toluenesulfonic acid monohydrate (10 mg). The mixture was stirred for 1.5 h at room temperature and then treated with Amberlite IRA-410 (OH^-) resin, to remove the acid; the resin was filtered off, and washed with methanol. The filtrate and washings were combined, and evaporated, and the syrupy residue was chromatographed on a column of silica gel (15 g) with chloroform and then 150:1 chloroform–methanol. The latter eluate gave compound **19** (1.1 g, 78%) as a syrup, $[\alpha]_D^{25}$ +141° (*c* 1.8, chloroform); ν_{max}^{film} 3300–3240 (OH, NH), 1630 and 1540 (amide), 855 and 845 (Me_2C), and 720 and 690 cm^{-1} (phenyl); n.m.r. data (in chloroform-*d*): δ 1.42

(s, 6 H, Me_2C), 1.87 (s, 3 H, AcN), 4.42 and 4.68 (2 d, 2 H, J_{gem} 11.5 Hz, benzyl methylene), 4.98 (d, 1 H, $J_{1,2}$ 3.5 Hz, H-1), 6.40 (d, 1 H, $J_{2,NH}$ 9.0 Hz, NH), and 7.38 (s, 5 H, Ph).

Anal. Calc. for $C_{18}H_{25}NO_6$: C, 61.52; H, 7.17; N, 3.99. Found: C, 61.32; H, 7.35; N, 3.76.

*Benzyl 2-acetamido-3-*O*-acetyl-2-deoxy-4,6-*O*-isopropylidene-α-*D*-galactopyranoside* (**20**). — Acetylation of **19** (600 mg) with acetic anhydride (0.5 mL)–pyridine (5 mL) at room temperature gave the product. Recrystallization from ether–hexane gave **20** (620 mg, 92%) as needles, m.p. 123°, $[\alpha]_D^{25}$ +139° (*c* 1.0, chloroform); ν_{max}^{Nujol} 3270 (NH), 1735 and 1240 (ester), 1640 and 1540 (amide), 860 and 855 (Me_2C), and 730 and 680 cm^{-1} (phenyl); n.m.r. data (in chloroform-*d*): δ 1.42 and 1.46 (2 s, 6 H, Me_2C), 1.87 (s, 3 H, AcN), 2.03 (s, 3 H, AcO), 3.50–3.55 (m, 1 H, H-5), 3.78 and 4.02 (2 dd, 2 H, $J_{6,6'}$ 12.0 Hz, $J_{5,6} = J_{5,6'} = 2.0$ Hz, H-6,6'), 4.25 (dd, 1 H, $J_{3,4}$ 3.8, $J_{4,5}$ 1.0 Hz, H-4), 4.49 and 4.51 (2 d, 2 H, J_{gem} 12.0 Hz, benzyl methylene), 4.50–4.80 (m, 1 H, H-2), 5.01 (d, 1 H, $J_{1,2}$ 3.8 Hz, H-1), 5.10 (dd, 1 H, $J_{2,3}$ 11.5, $J_{3,4}$ 3.8 Hz, H-3), 6.03 (d, 1 H, $J_{2,NH}$ 9.5 Hz, NH), and 7.27 (s, 5 H, Ph).

Anal. Calc. for $C_{20}H_{27}NO_7$: C, 61.05; H, 6.92; N, 3.56. Found: C, 61.23; H, 6.89; N, 3.51.

*Benzyl 2-acetamido-3-*O*-(*D*-1-carboxyethyl)-2-deoxy-α-*D*-galactopyranoside* (**22**). — To a stirred solution of **19** (500 mg) in dry 1,4-dioxane (2 mL) was added the sodium hydride reagent (200 mg), and the mixture was stirred for 30 min at 90–95°, and then cooled to 65°. L-2-Chloropropanoic acid (250 mg) was added, and the mixture was stirred for 3 h at 65°, and then processed as already described. The product was chromatographed on a column of silica gel (10 g) with (*a*) chloroform, (*b*) 50:1, and (*c*) 20:1 chloroform–methanol. Eluant (*c*) gave compound **22** (300 mg, 55%), m.p. 172–174°, $[\alpha]_D^{20}$ +141° (*c* 0.2, chloroform); ν_{max}^{Nujol} 3400 (OH), 3280 (NH), 2700–2600 (COOH), 1720 (C=O), 1620 and 1560 (amide), and 720 and 690 cm^{-1} (phenyl).

Anal. Calc. for $C_{18}H_{25}NO_8$: C, 56.39; H, 6.57; N, 3.65. Found: C, 56.52; H, 6.73; N, 3.61.

*Benzyl 2-acetamido-4,6-di-*O*-acetyl-2-deoxy-3-*O*-[*D*-1-(methoxycarbonyl)ethyl]-α-*D*-galactopyranoside* (**24**). — To a stirred solution of **19** (380 mg) in dry 1,4-dioxane (2 mL) was added the sodium hydride reagent (150 mg), and the mixture was stirred for 30 min at 90°, and then cooled to 65°; it was processed in the usual way, to give the crude acid (**21**). To a stirred solution of the acid in methanol (5 mL) was added Amberlite IR-120 (H^+) resin (3 g), and the mixture was stirred for 20 h at 50°. The resin was filtered off, and washed with methanol. The filtrate and washings were combined, and evaporated to a syrup which was acetylated with acetic anhydride–pyridine. The product, purified by chromatography on a column of silica gel (15 g) with 200:1 chloroform–methanol, was obtained as a syrup: yield 410 mg (79%), $[\alpha]_D^{20}$ +121° (*c* 0.49, chloroform); ν_{max}^{film} 3320 (NH), 1740 and 1220 (ester), 1660 and 1520 (amide), and 730 and 690 cm^{-1} (phenyl); n.m.r. data (in chloroform-*d*): δ 1.29 (d, 3 H, $J_{CH_3,CH}$ 6.0 Hz, MeCH), 2.01, 2.03, and 2.12 (3 s, 9 H, AcN, 2 AcO), 3.75

(s, 3 H, MeO), 4.49 and 4.65 (2 d, 2 H, J_{gem} 12.0 Hz, benzyl methylene), 5.35 (near d, 1 H, $J_{3,4}$ 3.5 Hz, H-4), 5.40 (d, 1 H, $J_{1,2}$ 4.0 Hz, H-1), and 7.28 (s, 5 H, Ph).

