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# Synthesis of allosamidin analogues

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**Abstract**

Two new isomers of allosamidin and allosamizoline, and two dimeric allosamidin analogues have been synthesized. The new compounds have been tested for inhibition of insect chitinase and for toxicity to insect larvae. © 1996 Elsevier Science Ltd.

*Keywords:* Allosamidin analogues, synthesis; Chitinase inhibition; Insecticides

## 1. Introduction

The allosamidins [1–3] are a family of naturally occurring pseudo-trisaccharides that inhibit chitinases from a variety of sources [4–7] and thus have potential utility as insecticides and fungicides. Since their discovery ten years ago [1,8], several research groups have undertaken the total synthesis of the parent compound **1** [9–12] and of a series of natural and non-natural analogues [10,12–17]. In addition, a number of independent approaches to the aglycon, allosamizoline (**2**) have been devised [18–22].

In a recent publication [12], we reported the synthesis of allosamidin (**1**) by a convergent route involving glycosyl acceptor **3** and donor **4**, which were derived from methyl α-D-glucopyranoside and 2-amino-2-deoxy-D-glucose, respectively. This route afforded, in addition to **1**, the linkage isomer **5**, due to the use of a diol as glycosyl acceptor. In this paper we describe the preparation of further allosamidin analogues, namely the dimer **6**, previously obtained by mild acid hydrolysis of allosamidin [23], its linkage isomer **7**, and the trimer **8**, as well as the monomer **9**, a regioisomer of allosamizoline (**2**). Ready access to these compounds was provided by the use of intermediates and byproducts of the original synthesis, in particular the monosaccharide trichloroacetimidate **10** and the carbocyclic carbamate **11**. The latter was formed in

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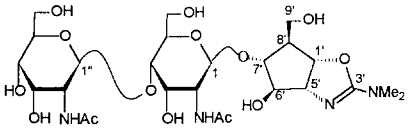
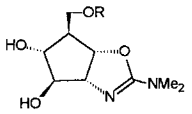
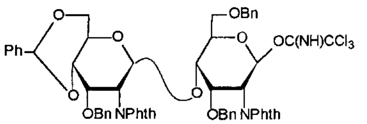
appreciable quantities as well as the required isomer **12** in the oxyamination of alkene **13**, the key step in the preparation of diol **3**.



**1**



**2** R = H
**3** R = Bn



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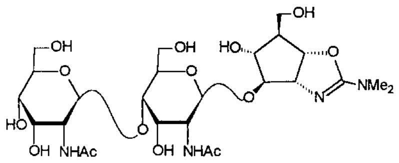
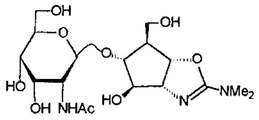
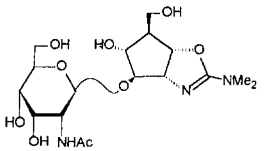


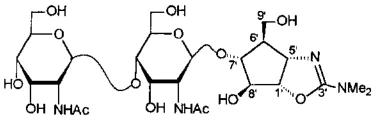
**5**



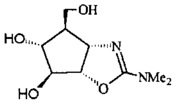
**6**



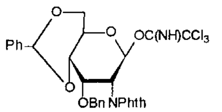
**7**



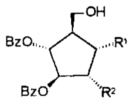
**8**



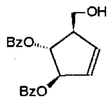
**9**



**10**



**11** $R^1 = NHCO_2Et$, $R^2 = OH$
**12** $R^1 = OH$, $R^2 = NHCO_2Et$

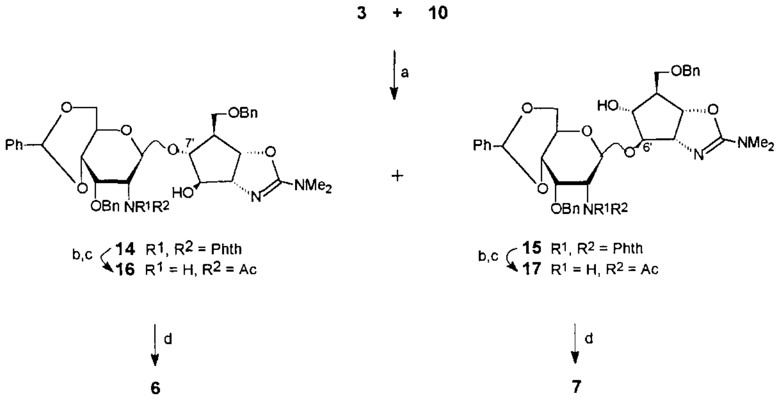


**13**

We were interested in examining the biological activity of these new compounds to obtain information on the structure–activity relationships in allosamidins. This, in combination with insights gained from the X-ray structure analysis of allosamidin bound to the plant chitinase hevamine [24], will allow us to begin to identify the structural elements necessary for chitinase inhibition and provide a basis for the design of simpler and possibly more potent inhibitors.

## 2. Results and discussion

Condensation of diol **3** with slightly more than one equivalent the monosaccharide trichloroacetimidate **10** gave the $\beta$-linked glycosides **14** and **15** as an inseparable



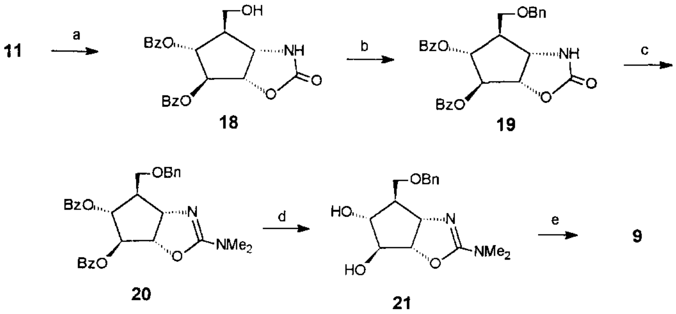
Scheme 1. (a) TMSOTf, $CH_2Cl_2$ [67% combined]. (b) $MeNH_2$, EtOH, $H_2O$. (c) $Ac_2O$, MeOH [**16**: 40% from **3**; **17**: 7.5% from **3**]. (d) $H_2$, Pd/C [80–85%].

mixture in the ratio 5:1 (by NMR spectroscopy) in 67% combined yield (Scheme 1). Using the procedures developed in our original work [12], the mixed isomers were *N*-dephthaloylated with methylamine in aqueous ethanol, and the amines were *N*-acetylated with acetic anhydride in methanol, at which stage chromatographic separation was possible. Hydrogenolysis of the *N*-acetates **16** and **17**, separately, afforded the pseudo-disaccharide allosamidin analogues **6** and **7**, respectively.

