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Synthesis of conformationally locked carbocyclic nucleosides built on an oxabicyclo[3.1.0]hexane system

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Abstract—The rigid 6-oxobicyclo[3.1.0]hexane scaffold, characteristic of the natural antibiotic neplanocin C (**3**), was used to build prototypes of conformationally locked deoxynucleosides in the North hemisphere of the pseudorotational cycle. The purine analogues **6** and **7** are conformationally equivalent to carbocyclic nucleosides built with the bicyclo[3.1.0]hexane template. The pyrimidine nucleosides were unstable and underwent a facile intramolecular epoxide ring-opening reaction leading to heterocycle **22**. Only the deoxyguanosine analogue **7** showed antiviral activity against EBV.

1. Introduction and background

As defined by the concept of pseudorotation, nucleosides in solution exist in a dynamic equilibrium characterized by the rapid change of the furanose ring between a North-type geometry and the corresponding antipodal South-type geometry.[1] Normally, a conformationally unrestricted furanose ring can adopt a number of envelope (E) or twist (T) forms, which can be conveniently described by the value of *P* in the pseudorotational cycle. By convention, a phase angle $P=0°$ corresponds to an absolute North conformation possessing a symmetrical twist form $^{3}T_{2}$, whereas its South antipode, $_{3}T^{2}$, is represented by $P=180°$ (Fig. 1). For the typical North conformation, the value of *P* can fluctuate between 342° and 18° ($_{2}E \rightarrow {}^{3}T_{2} \rightarrow {}^{3}E$) and for the typical antipodal South conformation *P* values range between 162° and 198° ($^{2}E \rightarrow {}^{2}T_{3} \rightarrow {}_{3}E$). Despite this dynamism in solution, only one preferred conformer is found in the solid state, and only one conformer is responsible for optimal molecular recognition in drug–enzyme complexes. In order to identify the preferred conformation in drug–enzyme interactions, we and others have proposed the use of cyclopentane nucleosides that are locked into one of the two extreme antipodes of the pseudorotational cycle by virtue of the presence of a fused three-member ring.[2] With this approach, we have shown that the bicyclo[3.1.0]hexane template can adopt a value of *P* that corresponds closely to either a North ($_{2}E$) or a South ($_{3}E$) conformation, as demonstrated in the case of North-methanocarba adenosine (**1**) and South-methanocarba adenosine (**2**).[3] A naturally occurring and conformationally equivalent template found in the antibiotic neplanocin C (**3**)[4] is 6-oxobicyclo[3.1.0]hexane. Neplanocin C is a minor component of the neplanocin family of antibiotics isolated from *Ampullariela regularis* which according to its X-ray structure corresponds to a conformationally locked nucleoside with an $_{2}E$ North envelope conformation ($P=338.03°$ and $\nu_{max}=21.89°$).[4e] An enantioselective synthesis of neplanocin C has been recently achieved employing D-ribonolactone as a chiral starting material.[5] A crucial point in this synthesis was the

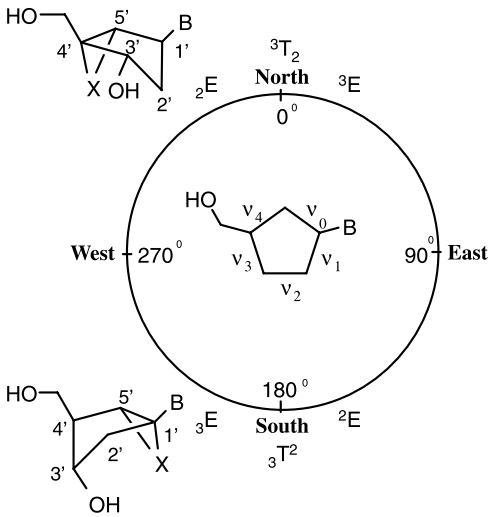

Figure 1. Fixed location of the bicyclo[3.1.0]hexane (X=CH_2) and 6-oxobicyclo[3.1.0]hexane (X=O) templates in the pseudorotational cycle (nucleoside numbering).

Keywords: carbocyclic nucleosides; conformationally locked nucleosides; 6-oxobicyclo[3.1.0]hexane; 2′-deoxyneplanocin C; antiviral activity, EBV.

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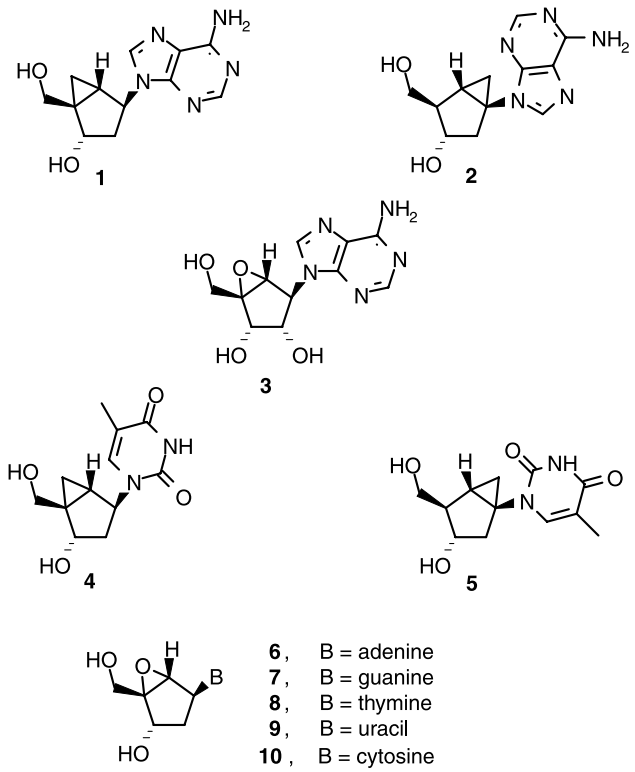
remarkable stability of the epoxide ring during the conversion of the 6-chloropurine ring into adenosine under harsh reaction conditions, such as methanolic ammonia at high temperature.[5] This remarkable stability of the epoxide ring was also observed in simpler, closely related carbanucleosides.[6]