Anal. Calc. for $C_{23}H_{31}NO_{10}$: C, 57.37; H, 6.49; N, 2.91. Found: C, 57.13; H, 6.65; N, 2.83.

*Benzyl 2-acetamido-2-deoxy-3-*O*-(*D*-2-propanoyl-*L*-alanyl-*D*-isoglutamine benzyl ester)-α-*D*-galactopyranoside* (**23**). — Coupling of **22** (100 mg) with the dipeptide (120 mg) in dry 1,4-dioxane (1.5 mL) in the presence of HOSu (40 mg), DCC (70 mg), and triethylamine (30 mg), as already described, afforded the product. It was chromatographed on a column of silica gel (15 g) with (*a*) 100:1, and (*b*) 10:1 chloroform–methanol. Eluant (*b*) gave compound **23** (160 mg, 91%), m.p. 213–214° (dec.), $[\alpha]_D^{20}$ +58.5° (*c* 0.2, methanol); ν_{max}^{Nujol} 3380, 3250 (OH, NH), 1720 and 1230 (ester), 1640, 1600, 1550, and 1540 (amide), and 740 and 680 cm^{-1} (phenyl).

Anal. Calc. for $C_{33}H_{44}N_4O_{11}$: C, 58.92; H, 6.59; N, 8.33. Found: C, 58.83; H, 6.57; N, 8.30.

*Benzyl 2-acetamido-2-deoxy-5,6-*O*-isopropylidene-α-*D*-mannofuranoside* (**25**). — To a stirred solution of benzyl 2-acetamido-2-deoxy-α-D-mannofuranoside[16] (8.0 g) in *N*,*N*-dimethylformamide (100 mL) were added 2,2-dimethoxypropane (10 mL) and *p*-toluenesulfonic acid monohydrate (200 mg), and the mixture was stirred for 2 h at room temperature, and treated with Amberlite IRA-410 (OH^-) resin. The mixture was processed as already described, to give the crystalline 5,6-*O*-isopropylidene derivative **25** (8.5 g, 94%), m.p. 165–166°, $[\alpha]_D^{20}$ +75.5° (*c* 1.0, chloroform); ν_{max}^{Nujol} 3300–3250 (OH, NH), 1640 and 1535 (amide), 855 (Me_2C), and 740 and 690 cm^{-1} (phenyl); n.m.r. data (in chloroform-*d*): δ 1.32 and 1.38 (2 s, 6 H, Me_2C), 1.95 (s, 3 H, AcN), 4.45 and 4.74 (2 d, 2 H, J_{gem} 12.0 Hz, benzyl methylene), 5.05 (d, 1 H, $J_{1,2}$ 1.5 Hz, H-1), 6.60 (d, 1 H, $J_{2,NH}$ 7.0 Hz, NH), and 7.30 (s, 5 H, Ph).

Anal. Calc. for $C_{18}H_{25}NO_6$: C, 61.52; H, 7.17; N, 3.99. Found: C, 61.51; H, 7.21; N, 3.99.

*Benzyl 2-acetamido-3-*O*-(*D*-1-carboxyethyl)-2-deoxy-5,6-*O*-isopropylidene-α-*D*-mannofuranoside* (**26**). — To a stirred solution of **25** (2.8 g) in dry 1,4-dioxane (30 mL) was added the sodium hydride reagent (920 mg), and the mixture was stirred for 1 h at 95°, and cooled to 65°. L-2-Chloropropanoic acid (1.8 g) was slowly added to the stirred mixture. After being kept for 3 h at 65°, with stirring, the mixture was processed as described before. The crude product was crystallized from ethyl acetate–hexane, to give **26** (3.2 g, 95%) as needles, m.p. 148–149°, $[\alpha]_D^{20}$ +49.5° (*c* 1.0, chloroform); ν_{max}^{Nujol} 3300 (NH), 2750–2600 (COOH), 1735 (C=O), 1640 and 1540 (amide), 860 (Me_2C), and 730 and 690 cm^{-1} (phenyl); n.m.r. data (in chloroform-*d*): δ 1.35 (d, 3 H, $J_{CH_3,CH}$ 7.0 Hz, MeCH), 1.35 and 1.47 (2 s, 6 H, Me_2C), 2.02 (s, 3 H, AcN), 4.10 (q, 1 H, $J_{CH_3,CH}$ 7.0 Hz, MeCH), 4.35 and 4.63 (2 d, 2 H, J_{gem} 11.5 Hz, benzyl methylene), 4.72 (d, 1 H, $J_{1,2}$ 1.0 Hz, H-1), 6.52 (d, 1 H, $J_{2,NH}$ 8.0 Hz, NH), and 7.23 (s, 5 H, Ph).

Anal. Calc. for $C_{21}H_{29}NO_8$: C, 59.56; H, 6.90; N, 3.31. Found: C, 59.35; H, 6.78; N, 3.27.