Table 1
$^{13}C$ NMR chemical shifts ($\delta$, ppm) of C-7 and C-6 or C-8 for cyclitols **2**, **3**, **9**, and **21** and for the cyclitol moieties of glycosides **1**, **5–7**, **8**, **16**, **17**, and **26**

| | Compound | $\delta$ C-7 | $\delta$ C-6 | $\delta$ C-8 |
|---|---|---|---|---|
| Compounds with free hydroxyl groups at C-7 and at C-6 or C-8 | **2** [a] | 77.8 | 84.6 | |
| | **3** [b] | 74.1 | 84.7 | |
| | **9** [a] | 76.7 | | 83.3 |
| | **21** [b] | 78.0 | | 83.1 |
| Compounds glycosylated at the 7-position of the cyclitol | **1** [a] | 85.9 | 81.3 | |
| | **6** [a] | 87.4 | 83.2 | |
| | **8** [a] | 86.8 | | 82.3 |
| | **16** [b] | 85.3 | 81.9 | |
| | **26** [b] | 85.4 | | 82.0 |
| Compounds glycosylated at the 6-position of the cyclitol | **5** [a] | 74.8 | 88.2 | |
| | **7** [a] | 76.4 | 90.2 | |
| | **17** [b] | 72.9 | 95.4 | |

[a] Spectrum recorded in $D_2O$.
[b] Spectrum recorded in $CDCl_3$.



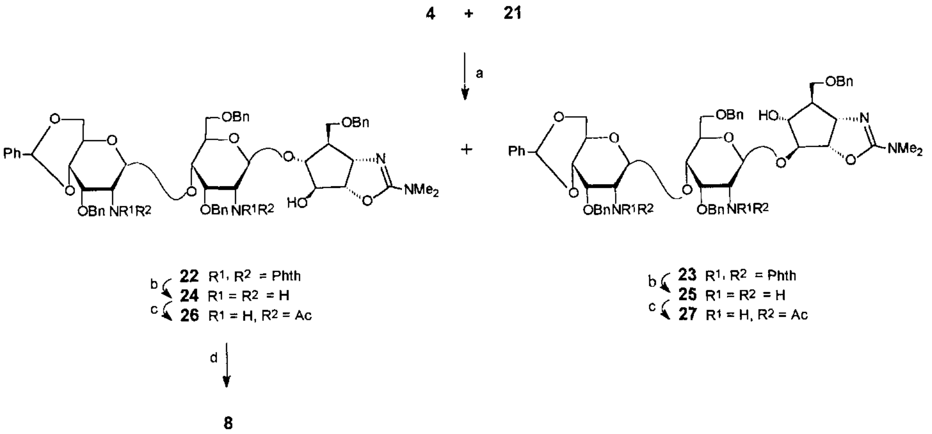
Scheme 2. (a) $(Bu_3Sn)_2O$ [68%]. (b) $BnOC(NH)CCl_3$, TfOH [74%]. (c) $Me_3^+ OBF_4^-$, $CH_2Cl_2$, then $MeNH_2$ [72%]. (d) $Et_3N$, MeOH, $H_2O$ [85%]. (e) $H_2$, Pd/C [90%].

The structures assigned to compounds **6**, **7**, **16**, and **17** were consistent with their NMR spectra, which closely matched those of the aglycon and non-reducing-end sugar moieties of the corresponding trimers **1** and **5** and their precursors [12]. Furthermore, the $^1H$ NMR spectral data for **6** agreed with those published [23]. Of particular interest was the downfield shift in the $^{13}C$ NMR spectra of the signals for C-7 (compounds **6** and **16**) and C-6 (compounds **7** and **17**) in the cyclitol moiety on glycosylation (see Table 1).

Sharpless oxyamination [25] of cyclopentene derivative **13**, available in seven steps from methyl $\alpha$-D-glucopyranoside [26], had furnished the required hydroxy carbamate **12** and its positional isomer **11** in the ratio 2:1 [12]. Compound **11** has now been converted to diol **21** in four steps (Scheme 2): (i) bis(tributyltin) oxide-promoted cyclization to the cyclic carbamate **18**; (ii) *O*-benzylation under acidic conditions [27] to give **19**; (iii) introduction of the *N*-dimethylamino group following the usual procedure [12,28] leading to **20**; and (iv) debenzoylation. Hydrogenolysis of diol **21** gave the allosamizoline isomer **9**. A sample of allosamizoline (**2**) itself was similarly obtained by debenzylation of diol **3**.

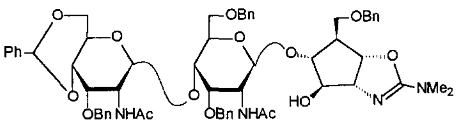
To prepare trimer **8**, diol **21** was glycosylated by use of disaccharide glycosyl trichloroacetimidate **4** (Scheme 3). The pseudo-trisaccharide diphthalimides **22** and **23** were obtained in the ratio 2.5:1 (by NMR spectroscopy) as an inseparable mixture. Consecutive treatment with methylamine in aqueous ethanol and methanolic acetic anhydride gave the mixed *N*,*N*′-diacetates **26** and **27** via the mixed diamines **24** and **25**. Neither of the mixtures could be separated by chromatography, but a sizeable proportion of pure **26** was obtained by fractional crystallization. Hydrogenolysis of **26** furnished **8** in high yield. Compounds **26** and **8** were identified as the (1 → 7′)-linked isomers on the basis of their $^{13}C$ NMR spectra, in particular the downfield shift of the C-7′ resonances relative to their position in diol **21** and triol **9** (see Table 1).