The use of bicyclo[3.1.0]hexane templates has already confirmed the existence of a conformational preference for a number of enzymes such as adenosine deaminase (ADA),[3b,7] HIV reverse transcriptase,[8] DNA (cytosine-C5) methyl transferase,[9] and several subtypes of adenosine receptors.[10] Furthermore, the conformationally locked antipodes of thymidine, North-methanocarba-T (**4**) and South-methanocarba-T (**5**), represent yet another example of a clear conformational discrimination, with the final outcome being that only the North antipode is endowed with activity against herpes infections caused by HSV-1 and HSV-2 viruses.[11]

Since for the most part North analogues have proven to be the more effective as antiviral agents, while the South derivatives have shown weakly inhibitory activities,[2e] we were encouraged to investigate the synthesis and antiviral activity of 2′-deoxycarbocyclic nucleosides **6**–**10**, which contain a similar pseudosugar template as that found in neplanocin C. Unfortunately, due to the extremely facile intramolecular ring opening of the epoxide in compounds with pyrimidine bases (thymine and uracil), only the target purines **6** and **7** were obtained.

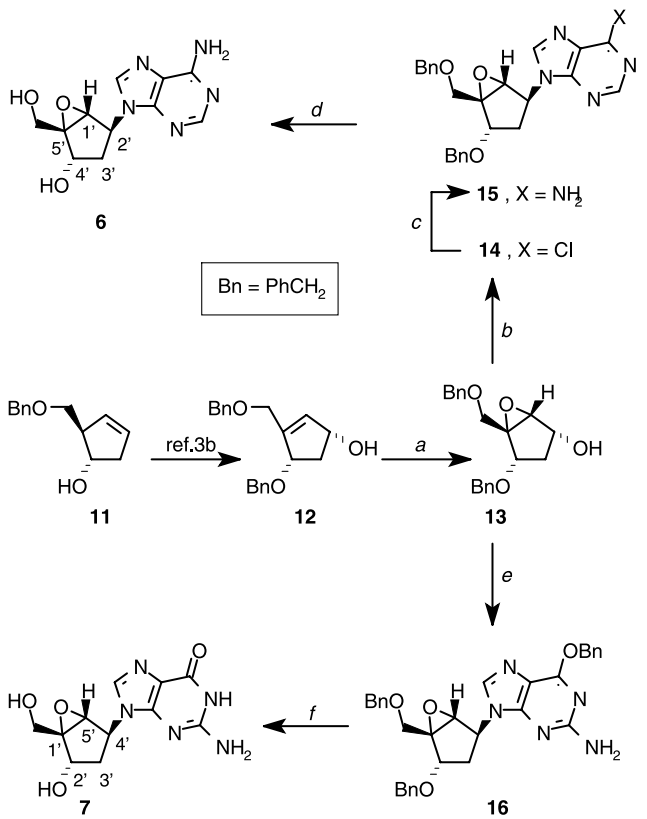
2. Results and discussion

The carbocyclic intermediate (1*R*,4*S*)-4-phenylmethoxy-3-[(phenylmethoxy)methyl]cyclopent-2-en-1-ol (**12**), which was prepared according to our published method,[3b] was employed for the preparation of all purine and pyrimidine 2′-deoxyneplanocin C analogues. This compound, in turn, was synthesized from the readily available chiral template (3*R*,4*S*)-4-phenylmethoxy-3-[(phenylmethoxy)methyl]-cyclopent-1-ene (**11**).[12]

2.1. Synthesis of purine analogues

The corresponding 2′-deoxy adenosine derivative **6** was synthesized following a convergent approach (Scheme 1). Treatment of **12** with *m*-chloroperbenzoic acid at 0°C gave exclusively the α-epoxy derivative **13**, which according to Hembest's rule resulted from the OH-directed attack of the incoming electrophile.[13] Despite the potential involvement of the epoxide ring, coupling 6-chloropurine to epoxy alcohol **13** under Mitsunobu conditions[14] successfully gave the desired product **14** in low but reproducible yields (ca. 36%). The desired N-9 alkylated product was the only compound isolated with no traces of the alternative N-7 alkylated product observed. Ammonolysis of **14** produced the adenosine analogue **15**, which under catalytic-transfer hydrogenation conditions with palladium black and formic acid afforded the expected target **6** in good yield.

The Mitsunobu reaction between **13** and 2-amino-6-benzyloxypurine was more efficient than that with 6-chloropurine, and the guanosine precursor **16** was obtained in 65% yield (Scheme 1). Cleavage of the benzyl groups by



Scheme 1. (a) *m*-CPBA, CH_2Cl_2, 0°C, 61%; (b) 6-chloropurine, PPh_3, DEAD, THF, 0°C→rt 2 h, 36%; (c) NH_3/MeOH, 70°C, 4 h, 47%; (d) Pd black, MeOH/96% HCO_2H (24:1),rt, 16 h, 64%; (e) 2-amino-6-benzyloxypurine, PPh_3, DEAD, THF, 0°C→rt 16 h, 65%; (f) H_2, 42 psi, 5% Pd/C, MeOH, rt, 4 h, 80%.

catalytic hydrogenation afforded the corresponding guanosine derivative **7** in 80% yield.

