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Synthesis of a tetrahydropyrano[2,3-*d*]oxazole analogue of trehazolin

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Abstract

Tetrahydropyrano[2,3-*d*]oxazole **6** as an analogue of trehazolin was synthesized from compound **3**, which is a protected 1,3-bis(β-D-glucopyranosyl)thiourea. In contrast, compound **10**, a protected 1,3-bis(α-D-glucopyranosyl)thiourea, and **15**, a mixed species having one α-D-glucopyranosyl group and one β-D-glucopyranosyl group, did not yield the corresponding pyranooxazolines as anticipated, giving instead, respectively, a furo[2,3-*d*]oxazole **14** and a complex mixture of unidentified products. © 1996 Elsevier Science Ltd.

Keywords: Trehazolin; Tetrahydropyrano[2,3-*d*]oxazole; Tetrahydrofuro[2,3-*d*]oxazole

1. Introduction

Carbohydrates govern a wide range of biological recognition phenomena, such as blood-group determinants, cell-surface receptors for proteins, toxins, pathogens, viruses, and binding sites for cell-adhesion molecules such as glycoconjugates. Moreover, in recent years interest in carbohydrate-based enzyme inhibitors continues to expand at an unabated pace. In particular, glycosidase inhibitors such as validamycins, acarbose, AO-128, deoxynojirimycin derivatives, and castanospermine analogues, have become an important group of glycosidase inhibitors [1]. Indeed some of them have been singled

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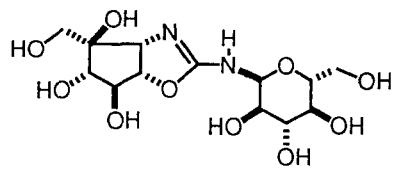
out as being effective for treatment of diabetes [2]. However, there is still considerable opportunity for finding even more effective glycosidase inhibitors, and certainly the search for such inhibitors having a potent efficacy in diabetes mellitus therapy and *N*-linked oligosaccharide processing glycosidase inhibition of HIV are no exceptions.

The natural occurrence of trehazolin, an inhibitor of trehalase, which is an enzyme of paramount ecological importance in the control of insects and certain fungi, was unveiled by workers at Sankyo [3] and Suntory [4] several years ago. Based on spectral data analyses [3] and synthetic studies [5], the structure was determined as shown below. The compound is a pseudodisaccharide consisting of an α-glucosyl group and a 5,5-ring-fused aminooxazoline. We [6] and other groups [7] have synthesized various analogues of trehazolin to find potential glycosidase inhibitors. However, to date, no inhibitors have been discovered. Therefore, modification of the 5,5-ring-fused aminooxazoline function of trehazolin is an exciting line of investigation that we wished to exploit further in terms of glycosidase inhibitory action. To this end, we considered the 5,6-ring-fused 2-amino-tetrahydropyrano[2,3-*d*]oxazole analogue of trehazolin as a viable target, and we sought to synthesize it.

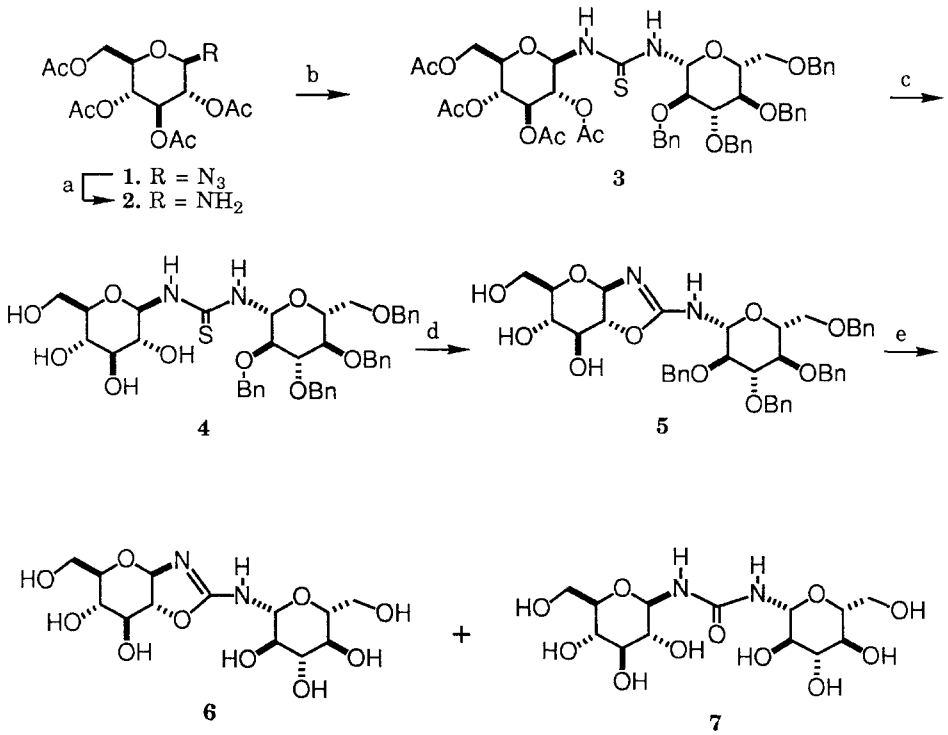


Trehazolin

However, we have recently revealed that 6,5-ring-fused 2-amino-hexahydrocyclopent[*d*][1,3]oxazine and 5,6-ring-*cis*-fused 2-amino-5*H*-pyrano[2,3-*d*]oxazole easily converted to the more stable 5,5-ring-fused tetrahydro-4*H*-cyclopentoxazole [8] and 5,5-ring-*cis*-fused 2-amino-tetrahydrofuro[2,3-*d*]oxazole [7,9], respectively. Therefore, we decided to investigate whether the 5,6-ring-*trans*-fused 2-amino-tetrahydropyrano[2,3-*d*]oxazole analogue of trehazolin exists, and whether it is active toward various glucosidases.