Insect chitinase inhibition studies, using *inter alia* the enzyme from *Chironomus tentans*, were carried out on compounds **1**, **2**, and **5**–**9**. It was shown that the activity of dimer **6** was similar to that of the parent compound **1**, whereas the monomer **2** was almost 1000 times less potent. The change in the linkage between saccharide and



Scheme 3. (a) TMSOTf, $CH_2Cl_2$ [59% combined]. (b) $MeNH_2$, EtOH, $H_2O$. (c) $Ac_2O$, MeOH [65% combined, over 2 steps]. (d) $H_2$, Pd/C [89%].

cyclitol moiety (compounds **5** and **7**) reduced the activity by a factor of 50, and the regioisomers **8** and **9** were less active than **1** and **2**, respectively, by a factor of 100.



**28**

Products **1**, **6**, and **8** and the less polar synthetic intermediates **16** and **28** [12] were assayed in vivo for toxicity to larvae of *Tineola bisselliella* (webbing cloth moth, a major pest in woollen materials) and *Epiphyas postvittana* (light brown apple moth, a major fruit crop pest) feeding on woolen cloth and alder leaves, repectively, treated with one of these compounds [1]. Allosamidin (**1**) and its dimeric analogue **6** showed very meaningful activity, blocking the ability of larvae to shed their cuticles during moulting and resulting in a high level of mortality.

## 3. Experimental

*General methods.*—NMR spectra were recorded in $CDCl_3$ (internal standard $Me_4Si$) with a Bruker AC300E spectrometer, unless otherwise stated. Spectra in $D_2O$ were

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[1] Insect assays were carried out by Dr. P. Gerard, AgResearch Limited, Ruakura, New Zealand and Dr. A. Tomkins, HortResearch Limited, Ruakura, New Zealand; details will be published elsewhere.

referenced to sodium 3-(trimethylsilyl)propanesulfonate using acetone as secondary standard [$\delta$ 2.217 ($CH_3$CO) and $\delta$ 33.174 ($C$H$_3$CO) for $^1$H and $^{13}$C NMR spectra, respectively]; assignments were made with the assistance of DEPT and COSY experiments. Optical rotations were, unless otherwise noted, determined for dichloromethane solutions (0.2–1.6 g/100 mL) with a Perkin–Elmer 241 automatic polarimeter. Melting points were measured on a Reichert Jung Thermovar hot stage apparatus and are uncorrected. For flash chromatography, Silica Gel S (Riedel–de Haën, 0.032–0.063 mm) was used, and TLC was performed on precoated aluminum sheets (E. Merck, Silica Gel-60 $F_{254}$). Accurate mass determinations were carried out at the Mass Spectroscopy Unit of the Horticulture and Food Research Institute of New Zealand Ltd at Palmerston North, New Zealand on a VG-70-250S double-focusing magnetic sector mass spectrometer (VG Analytical) equipped with either a standard VG 70S EI/CI ion source or with a standard LSIMS ion source, associated with a cesium ion gun.

*Preparation of (1S,5R)-3-dimethylamino-6-*exo*-7-*endo*-dihydroxy-8-*exo*-hydroxymethyl-2-oxa-4-azabicyclo[3.3.0]oct-3-ene (allosamizoline* **2***).*—The benzyl ether **3** [12] (30 mg, 0.098 mmol) in 10:1 MeOH–HOAc (1.0 mL) was hydrogenated over palladium-on-charcoal (10%, 20 mg) at 55 psi for 48 h. After addition of more catalyst (15 mg), the hydrogenolysis was continued for another 48 h. Removal of the solids by filtration through glass fibre and of the solvents by evaporation gave **2** (18.5 mg, 87%): $[\alpha]_D$ +5° ($c$ 2.0, $H_2O$), lit. +4.3° [12].

*(1S,5R)-8-*exo*-Benzyloxymethyl-3-dimethylamino-6-*exo*-hydroxy-2-oxa-4-azabicyclo[3.3.0]oct-3-en-7-*endo*-yl 3-O-benzyl-4,6-O-benzylidene-2-deoxy-2-phthalimido-β-D-allopyranoside* **(14)** *and (1S,5S)-8-*exo*-benzyloxymethyl-3-dimethylamino-7-*endo*-hydroxy-2-oxa-4-azabicyclo[3.3.0]oct-3-en-6-*exo*-yl 3-O-benzyl-4,6-O-benzylidene-2-deoxy-2-phthalimido-β-D-allopyranoside* **(15)**.—To a solution of diol **3** [12] (266 mg, 0.87 mmol) in dry $CH_2Cl_2$ (5 mL) at 0 °C under Ar, powdered 4 Å molecular sieves (0.25 g) were added, followed at 5 min intervals by a solution of trichloroacetimidate **10** [12] (580 mg, 0.92 mmol) in $CH_2Cl_2$ (5 mL) and TMSOTf (0.19 mL, 0.87 mmol). After 20 min at 0 °C and 20 min at ambient temperature, $Et_3N$ (0.25 mL) was added. After a further 10 min, the mixture was filtered through Celite, and the filtrate was concentrated. Purification of the residue by chromatography (EtOAc) gave a 5:1 mixture (by NMR spectroscopy) of **14** and **15** (453 mg, 67%): Selected $^1$H NMR data (**14**): $\delta$ 2.07 (m, 1 H, H-8′), 2.68 (s, 6 H, $NMe_2$), 3.22 (dd, 1 H, $J_{8',9'a}$ 5.3 Hz, $J_{gem}$ 9.5 Hz, H-9′a), 3.36 (dd, 1 H, $J_{8',9'b}$ 5.3 Hz, H-9′b), 4.65 (dd, 1 H, $J_{1',5'}$ 8.9 Hz, $J_{1',8'}$ 5.5 Hz, H-1′), 5.44 (s, 1 H, C$H$Ph), 6.04 (d, 1 H, $J_{1,2}$ 8.7 Hz, H-1); selected $^1$H NMR data (**15**): $\delta$ 2.15 (m, 1 H, H-8′), 5.00 (dd, 1 H, $J_{1',5'}$ 7.9 Hz, $J_{1',8'}$ 3.0 Hz, H-1′), 5.98 (d, 1 H, $J_{1,2}$ 8.7 Hz); $^{13}$C NMR data (**14**): $\delta$ 38.0 ($NMe_2$), 50.2 (C-8′), 56.3 (C-2), 63.8 (C-5′), 67.9 (C-9′), 68.9 (C-6), 72.6 (C-5), 73.7 ($C$H$_2$Ph), 74.3 (C-3), 74.1 ($C$H$_2$Ph), 79.7 (C-4), 82.4 (C-1′), 83.7 (C-6′), 84.9 (C-7′), 97.5 (C-1), 102.1 ($C$HPh), 122.9–138.5 (Ph), 161.7 (C-3′), 167.5, 167.7 (C=O, phthalimide). Mass spectrum: Calcd for $C_{44}H_{46}N_3O_{11}$ (M + 1): $m/z$ 776.3183; found: $m/z$ 776.3179.