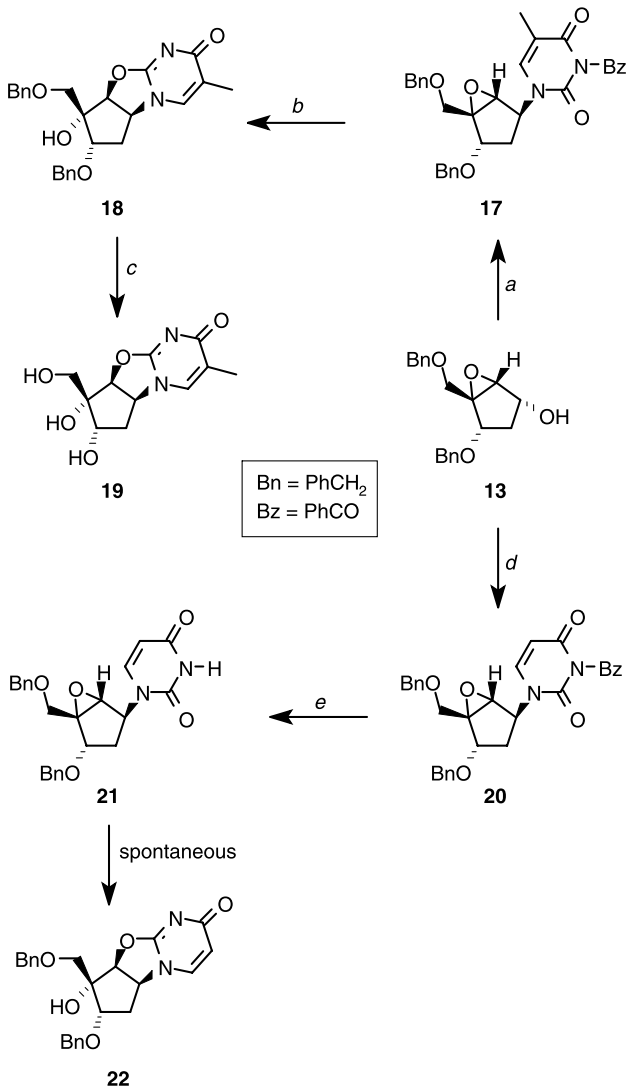
Both the deoxyadenosine (**6**) and deoxyguanosine (**7**) analogues appear to be conformationally equivalent to the bicyclo[3.1.0]hexane nucleosides as judged by the characteristic doublet (J=7.6 (**6**) and 7.7 Hz (**7**)) at δ 4.77–5.07 observed in the ^{1}H NMR spectra of these compounds. This doublet is typical of the pseudoanomeric proton (H-4′ (for compound **6**) and H-2′ (for compound **7**)) in this class of conformationally locked nucleosides. Additionally, these compounds display the distinctive epoxide singlet (δ 3.67 (for compound **6**) and at δ 3.57 (compound **7**)), which is unique to the 6-oxobicyclo[3.1.0]hexane system.

2.2. Synthesis of pyrimidine analogues

A similar attempt to synthesize pyrimidine derivatives **8**–**10** was complicated by this ring's tendency to participate in an intramolecular epoxide ring-opening reaction. Initially N^3-benzoylthymine[15] was coupled to **13** by a Mitsunobu reaction employing a large excess of triphenylphosphine and diethyl azodicarboxilate (Scheme 2). Under these conditions, only the desired N-1 alkylated product **17** was obtained. In contrast to other structurally related carbocyclic rings where competition between O-alkylation and N-alkylation is markedly noticeable,[2a,b] the O-alkylated product was not observed. Unfortunately, following the removal of the N^3-benzoyl group with methanolic ammonia, even at 0°C, an ensuing intramolecular nucleophilic opening of the epoxide ring resulted in the exclusive formation of the corresponding anhydride **18** (80%). Removal of the benzyl protecting groups by catalytic hydrogenation afforded the free anhydride **19** in 82% yield.

Under similar Mitsunobu conditions with N^3-benzoyluracil,[15] again only the N-alkylated product **20** was obtained (Scheme 2). However, the epoxy group in this compound appears to be slightly more stable than in the case of **17** since under the same deprotection conditions (methanolic ammonia at 0°C) some epoxide **21** could be obtained along with compound **22**. Confirmation that this intramolecular reaction is temperature dependent was revealed after the exclusive isolation of compound **21** when removal of the benzoyl group from **20** was performed at −45°C. The ^{1}H NMR singlet at δ 3.43 indicated that the epoxide ring in **21** was intact. Unfortunately, on standing the compound gradually converted to **22**. The instability of these compounds thwarted our hopes of obtaining the corresponding pyrimidine analogues **8**–**10**.