2. Results and discussion

Chemistry.—2,3,4,6-Tetra-*O*-acetyl-β-D-glucopyranosyl azide (**1**) obtained from 2,3,4,6-tetra-*O*-acetyl-α-D-glucopyranosyl bromide [10,11] or β-D-glucose pentaacetate [12] was used as starting material. Compound **1** was converted to an amine **2** by hydrogenolysis using $Pd(OH)_2$-on-charcoal as a catalyst. Treatment of **2** with 2,3,4,6-tetra-*O*-benzyl-β-D-glucopyranosyl isothiocyanate [13] at 25 °C for 2 days gave the *N*,*N*′-bis(β-D-glucopyranosyl)thiourea **3**. Deacetylation of **3** with a catalytic amount of KOH in EtOH at 25 °C gave the tetraol **4**. Treatment of **4** with 2-chloro-3-ethylbenzoxazolium tetrafluoroborate [14] gave a 5,6-ring-*trans*-fused 2-amino-tetrahydropyrano[2,3-*d*]oxazole **5**. Gratifyingly, compound **5** was stable enough at room

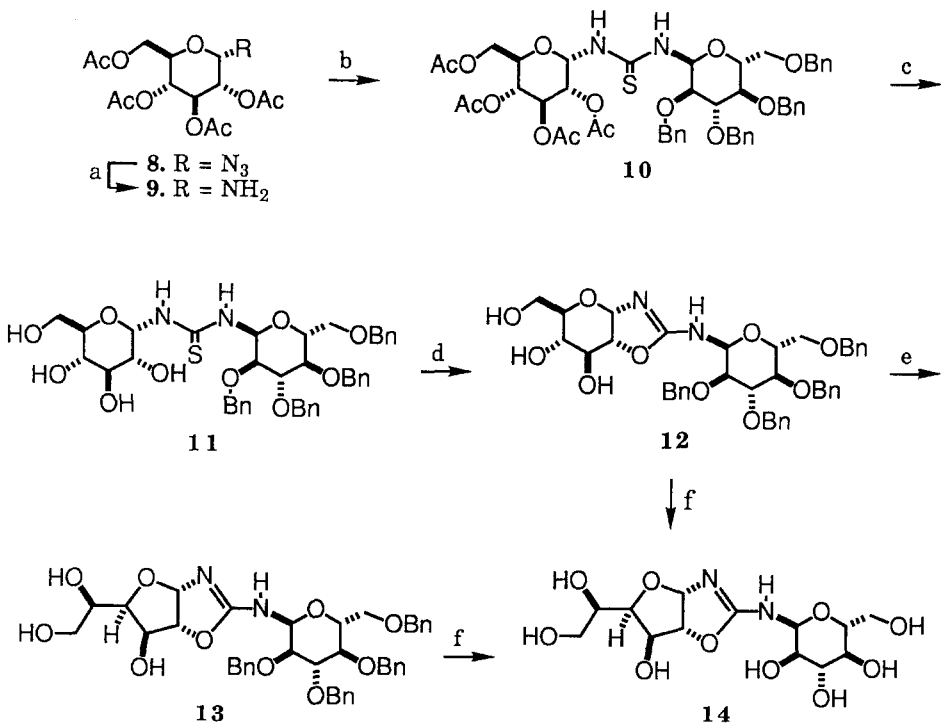


Scheme 1. Reagents and conditions: (a) H_2, $Pd(OH)_2$/C, 2:1 THF–EtOH, 24 °C, 3 h, 69%; (b) 2,3,4,6-tetra-*O*-benzyl-β-D-glucopyranosyl isothiocyanate, THF, rt, 2 days, two steps 47%; (c) cat. KOH, EtOH, 24 °C, 10 min, 84%; (d) 2-chloro-3-ethylbenzoxazolium tetrafluoroborate, MeCN, N_2, 0 °C, 2 h, then Et_3N, 0 °C, 10 min, and 24 °C, 30 min, 62%; (e) H_2, $Pd(OH)_2$/C, MeOH, 60 °C, 30 min, 17% (**6**), 32% (**7**).

temperature and did not rearrange to a furoxazole in *N*,*N*-dimethylformamide at 25 °C. Deprotection of the four benzyl groups of **5** gave the desired pyranoxazoline analogue **6** with an accompanying urea **7**. Thus we accomplished the synthesis of compound **6** in a stereocontrolled manner (see Scheme 1).

In the same way, 2,3,4,6-tetra-*O*-acetyl-α-D-glucopyranosyl azide **8** [15] was converted to an amine **9**, which was further converted to the *N*,*N*′-bis(α-D-glucopyranosyl)thiourea **10** by reaction with 2,3,4,6-tetra-*O*-benzyl-α-D-glucopyranosyl isothiocyanate [13]. Deacetylation and aminooxazolin formation on **10** gave a 5,6-ring-*cis*-fused 2-amino-tetrahydropyrano[2,3-*d*]oxazole **12** via **11**. The 5,6-ring-*cis*-fused compound **12** was unstable at room temperature and gradually rearranged to a furoxazole **13** in *N*,*N*-dimethylformamide solution. In this case, the conversion of a pyranose ring to a furanose ring occurred in an anticipated manner from the previous studies [8,9]. Deprotection of the four benzyl groups of both **12** and **13** gave a furoxazole analogue **14** (see Scheme 2).

