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A New Class of Nitrosoureas. VII.[1] Synthesis and Antitumor Activity of 3-Substituted 1-(2-Chloroethyl)-3-(methyl α-D-glucopyranosid-3-yl)-1-nitrosoureas

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A series of five 3,3-disubstituted nitrosoureas having the nitrosoureido group at the C-3 position of methyl glucoside were prepared and tested for antitumor activities. Heating of methyl 2,3-anhydro-α-D-allopyranoside (I) with various alkylamines followed by reaction with 2-chloroethyl isocyanate gave two regioisomers (II and III). The major product (II) and the minor product (III) were determined to be the ureido derivatives of methyl glucoside and methyl altroside, respectively. Nitrosation of II with dinitrogen tetroxide gave 3-substituted 1-(2-chloroethyl)-3-(methyl α-D-glucopyranosid-3-yl)-1-nitrosoureas (VI) in good yields. All the nitrosoureas obtained were remarkably active against leukemia L-1210 and Ehrlich ascites carcinoma. The structure–activity relationships of positional isomers with respect to the nitrosoureido group are discussed.

Keywords——chloroethyl nitrosoureas; 3,3-disubstituted nitrosoureas; methyl glucoside derivatives; antitumor activities; leukemia L1210; GANU; DCNU

In previous papers of this series,[2] we reported the synthesis and potent antitumor activity of various kinds of sugar derivatives of 3,3-disubstituted 1-(2-chloroethyl)-1-nitrosoureas in which the nitrosoureido group is attached to the C-1 position of the sugar moieties. The nitrosoureas possessing a nitrosoureido group at the C-2 position of methy glucoside were also synthesized, and the structure–activity relationships (SAR) of these positional isomers were discussed in the preceding paper.[1] Our studies on the SAR of the positional isomers with respect to the nitrosoureido group have now been extended to the synthesis of the nitrosoureas possessing the nitrosoureido group at the C-3 position of methyl glucoside. We describe herein the preparation and the potent antitumor activity of 3-substituted 1-(2-chloroethyl)-3-(methyl α-D-glucopyranosid-3-yl)-1-nitrosoureas.

Synthesis of Nitrosoureas and Discussion

The nitrosoureas (VIa—e) were prepared *via* the sequence outlined in Chart 1.

Little work has been done on the reaction of methyl 2,3-anhydro-α-D-allopyranoside[3] (I) with amines, though the possible formation of two regioisomers (C_2 or C_3 attack) is an interesting problem. The reaction of I with ammonia[4] has been reported to give only the amino compound of methyl altroside (C_2 attack), while the reaction with dimethylamine[5] gave the dimethylamino derivatives of methyl altroside and methyl glucoside (C_2 and C_3 attack) in an equal ratio. Therefore, the reaction of I with various alkylamines was anticipated to give a mixture of methyl altroside and methyl glucoside derivatives.

Various alkylamines were heated with I and the crude products were allowed to react with 2-chloroethyl isocyanate, because the isolation of the amino derivatives in a pure form was difficult. The resulting urea derivatives (II and III) could be purified by column chromatography. Thus, in a typical procedure, a mixture of I and *n*-butylamine was heated in a sealed tube at 150°C for 20 h then concentrated to dryness. The residue was allowed to react with 2-chloroethyl isocyanate in methanol to give a mixture of the ureas, and its thin-layer chromatography (TLC) showed two spots in a ratio of one to eight (Rf=0.51 and 0.62). The mixture was chromatographed on silica gel to give two compounds, which were