Ab initio energy calculations of optimized conformers for compounds **8** (target thymidine analogue) and the corresponding anhydride **19** with Gaussian 98W employing a HF/6-31Gdp basis set[16] indicated that **19** is 2.88 kcal/mol more stable than **8**. These results are in line with our inability to isolate stable pyrimidine analogues containing a 6-oxobicyclo[3.1.0]hexane pseudosugar. The corresponding pesudorotational parameters for **8** (P=350° ($_2$E–^{3}T$_2$), ν_{max}=17.7°) and **19** (P=24.6° (^{3}E–^{3}T$_4$), ν_{max}=39.8°) revealed substantial conformational differences between epoxides and anhydrides. These results are in agreement with the experimental ^{1}H NMR data where save for the



Scheme 2. (a) N^3-benzoylthymine, PPh_3, DEAD, THF, −45°C, 30 min→rt 16 h, 48%; (b) NH_3/MeOH, 0°C, 1 h, 80%; (c) H_2, 42 psi, 5% Pd/C, MeOH, rt, 4 h, 82%; (d) N^3-benzoyluracil, PPh_3, DEAD, THF, −45°C, 30 min→rt 16 h, 52%; (e) NH_3/MeOH, −45°C, 24 h, 84%.

signals of the nucleobase, the spectra of **17**, **20**, and **21** appeared nearly identical to those of closely related compounds with an intact epoxide ring.[2a,c,5,6] On the other hand, the ^{1}H NMR spectra of anhydrides **18** and **22** differed substantially. For example, the pseudoanomeric proton of anhydride **18** appeared as a triplet (δ=4.79, J=7.3 Hz) while the corresponding pseudoanomeric proton of epoxide **17** was a doublet of doublets (δ=4.96, J=6.1, 2.7 Hz) reflecting that important conformational changes had taken place.

2.3. Antiviral results

In contrast to North-methanocarba-T (**4**), anhydride **19** was devoid of antiviral activity against HSV-1 and HSV-2. The purine analogues **6** and **7** were also inactive against these viruses. Surprisingly, **19** showed good antiviral activity against EBV although with only a 40-fold selectivity index (EC_{50}=1.2 μg/mL, SI>40). The guanosine derivative **7** was much more potent than **19** against EBV and also less toxic (EC_{50}=0.34 μg/mL, SI>150). This value was ca. 6-fold

better than that obtained with the reference standard, acyclovir (EC_{50}=2 μg/mL). Anti-EBV activity in Daudi cells was determined by the viral capsid antigen (VCA) Elisa assay.[17]

3. Conclusion

We have achieved the convergent syntheses of deoxyadenosine (**6**) and deoxyguanosine (**7**) analogues of natural product neplanocin C via Mitsunobu coupling from a common precursor **13**. Conformationally, these compounds are equivalent to known carbocyclic nucleosides built with the bicyclo[3.1.0]hexane template. The epoxide in the 6-oxobicyclo[3.1.0]hexane ring was stable during the synthesis of purine analogues **6** and **7**. However, the corresponding pyrimidine nucleosides are unstable and subject to a facile intramolecular epoxide ring-opening reaction leading to the formation of anhydrides. Only the deoxyguanosine analogue **7** demonstrated important antiviral activity against EBV.

4. Experimental

4.1. General procedures

All moisture sensitive reactions were performed under a dry atmosphere of argon, and all the glassware used in air and/or moisture sensitive reactions was flame-dried. Unless otherwise noted, all chemicals were commercially available and used without further purification. Solvents were distilled before use. THF was distilled from sodium/benzophenone ketyl and CH_2Cl_2 was distilled from P_2O_5 and stored over freshly activated 4 Å molecular sieves. Anhydrous DMF was used as supplied. Column chromatography was performed on silica gel 60 (230–400 mesh) and analytical TLC was performed on commercial 0.2 mm aluminum coated silica gel plates (Kieselgel 60 F_{254}) and visualized by UV light (254 nm) or by immersion in ethanolic 5% H_2SO_4. Melting points are uncorrected. Routine optical rotations, IR and ^{1}H and ^{13}C NMR spectra were recorded using standard methods. Low-resolution mass spectra were obtained at 70 eV (direct inlet). Elemental analyses were conducted by Atlantic Microlab Inc., Norcross, Georgia.

4.1.1. (1*R*,2*R*,4*S*,5*S*)-6-Oxa-4-(phenylmethoxy)-5-[(phenylmethoxy)methyl]-bicyclo[3.1.0]hexan-2-ol (13). A stirred solution of **12** (2.00 g, 6.5 mmol)[3b] in methylene chloride (100 mL) was treated with a solution of 50% *m*-chloroperbenzoic acid (2.20 g) in methylene chloride (25 mL) at 0°C. The mixture was allowed to reach room temperature and stirred for a total of 3 h. The organic phase was washed with a saturated solution of $NaHCO_3$ (3×70 mL), water (3×70 mL) and dried ($MgSO_4$). The solvent was evaporated and the residue was purified by column chromatography (silica gel) eluting with hexanes/EtOAc (4:1) to give 1.30 g (61%) of pure **13** as a white solid: mp 101–103°C; $[\alpha]_D^{23}=-6.05°$ (*c* 1.1, $CHCl_3$); ^{1}H NMR ($CDCl_3$) δ 1.41 (ddd, *J*=12.5, 8.3, 4.1 Hz, 1H), 2.30 (ddd, *J*=12.3, 7.5, 5.0 Hz, 1H), 3.48 (s, 1H), 3.56 (d, *J*=11.6 Hz, 1H), 3.99 (d, *J*=11.6 Hz, 1H), 4.00 (distorted t, *J*=8.0 Hz, 1H), 4.03 (t, *J*=8.0 Hz, 1H), 4.49 (d, *J*=12.1 Hz, 1H), 4.53 (s, 2H), 4.59 (d, *J*=12.1 Hz, 1H), 7.30 (m, 10H); ^{13}C NMR ($CDCl_3$) δ 33.6, 62.0, 65.5, 66.9, 69.6, 71.8, 73.6, 75.0, 127.7, 127.8, 127.8, 128.4, 128.4, 137.7, 138.0. Anal. calcd for $C_{20}H_{22}O_4$: C, 73.60; H, 6.79. Found: C, 73.30; H, 6.72.