Finally, we tried to synthesize compound **18** from **15**, which had two D-glucopyranose parts connecting with a β-configuration on one *N*-atom and the α-configuration on the other *N*-atom. Treatment of the β-D-glucopyranosyl amine **2** with 2,3,4,6-te-



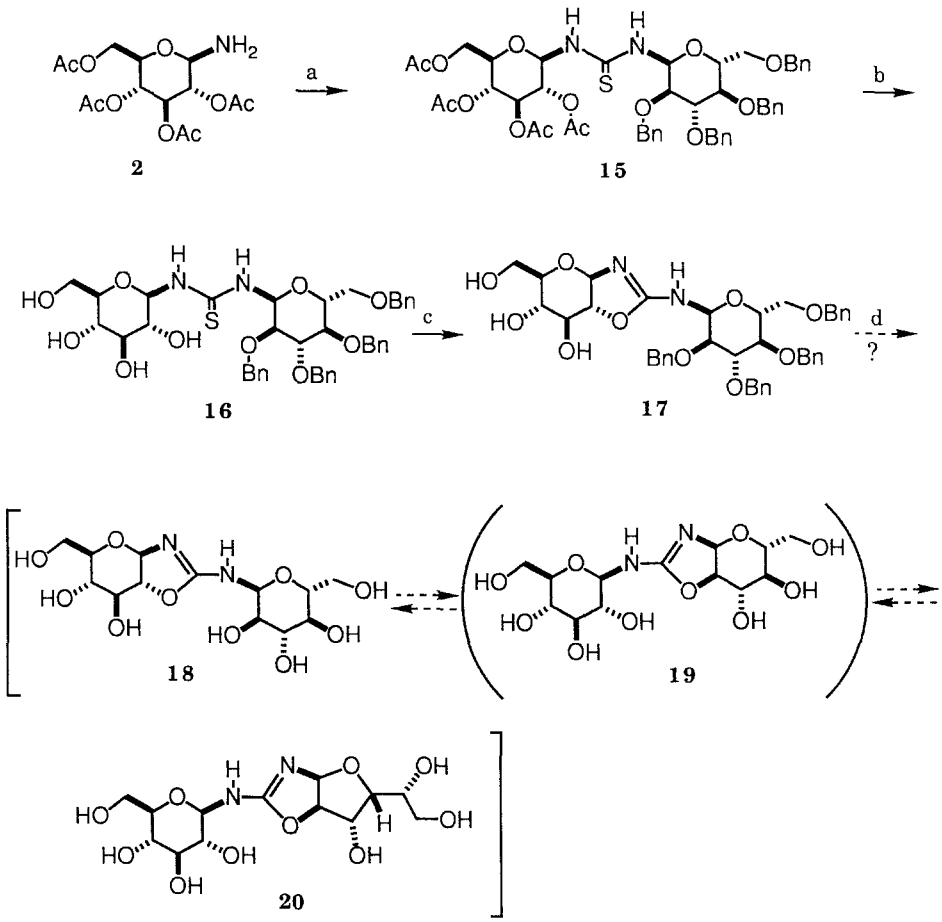
Scheme 2. Reagents and conditions: (a) H_2, $Pd(OH)_2$ /C, THF, 24 °C, 2 h; (b) 2,3,4,6-tetra-*O*-benzyl-α-D-glucopyranosyl isothiocyanate, THF, 24 °C, 2 days, two steps 32%; (c) cat. KOH, EtOH, 24 °C, 10 min, 82%; (d) 2-chloro-3-ethylbenzoxazolium tetrafluoroborate, MeCN, N_2, 0 °C, 2 h, then Et_3N, 0 °C, 10 min, and 24 °C, 30 min, 67%; (e) Me_2NCHO, rt, quant.; (f) H_2, $Pd(OH)_2$ /C, MeOH, 60 °C, 30 min, 59%.

tra-*O*-benzyl-α-D-glucopyranosyl isothiocyanate [13] gave a thiourea **15**. Deacetylation and aminooxazoline formation from **15** gave an 2-aminooxazoline **17** via the thiourea **16**. The *O*-benzyl protected compound **17** was deprotected to give **18** as an unidentified complex mixture. The mixture might have arisen from the rearrangement of the *trans*-oxazoline **18** to give the *cis*-pyranooxazoline **19**, then the furanooxazoline **20**, and so on. This scenario is predicted from the behavior of the previously mentioned compound **12** or that of the analogues recently reported [8,9]. We cannot offer a reasonable explanation why the thermodynamically more unstable *trans*-fused **5** and **6** can exist preferentially over the thermodynamically more stable *cis*-fused **12** (see Scheme 3).

Biological activity.—Compound **6** as well as **14** was disappointingly void of activity toward the various glycosidases tested.

3. Experimental

Melting points are uncorrected. ^1H NMR spectra (270, 400, and 500 MHz) were recorded using Me_4Si as an internal standard. Elemental analyses were performed by the



Scheme 3. Reagents and conditions: (a) 2,3,4,6-tetra-*O*-benzyl-α-D-glucopyranosyl isothiocyanate, THF, rt, 2 days, 63%; (b) cat. KOH, EtOH, 24 °C, 10 min, 82%; (c) 2-chloro-3-ethylbenzoxazolium tetrafluoroborate, MeCN, N_2, 0 °C, 2 h, then Et_3N, 0 °C, 10 min, and 24 °C, 30 min, 67%; (d) H_2, $Pd(OH)_2$/C, MeOH, 60 °C, 30 min, unknown mixture.