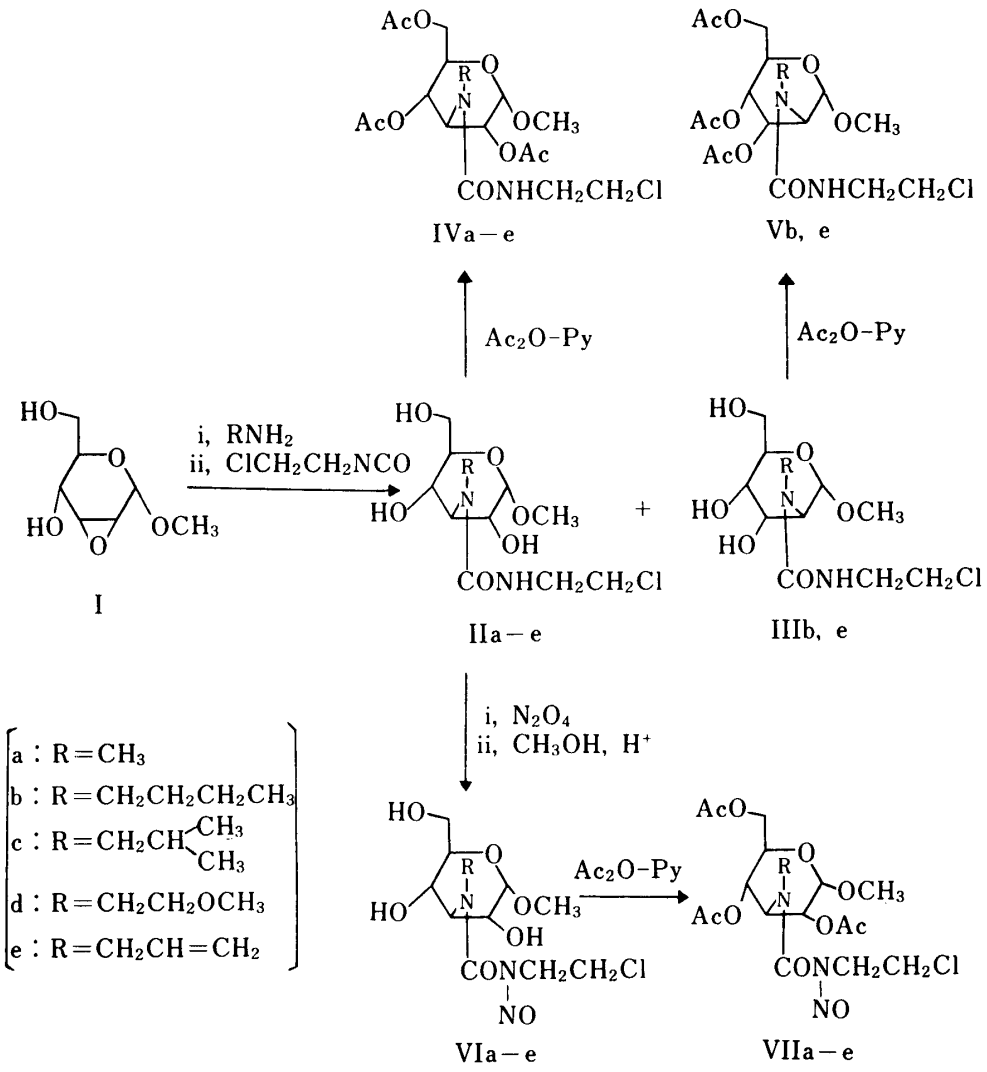


Chart 1

considered to be the urea derivatives of methyl altroside and mehtyl glucoside. The major product (IIb, Rf=0.62, $[\alpha]_D$ +106.5°) and the minor product (IIIb, Rf=0.51, $[\alpha]_D$ +29.1°) showed similar infrared (IR) and nuclear magnetic resonance (NMR) signals. They were acetylated to give the corresponding triacetates (IVb and Vb) which also gave similar spectroscopic data (see "Experimental"). However, the two isomers (IIb and IIIb) were found to differ greatly in their reaction with sodium metaperiodate. The minor product (IIIb) was oxidized rapidly with aqueous sodium periodate at room temperature, while the major product (IIb) remained unchanged under the same conditions. This seems to indicate that the minor product (IIIb) is the urea compound of methyl altropyranoside, possessing vicinal *cis* dihydroxyl groups, and the major product (IIb) is the urea of methyl glucoside. The reaction of I with other alkylamines followed by treatment with 2-chloroethyl isocyanate gave corresponding ureas (IIa, c—e and IIIe, respectively). Among these ureas, only IIIe, which was obtained as a minor product from the reaction of I with allylamine followed by treatment with 2-chloroethyl isocyanate, was readily oxidized with sodium metaperiodate, while the other ureas (IIa, c—e), obtained as major products, resisted the oxidation. These results seem to indicate that the reaction of I with alkylamines produces derivatives of glucopyranoside (II) (C_3 attack) predominantly, together with a small amount of those of altropyranoside (III) (C_2 attack). These results are considerably different from the reported