*Benzyl 2-acetamido-2-deoxy-5,6-*O*-isopropylidene-3-*O*-(*D*-2-propanoyl-*L*-alanyl-*

D-*isoglutamine benzyl ester*)-α-D-*mannofuranoside* (**27**). — Coupling of **26** (1.88 g) with the dipeptide (1.84 g) in dry 1,4-dioxane (25 mL) in the presence of HOSu (510 mg), DCC (1.1 g), and triethylamine (480 mg), as already described, gave the product; yield 2.71 g (86%), m.p. 151° (dec.), $[\alpha]_D^{20}$ +35° (*c* 1.0, chloroform); ν_{max}^{Nujol} 3300 and 3240 (NH), 1720 (C=O), 1660, 1630, and 1525 (amide), 850 (Me_2C), and 730 and 690 cm^{-1} (phenyl); n.m.r. data (in chloroform-*d*): δ 1.33 (d, 3 H, $J_{CH_3,CH}$ 7.0 Hz, MeCH), 1.38 and 1.45 (2 s, 6 H, Me_2C), 1.95 (s, 3 H, AcN), 4.27 and 4.55 (2 d, 2 H, J_{gem} 12.0 Hz, benzyl methylene), 4.70 (s, 1 H, H-1), 5.02 (s, 2 H, CO_2CH_2Ph), 6.05, 6.78, and 8.42 (3 d, 3 H, 3 NH), and 7.24 (s, 10 H, 2 Ph).

Anal. Calc. for $C_{36}H_{48}N_4O_{11}$: C, 60.66; H, 6.79; N, 7.86. Found: C, 60.53; H, 6.81; N, 7.79.

*Benzyl 2-acetamido-3-*O*-benzoyl-2-deoxy-5,6-*O*-isopropylidene-β-*D*-glucofuranoside* (**29**). — To a solution of benzyl 2-acetamido-2-deoxy-5,6-*O*-isopropylidene-β-D-glucofuranoside[17] (**28**; 3.0 g) in dry pyridine (5 mL) was added benzoyl chloride (1.5 g) at 0°. The mixture was kept for 1.5 h at 0°, and extracted with chloroform. The extract was successively washed with 2M hydrochloric acid, M sodium carbonate, and water, dried (sodium sulfate), and evaporated. The product, purified by chromatography on a column of silica gel (60 g) with 100:1 chloroform–methanol, was obtained as a syrup: yield 3.9 g (quantitative yield), $[\alpha]_D^{20}$ −35° (*c* 0.83, chloroform); ν_{max}^{film} 3260 (NH), 1730 and 1270 (ester), 1650 and 1540 (amide), 850 (Me_2C), and 750 and 700 cm^{-1} (phenyl).

Anal. Calc. for $C_{25}H_{29}NO_7$: C, 65.92; H, 6.42; N, 3.08. Found: C, 66.20; H, 6.69; N, 2.81.

*Benzyl 2-acetamido-3-*O*-benzoyl-2-deoxy-β-*D*-glucofuranoside* (**30**). — A solution of **29** (4.2 g) in 60% aqueous acetic acid (70 mL) was kept for 1.5 h at 50°, and then evaporated to a syrup which crystallized from ether–hexane to give **30** (3.45 g, 90%), m.p. 104°, $[\alpha]_D^{20}$ −31° (*c* 1.0, chloroform); ν_{max}^{Nujol} 3430 and 3240 (OH, NH), 1710 and 1265 (ester), 1670 and 1530 (amide), and 750 and 700 cm^{-1} (phenyl); n.m.r. data (in 1:1 chloroform-*d*–methanol-d_4): δ 1.99 (s, 3 H, AcN), 4.51 and 4.76 (2 d, 2 H, J_{gem} 12.0 Hz, benzyl methylene), 5.10 (s, 1 H, H-1), 5.54 (d, 1 H, $J_{2,3}$ 5.0 Hz, H-3), 7.28 (s, 5 H, Ph), and 7.20–7.98 (m, 5 H, $PhCO_2$).

Anal. Calc. for $C_{22}H_{25}NO_7$: C, 63.60; H, 6.07; N, 3.37. Found: C, 63.59; H, 6.05; N, 3.41.

*Benzyl 2-acetamido-3,6-di-*O*-benzoyl-2-deoxy-β-*D*-glucofuranoside* (**31**). — To a stirred solution of **30** (2.0 g) in dry pyridine (5 mL) was added benzoyl chloride (1.12 g) at −20°. After being kept for 5 h at −10°, the mixture was processed as described for the preparation of **29**. The product was chromatographed on a column of silica gel (40 g) with chloroform, and then 100:1 chloroform–methanol; the latter eluate afforded **31** (2.1 g, 84%) as needles, m.p. 193°, $[\alpha]_D^{20}$ −24° (*c* 1.0, chloroform); ν_{max}^{Nujol} 3340 and 3320 (OH, NH), 1715, 1695, and 1260 (ester), 1650 and 1510 (amide), and 740 and 700 cm^{-1} (phenyl); n.m.r. data (in 1:1 chloroform-*d*–methanol-d_4): δ 1.99 (s, 3 H, AcN), 4.49 and 4.67 (2 d, 2 H, J_{gem} 11.5 Hz, benzyl methylene),

4.52 (d, 1 H, $J_{2,3}$ 5.0 Hz, H-2), 5.13 (s, 1 H, H-1), 5.60 (dd, 1 H, $J_{2,3}$ 5.0, $J_{3,4}$ 1.2 Hz, H-3), 7.29 (s, 5 H, Ph), and 7.20–8.08 (m, 10 H, 2 $PhCO_2$).

Anal. Calc. for $C_{29}H_{29}NO_8$: C, 67.04; H, 5.63; N, 2.70. Found: C, 67.21; H, 5.38; N, 2.59.