Institute of Science and Technology, Inc. Preparative TLC was performed on silica gel plates (E. Merck, Silica Gel-60 F_{245}), and column chromatography was carried out on columns packed with E. Merck Silica Gel-60 (230–400 mesh) using a slightly increased pressure (1.5 atm) for elution. THF was distilled from $LiAlH_4$ and used immediately. CH_2Cl_2 was dried by being passed through ICN Alumina B-Super I. Me_2NCHO and pyridine were dried by storage over 4A molecular sieves. MeCN was dried by storage over 3A molecular sieves.

*Preparation of 2,3,4,6-Tetra-*O*-acetyl-β-D-glucopyranosyl azide* (**1**).—*Method A*. To 2,3,4,6-Tetra-*O*-acetyl-α-D-glucopyranosyl bromide (9.40 g, 22.9 mmol) was added a

stirred mixture of dry MeCN (50 mL) and NaN_3 (5.4 g, 83.0 mmol), and the mixture was refluxed for 12 h. The mixture was filtered, and the filtrate was concentrated in vacuo to yield **1** (7.14 g, 84%) after recrystallization from EtOH.

Method B. To a solution of β-D-glucose pentaacetate (10.0 g, 25.6 mmol) in CH_2Cl_2 (100 mL) was added Me_3SiN_3 (3.86 mL, 29.5 mmol) and $SnCl_4$ (5.34 g, 20.5 mmol), and the mixture was stirred for 30 min at 24 °C and diluted with EtOAc, which was washed with H_2O, aq 1 M HCl, and satd $NaHCO_3$, dried over $MgSO_4$, filtered, and concentrated in vacuo to give **1** (8.74 g, 91%) after recrystallization from hexane–EtOAc: mp 127–129 °C, lit. 123–124 °C [11]; $[\alpha]_D^{24}$ −29° (*c* 2.0, $CHCl_3$), lit. −22° (*c* 1.8, $CHCl_3$) [11]; IR ν_{max} (KBr) 2119, 1756, 1748, 1733 cm^{-1}; ^1H NMR ($CDCl_3$): δ 2.01 (3 H, s), 2.03 (3 H, s), 2.08 (3 H, s), 2.11 (3 H, s), 3.80 (1 H, ddd, *J* 2.6, 4.6, 9.9 Hz), 4.17 (1 H, dd, *J* 2.0, 12.5 Hz), 4.28 (1 H, dd, *J* 4.6, 12.5 Hz), 4.65 (1 H, d, *J* 8.6 Hz), 4.96 (1 H, t, *J* 8.6–9.2 Hz), 5.11 (1 H, t, *J* 9.2–9.9 Hz), 5.22 (1 H, t, *J* 9.2 Hz).

2,3,4,6-Tetra-O-acetyl-β-D-glucopyranosylamine (**2**).—A solution of **1** (1.0 g, 2.68 mmol) in 2:1 THF–EtOH (15 mL) was hydrogenolyzed using $Pd(OH)_2$-on-charcoal (Pd content 20%, wet, Degussa type, 60 mg) as a catalyst. The suspension was filtered, concentrated and chromatographed on a silica gel column. Elution with 1:1 hexane–EtOAc, and then EtOAc gave **2** (632 mg, 69%) after recrystallization from hexane–EtOAc: mp 121–123 °C; $[\alpha]_D^{24}$ +12.5° (*c* 1.8, $CHCl_3$); IR ν_{max} (KBr) 3412, 1755, 1735 cm^{-1}; ^1H NMR ($CDCl_3$): δ 1.74 (2 H, br, NH_2), 2.01 (3 H, s), 2.03 (3 H, s), 2.07 (3 H, s), 2.10 (3 H, s), 3.69 (1 H, ddd, *J* 2.0, 4.6, 9.9 Hz), 4.11 (1 H, dd, *J* 2.0, 12.5 Hz), 4.20 (1 H, d, *J* 9.2 Hz), 4.83 (1 H, t, *J* 9.2 Hz), 5.04 (1 H, t, *J* 9.2–9.9 Hz), 5.24 (1 H, t, *J* 9.2–9.9 Hz). Anal. Calcd for $C_{14}H_{21}NO_9$ (347.3): C, 48.41; H, 6.09; N, 4.03. Found: C, 48.27; H, 5.94; N, 3.99.

N-(2,3,4,6-Tetra-O-acetyl-β-D-glucopyranosyl)-N′-(2,3,4,6-tetra-O-benzyl-β-D-glucopyranosyl)thiourea (**3**).—A solution of 2,3,4,6-tetra-*O*-benzyl-β-D-glucopyranosyl isothiocyanate (286 mg, 0.492 mmol) in THF (10 mL) was added to a solution of **2** (180 mg, 0.518 mmol) in THF (10 mL). The mixture was allowed to stand at room temperature for 2 days, then it was concentrated in vacuo to give a residue, which was chromatographed on a silica gel column. Elution with 3:2 cyclohexane–EtOAc gave **3** (217 mg, 47%): IR ν_{max} (Nujol) 3300 (w), 1752 cm^{-1}; ^1H NMR ($CDCl_3$): δ 1.99 (3 H, s), 2.01 (6 H, s), 2.03 (3 H, s), 3.38 (1 H, t, *J* 8.7 Hz), 3.55–3.85 (6 H, m), 4.10 (1 H, dd, *J* 1.6, 12.2 Hz), 4.27 (1 H, dd, *J* 4.5, 12.2 Hz), 4.47–4.98 (10 H, m), 5.02 (1 H, t, *J* 9.5 Hz), 5.32 (1 H, t, *J* 9.5 Hz), 5.54 (1 H, d, *J* 8.7 Hz), 6.25 (1 H, br, NH), 7.14–7.19 (2 H, m), 7.26–7.43 (18 H, m). FABMS (positive): *m/z* 929 $[M + H]^+$. Anal. Calcd for $C_{49}H_{56}N_2O_{14}S$ (929.1): C, 63.35; H, 6.08; N, 3.02; S, 3.45. Found: C, 62.89; H, 6.02; N, 2.69; S, 3.46.