*(1S,5R)-8-*exo*-Benzyloxymethyl-3-dimethylamino-6-*exo*-hydroxy-2-oxa-4-azabicyclo[3.3.0]oct-3-en-7-*endo*-yl 2-acetamido-3-O-benzyl-4,6-O-benzylidene-2-deoxy-β-D-allopyranoside* **(16)** *and (1S,5S)-8-*exo*-benzyloxymethyl-3-dimethylamino-7-*endo*-hydroxy-2-oxa-4-azabicyclo[3.3.0]oct-3-en-6-*exo*-yl 2-acetamido-3-O-benzyl-4,6-*

*O-benzylidene-2-deoxy-β-*D*-allopyranoside* (**17**).—A solution of the mixed isomers **14** and **15** (396 mg, 0.51 mmol) in EtOH (8 mL) and aq $MeNH_2$ (40%, 5 mL) was kept at ambient temperature for 48 h. The solvent was removed under reduced pressure. The *N,N′*-dimethylphthalic diamide generated in the reaction was removed by chromatography (10:1 EtOAc–MeOH). The mixed diamines were treated with $Ac_2O$ (0.5 mL) in MeOH (5 mL) for 16 h at −10 °C. After methanolysis of the unchanged $Ac_2O$ by slow addition of $Et_3N$ (0.7 mL), partitioning between $CH_2Cl_2$ and water, drying ($MgSO_4$) and concentration of the organic phase, a syrup was obtained which was fractionated by chromatography (10:1 → 5:1 EtOAc–MeOH) to give the minor isomer **17** (37 mg, 11%): $[\alpha]_D$ −30°; $^1$H NMR data: δ 1.97 (s, 3 H, Ac), 2.12 (m, 1 H, H-8′), 2.89 (s, 6 H, $NMe_2$) 3.63 (dd, 1 H, $J_{8',9'a}$ 5.1 Hz, $J_{gem}$ 9.7 Hz, H-9′a), 3.69–3.81 (m, 5 H, H-4, H-6a, H-6′, H7′, H-9′b), 4.02–4.08 (m, 2 H, H-2, H-5′), 4.16 (dt, 1 H, $J_{5,6a}$ 9.9 Hz, $J_{5,6b}$ 5.1 Hz, H-5), 4.25 (t, 1 H, $J_{2,3} = J_{3,4}$ ca. 2 Hz, H-3), 4.33 (dd, 1 H, $J_{gem}$ 10.2 Hz, H-6b), 4.54 and 4.94 (ABq, 2 H, C*H*$_2$Ph), 4.72 (t, 1 H, $J_{1',5'} = J_{1,8'} = 7.0$ Hz, H-1′), 4.94 (d, 1 H, $J_{1,2}$ 7.7 Hz, H-1), 5.52 (s, 1 H, C*H*Ph), 5.65 (d, 1 H, *J* 10.8 Hz, NH), 7.29–7.50 (m, 15 H, Ph); $^{13}$C NMR data: δ 22.9 (Ac), 37.6 ($NMe_2$), 49.9 (C-8′), 53.1 (C-2), 64.2 (C-5′), 67.9 (C-9′), 68.9 (C-6), 71.5 (C-5), 72.9 (C-7′), 73.2, 74.7 (2 × *C*H$_2$Ph), 75.3 (C-3), 79.8 (C-4), 81.3 (C-1′), 95.4 (C-6′), 101.1 (C-1), 102.0 (Ph*C*H), 126.2–138.5 (Ph), 161.7 (C-3′), 170.2 (C=O, acetamide). Mass spectrum: Calcd for $C_{38}H_{46}N_3O_9$ (M + 1): *m/z* 688.3234; found: *m/z* 688.3247.

Compound **17** was followed by the major isomer **16** (209 mg, 60%): mp 203–205 °C; $[\alpha]_D$ −38°; $^1$H NMR data: δ 1.97 (s, 3 H, Ac), 2.27 (m, 1 H, H-8′), 2.92 (s, 6 H, $NMe_2$) 3.44 (dd, 1 H, $J_{8',9'a}$ 4.3 Hz, $J_{gem}$ 9.4 Hz, H-9′a), 3.53 (dd, 1 H, $J_{8',9'b}$ 4.1 Hz, H-9′b), 3.67–3.81 (m, 3 H, H-4, H-6a, H-7′), 3.95–4.15 (m, 5 H, H-2, H-3, H-5, H-5′, H-6′), 4.37 and 4.63 (ABq, 2 H, C*H*$_2$Ph), 4.39 (dd, 1 H, $J_{5,6b}$ 5.1 Hz, $J_{gem}$ 10.0 Hz, H-6b), 4.54 and 5.03 (ABq, 2 H, C*H*$_2$Ph), 4.62 (d, 1 H, $J_{1,2}$ 8.1 Hz, H-1), 4.84 (dd, 1 H, $J_{1',5'}$ 8.9 Hz, $J_{1',8'}$ 5.8 Hz, H-1′), 5.52 (s, 1 H, C*H*Ph), 5.65 (d, 1 H, *J* 10.8 Hz, NH), 7.12–7.55 (m 15 H, Ph); $^{13}$C NMR data: δ 22.9 (Ac), 37.8 ($NMe_2$), 49.9 (C-8′), 52.1 (C-2), 63.9 (C-5′), 66.7 (C-9′), 68.9 (C-6), 70.8 (C-5), 73.0, 74.9 (2 × *C*H$_2$Ph), 75.6 (C-3), 79.9 (C-4), 81.9 (C-1′), 81.9 (C-6′), 85.3 (C-7′), 101.1 (C-1), 102.1 (*C*HPh), 126.2–138.2 (Ph), 161.5 (C-3′), 169.2 (C=O, acetamide). Mass spectrum: Calcd for $C_{38}H_{46}N_3O_9$ (M + 1) *m/z* 688.3234; found: *m/z* 688.3232.