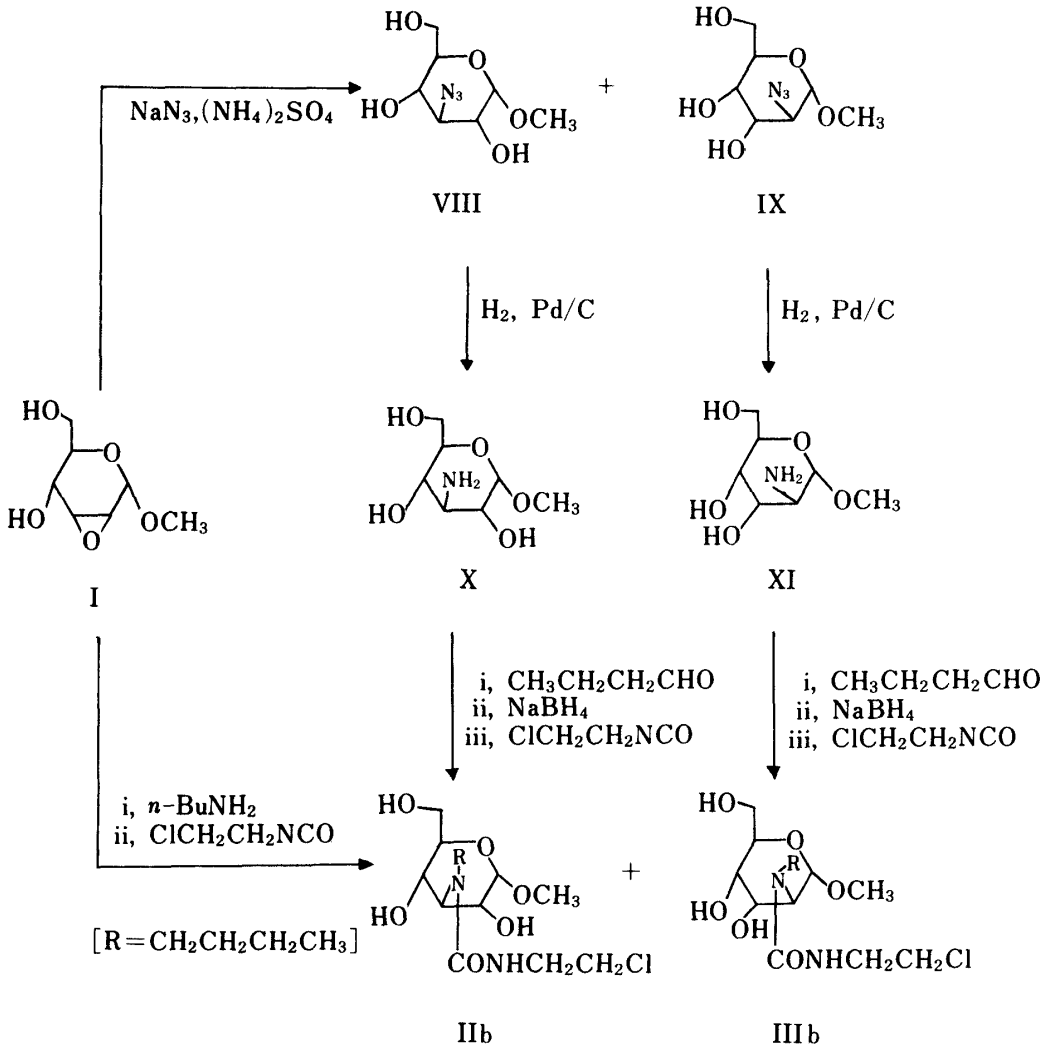


Chart 2

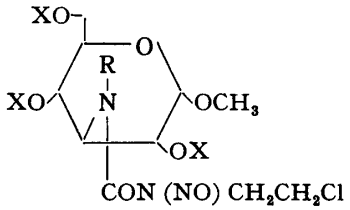
observation in the reaction of I with ammonia, in which the amino derivative of altropyranoside (C_2 attack) was the sole product. Therefore, to establish the structure of the isomeric ureas definitively, IIb and IIIb were synthesized by the sequences outlined in Chart 2.

The reaction of I with sodium azide gave two compounds in a ratio of one to two, and these were separated by column chromatography. The major product (VIII, mp 125—126°C, lit.,[6] mp 125—127°C) and the minor product (IX, mp 136—137°C, lit.,[7] mp 138—139°C) were determined to be methyl 3-azido-3-deoxy-α-D-glucopyranoside and methyl 2-azido-2-deoxy-α-D-altropyranoside, respectively. These azides were hydrogenated to give methyl 3-amino-3-deoxy-α-D-glucopyranoside (X, $[\alpha]_D$ +150.7°, lit.,[8] $[\alpha]_D$ +144.4°) and methyl 2-amino–2–deoxy-α-D-altropyranoside (XI·hydrochloride, $[\alpha]_D$ +40.8°, lit.,[4] $[\alpha]_D$ +39.7°), respectively. The amines (X and XI) were allowed to react with *n*-butyraldehyde to give the Schiff bases, which were reduced to the *n*-butylamino derivatives with $NaBH_4$ and then converted to the urea derivatives by treatment with 2-chloroethyl isocyanate in the same manner as described in the preceding paper.[1]

The urea having a glucopyranoside moiety was obtained from X in 38% yield and proved to be identical with IIb on the basis of its physical properties. The urea similarly obtained from XI in 42% yield proved to be identical with IIIb. Accordingly, IIb and IIIb were determined to be 3-*n*-butyl-1-(2-chloroethyl)-3-(methyl α-D-glucopyranosid-3-yl)urea, and 3-*n*-butyl-1-(2-chloroethyl)-3-(methyl α-D-altropyranosid-2-yl)urea, respectively.

Thus, five 3-substituted 1-(2-chloroethyl)-3-(methyl α-D-glucopyranosid-3-yl)ureas (IIa—e) were obtained by the reaction of I with various alkylamines followed by treatment with 2-chloroethyl isocyanate. They are unstable amorphous powders and have no definite melting points. Acetylation of IIa—e gave the corresponding triacetates (IVa—e).

TABLE I. Properties of 3-Substituted 1-(2-Chloroethyl)-3-(methyl α-D-glucopyranosid-3-yl)-1-nitrosoureas[a] and Their Acetates



Compound No.	R	X	mp (°C)	$[\alpha]_D$ (c, °C) in methanol	Yield (%)
VIa	CH_3	H	63	+92.3° (1.1, 17)	66
VIb	$CH_2CH_2CH_2CH_3$	H	50	+90.7° (1.1, 25)	71
VIc	$CH_2CH(CH_3)_2$	H	53	+84.2° (1.2, 18)	65
VId	$CH_2CH_2OCH_3$	H	55	+105.6° (1.0, 18)	67
VIe	$CH_2CH=CH_2$	H	52	+90.4° (1.3, 18)	63
VIIa	CH_3	$COCH_3$	Caramel	Not measured	73
VIIb	$CH_2CH_2CH_2CH_3$	$COCH_3$	Caramel	Not measured	75
VIIc	$CH_2CH(CH_3)_2$	$COCH_3$	Caramel	Not measured	72
VIId	$CH_2CH_2OCH_3$	$COCH_3$	Caramel	Not measured	70
VIIe	$CH_2CH=CH_2$	$COCH_3$	Caramel	Not measured	65