4.1.2. (1*S*,2*S*,4*S*,5*R*)-[4-(6-Chloropurin-9-yl)-6-oxa-2-(phenylmethoxy)bicyclo-[3.1.0]hexyl](phenylmethoxy)-methane (14). Diethyl azodicarboxylate (DEAD, 1.04 mL, 5.94 mmol) was added dropwise to a 0°C stirred solution of triphenylphosphine (1.71 g, 6.53 mmol) in anhydrous THF (12 mL). While still at 0°C, the resulting solution was added to a stirred suspension of **13** (0.925 g, 2.83 mmol) and 6-chloropurine (0.922 g, 5.94 mmol) in anhydrous THF (10 mL). The resulting mixture was stirred at 0°C for 30 min and at room temperature for 2 h. The solvent was evaporated and the product was purified by column chromatography (silica gel) eluting with hexanes/EtOAc (4:1) to give 1.10 g (36%) of **14** which was contaminated with traces of reduced DEAD: ^{1}H NMR ($CDCl_3$) δ 2.25 (m, 2H), 3.69 (s, 1H), 3.77 (d, *J*=10.7 Hz, 1H), 4.20 (d, *J*=10.7 Hz, 1H), 4.63 (m, 4H), 4.76 (t, *J*=7.9 Hz, 1H), 5.33 (d, *J*=7.6 Hz, 1H), 7.36 (m, 10H), 8.26 (s, 1H), 8.74 (s, 1H); ^{13}C NMR ($CDCl_3$) δ 34.7, 53.7, 59.3, 66.1, 67.0, 72.5, 73.6, 76.4, 127.6, 127.8, 128.2, 128.3, 131.8, 137.2, 137.5, 143.5, 151.1, 151.8. This compound was used directly in the next reaction.

4.1.3. (1*R*,2*S*,4*S*,5*S*)-9-{6-Oxa-4-(phenylmethoxy)-5-[(phenylmethoxy)methyl]-bicyclo[3.1.0]hex-2-yl}purine-6-ylamine (15). Compound **14** (1.00 g, 2.38 mmol) was treated with a saturated solution of methanolic ammonia (15 mL) in a sealed tube at 70°C for 4 h. The mixture was cooled to −70°C, the tube was opened, and nitrogen was bubbled to eliminate the ammonia. Analysis by TLC revealed that in addition to product some polar compounds were formed. The solvent was evaporated and the residue was purified by column chromatography (silica gel) with $CHCl_3$/MeOH (97:3) as eluant to give 0.50 g (47%) of pure **15** as a white solid: mp 172°C; $[\alpha]_D^{23}=-8.8°$ (*c* 0.4, $CHCl_3$); ^{1}H NMR (CD_3OD) δ 2.09 (m, 1H), 2.30 (dd, *J*=13.4, 7.8 Hz, 1H), 3.67 (s, 1H), 3.76 (d, *J*=11.0 Hz, 1H), 4.04 (d, *J*=11.0 Hz, 1H), 4.55 (m, 4H), 4.81 (t, *J*=7.8 Hz, 1H), 5.17 (d, *J*=7.6 Hz, 1H); 7.29 (m, 10H), 8.07 (s, 1H), 8.13 (s, 1H); ^{13}C NMR (CD_3OD) δ 35.6, 54.8, 61.1, 67.9, 68.2, 73.5, 74.5, 78.5, 120.2, 128.7, 128.8, 128.8, 129.0, 129.3, 129.4, 139.2, 139.5, 140.8, 150.4, 153.7. Anal. calcd for $C_{25}H_{25}N_5O_3$: C, 67.70; H, 5.68; N, 15.79. Found: C, 67.54; H, 5.72; N, 15.84.

4.1.4. (1*S*,2*S*,4*S*,5*R*)-4-(6-Aminopurin-9-yl)-1-(hydroxymethyl)-6-oxabicyclo-[3.1.0]hexane-2-ol (6). A solution of compound **15** (0.128 g, 0.29 mmol) in MeOH (24 mL) and 96% formic acid (1 mL) was stirred at room temperature overnight in the presence of palladium black (0.25 g) under argon. The solvent was evaporated and the residue was purified by column chromatography (silica gel) eluting with $CHCl_3$/MeOH (9:1) to afford 0.036 g (64%) of pure **6** as a white solid: mp 215–218°C (d); $[\alpha]_D^{23}=-33.0°$ (*c* 1.0, MeOH); ^{1}H NMR (DMSO-d_6) δ 1.91 (m, 1H), 2.17 (dd, *J*=14.0, 8.0 Hz, 1H), 3.63 (d, *J*=12.2 Hz, 1H), 3.67 (s, 1H), 4.04 (d, *J*=12.2 Hz, 1H), 4.73 (t, *J*=8.1 Hz, 1H), 5.07 (d, *J*=7.6 Hz, 1H), 7.26 (s, 2H), 8.12 (s, 1H), 8.19 (s, 1H);

^{13}C NMR (DMSO-d_6) δ 36.2, 52.2, 57.3, 59.3, 68.5, 69.0, 118.8, 138.7, 149.0, 152.3, 155.9. Anal. calcd for $C_{11}H_{13}N_5O_3$: C, 50.12; H, 4.98; N, 26.60. Found: C, 50.24; H, 4.91; N 26.38.