*Benzyl 2-acetamido-3,6-di-*O*-benzoyl-2-deoxy-5-*O*-mesyl-β-*D*-glucofuranoside* (**32**). — To an ice-cooled solution of **31** (1.0 g) in pyridine (2.5 mL) was added methanesulfonyl chloride (340 mg). The mixture was kept for 4 h at room temperature, and then processed in the usual way, to give **32** (1.05 g, 91%) as needles, m.p. 131°, $[\alpha]_D^{20}$ −39° (*c* 1.0, chloroform); ν_{max}^{Nujol} 3320 (NH), 1740, 1730, and 1260 (ester), 1670 and 1540 (amide), and 730 and 705 cm^{-1} (phenyl); n.m.r. data (in chloroform-*d*): δ 1.96 (s, 3 H, AcN), 4.54 and 4.78 (2 d, 2 H, J_{gem} 11.5 Hz, benzyl methylene), 5.22 (s, 1 H, H-1), 5.23–5.42 (m, 1 H, H-5), 5.67 (near d, 1 H, $J_{3,4}$ 3.0 Hz, H-3), 6.62 (d, 1 H, $J_{2,NH}$ 6.5 Hz, NH), 7.28 (s, 5 H, Ph), and 7.20–8.03 (m, 10 H, 2 $PhCO_2$).

Anal. Calc. for $C_{30}H_{31}NO_{10}S$: C, 60.29; H, 5.23; N, 2.34. Found: C, 60.35; H, 5.19; N, 2.35.

*Benzyl 2-acetamido-5,6-anhydro-2-deoxy-α-*L*-idofuranoside* (**33**). — To a solution of **32** (600 mg) in dry chloroform (6 mL), cooled to −15°, was added, with stirring, an ice-cooled solution of freshly prepared sodium methoxide in methanol (35 mg of sodium in 1.5 mL of methanol). The mixture was kept for 6 h at −5 to −10°, while the progress of the reaction was monitored by t.l.c., and then treated with Amberlite IR-120 (H^+) resin, to remove the base. The solution was evaporated to a syrup which was chromatographed on a column of silica gel (15 g) with (*a*) chloroform, (*b*) 50:1, and (*c*) 30:1 chloroform–methanol. Eluant (*c*) gave compound **33** (240 mg, 82%) as needles, m.p. 109°, $[\alpha]_D^{20}$ −82° (*c* 1.0, chloroform); ν_{max}^{Nujol} 3480 (OH), 3220 (NH), 1640 and 1560 (amide), and 750 and 690 cm^{-1} (phenyl); n.m.r. data (in chloroform-*d*): δ 1.93 (s, 3 H, AcN), 2.60–2.86 (m, 2 H, H-6,6'), 3.18–3.31 (m, 1 H, H-5), 3.90–4.34 (m, 3 H, H-2,3,4), 4.47 and 4.70 (2 d, 2 H, J_{gem} 11.5 Hz, benzyl methylene), 5.02 (s, 1 H, H-1), 6.93 (d, 1 H, $J_{2,NH}$ 6.8 Hz, NH), and 7.29 (s, 5 H, Ph).

Anal. Calc. for $C_{15}H_{19}NO_5$: C, 61.42; H, 6.53; N, 4.78. Found: C, 61.35; H, 6.29; N, 4.78.

*Benzyl 2-acetamido-5,6-anhydro-2-deoxy-3-*O*-(tetrahydropyran-2-yl)-α-*L*-idofuranoside* (**34**). — To a stirred solution of **33** (400 mg) in dry 1,4-dioxane (3 mL) were added dihydropyran (350 mg) and *p*-toluenesulfonic acid monohydrate (5 mg). The mixture was stirred for 2 h at room temperature, treated with Amberlite IR-45 (OH^-) resin to remove the acid, and evaporated to a syrup which was chromatographed on a column of silica gel (15 g) with chloroform, and then 70:1 chloroform–methanol. The latter eluant gave compound **34** (510 mg, quantitative) as a syrup; ν_{max}^{film} 3230 (NH), 1645 and 1540 (amide), and 740 and 690 cm^{-1} (phenyl).

Anal. Calc. for $C_{20}H_{27}NO_6$: C, 63.64; H, 7.21; N, 3.71. Found: C, 63.35; H, 7.47; N, 3.50.

*Benzyl 2-acetamido-2-deoxy-3-*O*-(tetrahydropyran-2-yl)-α-*L*-idofuranoside* (**35**). — A solution of **34** (590 mg) in 1,4-dioxane (2 mL) and 2M sodium hydroxide (10 mL)

was heated, with stirring, for 12 h at 45–50°, cooled, and then treated with Amberlite IRC-50 (H^+) ion-exchange resin to remove the base. The resin was filtered off, and washed successively with water and methanol, and the combined filtrate and washings were evaporated to a syrup which was purified by chromatography on a column of silica gel (15 g) with chloroform, and 20:1 chloroform–methanol. The latter eluant gave compound **35** (370 mg, 61%) as a syrup; ν_{max}^{film} 3400–3230 (OH, NH), 1650 and 1540 (amide), and 745 and 690 cm^{-1} (phenyl).

Anal. Calc. for $C_{20}H_{29}NO_7$: C, 60.74; H, 7.39; N, 3.54. Found: C, 60.50; H, 7.63; N, 3.45.

*Benzyl 2-acetamido-2-deoxy-α-*L*-idofuranoside* (**36**). — A solution of **35** (370 mg) in 70% aqueous acetic acid (3 mL) was heated for 2 h at 45–50°, and then evaporated to a syrup which was chromatographed on a column of silica gel (10 g) with chloroform, and then 10:1 chloroform–methanol. From the latter eluate, compound **36** was obtained as needles (220 mg, 76%) by recrystallization from ethanol–ether, m.p. 138°, $[\alpha]_D^{25}$ −74° (*c* 0.5, methanol); ν_{max}^{film} 3330–3240(OH, NH), 1650 and 1550 (amide), and 750 and 695 cm^{-1} (phenyl).