Compound No.	IR ν_{max} (cm^{-1})[b]	NMR δ (ppm) (J=Hz)[c]
VIa	3350 (br, OH), 1695 (CO) 1080, 1030 (–O–)	2.98 (3H, s, NCH_3), 3.33 (3H, s, OCH_3) 4.62 (1H, d, J=3, H-1)
VIb	3450 (br, OH), 1680 (CO) 1070, 1040 (–O–)	0.85—1.6 (7H, m, CH_2 CH_2CH_3), 3.32 (3H, s, OCH_3) 4.62 (1H, d, J=3, H-1)
VIc	3440 (br, OH), 1690 (CO) 1070, 1040, 1010 (–O–)	0.89 (6H, d, J=6.5, $CH(CH_3)_2$), 3.30 (3H, s, OCH_3), 4.64 (1H, d, J=2.5, H-1)
VId	3440 (br, OH), 1690(CO) 1040, 1010 (–O–)	3.23 (3H, s, OCH_3), 3.31 (3H, s, OCH_3) 4.65 (1H, d, J=2.5, H-1)
VIe	3450 (br, OH), 1690 (CO) 1050, 1010 (–O–)	3.33 (3H, s, OCH_3), 4.64 (1H, d, J=3, H-1)
VIIa	1740, 1700 (CO), 1230 1030 (–O–)	2.05 (3H, s, OAc), 2.11 (3H, s, OAc), 2.14 (3H, s, OAc) 2.99 (3H, s, N–CH_3), 3.44 (3H, s, OCH_3), 4.98 (1H, d, J=3, H-1)
VIIb	1750, 1690 (CO), 1230 1030 (–O–)	0.90—1.8 (7H, m, $CH_2CH_2CH_3$), 2.02 (3H, s, OAc), 2.05 (3H, s, OAc), 2.08 (3H, s, OAc), 3.43 (3H, s, OCH_3) 4.99 (1H, d, J=3.7, H-1)
VIIc	1745, 1690 (CO), 1230 1035 (–O–)	0.89 (6H, d, J=6.6, $CH(CH_3)_2$), 2.03 (3H, s, OAc), 2.08 (6H, s, OAc×2), 3.42 (3H, s, OCH_3), 5.04 (1H, d, J=3.3, H-1)
VIId	1750, 1690 (CO), 1235, 1040 (–O–)	2.03 (3H, s, OAc), 2.09 (6H, s, OAc×2), 3.33 (3H, s, OCH_3) 3.43 (3H, s, OCH_3), 5.04 (1H, d, J=3, H-1)
VIIe	1750, 1695 (CO), 1235, 1040 (–O–)	2.03 (3H, s, OAc), 2.09 (6H, s, OAc×2), 3.43 (3H, s, OCH_3), 5.05 (1H, s, J=3, H-1)

a) Elemental analyses of these compounds gave unsatisfactory results since these compounds are unstable amorphous powders including solvents (especially ethyl acetate) tightly.
b) Measured in Nujol mull (compounds VIa—e) or in $CHCl_3$ solution (compounds VIIa—e).
c) Measured in d_6-DMSO–D_2O (compounds VIa—e) or in $CDCl_3$ (compounds VIIa—e).

The nitrosation of the ureas was carried out by the use of dinitrogen tetroxide as described in our previous paper.[2] Thus, four equivalents of dinitrogen tetroxide was introduced into a mixture of the urea (IId) and anhydrous sodium acetate in tetrahydrofuran, and then methanol was added to decompose the nitrous ester groups in the glucoside moiety. After purification by silica gel chromatography, the nitrosourea (VId) was obtained in 67% yield as a yellow amorphous powder. It showed the IR signal due to the nitrosoureido group at 1690 cm^{-1} and NMR signals due to the two methoxyl protons at δ 3.23 and 3.31 and the equatorial anomeric proton at δ 4.65. Acetylation of VId gave the triacetate (VIId), which showed IR signals at 1750 ($OCOCH_3$) and 1690 (NCON) and NMR signals of the three acetoxyl protons at δ 2.03 and 2.09 in addition to the two methoxyl (δ 3.33 and 3.43) and the anomeric proton (δ 5.04) signals. Thus, VId was determined to be 1-(2-chloroethyl)-3-(2-methoxyethyl)-3-(methyl α-D-glucopyranosid-3-yl)-1-nitrosourea. The physical properties of the nitrosoureas (VIa—e) thus obtained are listed in Table I.

Antitumor Activities of Nitrosoureas and Discussion

The nitrosoureas (VIa—e) were tested for their antitumor activities against leukemia L1210 and Ehrlich ascites carcinoma by the methods described in the previous paper.[2] The results are summarized in Table II together with the comparative data for positive controls, GANU and DCNU. The data for the positional isomers (XIIa—d)[1] in which the nitrosoureido group is attached to the C-2 position of the glucoside moiety are also included for comparison.