4.1.5. (1*R*,2*S*,4*S*,5*S*)-6-Benzyloxy-9-(4-benzyloxy-5-benzyloxymethyl-6-oxabicyclo-[3.1.0]-hex-2-yl)-9*H*-purine (16). Diethyl azodicarboxylate (DEAD, 270 μL, 1.69 mmol) was added dropwise to a 0°C stirred solution of triphenylphosphine (0.444 g, 1.69 mmol) in anhydrous THF (5 mL). While still at 0°C, the resulting solution was added to a stirred suspension of **13** (0.240 g, 0.73 mmol) and 2-amino-6-benzyloxypurine (0.248 g, 1.03 mmol) in anhydrous THF (5 mL). The resulting mixture was vigorously stirred at 0°C for 30 min and at room temperature overnight. The solvent was evaporated and the residue was purified by column chromatography (silica gel) using hexanes/EtOAc (3:2) as eluant to give 0.26 g (65%) of the desired product **16** slightly contaminated with DEAD. An analytical sample was obtained after further purification by column chromatography using CH_2Cl_2/MeOH (99:1) as eluant to afford pure **16** as a colorless oil: $[\alpha]_D^{23}$=+45.1° (*c* 0.91, $CHCl_3$); ^{1}H NMR ($CDCl_3$) δ 2.12 (m, 2H), 3.54 (s, 1H), 3.65 (d, *J*=11.0 Hz, 1H), 4.08 (d, *J*=10.6 Hz, 1H), 4.52 (d, *J*=11.9 Hz, 1H), 4.55 (m, 2H), 4.60 (d, *J*=11.9 Hz, 1H), 4.67 (t, *J*=8.0 Hz, 1H), 5.01 (t, *J*=4.4 Hz, 1H), 5.55 (m, 2H), 7.23–7.36 (m, 13H), 7.49 (d, *J*=7.2 Hz, 2H), 7.61 (s, 1H); ^{13}C NMR ($CDCl_3$) δ 34.6, 52.7, 60.1, 66.6, 66.9, 68.0, 72.9, 73.7, 76.6, 127.8, 127.8, 127.9, 128.2, 128.3, 128.4, 128.5, 136.4, 137.5, 137.9, 153.7, 159.1, 161.1. Anal. calcd for $C_{32}H_{31}O_4N_5{\cdot}0.25H_2O$: C, 69.36; H, 5.73; N, 12.64. Found: C, 69.40; H 5.96; N, 12.21.

4.1.6. (1*R*,2*S*,4*S*,5*S*)-2-Amino-9-[4-hydroxy-5-(hydroxymethyl)-6-oxabicyclo-[3.1.0]hex-2-yl]hydropurin-6-one (7). A solution of **16** (0.069 g, 0.13 mmol) in methanol (5 mL) was in a hydrogenated at 42 psi in the presence of 5% Pd/C (5 mg). After 4 h, the mixture was filtered off and the solvent evaporated. The residue was purified by reverse phase column chromatography using water as eluant to give 0.030 g of pure **7** (80%) as a white solid: mp >270°C; $[\alpha]_D^{23}$=−36.4° (*c* 0.5, MeOH); ^{1}H NMR (DMSO-d_6) δ 1.78 (m, 1H), 2.04 (dd, *J*=14.1, 8.0 Hz, 1H), 3.55 (d, *J*=12.3 Hz, 1H), 3.57 (s, 1H), 3.99 (d, *J*=12.1 Hz, 1H), 4.59 (t, *J*=8.0 Hz, 1H), 4.77 (d, *J*=7.7 Hz, 1H), 6.49 (s, 2H), 7.68 (s, 1H); ^{13}C NMR (DMSO-d_6) δ 35.3, 53.4, 57.0, 58.9, 68.1, 69.3, 110.0, 135.4, 149.8, 154.1, 155.0. Anal. calcd for $C_{11}H_{13}O_4N_5{\cdot}1.25H_2O$: C, 43.78; H, 5.18; N, 23.21. Found: C, 44.01; H, 5.16; N 23.02.