N-(β-D-Glucopyranosyl)-N′-(2,3,4,6-tetra-O-benzyl-β-D-glucopyranosyl)thiourea (**4**). —A solution of **3** (194 mg, 0.209 mmol) in 99.5% EtOH (12 mL) containing KOH (12 mg) was stirred for 30 min at 25 °C. The solution was concentrated in vacuo to give a residue, which was chromatographed on a silica gel column. Elution with 5% MeOH in EtOAc gave **4** (133 mg, 84%): IR ν_{max} (Nujol) 3200–3300 cm^{-1}; ^1H NMR ($CDCl_3$ + D_2O): δ 3.12–3.16 (1 H, m), 3.38–3.69 (12 H, m), 4.21–4.80 (3 H, m), 4.63–4.76 (6 H, m), 5.55 (1 H, bs), 5.75 (1 H, bs), 6.98–7.05 (2 H, m), 7.12–7.22 (18 H, m). FABMS (positive): *m/z* 761 $[M + H]^+$. HRFABMS; *m/z* Calcd: 761.3108. Found: 761.3079.

Anal. Calcd for $C_{41}H_{48}N_2O_{10}$ (760.9): C, 64.72; H, 6.36; N, 3.68; S, 4.21. Found: C, 64.90; H, 6.19; N, 3.55; S, 4.19.

[3aR-(3aα,5β,6α,7β,7aβ)]-5-(Hydroxymethyl)-3a,6,7,7a-tetrahydro-2-[(2,3,4,6-tetra-O-benzyl-β-D-glucopyranosyl)amino]-5H-pyrano[2,3-d]oxazole-6,7-diol (**5**).—To a solution of **4** (124 mg, 0.163 mmol) in MeCN (10 mL) was added 2-chloro-3-ethylbenzoxazolium tetrafluoroborate (124 mg, 0.460 mmol) under nitrogen at 0–5 °C. After 2 h of stirring at 0–5 °C, Et_3N (124 mg, 1.23 mmol) was added. After 30 min of stirring at 0–5 °C, the reaction mixture was concentrated in vacuo and chromatographed on a silica gel column. Elution with 5% MeOH in EtOAc gave a fraction of product, which was concentrated in vacuo and dissolved in EtOAc. The solution was washed with water, dried over $MgSO_4$, and concentrated to give **5** (73 mg, 62%): IR ν_{max} (Nujol) 3300, 1690 (w), 1648 cm^{-1}; ^1H NMR ($CDCl_3$): δ 2.40 (1 H, br, NH), 3.40–3.96 (12 H, m), 4.39–4.54 (3 H, m), 4.67–4.87 (7 H, m), 7.06–7.10 (2 H, m), 7.22–7.32 (18 H, m). FABMS (positive): *m/z* 727 $[M + H]^+$. HRFABMS; *m/z* Calcd: 727.3231. Found: 727.3253. Anal. Calcd for $C_{41}H_{46}N_2O_{10}$ (726.8): C, 67.75; H, 6.38; N, 3.85. Found: C, 67.97; H, 6.41; N, 3.91.

[3aR-(3aα,5β,6α,7β,7aβ)]-5-(Hydroxymethyl)-3a,6,7,7a-tetrahydro-2-[(β-D-glucopyranosyl)amino]-5H-pyrano[2,3-d]oxazole-6,7-diol (**6**) *and 1,3-bis(β-D-glucopyranosyl)urea* (**7**).—A solution of **5** (30 mg, 0.041 mmol) in MeOH (6 mL) containing $Pd(OH)_2$-on-charcoal (600 mg, Pearlman's catalyst) was hydrogenolized at 60 °C for 35 min. The reaction mixture was filtered, concentrated in vacuo, and chromatographed on an Amberlite CG-50 (NH_4^+ type–H^+ type = 3:2, 10 mL) column. Elution with H_2O (2 mL each) gave an inseparable mixture (7.7 mg) of **6** (17%) and **7** (32%), and a small amount of unknown products in fraction no. 3. Physical data for the mixture is as follows: IR ν_{max} (Nujol) 3400 (br), 1650, 1580 cm^{-1}; ^1H NMR (500 MHz, D_2O) of **6** (numbering is according to that of glucose); aglycon part: δ 3.44 (1 H, dd, *J* 8.1, 9.6 Hz, H-4′), 3.65, (1 H, m, H-5′), 3.71 (1 H, dd, *J* 9.8, 10.4 Hz, H-2′), 3.75 (1 H, m, H-6′), 3.90 (1 H, m, H-6′), 4.04 (1 H, dd, *J* 8.1, 10.4 Hz, H-3′), 4.89 (1 H, d, *J* 9.8 Hz, H-1′), 4.37 (1 H, d, *J* 9.0 Hz, H-1), 4.78 (1 H, bs, OH), 5.08 (1 H, bs, OH); glucose part: 3.38 (1 H, dd, *J* 8.7, 9.1 Hz, H-2), 3.40 (1 H, dd, *J* 8.7, 9.1 Hz, H-4), 3.51 (1 H, ddd, *J* 2.0, 5.5, 8.7 Hz, H-5), 3.54 (1 H, dd, *J* 8.7, 9.1 Hz, H-3), 3.71 (1 H, dd, *J* 5.5, 12.3 Hz, H-6), 3.89 (1 H, dd, *J* 2.0, 12.3 Hz, H-6), 4.75 (1 H, d, *J* 9.1 Hz, H-1). ^{13}C NMR (D_2O); aglycon part: 61.9 (C-6′), 70.5 (C-4′), 74.6 (C-3′), 82.3 (C-5′), 86.1 (C-2′), 95.1 (C-1′), 163.8 (C-7′); glucose part: 61.9 (C-6), 70.6 (C-4), 73.1 (C-2), 77.7 (C-3), 78.4 (C-5), 83.0 (C-1). FABMS (positive): *m/z* 367 $[M + H]^+$, 349 $[M + H - H_2O]^+$. HRFABMS; *m/z* Calcd: 367.1353. Found: 367.1352. Physical data for **7**: ^1H NMR (500 MHz, D_2O) (numbering is according to that of glucose): δ 3.38 (1 H, dd, *J* 8.7, 9.1 Hz, H-2), 3.40 (1 H, dd, *J* 8.7, 9.1 Hz, H-4), 3.51 (1 H, ddd, *J* 2.0, 5.5, 8.7 Hz, H-5), 3.54 (1 H, dd, *J* 8.7, 9.1 Hz, H-3), 3.71 (1 H, dd, *J* 5.5, 12.3 Hz, H-6), 3.89 (1 H, dd, *J* 2.0, 12.3 Hz, H-6), 4.87 (1 H, d, *J* 9.1 Hz, H-1). ^{13}C NMR (D_2O): 61.9 (C-6), 70.6 (C-4), 73.1 (C-2), 77.7 (C-3), 78.4 (C-5), 82.1 (C-1), 160.3 (urea carbonyl carbon).