All the nitrosoureas in which the nitrosoureido group is attached to the C-3 position of the glucoside moiety were remarkably active against both leukemia L1210 and Ehrlich ascites carcinoma and showed greater therapeutic ratios than the two positive controls. Sixty-day survivors against leukemia L1210 were found at the optimal dose for these nitrosoureas

Table II. Antitumor Activities of Nitrosoureas

Compd. No.	Anti-L1210 activity[a]				Anti-Ehrlich activity[b]		
	ILS_{30}[c] (mg/kg/d)	OD[d] (mg/kg/d)	ILS_{max} (%)	Therapeutic[e] ratio	MED[f] (mg/kg/d)	MTD[g] (mg/kg/d)	Therapeutic[h] ratio
GANU	0.80	6.25	>198.6	7.8	0.39	12.5	32
DCNU	1.4	12.5	>240.0	8.9	0.78	12.5	16
VIa	0.26	12.5	>614.3[i]	48.1	0.39	25	64
VIb	0.65	25	>745.1[i]	38.5	0.78	50	64
VIc	0.67	12.5	>597.7[i]	18.7	0.195	25	128
VId	0.75	25	>344.0	33.3	0.195	25	128
VIe	0.39	12.5	>597.7[i]	32.1	0.195	25	128
XIIa[j]	1.9	50	>700.0[i]	26.3	0.78	100	128
XIIb[k]	1.5	50	>745.1[i]	33.3	1.56	100	64
XIIc[l]	1.4	50	>689.5[i]	35.7	0.78	50	64
XIId[m]	0.45	25	>650.0[i]	55.6	0.39	50	128

a) Anti-leukemic activity on L1210 in mice; leukemic cells (10^6) were inoculated *i.p.* into male BDF_1 mice and *i.p.* administration was begun 24 h after the inoculation and performed once daily for 5 d.
b) Growth-inhibitory effect on Ehrlich ascites tumor cells in mice; the ascites cells (10^6) were inoculated *i.p.* into female ICR mice and *i.p.* administration was begun 24 h after the inoculation and performed once daily for 5 d.
c) Daily dose providing 30% increase in life-span over the control. $ILS(\%)=(T/C-1)\times 100$
d) Optimal dose: the daily dose providing the maximum increase in life-span.
e) Therapeutic ratio=OD/ILS_{30}
f) Minimum effective dose: the minimum dose which shows 100% inhibition of the growth of the tumor.
g) Maximum tolerated dose: the maximum dose which shows 100% inhibition of the growth of the tumor without causing the death of mice.
h) Therapeutic ratio=MTD/MED.
i) All treated mice survived for more than sixty days.
j) 3-Methyl-1-(2-chloroethyl)-3-(methyl α-D-glucopyranosid-2-yl)-1-nitrosourea.
k) 3-*n*-Butyl-1-(2-chloroethyl)-3-(methyl α-D-glucopyranosid-2-yl)-1-nitrosourea.
l) 3-Isobutyl-1-(2-chloroethyl)-3-(methyl α-D-glucopyranosid-2-yl)-1-nitrosourea.
m) 3-(2-Methoxyethyl)-1-(2-chloroethyl)-3-(methyl α-D-glucopyranosid-2-yl)-1-nitrosourea.

(VIa—c, e). Comparison of the positional isomers (VIa—d and XIIa—d) showed that the effects of the alkyl substituent (R) at the N-3 position on antitumor activity against leukemia L1210 were considerably different in the two series. Thus, in the series of nitrosoureas (VIa—e), the optimal substituent was a methyl group (VIa), and isobutyl (VIc) and methoxyethyl (VId) groups conferred reduced activity. On the other hand, in the series of nitrosoureas (XIIa—d), a methoxyethyl group (XIId) was the optimal, and methyl substitution (XIIa) gave somewhat reduced acivity. Furthermore, the nitrosoureas (VIa—e) generally showed more potent antitumor activities than the nitrosoureas (XIIa—d) (*cf.* ILS_{30} and MED values), but they (VIa—e) were more toxic than XIIa—d (*cf.* OD and MTD values).

Further studies on the synthesis and antitumor activity of this new class of nitrosoureas are in progress.

Experimental

IR spectra were recorded with a Hitachi IR-215 spectrometer, and NMR spectra with a JEOL PMX-60 spectrometer using TMS as an internal standard in DMSO-d_6 or $CDCl_3$. The optical rotations were measured in a 0.5 dm tube with a Jasco DIP-180 polarimeter. Column chromatography was carried out on Merck silica gel 60. TLC was performed on Merck TLC plate silica gel 60 F254 and 30% sulfuric acid was used as the spray reagent. Organic solutions were generally concentrated by evaporation *in vacuo* below 40°C.

Preparation of 1-(2-Chloroethyl)-3-methyl-3-(methyl α-D-glucopyranosid-3-yl)-urea (IIa)——A mixture of 1.8 g of methyl 2,3-anhydro-α-D-allopyranoside (I)[3] and 30 ml of 30% methanolic methylamine solution was heated in a sealed tube at 150°C for 15 h, then concentrated. The residue was dissolved in 30 ml of methanol, and 1.6 g of 2-chloroethyl isocyanate was added to the solution under cooling. After being stirred at room temperature for 30 min, the mixture was concentrated. The residue was chromatographed on silica gel (solvent: chloroform–benzene–methanol=5: 2: 1) to give IIa in 58% yield as an amorphous powder. $[\alpha]_D^{18}$ +101.9° (c=1.1, methanol). IR ν_{max}^{Nujol} cm^{-1}: 3300, 1680, 1620, 1530, 1040. NMR (DMSO-d_6) δ: 2.71 (3H, s, NCH_3), 3.32 (3H, s, OCH_3), 4.67 (1H, d, J=2.5 Hz, H-1), 6.38 (1H, br, NH).