*(1S,5R)-3-Dimethylamino-6-*exo*-hydroxy-8-*exo*-hydroxymethyl-2-oxa-4-azabicyclo[3.3.0]oct-3-en-7-*endo*-yl 2-acetamido-2-deoxy-β-*D*-allopyranoside* (**6**).—Compound **16** (51 mg. 0.074 mmol) was hydrogenated as described above for the preparation of **2**, to give product **6** (26 mg, 84%): $[\alpha]_D$ −21° (*c* 0.61, $H_2O$); $^1$H NMR data ($D_2O$): δ 2.05 (s, 3 H, Ac), 2.53 (m, 1 H, H-8′), 3.07 (s, 6 H, $NMe_2$), 3.63 (dd, 1 H, $J_{3,4}$ 2.9 Hz, $J_{4,5}$ 9.8 Hz, H-4), 3.68–3.72 (m, 2 H, H-6a, H-9′a), 3.78–3.92 (m, 4 H, H-2, H-5, H-7′, H-9′b), 3.92 (dd, 1 H, $J_{5,6b}$ 2.0 Hz, $J_{gem}$ 12.0 Hz, H-6b), 4.05 (t, 1 H, $J_{2,3}$ 2.9 Hz, H-3), 4.29 (dd, 1 H, $J_{5',6'}$ 4.5 Hz, $J_{6',7'}$ 5.4 Hz, H-6′), 4.38 (dd, 1 H, $J_{1',5'}$ 8.8 Hz, H-5′), 4.72 (d, 1 H, $J_{1,2}$ ca. 8 Hz, H-1), 5.38 (dd, 1 H, $J_{1',8'}$ 4.9 Hz, H-1′); $^{13}$C NMR data ($D_2O$): δ 24.8 (Ac), 40.3 ($NMe_2$), 54.3 (C-8′), 55.9 (C-2), 62.1 (C-6), 64.2 (C-9′), 67.1 (C-5′), 69.8 (C-4), 72.7 (C-3), 76.7 (C-5), 83.3 (C-6′), 88.0 (C-7′), 89.6 (C-1′), 102.8 (C-1), 163.4 (C-3′), 176.6 (C=O, acetamide). Mass spectrum: Calcd for $C_{17}H_{30}N_3O_9$ (M + 1): *m/z* 420.1982; found: *m/z* 420.1984.

*(1S,5S)-3-Dimethylamino-7-*endo*-hydroxy-8-*exo*-hydroxymethyl-2-oxa-4-azabicyclo[3.3.0]oct-3-en-6-*exo*-yl 2-acetamido-2-deoxy-β-*D*-allopyranoside* (**7**).—Compound **17** (39 mg, 0.057 mmol) was hydrogenated as described above for the preparation of **2** to give product **7** (19.5 mg, 82%): $[\alpha]_D$ −30° (*c* 0.13, $H_2O$); $^1H$ NMR data ($D_2O$): δ 2.05 (s, 3 H, Ac), 2.47 (m, 1 H, H-8′), 3.09 (s, 6 H, $NMe_2$), 3.62–3.95 (m, 6 H, H-4, H-5, H-6a, H-6b, H-9′a, H-9′b), 3.92 (dd, 1 H, $J_{1,2}$ 8.7 Hz, $J_{2,3}$ 2.8 Hz, H-2), 4.00 (dd, $J_{6',7'}$ 6.4 Hz, $J_{7',8'}$ 7.4 Hz, H-7′), 4.10 (t, $J_{3,4}$ 2.8 Hz, H-3), 4.24 (dd, 1 H, $J_{5',6'}$ 3.7 Hz, H-6′), 4.43 (dd, 1 H, $J_{1',5'}$ 8.9 Hz, H-5′), 4.91 (d, 1 H, H-1), 5.37 (dd, 1 H, $J_{1',8'}$ 4.8 Hz, H-1′); $^{13}C$ NMR data ($D_2O$): δ 24.9 (Ac), 40.4 ($NMe_2$), 54.6 (C-8′), 55.2 (C-2), 61.9 (C-6), 63.9 (C-9′), 66.1 (C-5′), 69.4 (C-4), 72.8 (C-3), 76.8 (C-5), 76.9 (C-7′), 90.0 (C-6′), 90.2 (C-1′), 100.7 (C-1), 163.4 (C-3′), 176.4 (C=O, acetamide). Mass spectrum: Calcd for $C_{17}H_{30}N_3O_9$ (M + 1): *m/z* 420.1982; found: *m/z* 420.1983.

*(1S,5S)-7-*endo*-8-*exo*-Dibenzoyloxy-6-*exo*-hydroxymethyl-2-oxa-4-azabicyclo[3.3.0]octan-3-one* (**18**).—A solution of the ethyl carbamate **11** [12] (113 mg, 0.25 mmol) and bis(tributyltin) oxide (0.15 mL, 0.37 mmol) in toluene (10 mL) was heated under reflux with azeotropic removal of ethanol for 10 h. The solvent was evaporated; chromatography of the residue (1:1 EtOAc–$CH_2Cl_2$) afforded the cyclic carbamate **18** (69 mg, 68%) as white crystals: mp 169–171 °C; $[\alpha]_D$ −34.2° (acetone); $^1H$ NMR data [$(CD_3)_2O$]: δ 2.68 (m, 1 H, H-6), 3.76–3.92 (m, 2 H, H-9a, H-9b), 4.19 (t, 1 H, *J* 5.1 Hz, OH), 4.49 (dd, 1 H, $J_{1,5}$ 8.5 Hz, $J_{5,6}$ 4.1 Hz, H-5), 5.21 (dd, 1 H, $J_{1,8}$ 2.5 Hz, H-1), 5.66–5.74 (m, 2 H, H-7, H-8), 7.12 (s, 1 H, NH), 7.48–8.05 (m, 10 H, Ph); $^{13}C$ NMR data [$(CD_3)_2O$]: δ 55.0 (C-6), 57.0 (C-5), 60.7 (C-9), 77.6 (C-7), 82.9 (C-8), 83.1 (C-1), 129.4–134.4 (Ph), 158.4 (C-3), 165.8 and 166.1 (C=O, benzoates). Mass spectrum: Calcd for $C_{21}H_{20}NO_7$ (M + 1): *m/z* 398.1240; found: *m/z* 398.1252.