Anal. Calc. for $C_{15}H_{21}NO_6$: C, 57.86; H, 6.86; N, 4.50. Found: C, 57.79; H, 6.78; N, 4.41.

*Benzyl 2-acetamido-2-deoxy-5,6-*O*-isopropylidene-α-*L*-idofuranoside* (**37**). — To a stirred solution of **36** (95 mg) in *N,N*-dimethylformamide (2 mL) were added 2,2-dimethoxypropane (0.2 mL) and *p*-toluenesulfonic acid monohydrate (3 mg). The mixture was stirred for 2 h at room temperature, and then treated with Amberlite IRA-410 (OH^-) resin. The solution was processed in the usual way, to give the 5,6-*O*-isopropylidene derivative **37** (87 mg, 81%) as a syrup, $[\alpha]_D^{23}$ −62° (*c* 0.5, chloroform); ν_{max}^{film} 3400 and 3230 (OH, NH), 1640 and 1535 (amide), 840 (Me_2C), and 730 and 680 cm^{-1} (phenyl); n.m.r. data (in chloroform-*d*): δ 1.39 and 1.46 (2 s, 6 H, Me_2C), 1.96 (s, 3 H, AcN), 4.50 and 4.86 (2 d, 2 H, J_{gem} 12.0 Hz, benzyl methylene), 5.01 (s, 1 H, H-1), 6.05 (d, 1 H, $J_{2,NH}$ 9.0 Hz, NH), and 7.27 (s, 5 H, Ph).

Anal. Calc. for $C_{18}H_{25}NO_6$: C, 61.52; H, 7.17; N, 3.99. Found: C, 61.40; H, 7.35; N, 3.78.

*Benzyl 2-acetamido-3-*O*-(*D*-1-carboxyethyl)-2-deoxy-5,6-*O*-isopropylidene-α-*L*-idofuranoside* (**38**). — To a stirred solution of **37** (80 mg) in dry 1,4-dioxane (1 mL) was added the sodium hydride reagent (24 mg), and the mixture was stirred for 30 min at 95°, and then cooled to 65°. L-2-Chloropropanoic acid (40 mg) was added to the mixture, which, after being kept for 3 h at 65°, was processed as described before, to give **38** (59 mg, 61%) as a syrup, $[\alpha]_D^{20}$ −81° (*c* 0.6, chloroform); ν_{max}^{film} 3250 (NH), 2830–2700 (COOH), 1750 (C=O), 1660 and 1550 (amide), 840 (Me_2C), and 740 and 700 cm^{-1} (phenyl); n.m.r. data (in chloroform-*d*): δ 1.41 (d, 3 H, $J_{CH_3,CH}$ 6.4 Hz, MeCH), 1.41 and 1.46 (2 s, 6 H, Me_2C), 1.96 (s, 3 H, AcN), 4.32 (q, 1 H, $J_{CH_3,CH}$ 6.4 Hz, MeCH), 4.62 and 4.87 (2 d, 2 H, J_{gem} 12.0 Hz, benzyl methylene), 5.09 (s, 1 H, H-1), 6.16 (d, 1 H, $J_{2,NH}$ 5.0 Hz, NH), and 7.30 (s, 5 H, Ph).

Anal. Calc. for $C_{21}H_{29}NO_8$: C, 59.56; H, 6.90; N, 3.31. Found: C, 59.41; H, 7.22; N, 3.15.

Benzyl 2-acetamido-2-deoxy-5,6-O-isopropylidene-3-O-(D-2-propanoyl-L-alanyl-D-isoglutamine benzyl ester)-α-L-idofuranoside (**39**). — Coupling of **38** (50 mg) with the dipeptide (65 mg) in dry 1,4-dioxane (2 mL) in the presence of HOSu (25 mg), DCC (43 mg), and triethylamine (17 mg), as described for the preparation of **7**, yielded amorphous **39** (64 mg, 76%), m.p. 71–73° (dec.), $[\alpha]_D^{20}$ −37° (*c* 0.4, chloroform); ν_{max}^{KBr} 3280 (NH), 1730 (C=O), 1640 and 1520 (amide), and 730 and 690 cm^{-1} (phenyl); n.m.r. data (in 1:1 chloroform-*d*–methanol-d_4): δ 1.12 and 1.34 (2 d, 6 H, $J_{CH_3,CH}$ 7.0 Hz, 2 MeCH), 1.42 and 1.49 (2 s, 6 H, Me_2C), 1.94 (s, 3 H, AcN), 4.70 and 4.91 (2 d, 2 H, J_{gem} 12.0 Hz, benzyl methylene), 5.07 (s, 1 H, H-1), 5.10 (s, 2 H, CO_2CH_2Ph), and 7.31 (s, 10 H, 2 Ph).

Anal. Calc. for $C_{36}H_{48}N_4O_{11}$: C, 60.66; H, 6.79; N, 7.86. Found: C, 60.48; H, 6.92; N, 7.88.