Preparation of 3-*n*-Butyl-1-(2-chloroethyl)-3-(methyl α-D-glucopyranosid-3-yl)urea (IIb) and 3-*n*-Butyl-1-(2-chloroethyl)-3-(methyl α-D-altropyranosid-2-yl)urea (IIIb)——A mixture of **I** (2.6 g) and *n*-butylamine (30 ml) was heated in a sealed tube at 150°C for 15 h, then concentrated. The residue was reacted with 2-chloroethyl isocyanate (2.4 g) and worked up as described above. TLC of the mixture gave two spots (Rf=0.51 and 0.62, solvent: chloroform–benzene–methanol=5: 1: 1). The mixture was chromatographed on silica gel (solvent: chloroform–benzene–methanol=10: 4: 1) to give IIb (Rf=0.62) and IIIb (Rf=0.51). IIb was obtained in 46% yield. $[\alpha]_D^{25}$ +106.5° (c=1.2, methanol). IR ν_{max}^{Nujol} cm^{-1}: 3400, 1670, 1615, 1520, 1040. NMR (DMSO-d_6) δ: 0.75—1.8 (7H, m, $CH_2CH_2CH_3$), 3.32 (3H, s, OCH_3), 4.64 (1H, d, J=4 Hz, H-1). IIIb was obtained in 6% yield. $[\alpha]_D^{25}$ +29.1° (c=1.1, methanol). IR ν_{max}^{Nujol} cm^{-1}: 3300, 1680, 1620, 1535, 1030. NMR (DMSO-d_6) δ: 0.80—1.85 (7H, m, $CH_2CH_2CH_3$), 3.30 (3H, s, OCH_3), 6.30 (1H, br, NH).

Preparation of 1-(2-Chloroethyl)-3-isobutyl-3-(methyl α-D-glucopyranosid-3-yl)urea (IIc)——A mixture of I (1.8 g), isobutylamine (20 ml), and ammonium sulfate (1.3 g) was heated in a sealed tube at 110°C for 20 h. After filtration, the mixture was concentrated. The residue was reacted with 2-chloroethyl isocyanate (2 g) and the product was purified by silica gel chromatography to give IIc in 62% yield. $[\alpha]_D^{21}$ +63.4° (c=1.5, methanol). IR ν_{max}^{Nujol} cm^{-1}: 3300, 1660, 1560, 1510, 1040. NMR (DMSO-d_6) δ: 0.88 (6H, dd, J=6 Hz, $CH(CH_3)_2$), 3.33 (3H, s, OCH_3), 4.68 (1H, d, J=3 Hz, H-1).

Preparation of 1-(2-Chloroethyl)-3-(2-methoxyethyl)-3-(methyl α-D-glucopyranosid-3-yl)urea (IId)——A mixture of I (1.8 g), 2-methoxyethylamine (10 ml), ethanol (10 ml), and ammonium sulfate (1.3 g) was heated at 110°C for 20 h. After filtration and concentration, the residue was reacted with 2-chloroethyl isocyanate (2 g) and the product was purified by silica gel chromatography to give IId in 68% yield. $[\alpha]_D^{21}$ +49.8° (c=1.4, methanol). IR ν_{max}^{Nujol} cm^{-1}: 3300, 1670, 1625, 1590, 1515, 1040. NMR (DMSO-d_6) δ: 3.24 (6H, s, $OCH_3\times2$), 6.29 (1H, m, NH).

Preparation of 3-Allyl-1-(2-chloroethyl)-3-(methyl α-D-glucopyranosid-3-yl)urea (IIe) and 3-Allyl-1-(2-chloroethyl)-3-(methyl α-D-altropyranosid-2-yl)urea (IIIe)——A mixture of I (1.8 g), allylamine (10 ml), ethanol (10 ml), and ammonium sulfate (1.3 g) was heated in a sealed tube at 110°C for 20 h. After filtration and concentration, the residue was reacted with 2-chloroethyl isocyanate (2 g) and the crude product was purified by silica gel chromatography. IIe was obtained in 35% yield. $[\alpha]_D^{21}$ +102.3° (c=1.1, methanol). IR ν_{max}^{Nujol} cm^{-1}: 3300, 1670, 1640, 1510, 1040. NMR (DMSO-d_6) δ: 3.32 (3H, s, OCH_3), 4.67 (1H, d, J=3 Hz, H-1), 6.17 (1H, m, NH). IIIe was obtained in 7% yield. $[\alpha]_D^{21}$ +54.9° (c=1.0, methanol). IR ν_{max}^{Nujol} cm^{-1}: 3300, 1670, 1515, 1040. NMR (DMSO-d_6) δ: 3.27 (3H, s, OCH_3), 6.18 (1H, br, NH).

Periodate Oxidation of Ureas (IIa—e and IIIb, e)——The urea (0.1 mmol) was dissolved in 0.2 M sodium periodate solution (2 ml) at room temperature. After being stirred for 15 min, the reaction mixture was

monitored by TLC. The ureas IIIb (Rf=0.51) and IIIe (Rf=0.40), having an altropyranoside moiety, were oxidized rapidly to give the oxidation products (Rf=0.88 and 0.75, respectively), while other ureas (IIa—c) having a glucopyranoside moiety remained almost completely unchanged.