N-(2,3,4,6-Tetra-O-acetyl-α-D-glucopyranosyl)-N′-(2,3,4,6-tetra-O-benzyl-α-D-glucopyranosyl)thiourea (**10**).—A solution of 2,3,4,6-tetra-*O*-benzyl-α-D-glucopyranosyl isothiocyanate (582 mg, 1.00 mmol) in THF (10 mL) was added to a THF solution of **9**, which was obtained by filtration of the hydrogenolyzed (24 °C, 2 h) suspension of **8**

(373 mg, 1.00 mmol) in THF (10 mL) using wet $Pd(OH)_2/C$ (80 mg) as a catalyst. (Chromatography of **9** on silica gel causes the anomerization from the α anomer to the β anomer.) The mixture was allowed to stand for 2 days at room temperature, concentrated in vacuo, and chromatographed on a silica gel column. Elution with 5:3 cyclohexane–EtOAc gave **10** (297 mg, 32%) as a powder: IR ν_{max} (Nujol) 3310, 1740 cm^{-1}; ^1H NMR ($CDCl_3$): δ 1.82 (3 H, s), 2.01 (3 H, s), 2.02 (3 H, s), 2.05 (3 H, s), 3.46 (1 H, t, *J* 9.3 Hz), 3.56–3.86 (5 H, m), 3.87–3.93 (1 H, m), 3.97 (1 H, m), 4.07 (1 H, dd, *J* 2.6, 7.9 Hz), 4.25 (1 H, dd, *J* 4.3, 12.7 Hz), 4.44–4.91 (9 H, m), 5.10 (1 H, t, *J* 9.2–9.9 Hz), 5.20–5.37 (2 H, m), 6.06 (1 H, bs), 6.91 (1 H, bs), 7.02–7.05 (2 H, m), 7.23–7.32 (18 H, m). FABMS (positive): *m/z* 929 $[M + H]^+$. HRFABMS; *m/z* Calcd: 929.3530. Found: 929.3521. Anal. Calcd for $C_{49}H_{56}N_2O_{14}S \cdot 0.5H_2O$ (929.1 + 9.0): C, 62.74; H, 6.12; N, 2.98; S, 3.42. Found: C, 62.46; H, 6.09; N, 2.67; S, 3.31.

N-*(α-*D*-Glucopyranosyl)*-N′-*(2,3,4,6-tetra-*O*-benzyl-α-*D*-glucopyranosyl)thiourea* (**11**).—To a solution of **10** (430 mg) in 99.5% EtOH (27 mL) was added KOH (27 mg). After stirring for 10 min at 24 °C, the reaction mixture was concentrated in vacuo and chromatographed on a silica gel column. Elution with 19:1 EtOAc–MeOH gave **11** (288 mg, 82%) as a powder: IR ν_{max} (Nujol) 3300 cm^{-1}; ^1H NMR ($CDCl_3$): δ 3.2–4.0 (12 H, m), 4.0–4.9 (8 H, m), 5.58 (1 H, bs), 5.75 (1 H, bs), 7.00 (1 H, b, NH), 7.08 (2 H, bs), 7.12–7.30 (18 H, m), 7.70 (1 H, b, NH). Anal. Calcd for $C_{41}H_{48}N_2O_{10}S$ (760.9): C, 64.72; H, 6.36; N, 3.68; S, 4.21. Found: C, 64.29; H, 6.45; N, 3.48; S, 3.95.

*[3aS-(3aα,5α,6β,7α,7aα)]-5-(Hydroxymethyl)-3a,6,7,7a-tetrahydro-2-[(2,3,4,6-tetra-*O*-benzyl-α-*D*-glucopyranosyl)amino]-5*H*-pyrano[2,3-*d*]oxazole-6,7-diol* (**12**).—To a solution of **11** (70 mg, 0.092 mmol) in MeCN (3 mL) was added 2-chloro-3-ethylbenzoxazolium tetrafluoroborate (70 mg, 0.260 mmol) at 0–5 °C with stirring under nitrogen. The mixture was stirred for 2 h, and Et_3N (80 mg) was added. After stirring for 30 min at 0–5 °C, the reaction mixture was concentrated in vacuo and chromatographed on a silica gel column. Elution with 19:1 EtOAc–MeOH gave an unknown higher R_f isomer (10 mg) and **12** (44 mg, 67%) as a gum: IR ν_{max} (Nujol) 3600–3200, 1645 cm^{-1}; ^1H NMR ($CDCl_3$): δ 3.40–3.87 (14 H, m, containing 2 × OH), 4.45–4.92 (10 H, m, containing 2 × OH), 5.40 (1 H, bs), 5.67 (1 H, d, *J* 5.9 Hz), 7.06–7.09 (2 H, m), 7.22–7.27 (18 H, m). FABMS (positive): *m/z* 727 $[M + H]^+$. HRFABMS; *m/z* Calcd: 727.3231. Found: 727.3187.