2-Acetamido-2-deoxy-3-O-(D-2-propanoyl-L-alanyl-D-isoglutamine)-D-allopyranose (**40**). — A solution of **15** (300 mg) in 50% aqueous acetic acid (3 mL) was heated for 30 min at 45°, and then evaporated at 40° to a syrup. The residue was dissolved in methanol (10 mL), water (10 mL), and acetic acid (1 mL), and 10% Pd–C catalyst (300 mg) was added; hydrogen was bubbled through while the mixture was stirred at 40°, and, after 3 h, t.l.c. showed the hydrogenation to be complete. The catalyst was removed by filtration, and the filtrate was evaporated below 30°, to give a hygroscopic, amorphous mass (205 mg; quantitative yield), which showed a single spot in t.l.c., $[\alpha]_D^{20}$ −26.5° (*c* 0.5, methanol; equil.); ν_{max}^{KBr} 3370 (OH, NH), 2700–2650 (COOH), 1720 (C=O), and 1600, 1545, and 1530 cm^{-1} (amide); n.m.r. data (in D_2O): δ 1.29–1.41 (m, 6 H, 2 MeCH), 2.00, 2.02, 2.04, and 2.06 (4 s, 3 H, AcN), 4.97 (d, $J_{1,2}$ 8.6 Hz, H-1β in the $^4C_1$ form), 5.03 (d, $J_{1,2}$ 3.8 Hz, H-1α in the $^1C_4$ form), 5.14 (d, $J_{1,2}$ 1.6 Hz, H-1β in the $^1C_4$ form), and 5.33 (d, $J_{1,2}$ 4.8 Hz, H-1α in the $^4C_1$ form).

Anal. Calc. for $C_{19}H_{32}N_4O_{11}$: C, 46.33; H, 6.55; N, 11.38. Found: C, 46.01; H, 6.88; N, 11.05.

2-Acetamido-2-deoxy-3-O-(D-2-propanoyl-L-alanyl-D-isoglutamine)-D-gulopyranose (**41**). — A solution of **18** (280 mg) in 50% aqueous acetic acid (10 mL) was heated for 2 h at 40°, and evaporated at 40° to a syrup which was purified by chromatography on a column of silica gel (10 g) with 100:1, and 20:1, chloroform–methanol. The latter eluate afforded benzyl 2-acetamido-2-deoxy-3-*O*-(D-2-propanoyl-L-alanyl-D-isoglutamine benzyl ester)-α-D-gulopyranoside (240 mg, 92%); $[\alpha]_D^{20}$ +36° (*c* 1.0, methanol); ν_{max}^{KBr} 3450–3200 (OH, NH) 1720 (C=O), 1650 and 1510 (amide), and 730 and 680 cm^{-1} (phenyl). The benzyl glycoside (150 mg) was hydrogenated in the presence of 10% Pd–C catalyst (150 mg), in methanol (10 mL), water (10 mL), and acetic acid (1 mL) according to the procedure already described, to give **41** as an amorphous mass, in quantitative yield; $[\alpha]_D^{20}$ −45° (*c* 0.5, methanol; equil.); ν_{max}^{KBr} 3400–3200 (OH, NH), 2800–2700 (COOH), 1720 (C=O), and 1650 and 1520 cm^{-1} (amide); n.m.r. data (in D_2O): δ 1.34 and 1.41 (2 d, 6 H, $J_{CH_3,CH}$ 7.0 Hz, 2 MeCH), 1.99 and 2.00 (2 s, 3 H, AcN), 4.89 (d, $J_{1,2}$ 8.9 Hz, H-1β), and 5.07

(d, $J_{1,2}$ 3.8 Hz, H-1α); anomeric ratio (α:β) estimated, ~1:2 (from the ratio of the intensity of the H-1α and H-1β signals, and of that of AcN).

Anal. Calc. for $C_{19}H_{32}N_4O_{11}$: C, 46.33; H, 6.55; N, 11.38. Found: C, 46.05; H, 6.79; N, 10.98.

*2-Acetamido-2-deoxy-3-*O*-(*D*-2-propanoyl-*L*-alanyl-*D*-isoglutamine)-*D*-galactopyranose* (**42**). — Hydrogenation of **23** (90 mg), in the presence of 10% Pd–C catalyst (90 mg), in methanol (10 mL), water (10 mL), and acetic acid (1 mL), as already described, gave **42** (65 mg, quantitative) as an amorphous mass; $[\alpha]_D^{25}$ +41° (*c* 0.35, methanol; equil.); ν_{max}^{KBr} 3300 (OH, NH), 2800–2700 (COOH), 1720 (C=O), and 1640 and 1530 cm^{-1} (amide); n.m.r. data (in D_2O): δ 1.27 and 1.40 (2 d, 6 H, $J_{CH_3,CH}$ 7.0 Hz, 2 MeCH), 1.96 (s, 3 H, AcN), and 5.14 (d, $J_{1,2}$ 3.7 Hz, H-1α).

Anal. Calc. for $C_{19}H_{32}N_4O_{11}$: C, 46.33; H, 6.55; N, 11.38. Found: C, 45.99; H, 6.81; N, 11.03.

*2-Acetamido-2-deoxy-3-*O*-(*D*-2-propanoyl-*L*-alanyl-*D*-isoglutamine)-*D*-mannopyranose* (**43**). — A solution of **27** (2.5 g) in 60% aqueous acetic acid (50 mL) was heated for 1 h at 40–45°, and evaporated. The residue in methanol (20 mL), water (20 mL), and acetic acid (2 mL) was hydrogenated in the presence of 10% Pd–C catalyst (1.0 g) as described for the preparation of compound **40**, to give hygroscopic, amorphous **43** (1.7 g) in almost quantitative yield); $[\alpha]_D^{25}$ +37° (*c* 0.5, methanol; equil.); ν_{max}^{KBr} 3360–3250 (OH, NH), 2780–2700 (COOH), 1720 (C=O), and 1650 and 1530 cm^{-1} (amide); n.m.r. data (in D_2O): δ 1.37 and 1.46 (2 d, 6 H, $J_{CH_3,CH}$ 7.0 Hz, 2 MeCH), 2.05 and 2.08 (2 s, 3 H, AcN), 5.03 (d, $J_{1,2}$ 2.0 Hz, H-1β), and 5.18 (d, $J_{1,2}$ 1.5 Hz, H-1α).