General Procedure for the Preparation of 3-Substituted 1-(2-Chloroethyl)-3-(methyl 2,4,6-tri-*O*-acetyl-α-D-glucopyranosid-3-yl)ureas (IVa—e) and 3-Substituted 1-(2-Chloroethyl)-3-(methyl-3,4,6 tri-*O*-acetyl-α-D-altropyranosid-2-yl)urea (Vb, e)——Acetylation of the ureas (IIa—e and IIIb, e) was done as follows. A mixture of the urea (2 mmol), acetic anhydride (3 ml), and pyridine (6 ml) was stirred at room temperature overnight. The reaction mixture was poured into water and extracted with ethyl acetate. The extracts were washed with cold aqueous hydrochloric acid, water, aqueous sodium bicarbonate, and aqueous sodium chloride, successively. The organic layer was dried, filtered, and concentrated. The residue was chromatographed on silica gel. Triacetylated ureas were obtained in 75—85% yields as caramels. IVa; IR $\nu_{max}^{CHCl_3}$ cm^{-1}: 3300, 1740, 1650, 1520. NMR ($CDCl_3$) δ: 2.02 (3H, s, OAc), 2.06 (3H, s, OAc), 2.08 (3H, s, OAc), 2.70 (3H, s, NCH_3), 3.43 (3H, s, OCH_3), 4.98 (1H, d, J=3 Hz, H-1). IVb; IR ν_{max}^{liq} cm^{-1}: 3300, 1750, 1630, 1530. NMR ($CDCl_3$) δ: 0.9—1.7 (7H, m, $CH_2CH_2CH_3$), 2.03 (3H, s, OAc), 2.08 (6H, s, OAc×2), 3.39 (3H, s, OCH_3), 4.98 (1H, d, J=3 Hz, H-1). VIc; IR ν_{max}^{liq} cm^{-1}: 3300, 1750, 1640, 1530. NMR ($CDCl_3$) δ: 0.89 (6H, d, J=6.3 Hz, $CH(C\underline{H}_3)_2$), 2.03 (3H, s, OAc), 2.07 (6H, s, OAc×2), 3.38 (3H, s, OCH_3), 4.97 (1H, d, J=2.8 Hz, H-1), 5.82 (1H, broad, N$\underline{H}$). IVd; IR ν_{max}^{liq} cm^{-1}: 3340, 1740, 1650, 1530. NMR ($CDCl_3$) δ: 2.05 (3H, s, OAc), 2.10 (6H, s, OAc×2), 3.36 (3H, s, OCH_3), 3.40 (3H, s, OCH_3), 5.01 (1H, d, J=3 Hz, H-1), 6.44 (1H, m, NH). IVe; IR ν_{max}^{liq} cm^{-1}: 3430, 1750, 1650, 1530. NMR ($CDCl_3$) δ: 2.03 (3H, s, OAc), 2.12 (6H, s, OAc×2), 3.41 (3H, s, OCH_3), 4.96 (1H, d, J=3 Hz, H-1). Vb; IR ν_{max}^{liq} cm^{-1}: 3380, 1750, 1635, 1520. NMR ($CDCl_3$) δ: 0.9—1.7 (7H, m, $CH_2CH_2CH_3$), 2.03 (3H, s, OAc), 2.08 (6H, s, OAc×2), 3.39 (3H, s, OCH_3), 5.04 (1H, d, J=5 Hz, H-1). Ve; IR ν_{max}^{liq} cm^{-1}: 3430, 1750, 1650, 1530. NMR ($CDCl_3$) δ: 2.03 (3H, s, OAc), 2.12 (6H, s, OAc×2), 3.41 (3H, s, OCH_3), 4.96 (1H, d, J=5.1 Hz, H-1).

General Procedure for the Preparation of 3-Substituted 1-(2-Chloroethyl)-3-(methyl α-D-glucopyranosid-3-yl)-1-nitrosoureas (VIa—e)——The urea (0.01 mol) was dissolved in 40 ml of tetrahydrofuran and then anhydrous sodium acetate (0.04 mol) was added. Dinitrogen tetroxide (0.045 mol) was introduced into the mixture at −5°C for 10 min under stirring. After 10 min, 7 ml of methanol was added to the mixture, and the whole was stirred at the same temperature for 10 min. Cold ethyl acetate (40 ml), anhydrous sodium acetate (0.03 mol), and 10 ml of water were then added at −5°C. The whole was stirred vigorously for 10 min and the pH of the mixture was confirmed to be about 5. After filtration, the organic layer was collected, dried, filtered, and concentrated. The residue was purified by silica gel chromatography (solvent: ethyl acetate–benzene–methanol). The nitrosoureas (VIa—e) thus obtained were usually unstable yellow amorphous powders and the yields and physical properties are listed in Table I.

General Procedure for the Preparation of 3-Substituted 1-(2-Chloroethyl)-3-(methyl 2,4,6-tri-*O*-acetyl-α-D-glucopyranosid-3-yl)-1-nitrosoureas (VIIa—e)——Acetylation of these nitrosoureas (VIa—e) was done as described for the preparation of acetylated ureas (IVa—e and Vb, c). These acetylated nitrosoureas were obtained in 65—75% yields as yellow caramels and are listed in Table I with the yields and physical properties.