4.1.7. (1*R*,2*S*,4*S*,5*S*)-5-Methyl-1-{6-oxa-4-(phenylmethoxy)-5-[(phenylmethoxy)-methyl]bicyclo[3.1.0]hex-2-yl}-3-(phenylcarbonyl)-1,3-dihydropyrimidine-2,4-dione (17). A stirred solution of triphenylphosphine (0.569 g, 2.17 mmol) in anhydrous THF (8 mL) was treated with diethyl azodicarboxylate (0.34 mL, 2.17 mmol) at 0°C for 20 min. After cooling to −45°C, a solution of N^3-benzoylthymine (0.40 g, 1.74 mmol) and **13** (0.283 g, 0.87 mmol) in THF (8 mL) was added via canula over a period of 10 min. The reaction mixture was stirred at −45°C for 30 min and then at room temperature overnight. After volatiles were removed the residue was purified by column chromatography (silica gel) using hexanes/EtOAc (7:3) as eluant to give the desired *N*-alkylated product **17** (0.180 g, 48%) as a colorless oil: $[\alpha]_D^{23}$=+32.5° (*c* 1.3, $CHCl_3$); ^{1}H NMR (500 MHz, $CDCl_3$) δ 1.69 (d, *J*=0.9 Hz, 3H), 2.09 (m, 2H), 3.47 (s, 1H), 3.59 (d, *J*=10.5 Hz, 1H), 4.14 (d, *J*=10.2 Hz, 1H), 4.49–4.63 (m, 5H), 4.96 (dd, *J*=6.1, 2.7 Hz, 1H), 7.12 (d, *J*=0.9 Hz, 1H), 7.24–7.37 (m, 10H), 7.48 (t, *J*=7.9 Hz, 2H), 7.63 (t, *J*=7.5 Hz, 1H), 7.89 (dd, *J*=8.3, 1.0 Hz, 2H); ^{13}C NMR ($CDCl_3$) δ 12.2, 35.1, 55.6, 60.0, 67.0, 67.3, 72.5, 73.9, 76.2, 111.6, 127.7, 127.9, 127.9, 128.1, 128.5, 128.5, 129.1, 130.4, 131.6, 135.0, 137.1, 137.4, 149.7, 162.5, 168.8. Anal. calcd for $C_{32}H_{30}O_6N_2$: C, 71.36; H, 5.61; N, 5.20. Found C, 70.89; H, 5.74; N, 5.16.

4.1.8. (7*S*,8*S*,5a*S*,8a*S*)-8-Hydroxy-3-methyl-7-(phenylmethoxy)-8-[(phenylmethoxy)methyl]-5,6,7,8,5a,8a-hexahydro-5a*H*,8a*H*-cyclopenta[1,2-*d*]pyrimidino[2,1-*b*]1,3-oxazolidin-2-one (18). Compound **17** (0.15 g, 0.28 mmol) was treated with methanolic ammonia (5 mL, saturated at −78°C) and stirred in a pressure vessel at 0°C for 1 h. The solvent was evaporated and the residue was purified by column chromatography (silica gel) using EtOAc/MeOH (95:5) as eluant to give 0.097 g (80%) of pure **18** as a white solid: mp 166–167°C; $[\alpha]_D^{23}$=+37.0° (*c* 1.8, $CHCl_3$); ^{1}H NMR ($CDCl_3$) δ 1.95 (d, *J*=1.1 Hz, 3H), 2.22 (dd, *J*=13.3, 6.9 Hz, 1H), 2.31 (ddd, *J*=14.1, 10.0, 7.1 Hz, 1H), 3.06 (br s, 1H), 3.72 (m, 2H), 3.93 (dd, *J*=10.0, 6.8 Hz, 1H), 4.55 (m, 2H), 4.58 (m, 2H), 4.79 (t, *J*=7.3 Hz, 1H), 5.02 (d, *J*=7.7 Hz, 1H), 7.00 (d, *J*=1.1 Hz, 1H), 7.21−7.36 (m, 10H); ^{13}C NMR ($CDCl_3$) δ 14.1, 34.9, 59.1, 70.5, 73.2, 74.0, 78.7, 80.5, 85.2, 119.6, 127.8, 127.9, 127.9, 128.3, 128.5, 128.6, 130.3, 137.0, 137.4, 159.8, 172.1. Anal. calcd for $C_{25}H_{26}O_5N_2$: C, 69.11; H, 6.03; N, 6.45. Found: C, 68.98; H, 6.01; N, 6.54.

4.1.9. (7*S*,8*S*,5a*S*,8a*S*)-7,8-Dihydroxy-8-(hydroxymethyl)-3-methyl-5,6,7,8,5a,8a-hexahydro-5a*H*,8a*H*-cyclopenta[1,2-*d*]pyrimidino[2,1-*b*]1,3-oxazolidin-one (19). A solution of **18** (0.083 g; 0.19 mmol) in methanol (5 mL) was hydrogenated at 42 psi in the presence of 5% Pd/C (5 mg) for 4 h. The mixture was filtered and the solvent was evaporated. The residue was purified by column chromatography (silica gel) with CH_2Cl_2/MeOH (85:15) as eluant to give pure **19** (0.040 g, 82%) as a white solid: mp 230°C (d); $[\alpha]_D^{23}$=+82.9° (*c* 0.69, MeOH); ^{1}H NMR (D_2O) δ 1.87 (d, *J*=0.9 Hz, 3H), 2.23 (ddd, *J*=14.1, 11.2, 6.8 Hz, 1H), 2.33 (dd, *J*=14.1, 7.1 Hz, 1H), 3.70 (d, *J*=12.3 Hz, 1H), 3.81 (d, *J*=12.5 Hz, 1H), 4.03 (dd, *J*=11.4, 7.1 Hz, 1H), 5.04 (distorted t, *J*=7.4 Hz, 1H), 5.12 (d, *J*=8.0 Hz, 1H), 7.64 (d, *J*=0.9 Hz, 1H); ^{13}C NMR (CD_3OD) δ 13.9, 37.7, 60.8, 63.7, 72.3, 81.9, 87.6, 119.3, 135.1, 161.8, 175.4. Anal. calcd for $C_{11}H_{14}O_5N_2{\cdot}H_2O$: C, 48.53; H, 5.92; N, 10.29. Found: C, 48.17; H, 5.80; N, 9.99.