Anal. Calc. for $C_{19}H_{32}N_4O_{11}$: C, 46.33; H, 6.55; N, 11.38. Found: C, 46.08; H, 6.77; N, 11.10.

*2-Acetamido-2-deoxy-3-*O*-(*D*-2-propanoyl-*L*-alanyl-*D*-isoglutamine)-*L*-idopyranose* (**44**). — The 5,6-*O*-isopropylidene group of compound **39** (64 mg) was hydrolyzed by heating with 60% aqueous acetic acid (3 mL) for 2 h at 45°. By the procedure described in the previous section, the benzyl groups of the product thus obtained were removed, to give the amorphous *L-ido* analog **44** (45 mg, quantitative yield); $[\alpha]_D^{25}$ +22.3° (*c* 0.3, water; equil.); ν_{max}^{KBr} 3350–3200 (OH, NH), 2800–2700 (COOH), 1720 (C=O), and 1670, 1640, and 1540 cm^{-1} (amide); n.m.r. data (in D_2O): δ 1.33–1.42 (m, 6 H, 2 MeCH), 1.95, 1.96, 1.97, and 1.98 (4 s, 3 H, AcN), 5.04 (s, H-1α in the $^1C_4$ form), and 5.11 (d, $J_{1,2}$ 2.2 Hz, H-1β, in the $^4C_1$ form).

Anal. Calc. for $C_{19}H_{32}N_4O_{11}$: C, 46.33; H, 6.55; N, 11.38. Found: C, 46.08; H, 6.78; N, 11.03.

*3-*O*-(*D*-1-Carboxyethyl)-1,2:5,6-di-*O*-isopropylidene-α-*D*-glucofuranose* (**46**). — To a stirred solution of 1,2:5,6-di-*O*-isopropylidene-α-D-glucofuranose (650 mg) in dry 1,4-dioxane (6.5 mL) was added the sodium hydride reagent (480 mg), and the mixture was stirred for 1.5 h at 95°, and then cooled to 65°. L-2-Chloropropanoic acid (400 mg) was added to the stirred mixture, which was then stirred for 3 h at 65°; by the procedure already described, the title compound was obtained as a syrup. Purification by chromatography on a column of silica gel (10 g), with 200:1 chloro-

form–methanol, gave an analytical sample (800 mg, 96%); $[\alpha]_D^{20}$ +2° (*c* 1.0, chloroform); ν_{max}^{film} 2630–2600 (COOH), 1740 (C=O), and 850 cm^{-1} (Me_2C); n.m.r. data (in chloroform-*d*): δ 1.31–1.49 (m, 15 H, 2 Me_2C, MeCH), 4.80 (d, 1 H, 3.8 Hz, H-2), 5.91 (d, 1 H, $J_{1,2}$ 3.8 Hz, H-1), and 9.19 (s, 1 H, COOH).

Anal. Calc. for $C_{15}H_{24}O_8$: C, 54.21; H, 7.28; Found: C, 54.15; H, 7.33.

1,2:5,6-Di-O-isopropylidene-3-O-(D-2-propanoyl-L-alanyl-D-isoglutamine benzyl ester)-α-D-glucofuranose (**47**). — Coupling of **46** (300 mg) with the dipeptide (376 mg) in dry 1,4-dioxane (4 mL) in the presence of HOSu (125 mg), DCC (220 mg), and triethylamine (90 mg), as described for the preparation of **7**, gave **47** (410 mg, 73%) as a syrup, $[\alpha]_D^{20}$ −9.6° (*c* 1.46, chloroform); ν_{max}^{film} 3280 (NH), 1720 and 1250 (ester), 1650 and 1510 (amide), 840 (Me_2C), and 750 and 690 cm^{-1} (phenyl); n.m.r. data (in chloroform-*d*): δ 1.26–1.44 (m, 18 H, 2 Me_2C, 2 MeCH), 4.45 (d, 1 H, $J_{1,2}$ 4.0 Hz, H-2), 5.07 (s, 2 H, CO_2CH_2Ph), 5.86 (d, 1 H, $J_{1,2}$ 4.0 Hz, H-1), and 7.26 (s, 5 H, Ph).

Anal. Calc. for $C_{30}H_{43}N_3O_{11}$: C, 57.96; H, 6.97; N, 6.76. Found: C, 57.85; H, 7.15; N, 6.58.

3-O-(D-1-Carboxyethyl)-1,2:5,6-di-O-isopropylidene-α-L-glucofuranose (**48**). — To a stirred solution of 1,2:5,6-di-*O*-isopropylidene-α-L-glucofuranose[19] (94 mg) in dry 1,4-dioxane (1.8 mL) was added the sodium hydride reagent (35 mg), and the mixture was stirred for 1.5 h at 95°, and then cooled to 65°. L-2-Chloropropanoic acid (42 mg) was added to the stirred mixture, which was then stirred for 2 h at 65°. The mixture was processed as already described, and the product was purified by chromatography on a column of silica gel (10 g) with 100:1 chloroform–methanol, to afford **48** (89 mg, 74%) as a syrup; $[\alpha]_D^{25}$ +31° (*c* 0.75, chloroform); ν_{max}^{film} 2700–2600 (COOH), 1740 (C=O), and 850 cm^{-1} (Me_2C); n.m.r. data (in chloroform-*d*): δ 1.32–1.53 (m, 15 H, 2 Me_2C, MeCH), 4.50 (d, 1 H, $J_{1,2}$ 4.0 Hz, H-2), 5.92 (d, 1 H, $J_{1,2}$ 4.0 Hz, H-1), and 8.15 (broad s, 1 H, COOH).