The Preparation of Methyl 3-Azido-3-deoxy-α-D-glucopyranoside (VIII) and Methyl 2-Azido-2-deoxy-α-D-altropyranoside (IX)——A mixture of I (5.3 g, 0.03 mol), sodium azide (5.85 g, 0.09 mol), ammonium sulfate (4.0 g, 0.03 mol), and dimethyl formamide (60 ml) was heated at 110°C for 4 h with vigorous stirring, then cooled. Acetone (200 ml) was added to the mixture, and the whole was filtered. The filtrate was concentrated and the residue was chromatographed on silica gel (solvent: chloroform–benzene–methanol=6: 3: 1) to give VIII and IX. VIII was obtained in 41% yield as colorless crystals. mp 126—127.5°C (lit.,[6] 125—127°C). $[\alpha]_D^{21}$ +178.4° (c=1.0, water). IR ν_{max}^{KBr} cm^{-1}: 3400, 2100, 1040. NMR (DMSO-d_6) δ: 3.29 (3H, s, OCH_3), 4.53 (1H, d, J=3 Hz, H-1). *Anal.* Calcd for $C_7H_{13}N_3O_5$: C, 38.36; H, 5.94; N, 19.18. Found: C, 38.13; H, 5.87; N, 19.11. IX was obtained in 19% yield as colorless crystals. mp 138—139°C. $[\alpha]_D^{21}$ +69.1° (c=1, methanol). (lit.,[7] mp 140—141°C, $[\alpha]_D^{21}$ +63.8°). IR ν_{max}^{KBr} cm^{-1}: 3480, 2110, 1050. NMR (DMSO-d_6) δ: 3.32 (3H, s, OCH_3), 4.45 (1H, d, J=4.9 Hz, H-1). *Anal.* Calcd for $C_7H_{13}N_3O_5$: C, 38.46; H, 5.94; N, 19.18. Found: C, 38.47; H, 6.05; N, 19.08.

Preparation of Methyl 3-Amino-3-deoxy-α-D-glucopyranoside (X) and Methyl 2-Amino-2-deoxy-α-D-altropyranoside (XI)——The azide (VIII, 2.0 g) was dissolved in 2% methanolic hydrochloric acid solution (90 ml) and reduced with hydrogen at 3 atm pressure in the presence of 10% palladium–carbon catalyst for 20 h. After removal of the catalyst and concentration of the solution, X·hydrochloride was obtained in 92% yield as an amorphous powder. IR ν_{max}^{KBr} cm^{-1}: 3390, 1600, 1070, 1020. Free amine (X) was obtained by treating aqueous X·hydrochloride solution with IRA-400 ion exchange resin. X; mp 167—169°C. $[\alpha]_D^{21}$ +150.7° (c=1.0, water) (lit.,[8] mp 167—168°C, $[\alpha]_D^{18}$ +144.4°). *Anal.* Calcd for $C_7H_{15}NO_5$: C, 43.51; H, 7.82; N, 7.25. Found: C, 43.60; H, 7.86; N, 7.21.

XI-Hydrochloride was similarly obtained by hydrogenation of IX in 93% yield as an amorphous powder. mp 80°C (dec.). $[\alpha]_D^{21}$ +40.8° (c=1.0, water). (lit.,[4] sirup, $[\alpha]_D^{22}$ +39.7° (c=3.1, chloroform). *Anal.* Calcd for $C_7H_{16}NO_5Cl$: C, 36.60; H, 7.02; N, 6.09; Cl, 15.43. Found: C, 36.49; H, 7.12; N, 6.01; Cl, 15.27.

Preparation of Ureas from Amino Compounds (X and XI) of Glucopyranoside and Altropyranoside——Methyl 3-aminoglucoside (X·hydrochloride) (1.2 g, 0.005 mol) was stirred in a solution of sodium methoxide (0.005 mol) in 10 ml of methanol at room temperature for 1 h. After filtration and concentration, the residue

was dissolved in ethanol (20 ml). Butyraldehyde (0.5 g, 0.007 mol) was added to the solution. The whole was stirred for 5 min, then sodium borohydride (0.4 g, 0.01 mol) was added with cooling, and the mixture was stirred at room temperature for 2 h. After addition of methanol (10 ml), concentrated hydrochloric acid was added dropwise to the reaction mixture until the pH of the mixture reached about 4. After being stirred for 10 min, the mixture was basified with 20% potassium carbonate solution, filtered, and concentrated. The residue was dissolved in methanol (30 ml), and 2-chloroethyl isocyanate (1.0 g, 0.01 mol) was added dropwise at 5°C, then the mixture was stirred for 1.5 h at room temperature and concentrated. The residue was chromatographed on silica gel (solvent: chloroform–benzene–methanol=5: 2: 1) to give the urea in 38% yield; it was identical with IIb. $[\alpha]_D^{25}$ +106.1° (c=1.0, methanol). The urea having an altropyranoside moiety was similarly obtained from XI in 41% yield and proved to be identical with IIIb. $[\alpha]_D^{25}$ +28.9° (c=1, methanol).

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