*(1S,5S)-7-*endo*-8-*exo*-Dibenzoyloxy-6-*exo*-benzyloxymethyl-2-oxa-4-azabicyclo[3.3.0]octan-3-one* (**19**).—Trifluoromethanesulfonic acid (0.05 mL) was added to a stirred solution of the alcohol **18** (0.34 g, 0.85 mmol) and benzyl trichloroacetimidate (0.35 mL, 1.88 mmol) in dry 1,2-dichloroethane (10 mL). After 2 h at ambient temperature, more benzyl trichloroacetimidate was added (0.1 mL). After stirring for a further 2 h, the solution was washed with aq $NaHCO_3$, and the organic phase was dried ($MgSO_4$) and concentrated. Purification of the residue by chromatography gave the crystalline benzyl ether **19** (0.31 g, 74%): mp 80–81 °C; $[\alpha]_D$ −17.7°; $^1H$ NMR data: δ 2.62–2.71 (m, 1 H, H-6), 3.57 (dd, 1 H, $J_{6,9a}$ 5.0 Hz, $J_{gem}$ 9.5 Hz, H-9a), 3.70 (dd, 1 H, $J_{6,9b}$ 9.4 Hz, H-9b), 4.29 (dd, 1 H, $J_{1,5}$ 8.6 Hz, $J_{5,6}$ 5.2 Hz, H-1), 4.53 (ABq, 2 H, $CH_2Ph$), 5.03 (dd, 1 H, $J_{1,8}$ 2.9 Hz, H-1), 5.55 (t, 1 H, $J_{6,7} = J_{7,8} = 6.3$ Hz, H-7), 5.75 (dd, 1 H, H-8), 6.08 (s, 1 H, NH), 7.28–8.01 (m, 15 H, Ph); $^{13}C$ NMR data: δ 51.7 (C-6), 56.5 (C-5), 68.5 (C-9), 73.5 ($CH_2Ph$), 76.0 (C-7), 81.4 (C-8), 82.5 (C-1), 127.8–137.5 (Ph), 158.0 (C-3), 165.0 and 165.6 (C=O, benzoates). Mass spectrum: Calcd for $C_{28}H_{24}NO_7$ (M-1): *m/z* 486.1552; found: *m/z* 486.1543.

*(1S,5S)-7-*endo*-8-*exo*-Dibenzoyloxy-6-*exo*-benzyloxymethyl-3-dimethylamino-2-oxa-4-azabicyclo[3.3.0]oct-3-ene* (**20**).—To a solution of the oxazolidinone **19** (0.60 g, 1.23 mmol) in dry $CH_2Cl_2$ (5 mL), powdered 4 Å molecular sieves (0.5 g) and trimethyloxonium tetrafluoroborate (0.90 g, 6.3 mmol) were added. The mixture was stirred at ambient temperature under Ar for 24 h. A solution of dimethylamine (0.75 g, 16.5 mmol) in $CH_2Cl_2$ (5 mL), previously stirred with powdered 4 Å molecular sieves for 30

min, was added. After stirring for another 24 h, the mixture was diluted with $CH_2Cl_2$ (30 mL) and washed with aq $NaHCO_3$. The aqueous phase was reextracted with $CH_2Cl_2$ (2 × 20 mL), and the combined organic extracts were washed with water, dried ($MgSO_4$), the solvent was removed and the residue was purified by flash chromatography (EtOAc → 10:1 EtOAc–MeOH) to give **20** (0.45 g, 72%): $[\alpha]_D$ −47.3°; $^1H$ NMR data: $\delta$ 2.63 (m, 1 H, H-6), 2.93 (s, 6 H, $NMe_2$), 3.67 (dd, 1 H, $J_{6,9a}$ 4.9 Hz, $J_{gem}$ 9.6 Hz, H-9a), 3.75 (dd, 1 H, $J_{6,9b}$ 5.8 Hz, H-9b), 4.52 (dd, 1 H, $J_{1,5}$ 8.6 Hz, $J_{5,6}$ 4.7 Hz, H-5), 4.55 (s, 2 H, $CH_2Ph$), 4.92 (dd, 1 H, $J_{1,8}$ 3.5 Hz, H-1), 5.68–5.75 (m, 2 H, H-7, H-8), 7.20–7.55 and 7.95–8.00 (2 m, 15 H, Ph); $^{13}C$ NMR data: $\delta$ 37.7 ($NMe_2$), 52.2 (C-6), 68.6 (C-5), 68.2 (C-9), 73.7 ($CH_2Ph$), 76.2 (C-7), 82.5 (C-8), 85.8 (C-1), 127.5–138.2 (Ph), 161.3 (C-3), 165.4 and 165.5 (C=O, benzoates). Mass spectrum: Calcd for $C_{30}H_{30}N_2O_6$ (M): *m/z* 514.2104; found: *m/z* 514.2109.

*(1S,5S)-6-*exo*-Benzyloxymethyl-3-dimethylamino-7-*endo*-8-*exo*-dihydroxy-2-oxa-4-azabicyclo[3.3.0]oct-3-ene* (**21**).—A solution of dibenzoate **20** (290 mg, 0.56 mmol) in 8:2:1 $Et_3N$–MeOH–$H_2O$ (12 mL) was left for 72 h at ambient temperature. Removal of the solvent and purification by chromatography (200:1 MeOH–$Et_3N$) gave **21** (145 mg, 85%): $[\alpha]_D$ −24.1°; $^1H$ NMR data: $\delta$ 2.15 (m, 1 H, H-6), 2.89 (s, 6 H, $NMe_2$), 3.62 (dd, 1 H, $J_{6,9a}$ 6.4 Hz, $J_{gem}$ 9.2 Hz, H-9a), 3.80–3.84 (m, 2 H, H-7, H-9b), 4.02 (dd, 1 H, $J_{1,8}$ 4.2 Hz, $J_{7,8}$ 6.9 Hz, H-8), 4.11 (dd, 1 H, $J_{1,5}$ 8.9 Hz, $J_{5,6}$ 5.8 Hz, H-5), 4.52 (s, 2 H, $CH_2Ph$), 4.62 (dd, 1 H, H-1), 7.15–7.30 (m, 5 H, Ph); $^{13}C$ NMR data: $\delta$ 37.7 ($NMe_2$), 52.5 (C-6), 67.1 (C-5), 70.9 (C-9), 73.5 ($CH_2Ph$), 78.0 (C-7), 83.1 (C-8), 87.5 (C-1), 127.5–138.2 (Ph), 161.3 (C-3). Mass spectrum: Calcd for $C_{16}H_{22}N_2O_4$ (M): *m/z* 306.1580; found: *m/z* 306.1584.