Anal. Calc. for $C_{15}H_{24}O_8$: C, 54.21; H, 7.28. Found: C, 54.00; H, 7.43.

1,2:5,6-Di-O-isopropylidene-3-O-(D-2-propanoyl-L-alanyl-D-isoglutamine benzyl ester)-α-L-glucofuranose (**49**). — Coupling of **48** (89 mg) with the dipeptide (124 mg) in dry 1,4-dioxane (2 mL) by using HOSu (46 mg), DCC (83 mg), and triethylamine (32 mg), as already described, gave **49** (122 mg, 74%) as a syrup, $[\alpha]_D^{20}$ +27.5° (*c* 0.6, chloroform); ν_{max}^{film} 3300 (NH), 1710 and 1250 (ester), 1660 and 1520 (amide), and 840 cm^{-1} (Me_2C); n.m.r. data (in chloroform-*d*): δ 1.31–1.47 (m, 18 H, 2 Me_2C, 2 MeCH), 4.53 (d, 1 H, $J_{1,2}$ 4.0 Hz, H-2), 5.09 (s, 2 H, benzyl methylene), 5.87 (d, 1 H, $J_{1,2}$ 4.0 Hz, H-1), 6.35 and 7.04 (broad s, 2 H, 2 NH), 7.55 (2 H, NH_2), and 7.28 (s, 5 H, Ph).

Anal. Calc. for $C_{30}H_{43}N_3O_{11}$: C, 57.96; H, 6.97; N, 6.76. Found: C, 57.88; H, 6.97; N, 6.58.

3-O-(D-2-Propanoyl-L-alanyl-D-isoglutamine benzyl ester)-D-glucopyranose (**50**). — A solution of **47** (145 mg) in 90% aqueous trifluoroacetic acid (1 mL) was stirred for 1 h at room temperature, and evaporated below 30° to a syrup which was chromatographed on a column of silica gel (5 g) with 10:1 chloroform–methanol, to

afford **50** (60 mg, 48%) as a syrup; $[\alpha]_D^{20}$ +48° (*c* 0.3, methanol; equil.); ν_{max}^{film} 3300–3240 (OH, NH), 1720 and 1250 (ester), 1660 and 1540 (amide), and 750 and 690 cm^{-1} (phenyl); n.m.r. data (in methanol-d_4): δ 1.40 and 1.42 (2 d, 6 H, J_{CH_3CH} 7.0 Hz, 2 MeCH), 5.12 (s, 2 H, benzyl methylene), and 7.32 (s, 5 H, Ph).

Anal. Calc. for $C_{24}H_{35}N_3O_{11}$: C, 53.23; H, 6.51; N, 7.76. Found: C, 53.01; H, 6.80; N, 7.55.

3-O-(D-*2-Propanoyl*-L-*alanyl*-D-*isoglutamine*)-D-*glucopyranose* (**51**). — Hydrogenation of **50** (60 mg) in ethanol (10 mL) in the presence of 10% Pd–C catalyst (30 mg) for 30 min at room temperature gave compound **51** (50 mg) as a syrup, in quantitative yield; $[\alpha]_D^{20}$ +61° (*c* 0.5, methanol; equil.); ν_{max}^{KBr} 3400–3250 (OH, NH), 2750–2700 (COOH), 1720 (C=O), and 1650 and 1520 cm^{-1} (amide).

Anal. Calc. for $C_{17}H_{29}N_3O_{11}$: C, 45.23; H, 6.48; N, 9.31. Found: C, 44.90; H, 6.73; N, 9.08.

3-O-(D-*2-Propanoyl*-L-*alanyl*-D-*isoglutamine benzyl ester*)-L-*glucopyranose* (**52**). — Hydrolysis of the isopropylidene groups in **49** (103 mg) with 90% aqueous trifluoroacetic acid (1 mL), as already described, gave **52** (49 mg, 55%) as a syrup, $[\alpha]_D^{20}$ +2° (*c* 0.4, methanol; equil.); ν_{max}^{KBr} 3350–3230 (OH, NH), 1710 and 1240 (ester), 1650 and 1520 (amide), and 730 and 680 cm^{-1} (phenyl); n.m.r. data (in methanol-d_4): δ 1.40 and 1.42 (2 d, 6 H, $J_{CH_3,CH}$ 7.0 Hz, 2 MeCH), 5.12 (s, 2 H, benzyl methylene), and 7.32 (s, 5 H, Ph).

Anal. Calc. for $C_{24}H_{35}N_3O_{11}$: C, 53.23; H, 6.51; N, 7.76. Found: C, 52.98; H, 6.86; N, 7.59.

3-O-(D-*2-Propanoyl*-L-*alanyl*-D-*isoglutamine*)-L-*glucopyranose* (**53**). — Compound **52** (30 mg) was dissolved in ethanol (2 mL), 10% Pd–C catalyst (20 mg) was added, and hydrogen was bubbled through for 30 min at room temperature. The catalyst was removed by filtration, and the filtrate was evaporated below 30°, to give hygroscopic, amorphous **53** (25 mg; quantitative), which showed a single spot in t.l.c., $[\alpha]_D^{20}$ 0° (*c* 0.2, methanol; equil.); ν_{max}^{KBr} 3350–3250 (OH, NH), 2750–2700 (COOH), 1720 (C=O), and 1650 and 1520 cm^{-1} (amide).

Anal. Calc. for $C_{17}H_{29}N_3O_{11}$: C, 45.23; H, 6.48; N, 9.31. Found: C, 44.89; H, 6.85; N, 9.08.

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