[3aS-[3aα,5α(S),6α,6aα]]-5-[(1,2-Dihydroxy)ethyl]-3a,5,6,6a-tetrahydro-2-[(2,3,4,6-tetra-*O*-benzyl-α-*D*-glucopyranosyl)amino]-furo[2,3-*d*]oxazole* (**13**).—A solution of **12** (10 mg) in Me_2NCHO (1 mL) was allowed to stand overnight at room temperature, at the end of which time it was concentrated in vacuo to give **13** quantitatively. (NMR monitoring in Me_2NCHO-d_7.) ^1H NMR (400 MHz, Me_2NCHO-d_7) (numbering is according to that of glucose); aglycon part: δ 3.20 (1 H, m, H-6), 3.31 (1 H, H-4), 3.39 (1 H, H-6), 3.65, (1 H, H-5), 3.91 (1 H, OH-6), 3.99 (1 H, dd, *J* 1.0, > 10 Hz, H-3), 4.28 (1 H, bs, OH-5), 4.44 (1 H, dd, *J* > 1.0, 5.2 Hz, H-2), 4.90 (1 H, bs, OH-3), 5.58 (1 H, d, *J* 5.2 Hz, H-1); 1-amino-tetra-*O*-benzyl-glucose part: 3.22 (1 H, H-4), 3.35 (1 H, m, H-6), 3.40 (1 H, dd, *J* 5.0, 7.5 Hz, H-2), 3.42 (1 H, m, H-6), 3.52 (1 H, H-5), 3.81 (1 H, H-3), 4.22 (2 H, s, OCH_2Ph-6), 4.31, 4.50 (2 H, AB-q, OCH_2Ph-4), 4.34, 4.43 (2 H, AB-q, OCH_2Ph-2), 4.45, 4.60 (2 H, AB-q, OCH_2Ph-3), 5.36 (1 H, H-1). FABMS (positive): *m/z* 727 $[M + H]^+$.

*[3a*S*-[3aα,5α(S*),6α,6aα]]-5-[(1,2-Dihydroxy)ethyl]-3a,5,6,6a-tetrahydro-2-[(α-*D*-glucopyranosyl)amino]-furo[2,3-*d*]oxazole* (**14**).—A solution of **12** (20 mg) or **13** in MeOH (4 mL) containing wet $Pd(OH)_2$-on-charcoal (400 mg) was hydrogenolyzed at 60 °C for 30 min with vigorous stirring and filtered. The filtrate was concentrated in vacuo and chromatographed on an Amberlite CG-50 (NH_4^+ type–H^+ type = 3:2, 5 mL) column. Elution with H_2O (1.5 mL each) and lyophilization gave **14** (6.0 mg, 59%) in fraction nos. 6, 7, and 8: IR ν_{max} (KBr) 3370 (br), 1656, 1553, 1428, 1350, 1236, 1092, 1031 cm^{-1}; ^1H NMR (400 MHz, Me_2NCHO-d_7) (numbering is according to that of glucose); aglycon part: δ 3.65 (1 H, m, H-6), 3.68 (1 H, dd, *J* 2.7, 8.7 Hz, H-4), 3.80 (1 H, m, H-6), 3.96, (1 H, ddd, *J* 2.8, 5.9, 8.7 Hz, H-5), 4.45 (1 H, d, *J* 2.7 Hz, H-3), 4.95 (1 H, d, *J* 5.1 Hz, H-2), 5.98 (1 H, d, *J* 5.1 Hz, H-1); 1-amino-tetra-*O*-benzyl-glucose part: 3.43 (1 H, dd, *J* 9.5, 10.3 Hz, H-4), 3.56 (1 H, ddd, *J* 1.0, 2.8, 9.5 Hz, H-5), 3.68 (1 H, dd, *J* 10.3, 10.5 Hz, H-3), 3.73 (1 H, m, H-6), 3.80 (1 H, dd, *J* 4.8, 10.5 Hz, H-2), 3.81 (1 H, m, H-6), 5.37 (1 H, d, *J* 4.8, H-1). ^{13}C NMR (Me_2NCHO-d_7) (numbering is according to that of glucose); aglycon part: δ 65.7 (C-6), 70.5 (C-5), 75.2 (C-3), 79.7 (C-4), 88.8 (C-2), 99.3 (C-1); glucose part: δ 62.8 (C-6), 71.7 (C-4), 72.0 (C-2), 74.4 (C-5), 75.1 (C-3), 82.6 (C-1). FABMS (positive): *m/z* 389 $[M + Na]^+$, 367 $[M + H]^+$. HRFABMS; *m/z* Calcd: 367.1353. Found: 367.1361. Anal. Calcd for $C_{13}H_{22}N_2O_{10} \cdot 1.25H_2O$ (388.8): C, 40.15; H, 6.35; N, 7.20. Found: C, 40.63; H, 6.29; N, 6.86.