*(1S,5S)-3-Dimethylamino-7-*endo*-8-*exo*-dihydroxy-6-*exo*-hydroxymethyl-4-oxa-2-azabicyclo[3.3.0]oct-3-ene* (**9**).—Diol **21** (30 mg, 0.098 mmol) was hydrogenated as described above for the preparation of **2**, to give **9** (19 mg, 90%): $[\alpha]_D$ +23° (*c* 0.3, $H_2O$); $^1H$ NMR data ($D_2O$): $\delta$ 2.23 (m, 1 H, H-6), 3.10, 3.15 (2 s, 6 H, $NMe_2$), 3.73 (dd, 1 H, $J_{6,9a}$ 6.7 Hz, $J_{gem}$ 10.3 Hz, H-9a), 3.82–3.87 (m, 2 H, H-4, H-9b), 4.28 (dd, 1 H, $J_{1,8}$ 4.8 Hz, $J_{7,8}$ 7.9 Hz, H-8), 4.46 (dd, 1 H, $J_{1,5}$ 9.2 Hz, $J_{5,6}$ 6.3 Hz, H-5), 5.22 (dd, 1 H, H-1); $^{13}C$ NMR data ($D_2O$): $\delta$ 40.1 40.3 ($NMe_2$), 54.9 (C-6), 60.9 (C-5), 62.6 (C-9), 76.7 (C-7), 83.3 (C-8), 92.5 (C-1) 163.5 (C-3). Mass spectrum: Calcd for $C_9H_{16}N_2O_4$ (M): *m/z* 216.1110; found: *m/z* 216.1110.

*(1S,5S)-6-*exo*-Benzyloxymethyl-3-dimethylamino-8-*exo*-hydroxy-2-oxa-4-azabicyclo[3.3.0]oct-3-en-7-*endo*-yl 3,6-di-O-benzyl-4-O-(3-O-benzyl-4,6-O-benzylidene-2-deoxy-2-phthalimido-β-*D*-allopyranosyl)-2-deoxy-2-phthalimido-β-*D*-allopyranoside* (**22**) and *(1S,5S)-6-*exo*-benzyloxymethyl-3-dimethylamino-7-*endo*-hydroxy-2-oxa-4-azabicyclo[3.3.0]oct-3-en-8-*exo*-yl 3,6-di-O-benzyl-4-O-(3-O-benzyl-4,6-O-benzylidene-2-deoxy-2-phthalimido-β-*D*-allopyranosyl)-2-deoxy-2-phthalimido-β-*D*-allopyranoside* (**23**).—To a stirred solution of diol **21** (130 mg, 0.42 mmol) under Ar at 0 °C, powdered 4 Å molecular sieves (0.25 g) were added, followed at 5 min intervals by a solution of trichloroacetimidate **4** (492 mg, 0.45 mmol) in $CH_2Cl_2$ (5 mL) and TMSOTf (81 μL, 0.42 mmol). After 20 min at 0 °C and 20 min at ambient temperature, $Et_3N$ (0.15 mL) was added. After a further 10 min, the mixture was filtered through Celite, and the filtrate was concentrated. Purification of the residue by chromatography (20:1 $CH_2Cl_2$–MeOH) gave a 2.5:1 mixture (by NMR spectroscopy) of **22** and **23** (310 mg,

59%): Selected $^1$H NMR data (**22**): δ 1.93 (m, 1 H, H-6′), 2.86 (s, 6 H, NMe$_2$), 3.06 (dd, 1 H, $J_{6',9'a}$ 5.3 Hz, $J_{gem}$ 9.5 Hz, H-9′a), 3.1–3.6 (m, 3 H, H-6a, H-6b, H-9′b), 4.68 (dd, 1 H, $J_{1',5'}$ 9.3 Hz, $J_{1',6'}$ 5.1 Hz, H-1′), 5.45 (d, 1 H, $J_{1,2}$ 8.7 Hz, H-1), 5.86 (d, 1 H, $J_{1'',2''}$ 8.5 Hz, H-1″), 6.13 (s, 1 H, C*H*Ph); selected $^1$H NMR data (**23**): δ 6.15 (d, 1 H, $J_{1'',2''}$ 8.7 Hz, H-1″); selected $^{13}$C NMR data (**22**): δ 38.0 (NMe$_2$), 50.8 (C-6′), 56.5, 56.8 (C-2, C-2″), 63.8 (C-5), 66.9 (C-9′), 69.1 (C-6″), 70.1 (C-6), 89.4 (C-1′), 97.0 (C-1), 98.7 (C-1″), 102.1 (*C*HPh), 160.8 (C-3′), 167.6, 167.8 (C=O, phthalimides).