4.1.10. (1*R*,2*S*,4*S*,5*S*)-1-{6-Oxa-4-(phenylmethoxy)-5-[(phenylmethoxy)methyl]-bicyclo-[3.1.0]hex-2-yl}-3-(phenylcarbonyl)-1,3-dihydropyrimidine-2,4-dione (20). According to the procedure used for **17**, triphenylphosphine (0.444 g, 1.69 mmol), DEAD (0.27 mL, 1.69 mmol), N^3-benzoyluracil (0.294 g, 1.36 mmol) and **13** (0.221 g, 0.68 mmol), were reacted to give a crude product which was purified by column chromatography (silica gel) using hexanes/EtOAc (6:4) as eluant to give compound **20** still contaminated with reduced DEAD and triphenylphosphine

oxide. The product was repurified by column chromatography (silica gel) using a mixture of CH_2Cl_2/MeOH (99:1) to afford pure **20** (0.186 g, 52%) as a colorless oil: $[\alpha]_D^{23}=+25.3°$ (*c* 0.97, $CHCl_3$); ^1H NMR ($CDCl_3$) δ 2.10 (m, 2H), 3.48 (s, 1H), 3.59 (d, *J*=10.7 Hz, 1H), 4.11 (d, *J*=10.5 Hz, 1H), 4.45–4.60 (m, 5H), 4.95 (s, 1H), 5.66 (d, *J*=8.0 Hz, 1H), 7.24 (d, *J*=8.0 Hz, 1H), 7.29–7.35 (m, 10H), 7.48 (t, *J*=7.7 Hz, 2H), 7.65 (t, *J*=7.5 Hz, 1H), 7.91 (d, *J*=8.2 Hz, 2H); ^{13}C NMR ($CDCl_3$) δ 35.1, 56.1, 59.8, 66.8, 67.5, 72.5, 73.8, 76.3, 102.9, 127.7, 127.8, 128.0, 128.1, 128.5, 128.5, 129.2, 130.5, 131.4, 135.2, 137.5, 137.7, 141.4, 149.6, 161.7, 168.5. Anal. calcd for ($C_{31}H_{28}O_6N_2$): C, 70.98; H, 5.38; N, 5.34. Found: C, 70.57; H, 5.45; N, 5.10.

4.1.11. (1*R*,2*S*,4*S*,5*S*)-1-{6-Oxa-4-(phenylmethoxy)-5-[(phenylmethoxy)methyl]-bicyclo-[3.1.0]hex-2yl}-1,3-dihydropyrimidine-2,4-dione (21) and (7*S*,8*S*,5a*S*,8a*S*)-8-hydroxy-7-(phenylmethoxy)-8-[(phenylmethoxy)-methyl]-5,6,7,8,5a,8a-hexahydro-5a*H*,8a*H*-cyclopenta[1,2-*d*]pyrimidino[2,1-*b*]1,3-oxazolidin-2-one (22). Compound **20** (0.018 g, 0.03 mmol) was treated with methanolic ammonia (1.0 mL, saturated at −78°C) and the reaction mixture was stirred in a sealed tube at −45°C for 24 h. The solvent was evaporated and the residue was purified by preparative TLC (silica gel 60, Merck) using EtOAc/hexanes (4:1) as eluant to give 0.012 g of **21** (84%) and 0.002 g (13%) of **22** as colorless oils.

Compound **21**. ^1H NMR ($CDCl_3$) δ 2.04–2.08 (m, 2H), 3.43 (s, 1H), 3.59 (d, *J*=10.5 Hz, 1H), 4.11 (d, *J*=10.5 Hz, 1H), 4.45 (t, *J*=7.8 Hz, 1H), 4.50 (d, *J*=11.8 Hz, 1H), 4.52 (d, *J*=11.5 Hz, 1H), 4.56 (d, *J*=11.5 Hz, 1H), 4.59 (d, *J*=11.8 Hz, 1H), 4.92 (d, *J*=6.8 Hz, 1H), 5.56 (dd, *J*=8.0, 2.1 Hz, 1H), 7.19 (d, *J*=8.2 Hz, 1H), 7.26–7.35 (m, 10H), 8.26 (br s, 1H); ^{13}C NMR ($CDCl_3$) δ 35.0, 55.8, 59.9, 66.8, 67.5, 72.5, 73.8, 76.3, 102.8, 127.7, 128.0, 128.0, 128.1, 128.5, 128.5, 137.5, 137.8, 141.7, 150.3, 162.4. On standing this compound gradually converts to **22**.

Compound **22**. $[\alpha]_D^{23}=+28.6°$ (*c* 0.84, $CHCl_3$); ^1H NMR ($CDCl_3$) δ 2.32 (m, 2H), 3.64 (d, *J*=9.9 Hz, 1H), 3.73 (d, *J*=10.2 Hz, 1H), 3.92 (t, *J*=8.2 Hz, 1H), 4.55 (m, 4H), 4.98 (m, 1H), 5.10 (d, *J*=8.0 Hz, 1H), 5.99 (d, *J*=7.3 Hz, 1H), 7.27 (m, 10H), 7.68 (d, *J*=7.3 Hz, 1H); ^{13}C NMR ($CDCl_3$) δ 34.9, 61.0, 72.6, 73.7, 74.8, 80.1, 81.8, 88.2, 109.6, 128.7, 128.9, 129.0, 129.1, 129.4, 139.3, 158.2, 162.4. Anal. calcd for ($C_{25}H_{26}O_6N_2O_5\cdot1.5H_2O$): C, 65.06; H, 6.33; N, 6.07. Found: C, 64.93; H, 6.63; N, 6.07.

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