N-*(2,3,4,6-Tetra*-O-*acetyl-β*-D-*glucopyranosyl)*-N′-*(2,3,4,6-tetra*-O-*benzyl-α*-D-*glucopyranosyl)thiourea* (**15**).—A solution of 2,3,4,6-tetra-*O*-benzyl-α-D-glucopyranosyl isothiocyanate (586 mg, 1.01 mmol) in THF (10 mL) was added to a THF (10 mL) solution of **2** (347 mg, 1.00 mmol). The mixture was allowed to stand for 2 days at room temperature, concentrated in vacuo, and chromatographed on a silica gel column. Elution with 5:3 cyclohexane–EtOAc gave **15** (585 mg, 63%) as a powder: IR ν_{max} ($CHCl_3$) 1750, 1620 (br) cm^{-1}; ^1H NMR ($CDCl_3$): δ 2.01 (3 H, s), 2.02 (3 H, s), 2.04 (3 H, s), 2.13 (3 H, s), 3.65–3.84 (7 H, m), 4.15 (1 H, dd, *J* 1.0, 11.0 Hz), 4.29 (1 H, dd, *J* 4.0, 11.0 Hz), 4.46–4.94 (9 H, m), 5.08 (1 H, t, *J* 9.2–9.9 Hz), 5.11 (1 H, bs), 5.37 (1 H, t, *J* 9.2–9.9 Hz), 5.95 (1 H, t, *J* 9.2 Hz), 6.75 (1 H, d, *J* 2.6 Hz), 7.11–7.14 (2 H, m), 7.26–7.37 (19 H, m). Anal. Calcd for $C_{49}H_{56}N_2O_{14}S$ (929.1): C, 63.35; H, 6.08; N, 3.02; S, 3.45. Found: C, 63.12; H, 6.10; N, 2.90; S, 3.29.

N-*(β*-D-*Glucopyranosyl)*-N′-*(2,3,4,6-tetra*-O-*benzyl-α*-D-*glucopyranosyl)thiourea* (**16**).—To a solution of **15** (430 mg) in 99.5% EtOH (27 mL) was added KOH (27 mg). After stirring for 10 min at 24 °C, the reaction mixture was concentrated in vacuo and chromatographed on a silica gel column. Elution with 19:1 EtOAc–MeOH gave **16** (288 mg, 82%) as a powder: IR ν_{max} (Nujol) 3300 cm^{-1}; ^1H NMR ($CDCl_3$): δ 3.2–3.9 (12 H, m), 4.25–4.90 (9 H, m), 5.58 (1 H, bs), 7.06 (2 H, bs), 7.2–7.3 (19 H, m), 7.68 (1 H, bs, NH). FABMS (positive): *m/z* 761 $[M + H]^+$. HRFABMS; *m/z* Calcd: 761.3108. Found: 761.3095. Anal. Calcd for $C_{41}H_{48}N_2O_{10}S$ (760.9): C, 64.72; H, 6.36; N, 3.68; S, 4.21. Found: C, 64.56; H, 6.16; N, 3.31; S, 4.20.

*[3a*R*-(3aα,5β,6α,7β,7aβ)]-5-(Hydroxymethyl)-3a,6,7,7a-tetrahydro-2-[(2,3,4,6-tetra*-O-*benzyl-α*-D-*glucopyranosyl)amino]-5*H*-*pyrano[2,3-*d*]oxazole-6,7-diol* (**17**).—To a solution of **16** (288 mg, 0.378 mmol) in MeCN (20 mL) was added 2-chloro-3-ethylbenzoxazolium tetrafluoroborate (288 mg, 1.07 mmol) at 0–5 °C with stirring under

nitrogen. The mixture was stirred for 2 h, and Et_3N (300 mg) was added. After stirring for 30 min at 0–5 °C, the reaction mixture was concentrated in vacuo and chromatographed on a silica gel column. Elution with 19:1 EtOAc–MeOH gave **17** (183 mg, 67%) as a gum: IR ν_{max} (Nujol) 3325, 1640 cm^{-1}; ^1H NMR (400 MHz, Me_2NCHO-d_7) (numbering is according to that of glucose); aglycon part: δ 3.06 (1 H, dd, *J* 7.5, 8.5 Hz, H-4), 3.14, (1 H, dm, *J* 8.5 Hz and multiplet, H-5), 3.16 (1 H, dd, *J* 9.0, 10.5 Hz, H-2), 3.34 (1 H, m, H-6), 3.49 (1 H, m, H-6), 3.53 (1 H, dd, *J* 7.5, 10.3 Hz, H-3), 4.17 (1 H, bs, OH), 4.37 (1 H, d, *J* 9.0 Hz, H-1), 4.78 (1 H, bs, OH), 5.08 (1 H, bs, OH); 1-amino-tetra-*O*-benzyl-glucose part: 3.20 (1 H, t, *J* 9.5 Hz, H-4), 3.35 (1 H, m, H-6), 3.38 (1 H, dd, *J* 5.0, 7.5 Hz, H-2), 3.41 (1 H, m, H-6), 3.56 (1 H, m, H-5), 3.82 (1 H, dd, *J* 7.5, 9.5 Hz, H-3), 4.20 (2 H, s, OCH_2Ph-6), 4.30, 4.50 (2 H, AB-q, OCH_2Ph-4), 4.33, 4.39 (2 H, AB-q, OCH_2Ph-2), 4.45, 4.59 (2 H, AB-q, OCH_2Ph-3), 5.32 (1 H, d, *J* 5.0, H-1). FABMS (positive): *m/z* 727 $[M + H]^+$. HRFABMS; *m/z* Calcd: 727.3231. Found: 727.3187.

Hydrogenolysis of **17**.—Treatment of **17** as described in the formation of **6** from **5** gave a mixture of unidentified complex compounds.

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