*(1S,5S)-6-*exo*-Benzyloxymethyl-3-dimethylamino-8-*exo*-hydroxy-2-oxa-4-azabicyclo[3.3.0]oct-3-en-7-*endo*-yl 2-acetamido-4-*O*-(2-acetamido-3-*O*-benzyl-4,6-*O*-benzylidene-2-deoxy-β-*D*-allopyranosyl)-3,6-di-*O*-benzyl-2-deoxy-β-*D*-allopyranoside* (**26**).—A solution of the mixed isomers **22** and **23** (160 mg, 0.13 mmol) in EtOH (8 mL) and aq MeNH$_2$ (40%, 5 mL) was kept at ambient temperature for 48 h. The solvent was removed under reduced pressure. The *N*,*N*′-dimethylphthalic diamide generated in the reaction was removed by chromatography (10:1 → 5:1 EtOAc–MeOH). The mixed diamines **24** and **25** (105 mg) were treated with Ac$_2$O (0.5 mL) in MeOH (5 mL) for 16 h at −10 °C. Methanolysis of the unchanged Ac$_2$O by slow addition of Et$_3$N (0.7 mL), partitioning between CH$_2$Cl$_2$ and water, drying (MgSO$_4$) and concentration of the organic phase, gave a mixture of **26** and **27** (91 mg, 65%) as a crystalline solid from which pure **26** (40 mg) was obtained by fractional crystallization from MeOH: mp 177–179 °C; $[\alpha]_D$ −40.0°; $^1$H NMR data: δ 1.69 (s, 6 H, 2 Ac), 1.97 (m, 1 H, H-6′), 2.88 (s, 6 H, NMe$_2$), 3.37 (dd, 1 H, $J_{6',9'a}$ 2.8 Hz, $J_{gem}$ 9.5 Hz, H-9′a), 3.57–3.71 (m, 7 H, H-4, H-4″, H-6a, H-6b, H-6″a, H-7′, H-9′b), 3.74 (t, 1 H, $J_{5'',6''a} = J_{gem} = 10.3$ Hz, H-6″b), 3.95 (dt, 1 H, $J_{1,2} = J_{N,H} = 8.5$ Hz, $J_{2,3}$ 2.9 Hz, H-2), 4.04–4.14 (m, 6 H, H-2″, H-3, H-3″, H-5, H-5″, H-8′), 4.26 (dd, 1 H, $J_{1',5'}$ 8.5 Hz, $J_{5',6'}$ 2.9 Hz, H-5′), 4.31 (d, 1 H, C$H_2$Ph), 4.41–4.60 (m, 6 H, H-1, H-1′, H-6″b, 3 × C$H_2$Ph), 4.63 (d, 1 H, C$H_2$Ph), 4.65 (d, 1 H, $J_{1'',2''}$ 8.2 Hz, H-1″), 4.66 (d, 1 H, C$H_2$Ph), 4.98 (d, 1 H, C$H_2$Ph) 5.01 (d, 1 H, C$H_2$Ph), 5.32 (d, *J* 9 Hz, NH), 5.52 (s, 1 H, C*H*Ph), 5.61 (d, 1 H, NH″), 7.15–7.49 (m, 25 H, Ph); $^{13}$C NMR data: δ 22.6 and 22.7 (2 × Ac), 37.6 (NMe$_2$), 51.4 (C-6′), 51.6 (C-2), 52.2 (C-2″), 64.8 (C-5′), 66.2 (C-9′), 69.0 (C-6 and C-6″), 70.7 (C-5), 71.7 (C-5″), 72.4, 73.9, 74.6, 74.8 (4 × *C*H$_2$Ph), 75.3, 76.5 (C-3, C-3″), 80.0 (C-4″), 77.9 (C-4), 82.0 (C-8′), 85.4 (C-7′), 85.8 (C-1′), 100.8 (C-1), 102.0 (*C*HPh), 102.4 (C-1″), 126.0–138.8 (Ph), 161.1 (C-3′), 168.9, 169.0 (C=O, acetamides). Mass spectrum: Calcd for $C_{60}H_{71}N_4O_{14}$ (M + 1): *m/z* 1071.4967; found: *m/z* 1071.4966.

*(1S,5S)-3-Dimethylamino-8-*exo*-hydroxy-6-*exo*-hydroxymethyl-4-oxa-2-azabicyclo[3.3.0]oct-2-en-7-*endo*-yl 2-acetamido-4-*O*-(2-acetamido-2-deoxy-β-*D*-allopyranosyl)-2-deoxy-β-*D*-allopyranoside* (**8**).—Compound **26** (20.0 mg, 0.019 mmol) was hydrogenated as described above for the preparation of **2**, to give **8** (10.5 mg, 89%) as a foam: $[\alpha]_D$ +4.7° (*c* 0.15, H$_2$O); $^1$H NMR data (D$_2$O): δ 2.12 and 2.15 (2 s, 6 H, 2 Ac), 2.38 (m, 1 H, H-6′), 3.14 and 3.17 (2 s, 6 H, NMe$_2$), 3.67–3.98 (m, 12 H, H-2, H-2″, H-4, H-4″, H-5, H-5″, H-6a, H-6b, H-6″a, H-6″b, H-7′, H-9″a), 3.76 (dd, 1 H, $J'_{6',9'b}$ 3.2 Hz, $J_{gem}$ 11.8 Hz, H-9′b), 4.13 and 4.43 (2 t, 2 H, $J_{2,3} = J_{2'',3''} = J_{3,4} = J_{3'',4''} = 2.8$ Hz, H-3 and H-3″), 4.50 (dd, 1 H, $J_{1',8'}$ 4.4 Hz, $J_{7',8'}$ 6.5 Hz, H-8′), 4.54 (dd, 1 H, $J_{1',5'}$ 9.2 Hz, $J_{5',6'}$ 6.0 Hz, H-5′), 4.85 (d, 2 H, $J_{1,2} = J_{1'',2''} = 8.5$ Hz, H-1 and H-1″), 5.29 (dd, $J_{1'5'}$ 9.1 Hz, $J_{1',8'}$ 4.4 Hz, H-1′); $^{13}$C NMR data (D$_2$O): δ 22.7, 23.5 (2 × Ac), 38.2 (NMe$_2$), 54.6 (C-6′), 55.3 and 55.6 (C-2 and C-2″), 60.8 (C-5′), 62.3 (C-9′) 63.7 (C-6

and C-6″), 69.1 (C-4″), 71.8 and 72.8 (C-3 and C-3″), 75.3 and 76.3 (C-5 and C-5″), 79.7 (C-4), 82.3 (C-8′), 86.8 (C-7′), 92.7 (C-1′), 102.7 (C-1), 103.5 (C-1″), 162.9 (C-3′), 176.5 (C=O, acetamides). Mass spectrum: Calcd for $C_{25}H_{43}N_4O_{14}$ (M + 1): *m/z* 623.2776; found: *m/z* 623.2787.

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